Trochus: Status, Hatchery Practice and Nutrition

Proceedings of a workshop held at Northern Territory University, 6–7 June 1996

Editors: **Chan L. Lee and Peter W. Lynch**

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Overview of Trochus Project

It gives me great pleasure to address the Trochus End-of-project Workshop.

This project, funded by the Department of Employment, Education and Training (DEET) under the Targeted Institutional Links (TIL) Program, was a direct result of the Northern Territory University (NTU) Memorandum of Cooperation (MOC) with universities in the eastern Indonesian region. The project has provided an excellent example of the mutual benefits that can be achieved from the MOC, not only for the universities involved but for the community and the region. In this instance NTU has been working with the University Nusa Cendana (UNDANA) in Kupang.

Two Indonesian staff from UNDANA have been involved in the **Phase 1 Trochus Hatchery and Nutrition Project**, Dr Felix Rebhung, a postdoctoral fellow working on trochus nutrition and Mr Ricky Gimin, undertaking a Master of Science with a scholarship provided by AUSAID, working on trochus reproduction and hatchery studies. As part of the project, Dr Rebhung and Mr Gamin respectively undertook research at NTU for approximately 12 and 24 months.

Milestones of the Project

Program 1	Literature review and dissemination of information.
Result	Successfully completed: a compendium of selected reprints on trochus was published.
Program 2	Maturation, spawning and juvenile production.
Result	Successfully completed: a state-of-the-art hatchery was developed for spawning and producing juvenile trochus.
Program 3	Use of microalgae and seaweeds as food.
Result	Seaweeds in Darwin Harbour were identified and tested.
Program 4	Nutritional requirements of trochus.
Result	Nutritional value and requirements of the trochus were determined.

In addition to these activities, the project also conducted two fellowship training courses for staff from UNDANA and a training workshop for the Aboriginal communities in King Sound, Western Australia.

The success of this project led to the Australian Centre for International Agricultural Research (ACIAR) funding a second phase of the trochus project, 'Reef reseeding research of the topshell *Trochus niloticus* in northern Australia, eastern Indonesia and the Pacific' in July 1995, a year before the completion of the phase 1 project.

Phase 1 of the project had an operating grant of $250 000. A total budget of $1.49 million was committed to Phase 2 with the entire cash component of $680 000 being contributed by ACIAR. Phase 2 commenced in July 1995 and is due for completion in June 1998.

The project leader, Dr Chan Lee, Senior Lecturer in Aquaculture, said that stock enhancement of wild fisheries with hatchery-produced seeds is one of the most exciting and difficult areas of research at present. With the world capture fishery at a standstill, stock enhancement via aquaculture could be a possible solution to overcoming the dwindling seafood supply in the 21st century.

I would like personally to thank you for the invitation to open this workshop; this is certainly an exciting and relevant research project, and I congratulate Dr Chan Lee and all those involved.

R. Holmes
Vice-Chancellor
Northern Territory University

Cooperative Research into the Biology of the Topshell *Trochus niloticus* from Northern Australia and Eastern Indonesia—an End-of-project Review

C. L. Lee* and M. T. Toelihere†

Abstract

Four research programs involving (a) literature review, (b) spawning, hatchery system development and juvenile production, (c) use of microalgae and seaweeds as food and (d) nutritional studies were implemented as part of the cooperative research on trochus, *Trochus niloticus,* involving Australia and eastern Indonesia. During the three-year project, a closed recirculating hatchery system using saline bore water was successfully developed for spawning and producing juvenile trochus at the Northern Territory University (NTU). The trochus life-cycle was successfully closed in the NTU hatchery in early 1996. In the same period, the nutritional value of (a) adult trochus, (b) hatchery-reared trochus up to 18 months old and (c) 17 species of macroalgae and mixed diatoms were studied. As part of the nutritional study, the types and occurrence of different species of macroalgae found in Darwin and Kupang harbours were determined. Preliminary trials indicated that some of the macroalgae may have potential as feed for juvenile trochus. The success of the research work led to the project's receiving additional funding from the Australian Centre for International Agriculture Research to expand the work into a larger regional project involving reef reseeding research with hatchery-produced juveniles in Australia, Indonesia and Vanuatu.

THE research project 'Cooperative research into the biology of the topshell *Trochus niloticus* from northern Australia and eastern Indonesia', commonly referred to as the 'Trochus hatchery and nutrition project' was initiated in July 1993. The three-year research project was funded under Round 1993 of the Targeted Institutional Links (TIL) Program, Department of Employment, Education and Training (DEET). This end-of-project review provides an overview and summary of its progress and achievements from 1 July 1993 to 30 June 1996.

The Trochus Hatchery and Nutrition Project involved two institutions, the Northern Territory University (NTU), Darwin, Northern Territory, Australia the commissioned organisation and the University Nusa Cendana (UNDANA), Kupang, eastern Indonesia, as the collaborating university. Academic staff involved in the project are:

* School of Biological Sciences, Northern Territory University, Darwin 0909, NT, Australia.

† University Nusa Cendana, Kupang, NTT, Indonesia.

NTU

Dr C. L. Lee	Project Coordinator; Research Programs 1 and 2
Associate Professor D. Parry	Chemist, nutrition studies; Research Program 4
Mrs S. Renaud	Chemist, nutrition studies; Research Program 4
Dr J. Luong-Van	Botanist, algae as food; Research Program 3

UNDANA

Professor M. T. Toelihere	Project Coordinator, UNDANA
Dr Felix Rebhung	Biochemist, nutrition studies; Research Program 4
Mr Burhanuddin	Nutritionist; Research Program 4
Mr R. Gimin/ Mrs F. Risamasu	Biologists; Research Programs 2 and 3

Research Scope and Outcomes

The four major research programs listed for implementation and investigation were:

1. Literature review and dissemination of information on trochus.
2. Maturation, spawning and juvenile production.
3. Use of microalgae and seaweeds as food.
4. Nutritional requirements of trochus.

The research programs were under the control of academic staff based in the two universities. The milestones, progress and outcomes for each of the four programs are given in the following sections of the review.

Program 1: Literature review and dissemination of information

Milestones

To finalise a literature review and produce an annotated compendium of selected reprints on *T. niloticus.*

Progress and outcome

The work is completed and an annotated compendium has been produced. The project now operates a 'Trochus library' and is sending on request copies of trochus publications to Australian and overseas scientists. The library has proved particularly useful for staff from the eastern Indonesian institutions.

Future activities

Satisfactory progress resulted in funding being made available by the Australian Centre for International Agriculture Research (ACIAR) to expand the project into a larger Phase 2 research program involving reseeding research of hatchery-produced trochus juveniles in selected sites in northern Australia (King Sound), Indonesia (Ambon and Kupang) and the Republic of Vanuatu (Port Vila). The TIL Program library will continue to be maintained and the resource made available to all interested researchers till the end of the ACIAR-funded Trochus Reseeding Research Project scheduled to finish in June 1998.

Program 2: Maturation, spawning and juvenile production

Milestones

To develop simple methods for spawning and develop a standardised hatchery production system for juvenile trochus.

Progress and outcomes

The program involves six separate but closely interrelated activities:

(a) identify a source of broodstock for spawning,

(b) develop a suitable method for transporting broodstock to NTU,

(c) establish the reproductive cycle of trochus in King Sound Peninsula, WA,

(d) develop a protocol for (i) selecting spawners and (ii) induced spawning,

(e) develop a protocol for larval rearing and juvenile production, and

(f) close the life-cycle of trochus.

Outcomes of these activities are summarised below.

Source of broodstock. King Sound Peninsula, Broome, WA was identified as a good source of broodstock for the project. There were many reasons for choosing this site. King Sound has a viable and successful ongoing small trochus fishery run by the Bardi Aboriginal Association at One Arm Point. The Aboriginal communities involved in the fishery were very concerned about overfishing of their trochus fishery, and were keen to support research to develop mass hatchery production techniques for trochus juveniles followed by reseeding research in the reefs under their control. Consequently, they offered their support to the project by providing all broodstock needed for the hatchery and nutrition work at NTU, Darwin. The Western Australia Fisheries Department and its Broome-based aquaculture development officer, Mr Colin Ostle, were also strongly supportive of the research and largely responsible for coordinating the shipment of trochus broodstock from King Sound to NTU. Consequently, King Sound Peninsula was selected as the site to supply broodstock for the research project.

Method of transporting broodstock. A successful method for shipping trochus broodstock using road and air transport from One Arm Point, King Sound to NTU was successfully developed. This involved packing the shells in 'Nelly' bins or similar-sized containers and keeping them out of water but moist during road transport to Broome. Air transport from Broome to NTU used oxygen-filled clear double-polythene bags. The shells in the bags were maintained dry but under moist conditions during transport. Further details on the method are given in Lee and Ostle (these Proceedings).

Establish the reproductive cycle of trochus in King Sound. The reproductive cycle of trochus in King Sound was established by sampling 25 female and 15 male trochus each month over a 12-month period and examining the gonads of the mature broodstock histologically in the laboratory. Results indicated that the trochus are asynchronous spawners with spawning concentration occurring during July–August and January–February (Gimin and Lee, these Proceedings). This is in agreement with studies conducted by other workers in the Pacific (Hahn 1993; Bour 1988).

Protocol for selecting spawners for induced spawning. A cannula or a similar device is used to remove some eggs directly from the ovary for determining the stage of development of the ova prior to induced spawning. A similar method of gonad biopsy for giant clams using a hypodermic needle was successfully developed to determine their ripeness before spawning trials (Braley 1992). In trochus, however, the gonad is closely attached to the digestive gland and located at the apex of the shell, making it inaccessible to the cannula, needles or other forms of sampling. The development of a 'window' method by Dobson and Lee (1996) enabled the gonad to be viewed under a light microscope, and the sex of the animal determined, but sampling eggs through the window invariably resulted in the death of the females. Consequently, a new method of staging ovarian development using a dissecting microscope provided with a cool light source was successfully developed (Gimin and Lee, these Proceedings). Studies indicated that for successful spawning, two criteria must be satisfied. Ova density must be at least 4/mm^2 and each ovum must reach a size 150–160 µm in diameter before successful induction of spawning can be carried out.

Protocol for induced spawning. During the three-year project, numerous physical and chemical stimuli were tested for their suitability for inducing the trochus to spawn. They were:

—temperature (elevation),

—salinity stress,

—water change,

—desiccation,

—UV-treated water,

—hydrogen peroxide, and

—seratonin.

During the early phase of the research, some encouraging results were obtained in induced spawning experiments (Dobson and Lee 1996). However, at the beginning of the project, the inability to stage the ripeness and determine the size of the ova in the gonad made it impossible to standardise the results of induced spawning. With the development of the window method of sexing the animals, the problem was resolved.

Protocol for larval rearing and juvenile production. The closed recirculating trochus hatchery developed at NTU has been very successful in producing the juveniles needed for this and the ACIAR-funded reseeding project. The system was found to be adequate for the production of juveniles and very efficient in labour and water utilisation. Up to 50 000 three-to-four-week-old juveniles (1–2 mm in basal diameter) can be produced in such a system. Estimated cost of production varies from <0.1–2.6 cents/juvenile.

The production system developed at NTU for producing juveniles was based on a modular system design. Each module was made up of a fibreglass tank measuring 2.0 m wide × 3.5 m long × 0.9 m high and divided into a large (2.0 m wide × 2.5 m long × 0.9 m high) spawning and juvenile growing compartment and a small (2.0 m wide × 1.0 m long × 0.9 m high) biological filter compartment. A full description of the hatchery and its operation is given by Lee (these Proceedings).

Closing the life-cycle of trochus. The life-cycle of trochus was closed in the NTU hatchery in 1996. Fifty of the first batch of F_1 that were spawned in NTU in September 1993 were successfully induced to spawn in March 1996 (Lee, these Proceedings). With the production of F_2 individuals, the trochus was domesticated and its life-cycle closed within the land-based hatchery set up at NTU.

Program 3: Use of microalgae and seaweeds as food for trochus

Milestones

To determine the types, occurrence and suitability of various seaweeds for feeding juveniles and adult trochus.

Progress and outcome

The types and occurrence of seaweeds along the intertidal areas of Darwin Harbour were collected and identified (Wynne and Luong-Van, these Proceedings). The seaweeds are represented by seven orders of Rhodophyta and four orders of Phaeophyta. Similar species were found in Kupang Harbour.

Ten species of Darwin Harbour seaweeds (*Halimeda borneensis*, *Symploca* sp., *Dictyota ciliolata*, *Padina* *australis, Padina boryana, Rosenvingea nhatrangensis, Sargassum* sp., *Acanthophora muscoides, Tolypiocladia glomerulata* and *Hypnea* sp.) were used in a series of experiments to determine the food preference of adult trochus and to compare the growth rates of juvenile trochus fed on the seaweeds and microalgae. It was found that adult trochus preferred soft filamentous seaweeds (e.g. *Hypnea* sp. *Symploca* sp., and *Tolypiocladia glomerulata*) and the corticated *Acanthophora muscoides* were most eaten. Microalgae were found to support the highest growth rates compared to the seaweeds. Juvenile trochus grew at a rate of 0.055 mm diameter per day, while those fed seaweeds grew on average 0.022–0.028 mm diameter per day (Lambrinidis et al., these Proceedings).

Milestones

To develop methods for mass culture of suitable seaweeds and diatoms identified to contain high PUFA (polyunsaturated fatty acids) contents as ingredients for producing artificial diet plates.

Progress and outcomes

Preliminary work was carried out to grow and maintain *Caulerpa racemosa, C. serrulata, C. peltata, C. prolifera, Gracilaria crassa, Sargassum* sp. and *Jania* sp. in the NTU hatchery. These seaweed will grow in diluted bore water (35 ppt salinity), as long as the temperature and light intensity are maintained in the range 25–32°C and 400–700 µmoles photons m^2/s, respectively. Under these conditions, *Caulerpa* spp. grew at a rate of 2–3 cm/day, while *Gracilaria crassa* grew much slower at 0.05–0.2 cm/day.

Program 4: Nutritional requirements of trochus

Milestones

To determine the nutritional composition, including lipid, long-chain polyunsaturated fatty acids, carbohydrate and protein contents of macroalgae and microalgae identified as preferred food species for trochus.

Progress and outcomes

The gross chemical composition, including total carbohydrate, lipid, protein and ash content of 10 macroalgae (seaweeds) collected from Darwin Harbour and one mixed benthic microalgal culture used in feed preference trials with trochus were determined. The samples included five brown algae species (Phaeophyta), three red algae (Rhodophyta) and one species each of green algae, blue-green algae and diatoms. All macroalgal species studied contained adequate levels of protein, lipid and carbohydrate for gastropod nutrition. The lipid contents of another seven species of macroalgae from Darwin Harbour were determined as part of this study.

The percentage of each of 26 fatty acids were determined in all macroalgal and microalgal samples. Among the macroalgae, the highest level of the essential fatty acid eicosapentaenoic acid (EPA) was found in red algae, followed by brown algae. All macroalgae were deficient in essential fatty acid docosahexaenoic acid (DHA), but the mixed benthic microalgae did contain small amounts of this fatty acid.

Milestones

To investigate an artificial diet for trochus.

Progress and outcomes

Seven artificial diets for trochus were tested for 16-hour water stability. Lowest turbidity was found in the diet containing wholemeal wheat flour (30% dry weight), soybean meal (30%), fishmeal (20%), gelatine (15%) and agar (5%). The feeding attraction of this diet, modified to include one of the attractants black pepper, cinnamon, powdered *Spirulina* sp. or peanut butter, to juvenile and mature *T. niloticus* was tested, and the chemical composition of the most attractive diet investigated.

Milestones

To investigate the baseline biochemical composition of trochus including:

(a) comparing the biochemical composition of wild trochus with those kept under aquaculture for 12 months;

(b) investigating the changes in fatty acid composition related to age of hatchery-reared trochus;

(c) investigating the concentrations of arsenic and the metals cadmium, copper, iron, lead, manganese and zinc within the whole body of wild trochus.

Progress and outcomes

The gross chemical composition (carbohydrate, lipid, protein and ash) of the muscle tissue (foot) of wild and hatchery-held trochus were completed. Animals held for one year in the hatchery and fed on naturally grown benthic microalgae had slightly lower total lipid than those recently collected from the wild. Levels of protein, carbohydrate and ash did not change much during hatchery culture.

There were significant changes in the fatty acid composition of foot and viscera of hatchery-held animals. These included significant decreases in the percentages of saturated fatty acids 16:0 and 18:0, and increases in the polyunsaturated fatty acids including Linoleic acid (LA), EPA, and DHA. These changes were believed to be related to dietary fatty acids of benthic algae growing in the hatchery tank.

The fatty acid compositions of 3, 12 and 18-month-old juvenile hatchery-reared trochus were investigated to follow the changes in fatty acid biosynthesis with age. Generally, there was an increase in many of the long-chain polyunsaturated fatty acids of hatchery-reared animals as they grew older (Rebhung et al., these Proceedings).

X-ray analysis of sections of trochus shell using EDAX on the scanning electron microscope (SEM) clearly showed there were no significant inclusions of any other elements in the $CaCO_3$ matrix. The SEM photographs showed significant differences in the patterns of deposition and growth of the shell.

The concentrations of arsenic and the metals cadmium, copper, iron, lead, manganese and zinc were determined on whole-body *T. niloticus*. The concentrations of all elements except As and Cd were below the recommended maximum permissible concentration for molluscs. Cadmium was higher than the recommended levels of 2 mg/kg and there was a positive correlation between size and Cd concentration.

Other Activities

Workshops

Apart from the research programs, three training workshops were conducted at NTU during the three years of the project. The fellowship workshops were conducted for staff from UNDANA and a special training workshop carried out on behalf of the Aboriginal communities, King Sound, Western Australia.

Fellowships and postgraduate studies

In the second year of the project, a postdoctoral staff member Dr Felix Rebhung from UNDANA spent 12 months working at NTU on nutrition aspects of the project. He was followed by another UNDANA staff member, Mr Ricky Gimin, who was awarded an AusAID scholarship under an Australian-Indonesian Eastern University project to take up MSc research into trochus reproduction, spawning and growth, under the supervision of Dr C. Lee. In addition to overseas staff, two NTU students completed BSc Honours working on the trochus project.

Publicity and publications

The project was given a very high profile and attracted strong public support. During the three years, numerous interviews were given by project staff to the Darwin Top FM, ABC and ABC national radio stations. The project also appeared on the ABC 'Quantum' program in June 1995 and in the Territory *Business and Asian Business Review.*

The project produced 21 publications which were reported during the End-of-project Workshop attended by researchers from Australia and the Indo-Pacific region.

Other

In addition to the work at NTU, the most pleasing and encouraging outcome not part of the project brief was the involvement of the Aboriginal communities in King Sound and the staff of WA Fisheries in Broome. Without the strong support of the Aboriginal communities in King Sound and their supply of broodstock, the project would have faced huge costs and additional difficulties trying to obtain supply from Queensland. The success of the project in King Sound has led to the Aboriginal and Torres Strait Islander Commission (ATSIC) Regional Aboriginal Council in Broome incorporating the Kimberley Aboriginal Aquaculture Corporation (KAAC) to advance the aquaculture interests of all Aboriginal communities in the Kimberley.

The positive and supportive interactions among the staff of NTU, UNDANA, the Aboriginal Communities of King Sound and WA Fisheries were probably the best unplanned outcome of this project.

Performance Indicators

Five performance indicators were listed for evaluating the success of the research project. They are:

(a) number of research publications,

(b) number of training workshops and consultancies,

(c) the type and number of culture techniques developed,

(d) expressions of interest by other universities in eastern Indonesia to participate in Phase 2 of the research project, and

(e) the response and interest of Australian and Indonesian fishing communities in taking up the culture of trochus shell.

The performance indicators and the outcomes of the evaluation are summarised below. In all instances, the project performed very well against the indicators set down in the project document.

Discussion and Conclusion

The trochus hatchery and nutrition project was successfully concluded in June 1996, three years after it was implemented. During the three years, all four research programs were completed, with some areas exceeding the targets set.

Research Program 1, setting up a trochus project library in NTU, was completed successfully and is being improved continuously by the ACIAR-funded trochus reseeding research project. The library will grow as more recent literature is accumulated and collected by project staff. At the end of the ACIAR phase of the project in 1998, the library will be handed over to the NTU library.

Research Program 2 has succeeded in many different areas. It has (a) developed a new and non-destructive method of differentiating the sexes of the trochus, (b) established the advantage of using saline bore water supply in a trochus hatchery, (c) developed a successful system design for a closed recirculating system for producing trochus juveniles and (d) standardised the method of inducing trochus to spawn. Unexpected outcomes of the program have been the participation of the Aboriginal communities from King Sound, WA in the project and the training workshop conducted at NTU to train Aboriginal people in trochus spawning and hatchery management.

Research Program 3 has identified many species of macroalgae found in the Darwin and Kupang harbours. Some preliminary work was carried out to grow at least one species of macroalgae (*Caulerpa* spp.). Considerable work can still be carried out to grow different macroalgae, benthic diatoms and microalgae suitable as food for juvenile trochus.

Research Program 4 has completed: (a) determination of the nutritional composition of macroalgae from Darwin Harbour and the microalgae found growing in NTU hatchery tanks; (b) determination of the baseline biochemical composition of trochus broodstock from the wild, and wild broodstock held for 12 months in the hatchery; and (c) investigation of the changes in fatty acid composition related to the age of hatchery-reared juveniles. Preliminary work began on the production of algae plate to feed trochus juveniles, work requiring considerable future attention.

Performance Indicators	Outcomes
Research publications	20 publications, plus two BSc Hons theses and one MSc due for completion in 1996
Workshops and consultancies	Two fellowships conducted for Indonesian staff; one Aboriginal training workshop; end-of-project workshop and regional conference.
Development of new technologies	A new method of determining the sexes of trochus; a state-of-the-art closed recirculating hatchery system for the production of trochus juveniles; the establishment of the use of saline bore water as a good source of hatchery water supply.
Expansion into Phase 2 project	The success of the project led to ACIAR funding a further phase of the project involving reef reseeding research with hatchery-produced juveniles. Three countries (Australia, Indonesia and Vanuatu) with six institutions are involved in the project. A total research budget of $1.49 million is committed with ACIAR providing all the cash component ($680 000).
Interest generated	The success of the project led to (a) the establishment of the first private commercial trochus hatchery in Indonesia and (b) incorporation of the Kimberley Aboriginal Aquaculture Corporation. KAAC is funding the establishment of a multispecies hatchery with trochus as the first species to be produced.

Based on the evaluation of the five performance indicators set by the project document, the staff has done very well in completing its missions.

Acknowledgments

We wish to thank all project staff from the NTU and UNDANA for their support and hard work during the three years of the project. Without their dedication and effort much less would have been achieved. Special thanks to the Aboriginal communities in King Sound and in particular to Mr Colin Ostle of WA Fisheries for their support and efforts. NTU technical staff Kathy Kellam and Nick Ryan helped set up the trochus hatchery and looked after the animals in the hatchery. The former Vice-Chancellor of NTU, Professor Nairn and the Dean of Science provided invaluable support to the project. Finally, sincere thanks to TIL Program, DEET for funding the research project.

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A Review of Trochus Fisheries in Eastern Indonesia

Burhanuddin*

Abstract

Trochus shell is a high-value commodity for domestic use and export for Indonesia. *Trochus niloticus* is the most important species, but another species, *T. makassar*, is said to produce shell of very high quality.

Trochus grow in eastern Indonesian waters. This paper briefly describes the distribution, harvesting, and culture of *T. niloticus*, and gives basic production and trade statistics for the trochus industry.

Catch rates indicate serious over-exploitation of the trochus resource, and various research projects have sought to develop improved management and conservation practices in the industry.

EASTERN Indonesian waters have specific topography and substrate conditions for various marine biota. Various molluscs are found in the coral reef area, one being *Trochus niloticus*, also known as *lola*.

T. niloticus shell has high economic value as an export commodity. The shells are exported to many countries including Japan, Hong Kong, Singapore and South Korea. The shell is used as raw material in paint, buttons, cosmetics, furniture and other accessory industries (Ali et al. 1992). Furthermore, according to Bour (1990), an Indonesian trochus known as *T. makassar* has the best quality shell, compared to trochus shells from South Pacific countries, and is used by the Japanese as a guide to determine the price of different quality shells.

In Indonesia, trochus shell is used in home industries to make jewellery such as earrings, necklaces, bracelets, brooches and rings (Dharma 1988). The meat of the trochus is also used by fishermen as a source of animal protein. The increase in the economic value of *T. niloticus* as export goods and as a commodity within the country causes increased intensity in trochus utilisation, which results in the harvesting area expanding from coastal waters. Indonesian fishermen sometimes enter Australian waters.

The extension in harvesting area may also be due to the fact that some harvesting areas in Eastern Indonesian waters have been exploited intensively, especially near the coast which has a high fishermen population. Areas which already have small endangered populations of *T. niloticus* include south and southeast Sulawesi waters (Ali et al. 1992). Data show that the production of *T. niloticus* tended to decrease because its natural population decreased.

Distribution of *T. niloticus*

T. niloticus lives in a coral ecosystem of dead coral remains layered by algae. The existence of algae layers on coral is an indicator of the presence of *T. niloticus*. Generally it lives in intertidal zones to a depth limit of 8–10 m (Nontji 1987; Sugiarto 1990), 17–20 m (Risamasu et al. 1995).

T. niloticus is found in the Indo-Pacific region, its origin in Indo-Malaya, Melanesia and Micronesia. It has been introduced to eastern islands. In Indonesia, *T. niloticus* is widely distributed in coral regions, especially in coral waters in Eastern Indonesia (Sudradjat and Tonnek 1992).

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In Maluku, *T. niloticus* is found in the islands of Saparua, East Seram, Tanimbar, Barbar and Wetar. In Nusa Tenggara Timur (NTT), it is found in East Sumba, Alor and Kupang waters. In Kupang coastal waters, it is found in Tablolong, Kulit Kuma Cape, Slupri and Air Cina, Semau Island, Rote Island and Landu Island (Risamasu et al. 1995). In east and southeast Sulawesi it is found in Sembilan, Spermonde and Pangkep island (Ali et al. 1992).

Management of Harvesting

The exploitation level of *T. niloticus* has shown critical signs, from both number and size and the aspect of marine life resource conservation. Overfishing has been seen in some areas, and in others the extinction sign of trochus is clearly seen. In National Marine Park in southeast Sulawesi, trochus density is only about 0.42/ha while the total population is about 92 632 shells (Nessa, Rachman and Hatta 1995). This condition forces Buton and Muna (southeast Sulawesi) fishermen to enter Australian waters, which in turn causes problems in the relationship between Indonesian and Australian governments.

The increase in habitat depression (due to coral harvesting and fish bombing) and trochus harvesting will undoubtedly decrease the trochus population and finally will affect other biota.

One effort to conserve *T. niloticus* is to restrict harvesting in certain areas or to determine the size which can be harvested. In Buton, for example, a system is used where harvesting area is auctioned by village authorities.

In Maluku, a traditional conservation system called *sasi* has been developed to anticipate biota extinction. Sasi is applied once every 2–3 years and the season opens when the biota are abundantly available. It lasts for one week (Risamasu et al. 1995). In Palau, local government or village authorities stop harvesting voluntarily for 1–2 years after one month harvesting (Sudradjat and Tonnek 1992). Another management system applied in Palau is seed restocking of natural waters followed by regulating the time, size and number of harvests (Nessa, Ali and Hatta 1993).

In NTT, *T. niloticus* harvesting is in the wet season because the sea is calm, there is no tide and the current is weak. Harvesting is by diving when the sea is at low tide (Risamasu et al. 1995). Harvesting when the sea is calm and at low tide also takes place in Maluku, September to November and February to April (Pieris 1988).

Culturing *T. niloticus*

T. niloticus culture applied at present is still very simple and based on temporary keeping by many fishermen. Seed supply is a main condition for culture and still depends on seed collection from nature. Its availability in terms of number, time and quality varies highly (Sudradjat and Tonnek 1992).

Seed collection techniques vary depending on location. In a relatively flat coral reef with a wide distribution of dead coral, seeds can be found under the coral pieces during the day. At night the seeds are often scattered to graze. Where the coral reef is narrow and very steep, seeds can be obtained by putting papaya and banana stems on coral slopes, where at night the seeds gather and graze around the stems. Many fishermen in Buton and Muna regency adopt this technique. In Selayar regency (south Sulawesi), trochus seed collection uses a simple technique — at night when the tide is low, fishermen bring kerosene lamps to the reef and collect grazing seeds (Nessa, Ali and Hatta 1993).

T. niloticus culture adopted by fishermen is a trial-and-error system and generally in the form of temporary keeping, the keeping instrument varying according to materials available. In Southeast Sulawesi, systems adopted have been fairly developed from using tunnel-net for temporary keeping to the kurung-kurung system and the utilisation of trochus behaviour in nature.

In these culture systems the feed given to trochus is various, including papaya stem, banana stem, animal skin and organic matters which rot in a short time. None of this feed has been well identified scientifically to support the development of trochus commodity (Sudradjat and Tonnek 1992).

The spawning process of male and female trochus occurs (in nature) at night or at sunset for 1–2 days at the time of new moon or full moon. The size of trochus babies can be 3 cm in the first year, about 6.5–8 cm prior to maturity and about 9–15 cm at maturity (10 years old). The size of trochus taken in Indonesia is 5–13 cm.

Trochus Production

In Indonesia, all trochus caught and traded is from nature. In NTT, trochus are collected to a sufficient number, sold to merchants from other islands and finally transported to Ujung Pandang. The cost for one kilogram shell of 5–13 cm is Rp. 10 000–14 000 in Ambon and Rp. 15 000 in Kupang, while the cost of exported shell is Rp. 30 000/kg in Ujung Pandang.

Total trochus export in Indonesia in 1986 was 1 567 930 kg, and in 1990, 165 032 kg (Table 1). Table 1 clearly shows that export volume decreased from year to year (Sudradjat and Tonnek 1992). Specifically, NTT trochus production has fluctuated in recent years (Table 2).

Table 1. Trochus export in 1986–90.

Year	Volume (kg)	Value (US$)
1986	1567930	3808320
1987	1816890	6005425
1988	887230	3928280
1989	460631	2386954
1990	165032	1307539

Table 2. Trochus production (*T. niloticus*) in NTT.

Year	Production (ton)
1988	16.0
1989	17.4
1990	30.7
1991	7.7
1992	4.9
1993	12.65
1994	6.65

The data in these tables may indicate decreasing trochus resources in nature, decreasing harvesting activity due to new government regulation which considers trochus a protected wild animal, or overfishing.

Research into *T. niloticus*

Decreased production from year to year has encouraged efforts of production improvement and conservation, including the following research:

- Trochus (*T. niloticus*) Resources and Culture Management (Ali et al.1992);
- The effect of substrate and fertilization on the survival and growth rate of trochus *T. niloticus* (Nessa et al. 1993);
- Study on reproduction, distribution and density of *Trochus* spp. and *Tectus* spp. in Taka Bonerate National Marine Park (Nessa et al.1995);
- Study on the Distribution Area of *T. niloticus* in NTT coastal waters (Risamasu et al. 1995).

Conclusion

Trochus shell is a high-value economic commodity taken from nature. Overfishing occurs and as trochus becomes a protected animal, its production decreases. But trochus harvesting in areas away from control will result in its extinction.

Some efforts to improve production and conservation have been made by both fishermen and the government, but have not shown any sign of success, possibly because the efforts are very simple and traditional.

Professional efforts that must be made soon include trochus culture, the determination of conservation areas, harvested size and harvesting season, and research into *T. niloticus*. These efforts should be made as soon as possible to limit the effects of continuous harvesting due to the high price of trochus.

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Collation of Natural History and Ethnobiological Studies Relevant to Trochus Fishing in King Sound, Northwestern Australia

K. L. Magro*

Abstract

The aim of this paper is to provide relevant background material for studies on trochus in King Sound (16°00' to 17°07'S and 123°18' to 123°23'E), including a general description of the area and documentation of the available meteorological and oceanographical information. Emphasis is given to collating information from both European and Aboriginal sources. The climate follows the typical wet (December to March) and dry (April to November) seasons of the tropics. The warmest months are November to January and high rainfall occurs from December to March. The Bardi people recognise six seasons during each year according to the direction and intensity of the wind and the amount of rainfall and times of resource availability, particularly marine resources. Waters in King Sound follow a semi-diurnal tide with variations in range between springs and neaps of up to 11 m. Words associated with tide terminology in the Bardi language are complex and reflect their detailed knowledge of water movement in King Sound. The trochus fishery extends throughout the northern waters of King Sound. Trochus are collected from inter-tidal reef platforms by hand at low tide. King Sound is biologically diverse and culturally significant. Collation of the previous research conducted in King Sound provides relevant background material for future research in King Sound.

CONDUCTING research in remote, isolated locations requires an understanding of factors that are likely to affect the research. For marine research in particular, it is important to understand weather conditions and tidal information as these can affect accessibility to field sites and the suitability of environmental conditions to support the planned research. The aim of this paper is to provide relevant background material for studies on trochus in King Sound. This includes a general description of the area and documentation of the available meteorological and oceanographical information and a description of the King Sound trochus fishery. Emphasis is given to collating information from both European and Aboriginal sources.

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Methods

Relevant literature from scientific, ethnobiological and natural history studies were collated. Additionally, interviews with members of the Bardi Aboriginal Community, Fisheries Department of Western Australia and other people with a history of conducting research in King Sound were conducted.

Results

Area description

King Sound is a large inlet extending 145 km by 60 km in the north-west Kimberley region of Western Australia. It extends from latitude 16°00' to 17°07'S and from longitude 123°18' to 123°23'E (Australian Gazetteer 1975). The group of islands at the entrance of King Sound belong to the Buccaneer Archipelago (15°57' to 16°12'S and 123°13' to 123°26'E). The Buccaneer Archipelago contains an extensive island network of over 100 islands, depending on the tidal height. The islands vary in size, the largest being MacLeay Island covering an area of 2400 hectares. There is a diversity of interesting features on the islands in King Sound; bird rookeries are found on the Swan Islands, rock wallabies on Long Island and rich iron-ore deposits on Koolan and Cockatoo Islands (Wright pers. comm.). The islands in King Sound were formed during the Devonian geological period (Semeniuk 1986). The climate and biota of these islands are characteristic of the Western Australian tropical biogeographical province. The Bardi Aborigines reside at One Arm Point (16°26'S and 123°05'E) on the eastern side of the Dampierland Peninsula.

Meteorology in King Sound

Meteorological information was obtained from the Western Australian Regional Office of the Bureau of Meteorology. A weather station was established in 1985 at Cygnet Bay, approximately 5 km west of One Arm Point at 16°27'S and 123°00'E (1:50000 map Sunday Island). This station records rainfall, daily minimum and maximum temperatures and other meteorological data.

The average monthly minimum and maximum temperatures are shown in Figure 1. The warmest months are November to January. The hottest daily maximum temperature of 34.5°C occurs in December and the hottest daily minimum temperature of 24.0°C occurs in November. The coolest months are June to August. The coldest daily maximum temperature of 27.5°C occurs in July and the coldest daily minimum temperature of 14.9°C occurs in August.

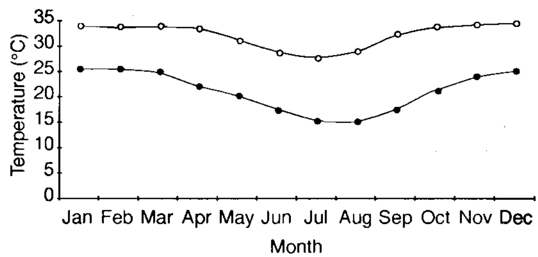

Figure 1. Average monthly minimum (closed symbols) and maximum (open symbols) temperature (°C) recorded by the Bureau of Meteorology at Cygnet Bay (lat. 16°27'S, Long 123°00'E) from 1985 to 1990.

Figure 2 shows the average monthly rainfall data. The rain season extends from December to March. The highest average monthly rainfall is 224 mm in January. South-east trade winds and northerly monsoons contribute to the heavy, seasonal rainfall which can accompany tropical cyclones and thunderstorm activity. These meteorological patterns conform to the wet and dry seasons of tropical areas. Low precipitation occurs during winter months from April to November. The wet season occurs between December to March and the dry season occurs between April to November.

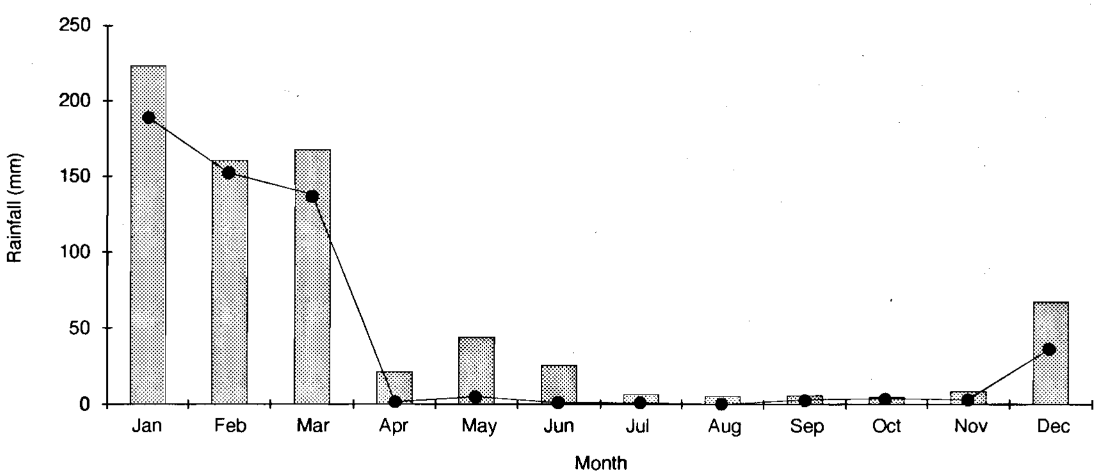

Figure 2. Average monthly (bars) and median (lines) rainfall (mm) recorded by the Bureau of Meteorology at Cygnet Bay (lat. 16°27'S, Long 123°00'E) from 1963 to 1990.

The Bardi people recognise six seasons during each year according to the direction and intensity of the wind, the amount of rainfall and times of resource availability, particularly marine resources (Smith 1985). The Bardi people would traditionally use reefs around One Arm Point during two of the more favourable seasons: the *Iralboo* or king tide season, between March to May, where there is usually no wind at the start of this season but south east winds develop in April; the *Djalalayi* or warming up season, between August and September, where there are strong west winds and unusually low tides (Smith 1987). Reefing activities, including trochus collecting, are more likely to occur during these favourable weather seasons because the low tides exposure large reef areas and the winds are generally favourable for small boats.

Oceanography in King Sound

There is very little oceanographic information available for waters in King Sound. The water in King Sound originates from Indonesian flow-through; the "flow of warm, low salinity water from the Western Pacific through the Indonesian archipelago" (Pearce and Cresswell 1985). There are no water temperature depth profiles available. Rand Dybdahl (Formerly Fisheries Department of Western Australia) recorded sea surface water temperature from a depth of 20 m at Deep Water Point, approximately 25 km south from One Arm Point at 16°40'S and 123°05'E. The sea surface water temperature varied from 30.4°C in February to 24.3°C in July (Figure 3).

The tidal height and range in King Sound are characteristic of the region. It is a semi-diurnal tide with variations in range between springs and neaps of up to 11 m (Australian National Tide Tables 1986). There are few references to measured tidal elevations within King Sound and there are no tidal charts for locations within King Sound, except Derby. The Australian National tide Tables (1986) allow calculation of tides at secondary ports, such as Sunday Island, using Port Hedland as a standard port. These calculations, however, indicate a maximum tidal range of 6 m for Sunday Island. In the authors view this under-estimates the actual tidal range.

Easton (1970) suggested that many islands in King Sound modify the tides producing variable changes in tidal amplitude and a time difference of several hours compared to standard ports in the Kimberley. The tides and powerful tidal flow dominate the major water currents within King Sound where there are many narrow straits between the coastline and the numerous islands. The moving tides can produce currents of up to 10 to 30 knots with whirlpools, eddies and vertical walls on the water surface. The Bardi Aborigines have a highly accurate and precise knowledge of the oceanography in King Sound and the Buccaneer Archipelago. This knowledge assists navigation of traditional double rafts, known as Kalwa, throughout the Dampierland Peninsula (Ackerman 1975) and more recently to navigate boats and dinghies with outboard engines.

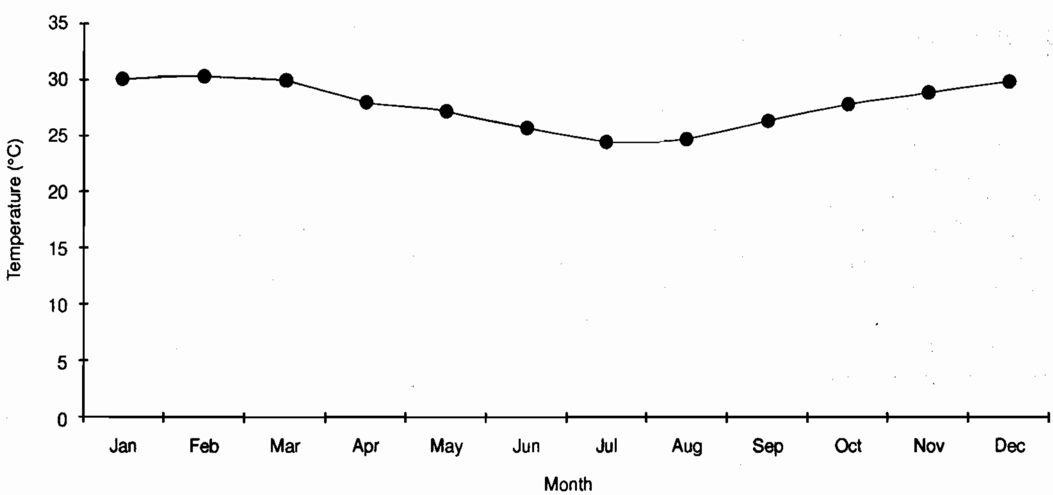

Figure 3. Average monthly sea surface temperature (°C) recorded from 20 m water depth near Deep Water Point (Lat 16°40'S, Long 123°05'E) between 1982 to 1984.

Aklif (pers. comm.) recorded tide terminology as part of a Bardi dictionary that was collated in 1993. *Ganyginy* and *nalan* are Bardi words describing the two major descriptive components of a tidal regime, neap and spring tides respectively. However, the words associated with tides in the Bardi language are complex and reflect their detailed knowledge of water movement. A strong spring tide *(goorlornoo)* can be dangerous for boating activity because the whirlpools *(jiidid)*, strong currents (*jarrany* and *loo*) and waves *(alalgoord)* can carry boats into the ocean, making boats difficult to move, tipping them over or pushing them against rocks.

There are Bardi words to describe the movement of the tides during each cycle, regardless of the tide being spring or neap. The different phases during each tidal cycle are useful for different activities. The safest opportunity for boating occurs with the reduced currents during the middle tide (between full and low tide) known as *noorrngoorrngoo*. *Iwooloongan* describes a tide moving in towards a full. *Imbooloongij* describes a tide that is nearly full and this is a good time for swimming. A tide at the maximum height (full tide) is known as *boorrnginyjin* and when it is going out towards a low tide it is known as *iyoordin*. *Inyjoordij* describes a low tide which is a good time for walking on exposed reef flats or fishing off the edge of the reef flat, although there may be water on the reef flat *(ooloolarridin)* before the reef is fully dry. Trochus collecting often occurs during *inyjoordij*. A very full tide *(Inoomboorngij)* is an especially good time for reefing activities, including trochus collecting, when the reefs are "dry" during *inyjoordij*.

Description of the Trochus Fishery at One Arm Point

The trochus fishery extends throughout the northern waters of King Sound where approximately 60 of the 350 Bardi community members hold a permit to collect trochus. A summary of the fishery is given in Table 1. Trochus are collected from inter-tidal reef platforms by hand at low tide. This form of collection is known as dry picking. There is no swimming or diving for shell, possibly because of poor visibility and strong tides. Shells are collected during appropriate low spring tides when the reefs are exposed. Fishers suggested the tides are the main influence on fishing activity and other seasonal variation are less important. Trochus permit holders get to the inter-tidal reefs by boats, usually 14 ft outboard-powered aluminium dinghies. The crew size varies between 1 and 5, but is usually 2. The boats are left on the reef while the crew walk around the reef and gather shells into 2 gallon buckets.

Table 1. Summary of modern trochus fishery.

Resource species	Trochus niloticus
Distribution	Unknown accurately
Fishing grounds	Northern king sound, buccaneer archipelago
Catch composition	Single species fishery
Annual production	Has fluctuated from 135 to 29 metric tons
Fishing boats	Aluminium dinghies
Fishing gear	Hand gathered (no diving)
Crew on boats	1 to 5 people per boat
Fishing effort	Hours spent gathering during appropriate tides
Fishing season	Throughout the year according to tides
Fishing base	One arm point
Management regulations	Only size limits (65 to 100 mm)
Disposal of catch	Sold as raw shell or processed at one arm point
Channel of disposal	Through the Community Council

The shells are processed at the end of a day's fishing to avoid them becoming "stinky". The fishers clean the shells on the beach in 44 gallon drums filled with boiling sea water and then tap the flesh out of the shell. The shells are then sold to the community council, where the sizes are inspected. The shells are weighed and placed into hessian bags awaiting shipment. One hessian bag contains approximately 70 kg of raw shell. The majority of shell is exported but a proportion of raw shell is used at One Arm Point for polishing and jewellery making.

The only management restriction for the trochus fishery is a size limit. The size limit corresponds to the measurement across the base of the shell from edge to edge. The size limits have changed over the last 11 years. The minimum size limit was 64 mm (2.5 inches) when the fishery first opened (Commonwealth Gazette 1970). The minimum size limit increased to 76 mm in 1983 (Government Gazette 1983) and was reduced to 65 mm in 1984 with a maximum size limit of 100 mm introduced (Government Gazette 1984). The 65 to 100 mm size range was decided by the Bardi Council and the Fisheries Department.

Trochus are dry picked from exposed reef flats during low tide periods in King Sound by the Bardi Aboriginal Community (Jones 1986) generating an annual revenue of $500 000 (Altman et. al. 1993). The fishery at One Arm Point began in mid-1979 when the Fisheries Department of Western Australia issued a licence to the Bardi Aborigines Association to collect trochus. The Bardi Aborigines Association held the only license to collect trochus in Western Australia until September 1992 when a second trochus license was issued to the Lombadina Aboriginal Community and their joint venture. A third trochus fishing license was issued in early 1993 to a group of Torres Strait fishers.

The Bardi Council impose strict penalties on fishers caught breaking management regulations. Fishers have lost their permits for collecting too many undersized shells, so they check shell sizes with special gauges. The need for appropriate management was expressed by all community members interviewed.

Discussion

King Sound is biologically diverse and culturally significant. The depth of both is unknown due to the limited studies that have been conducted there. Collation of the previous research conducted in King Sound provides relevant background material for future research in King Sound, and an understanding of the previous ethnobiological research is useful reading for anyone interested in working within the aboriginal communities.

The trochus resource has been harvested over a long period and has cultural significance to the Bardi community. Harvesting of trochus in the future is desired by community members and this will require a more detailed understanding of the trochus populations in King Sound. Also, there is a need to investigate the potential for aquaculture of trochus as a means of both reseeding reefs that have been overfished and to support an industry of importance to the roots and independence of the Bardi people.

Acknowledgments

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A Review of the Trochus Fishery in King Sound, Western Australia

C. Ostle*

Abstract

The trochus fishery on the northwestern coast of Australia has a long history of exploitation. At its peak the fishery produced 135 tonnes of shell in one year. Continued exploitation with increasing size and effort restrictions have resulted in the fishery possibly stabilising at an annual production level of 30 tonnes. To guarantee safe harvest levels of this order stock enhancement and management intervention is required.

THE natural range of trochus or topshell, *Trochus niloticus*, extends along the northwestern Australian coast approximately between latitudes 3°30′S and 20°30′S.

Viable populations also occur on off shore reef complexes within that range. King Sound is the only area of the coast supporting a commercial stock of trochus. The fishery is located within a 20 nautical miles radius of position 16°20′S; 123°20′E and extends across approximately 600 square nautical miles at the mouth of the Sound.

King Sound

The entrance to King Sound is 30 nautical miles (52km) wide, contains chains of small silt stone islands with adjoining and free standing reef areas of generally the same silt stone composition and coral with areas of weathering soft quartz-gneiss and granites.

Extreme twice daily tides provide a range of 6 to >9 metres and deliver high energy flows of both sea and estuarine type water to the area through major flow channels that dissect the mouth. The tidal influence is maintained to the mouths of the rivers that feed into the base of the sound, some 75 nautical miles (120km) inland.

Water at the base of the sound is subject to a wide salinity range due to seasonal flooding from the rivers and the effects of high evaporation rates and extensive tidal flats. High levels of turbidity are also maintained in the southern half of the sound and are influenced by the big silt load of the rivers and high energy tidal flows across the mud flats.

Little is known of the actual water body movements or rate of exchange occurring in the southern Sound due to tidal influences. Observations indicate a low rate of exchange and a high sedimentation level that limits the establishment of aquatic vegetation or corals and associated life forms.

The interface zone, between the outer and southern sections of the Sound, is an area of low to marginal productivity for trochus, probably due to poor conditions for recruitment and growth.

Coastal areas to the north of King Sound are subject to turbid conditions and do not support large populations of trochus.

The trochus fishery of King Sound is limited to the high energy tidal, clean sea water movement area that gives rise to healthy clean reefs clear of silt deposits. Despite this, the animals still maintain distinct areas of preferred habitat within the greater area of the fishery that relate more to location in relation to water

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movement than to reef type although the latter is thought to have some effect.

Harvesting

The trochus stock in King Sound is harvested entirely by the region's Aboriginal communities with one particular group, the Bardi at One Arm Point, holding the main harvesting authorisation. The group purchases and markets the bulked catch. There are two other non connected groups holding restricted licenses to gather shell for value adding and sale within their communities.

The Bardi authorisation has enabled the community to sub authorise a certain number of community members to gather trochus each year. This arrangement attempts to manage the resource to maintain viable populations by taking into account seasonal conditions, the limited technology available and the wide distribution of the stock.

The number of fishers or collectors has varied over the years as has the criteria for holding an authorisation. However, an authorisation has always allowed for expansion, because family groups often work under what is regarded as being a single unit of effort or authorisation.

Trochus has provided the major source of individual employment and economic return for this community group since the late 1970s when the fishery recommenced after a break of approximately 40 years. The social value of the trochus fishery has been very high, as it provides self employment, a business structure and non-welfare dollar returns within the community.

Production

From a peak of 135 tonnes of shell harvested from a virgin fishery with no size or effort restrictions, production has progressively declined to 30 tonnes last year, albeit with rigidly enforced size and effort restrictions. Indications are that this year's catch will be of the same order, again with a rigidly enforced size limit of 65mm and a reduced number of authorisations.

An annual catch of 40 tonnes had been considered sustainable. However the failure of some areas to recover or recruit juveniles is beginning to indicate to some fishers that areas are being over harvested. As a consequence, fishers are moving further afield to gather shell but this is not an answer to declining catches.

Harvesting methods have remained constant over the years, with fishing being confined to searching for and gathering by hand into buckets when the animals and reefs are exposed at low spring tides. No diving has been allowed or used as a collection technique for harvesting trochus from the submerged reef faces. This allows the trochus in these areas to remain unfished while they remain there.

Fishers are beginning to recognise population decline in areas of previous abundance. As a consequence, the community is now looking at further reducing the number of authorisations and investigating the possibility of stock enhancement through reseeding with hatchery-reared juveniles.

Resource Management

Overall management of trochus in Western Australia rests with the Fisheries Department and that agency is at present reviewing its policy and options for the King Sound fishery. Options being considered include:

- a reduction in fishing effort
- protection of certain areas or sections of the population to maintain a viable brood stock
- raising the minimum harvest size to 70 or 75mm to allow more animals to achieve at least one spawning before capture
- exploring value-adding as a means of reducing reliance on volume to provide maximal financial returns
- a total yearly catch limitation.

All of these options require the full support of the fishing community to have the desired effect. Constant supervision by departmental enforcement personnel and/or community rangers is not possible nor would it be effective given the extensive area the fishery and consequently the cost of surveillance.

Seasonal environmental conditions such as the extreme tidal influences and the lack of suitable boats to continually fish the more remote sections of the fishery has protected these sections from over exploitation. As a consequence there are areas with sufficient numbers of trochus to provide viable spawning groups.

Poaching has been a problem in the past and at its peak certainly influenced population levels on the islands and reefs in the mouth of the Sound. Odd instances still occur but this activity is not considered a major threat at this time. This situation could change should reseeding lead to the establishment of large numbers of animals on the reefs.

Conclusion

The trochus population of King Sound is showing signs of decline and this is probably due to over-exploitation. It is possible that environmental factors are contributing to the decline and may suppress or limit recovery. These environmental effects may be short-term and capable of correction through stock enhancement and/or strict management controls and interventions.

Hopefully the catch has stabilised again.This will allow adjustments to be made to conserve the stock while it still remains economically viable and enable the development of a better understanding of its maintenance requirements.

Current Practice and Tradition Related to Trochus Fisheries in Eastern Indonesia

J. C. Dangeubun*

Abstract

The only system currently practised in Eastern Indonesia in managing the trochus resource is *sasi* (closed area or seasons). Sasi is the traditional system of protecting natural resources, a system banning fishing activity in a specific resource at particular areas and periods. Besides regulating the resources, sasi also regulates community relationships by organising and controlling behaviour and attitude. Study of the annual harvesting of trochus in the last 15 years in Eastern Indonesia indicates that the sasi system by itself is not adequate to protect trochus resources.

ALL Moluccans are familiar with the word 'sasi', which plays an important role in the traditional way of managing natural resources in this eastern part of Indonesia. Everyone in this area knows exactly what sasi is, how it works in the community, and how far this system benefits them.

Sasi, (closed areas or seasons) is both written and spoken regulation to ban the fishing activity of a particular resource at a particular area and in a particular period. Besides regulating the resources, sasi also regulates community relationships by organising and controlling behaviour and attitude. Related to sasi is the *Lembaga Adat*, a community-based board which elect a suitable person who is given the responsibility of controlling the operation of the sasi system.

Almost all the coastal villages in Kei Besar harvest trochus as their main (in most villages the only) source of cash income. Harvesting trochus is an activity in which all the families in the village, as part of the community, fully participate. Although none of the fishermen in Kei Besar could remember exactly when they first harvested this resource, they all agree that trochus exploitation was already taking place through a number of generations in the past. Previously the trochus resource was harvested once or twice annually. However, in recent years, the frequency of harvesting has been reduced to once a year or once in two years. This condition applies in almost all the trochus fishing grounds in Kei Besar.

Like other natural resources, one could expect a significant reduction in density as well as size distribution of the trochus population in this area. However, the trochus population in Kei Besar is considered to be in a healthier condition than any other trochus population in Maluku. It is believed that one reason for this condition is that all villages in Kei Besar strongly practise 'Trochus Sasi'.

This paper discusses current practice and tradition related to *Trochus* fisheries in Kei Besar, Eastern Indonesia.

Materials and Methods

Ten villages of more than 100 coastal villages around Kei Besar were randomly chosen as study sites. From each village, 10% of the population was randomly sampled for interview and filling in questionnaire sheets. The questions concerned how sasi is practised on the trochus resource, the annual harvesting of trochus, the socioeconomic aspects related to the utilisation of the trochus resource, other sources of cash income, indigenous environmental knowledge and awareness and community perception of sasi of the

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trochus. All the information was then descriptively presented and discussed.

Results

In Kei Besar, there are three types of sasi in trochus resources. First, fishing trochus is banned in particular trochus fishing grounds for one or two years. In this type of sasi, other fishing activities are allowed to take place. However, certain fishing methods usually practised by local people in collecting small fish and shell (for example, overturning boulders or rocks) are not allowed. Second, all fishing activities are banned. In this type of sasi, no fishing activity at all can be allowed to take place in this particular area of trochus population. This is usually applied to the trochus fishing ground which according to local people is the prime trochus fishing ground or the highest density trochus population. The period of this type of sasi is about the same as the first, for one or two years. Third, fishing is banned to the different trochus fishing grounds, alternatively. In this type of sasi, fishing trochus is allowed in one or two fishing grounds while others are closed, then vice versa in the next period. This fishing banning period is known as 'tutup sasi'. Regulations and sanctions are set up by the village leaders for the sasi system.

After the closed period, fishing activity is opened (called 'buka sasi') with an opening ceremony by religious leaders in the presence of the village leaders and the traditional institution leaders. 'Buka sasi' takes place for about two weeks to one month. All members in the community have the right to harvest trochus. In some villages, there is a small fee for administration collected from each member who harvests the trochus. In most villages there is no fee, but they allocate certain trochus fishing grounds to be harvested only for village administration purposes, some only for the church or for other religious bodies in the villages.

In most villages in Kei Besar there is declining annual production of trochus. This is due to many factors, including the reduction in the size and density of the trochus population, the marketing system which does not allow legal marketing for wild trochus, the economic pressure which caused 'sasi' to open more often, and other socioeconomic problems such as migration or young people becoming accustomed to modern ways.

One of the biggest problems faced by the local community in maintaining its trochus population is economic pressure. Most of the village community (95%) fully depend on the availability of natural resources, while the only cash resource for the villages is from coconuts and trochus. This condition puts a lot of pressure on the trochus population because, unlike coconuts which are owned by families or at least by clans, trochus is communally owned. In addition, market demand for trochus increases while the price of coconut decreases. Because of these pressures, the sasi system in most villages has become more flexible and allows more frequent trochus fishing during the banning periods.

Discussion

Nationally, the regulation about the conservation of natural resources and ecosystems is very new (1990), while regionally, this regulation is already part of life for the people in Kei Besar from generations of practising sasi as their traditional way of managing resources. If the local community is given the choice, it would rather be regulated by sasi than by national regulation. Sasi is part of their culture and their identity, while national regulation is not fully part of community life activities. One basic reason why the national regulation is not fully part of community life activities is that it does not fully spread to all parts, especially remote communities, in the nation.

In addition, through sasi each person in the community learns to acknowledge the communal ownership of the resources. For example, at the opening of sasi of the particular resource, where all the sasi regulations are withdrawn, the resource will be harvested by everybody in the community. The harvested resource will then be divided equally among the whole community after a small percentage is allocated for village administration purposes. In utilising the resource each individual in the community has equal responsibility for protecting the resource. Besides that, all of the conservation regulations are applied at the time of harvesting, for example, only certain sizes can be harvested, or at certain times.

Because sasi is the culture and the identity of the community, all of the regulations (including its sanctions) will be fully part of its life, to be respected and abided by by the traditional community. The regulation not to harvest or utilise a particular resource at a particular area and time will be fully observed by the community. In this case, sasi is ecologically a very significant tool in supporting sustainable resources development in any area maintained by a traditional community.

Design and Operation of a Land-based Closed Recirculating Hatchery System for the Topshell, *Trochus niloticus*, using Treated Bore Water

C. L. Lee*

Abstract

Saline bore water high in iron was successfully treated and found to be ideal as a source of seawater for a land-based research hatchery. After aeration, sedimentation and dilution with fresh water, the treated saline bore water was used successfully to maintain trochus broodstock to produce juvenile trochus in the hatchery of the Northern Territory University. Wild trochus collected from King Sound, Western Australia were successfully translocated and maintained in the closed recirculating tank system developed there. The wild broodstock spawned and the F_1 matured and produced F_2 successfully after 2.5 years, thereby 'closing' the life-cycle. The closed recirculating system was highly efficient in water utilisation and low in labour input. The system was adequate for producing the juveniles needed for the nutrition and related growth rate and reseeding studies. Over the three years, several hundred thousand juveniles of different size classes ranging 1–25 mm were produced in the NTU hatchery. Average estimated cost of production for the smaller 1–3 mm size class juveniles varied <1.0–3.3 cents/juvenile.

THE marine topshell *Trochus niloticus* forms a small artisanal fishery in Australia and many Indo-Pacific countries. Increase in the price of the shell in the last 10 years led to overfishing and serious depletion of the stock in many countries. As a result, in Indonesia for example, trochus fishing was banned in 1987 under a decree of the Ministry of Forestry (Arafin and Purwati 1993). In many Pacific countries and Australia, management measures including catch quotas and catchable size limit (lower and upper limits) at harvest were implemented, and more recently stock enhancement of trochus through hatchery-produced juveniles was considered as one of the options for sustaining the trochus fishery.

In July 1993, the Northern Territory University (NTU), Darwin and the University Nusa Cendana (UNDANA), Kupang, eastern Indonesia were funded by the Commonwealth Department of Employment, Education and Training (DEET) to commence a joint research project on trochus hatchery and nutrition under its Targeted Institutional Links (TIL) Program. Since trochus was first recorded in the 1980s to spawn in the hatchery (Heslinga 1980; Heslinga and Hillmann 1981), numerous papers on the method of natural spawning and juvenile production have been published (Nash 1989; Amos 1991; Kikutani and Patris 1991). All the hatcheries mentioned were based on flowthrough systems using either untreated or filtered natural seawater; they were mostly sited on atolls or coral reef islands where high-quality seawater was readily available.

The NTU hatchery is approximately 1.5 km from the coast where the seawater quality is at best highly silty and subject to violent fluctuations in salinities during the annual monsoon season. Consequently, seawater obtained directly from the sea is unsuitable as a source of hatchery water. The hatchery in the university therefore sourced its seawater supply from an underground bore located beside the hatchery complex. This paper reports on the treatment of the saline

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bore water prior to its use and describes the closed recirculating tank system developed for producing trochus juveniles in the hatchery in NTU using the treated saline bore water.

Materials and Methods

Water supply and treatment

The NTU hatchery seawater was obtained by drilling an underground bore to a depth of 55 m beside the hatchery site. Internal diameter of the bore was 146 mm and water from the bore was delivered to the hatchery via a 50-mm PVC pipe. Its flow was rated at 2.25 L/second. The raw bore water has a very high salinity of 51–52 parts per thousand (ppt), is very rich in iron and has a low pH (Table 1). It had to be treated and the iron removed prior to its being used in the hatchery. The five steps involved in the treatment of the saline bore water were:

—water was pumped to a circular tank of 12 000 L capacity;

—water was aerated vigorously for 24 hours; during aeration, the ferrous iron in the water was oxidised to form precipitate of ferric iron;

—precipitates were allowed to settle for 72 hours to the bottom of the tank;

—cleared saline water was pumped to a mixing tank of 9000 L capacity;

—saline water was mixed with tap water until a salinity of 35 ppt was obtained. The water was then pumped up to an overhead tank which supplied the entire hatchery complex.

Spawning and larval rearing tank

Trochus were spawned either in rectangular glass tanks each 800 mm × 570 mm × 470 mm high or in plastic basins 600 mm × 400 mm × 330 mm high; each tank was able to hold up to 50 broodstock. The method of inducing the trochus to spawn in the NTU hatchery is described by Gimin and Lee (these Proceedings) and Dobson (these Proceedings). After spawning, the eggs were transferred to the larval tank for hatching and for ongrowing to postlarvae (P/L) and juveniles (Js).

Each rectangular larval tank was made of fibreglass material and measured 3500 mm × 2000 mm × 900 mm high. The tank was divided into two compartments, a smaller filter compartment (SFC) measuring 900 mm × 2000 mm × 900 mm high and a larger spawning and P/L and Js rearing compartment (LRC) measuring 2600 mm × 2000 mm × 900 mm high (Fig. 1). The two compartments were connected via two sets of five 50-mm holes drilled through the partitioning wall; one set of holes is located at the bottom and the other two-thirds up the partitioning wall. The five upper holes allowed up to five 40-mm air-lift pumps (ALP) to be installed. These were used to circulate the water from the LRC through the SFC. Water returned to the LRC via the holes at the bottom of the partition.

Table 1. Quality of bore water supply before and after treatment by aeration and sedimentation — NTU hatchery.

	Bore water		Seawater
	Before	After	
Sodium	16400	10700	10560
Calcium	689	447	400
Potassium	514	339	380
Magnesium	2059	1349	1272
Iron	20	030	0.02
Silica	20	15	8.6
Chloride	30250	20000	18980
Sulfate	4800	2850	2560
pH	6.2	7.8	8.2
Salinity	51	35	35

All ions in ppm, salinity in ppt; before = saline raw bore water; after = bore water after 24 hours aeration, 72 hours sedimentation and dilution to 35 ppt.

The SFC served as a biological filter bed and consisted of a 'false' bottom filled with layers of graded shell grits 3–12 mm diameter to a depth of about 400 mm. The coarse shell grits were placed on the surface of the filter bed with the finer shells placed in subsequent lower layers. During the period of larvae rearing, the tank was filled with water to a height of 700 mm thus giving a water capacity (SFC and LRC) of about 3500 L. Each LRC was able to hold up to 1–2 million eggs although much higher egg densities had been stocked in the tank.

Water from the LRC was transferred to the biological filter bed of the SFC by using a series of three 40-mm ALPs installed via the 50-mm holes located two-thirds up the partitioning wall. The other two holes are used to install another two ALPs for closed circulation of the SFC (Fig. 1). The ALPs pumped water from the LRC to the SFC via a closed-loop 40-mm PVC pipe placed on the bottom of the LRC. Approximately 50 8-mm holes were drilled along the closed-loop

PVC pipe at the point furthest from the partitioning wall. This arrangement allows optimum movement and efficient filtration of the water in the LRC (Fig. 1). The water flowed through the filter bed and returned to the LRC via a series of five 50-mm holes placed below the false bottom of the filter bed. The flow rates of the pumps were regulated so that water passed through the filter bed at least six times a day. This closed recirculating larval rearing system (CRLRS) for producing P/L and Js allowed great flexibility in its operation and was extremely efficient in producing the juveniles needed.

Results and Discussion

Water quality

The treatment of the raw bore water by aeration to oxidise the ferrous iron into ferric followed by sedimentation of the precipitate was found to be a cheap and easy means of removing the high iron content. After aeration, precipitation and dilution to 35 ppt, the iron content was reduced to 0.3 ppm which was found to be acceptable for use in the hatchery. The pH of the water after treatment had also improved mark-

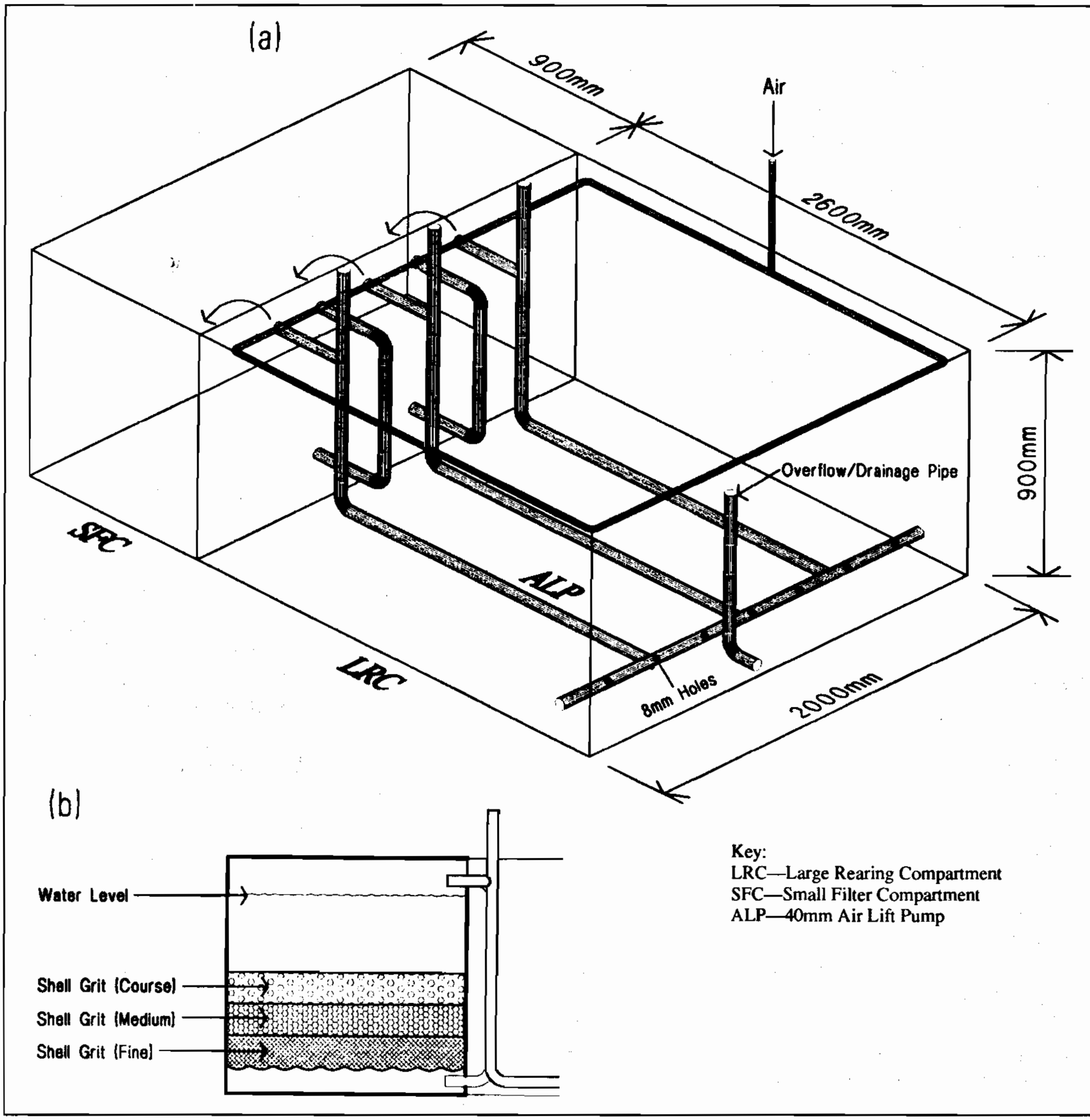

Figure 1. Closed recirculating larval rearing and juvenile production system for trochus in NTU; (a) overall view, (b) cross-section of SFC.

edly and all the major ions present in the water were within the range suitable for growing marine organisms (Table 1). An added bonus of using treated bore water was the absence of suspended particles, fouling marine organisms, planktons and organic matters. The treated water was sterile and could be held in the delivery system (consisting of 50-mm diameter PVC pipe) supplying the hatchery for up to a week or more without deterioration or fouling. In the three years of using the treated saline bore water to supply the NTU hatchery, no undesirable side effect or problem was recorded for the different marine species maintained in the hatchery, i.e. trochus, barramundi and algae.

Larval tank and system operation

The CRLRS design was found to be adequate for producing the juveniles needed. It had many features that made it ideal for producing juveniles of benthic grazing species such as trochus. The system design allowed tremendous flexibility in its operation. For example, trochus were induced to spawn in glass tanks and the fertilised eggs or trochophore larvae transferred directly into the SFC where they hatched into P/L. Alternately, the broodstock were induced to spawn in the LRC and the fertilised eggs and trochophore larvae gradually air-lifted by the ALPs to the SFC over 2–3 days before they settled as P/L and grew on the shell grits in the filter bed of the SFC. When required, they were harvested and transferred together with the shell grits to other systems for ongrowing or experimentation.

If it was preferable to allow the P/L to metamorphose and grow in the LRC instead of the SFC, the system design allowed the LRC to be closed off from the rest of the system. This was done by simply switching off the three 40-mm ALPs from the LRC and turning on the two 50-mm ALPs to recirculating mode in order to allow the biological filter bed to run without impairment and deterioration. After the larvae had settled as P/L in the LRC, the SFC could be reconnected to the LRC to provide the biological filtration function needed to maintain the water quality in the LRC of the CRLRS.

This CRLRS system reduced the need for manually handling a large number of eggs, larvae or P/L and was a valuable labour-saving system for producing P/L and Js in a high-labour-cost country like Australia.

Juvenile production

The larvae settled as P/L within 4–5 days after spawning. At the end of about three weeks, 50 000–100 000 juveniles could be produced. After this period, food in the form of benthic diatoms growing on the surfaces of the shell grits was exhausted and juveniles suffered an increasing rate of mortality. By week 10, the highest number of juveniles recorded in the LRC was 11 900 (Table 2).

To reduce mortality and to improve growth rates, soon after 2–3 weeks the juveniles growing on the shell grits in the SFC needed to be dispersed into the LRC, where a larger growing area and food were

Table 2. Hatching rate and larval settlement of trochus induced to spawn in the NTU hatchery.

Spawning	Fecundity	Survival to settlement (%)	Survival to 3 weeks	Survival to 10 weeks
5/9/93	>3 000 000	82.0	50 000[a]	c
3/8/94	750 000	70.5	d	d
8/8/94	250 000	83	105 000	11 900
24/11/94	50 000	71.4	d	1 700[b]
16/8/95	13 000 000	mostly died[c]	d	2 700
12/11/95	1 600 000	72	57 000	9 500

[a] Very high settlement and survival; more than 50 000 after 3 months; no accurate record kept; 50 F_1 juveniles reared to maturity and most of them spawned on 12/3/96.
[b] Numbers after eight months.
[c] Spontaneous mass spawning resulting in overstocking and subsequent fouling of water.
[d] No sampling carried out.
Fecundity rounded up to the nearest thousand.
Survival to settlement (%), survival to 8 and 13 weeks, rounded up to nearest 0.1%.

available. This was easily achieved by collecting and transferring the shell grits (together with the attached juveniles) manually to the LRC. The production and growth of the benthic diatoms, e.g. *Nitzschia* sp. and *Navicula* sp. in the LRC were accelerated by inoculation with diatom culture followed by fertilisation. Initially, the hatchery used fibreglass plates to provide increased surface area for growing diatoms. This was discarded when it was found that Js grown on plates in the hatchery were not adapted to grazing on coral rubbles. It was observed that these Js avoided using the coral rubbles as their habitats. Consequently, the hatchery design was improved by replacing the fibreglass plates and introducing large coral pieces and coral rubble into the LRC and fertilising and inoculating them with diatoms before the newly settled P/L were released.

The NTU hatchery used a commercially available water-soluble fertiliser Aquasol™ applied at a rate of up to 10 ppm weekly to the rearing tanks. Aquasol is a fast-acting soluble fertiliser with added trace elements developed for the horticulture industry; its composition is given in Table 3.

Table 3. Chemical composition of the commercially available Aquasol™.

Composition	(%)
N as mono-ammonium phosphate	1.8
N as potassium nitrate	2.6
N as urea	18.6
Total P as mono-ammonium phosphate	4.0
K as potassium nitrate	7.8
K as potassium chloride	10.2
Zn as zinc sulfate	0.05
Cu as copper sulfate	0.06
Mo as molybdenum as sodium molybdate	0.0013
Mn as manganese sulphate	0.15
Fe as sodium ferric EDTA	0.06
B as sodium borate	0.011
Maximum biuret	0.40

Using the CRLRS developed in NTU, a few hundred thousand 2–25 mm juveniles trochus were produced over the last three years. Many were sacrificed for nutrition studies, some were used for reseeding research in King Sound, Western Australia, a few were kept to produce future broodstock, and the rest preserved in formalin for further studies. It was encouraging to record that the 59 F_1 born in the NTU hatchery in September 1993 matured and spawned in March 1996 to produce viable F_2, thereby closing the trochus spawning cycle in the CRLRS of the NTU hatchery.

Cost of production

The estimated cost of producing small (1–3 mm) and medium (6–10 mm) size classes of juveniles are given in Table 4. The production costs are derived by using the following assumptions.

- —The large fibreglass tank can be used six and 2.4 times a year for the small and medium size juveniles respectively with the value depreciated to zero over five years.
- —Cost of fibreglass tank plus fittings is calculated to be $2500.
- —The two glass spawning tanks can be used six times a year with the value depreciated to zero over five years.
- —Cost of glass tanks plus fittings is calculated to be $300.
- —Cost of fertiliser Aquasol™ applied at a dose of five ppm is $1.50/month.
- —Power supply is estimated at $12/month.
- —Water supply is estimated at $10/month.
- —Labour 10 hours/month at $20/hour.
- —1–3 mm size juveniles required one month to produce and 6–10 mm size juveniles required six months to produce.

Based on the calculations given in Table 4, depending on the size of the juvenile, the estimated production cost may be as low as 0.7 cents for 1–3 mm size individuals, increasing to 29–71 cents/juvenile for 6–10 mm size class juveniles.

Acknowledgments

I wish to thank the technical staff, Kathy Kellam and Nick Ryan, for help in setting up the hatchery tanks. Kathy Kellam was also actively involved in looking after the juveniles in the hatchery complex. Many NTU students were involved in helping with hatchery work and setting up recirculating hatchery systems. Their efforts are appreciated. The help of the Power and Water Authority, NT in analysing the bore water is gratefully acknowledged. This work was funded by the Targeted Institutional Links Program, Commonwealth Department of Employment, Education and Training.

Table 4. Estimated production costs for 1–3 mm and 6–10 mm size-class juvenile at different densities.

Descriptions	Production cost ($) (1–3 mm size class)		Production cost ($) (6–10 mm size class)	
Items	10 000/tank	50 000/tank	2000/tank	5000/tank
Fibreglass tanks	83.30	83.30	250.00	250.00
Glass tanks	10.00	10.00	10.00	10.00
Fertiliser	1.50	1.50	7.50	7.50
Labour	200.00	200.00	1000.00	1000.00
Power	12.00	12.00	60.00	60.00
Water	15.00	15.00	75.00	75.00
Miscellaneous items	10.00	10.00	25.00	25.00
Total:	$331.80	$331.80	$1427.50	$1427.50
Cost/juvenile	$0.033	$0.007	$0.71	$0.29

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A Hatchery for the Topshell (*T. niloticus*) in Eastern Indonesia

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Abstract

The topshell (*Trochus niloticus* L.) is one of the most valuable marine resources in Indonesia. Due to its fishing pressure, trochus is categorised as a threatened species. In order to enhance its population, seed production of this topshell was initiated in eastern Indonesia in 1994.

Seven batches of trochus have been produced artificially. Strong aeration followed by UV-treated seawater is shown effective to induce the broodstock to spawn. The larvae and juveniles are fed mainly on a cultured sessile diatom, *Navicula* spp. Growth rate is relatively high but the survival rate is low.

In spite of good growth of the seeds, improvements in the rearing techniques of this species are needed. The result of the seed production techniques is presented and discussed.

TOPSHELL (*Trochus niloticus*) is an important marine resource in eastern Indonesian waters because of its valuable mother-of-pearl shell. During the last two decades, fishing was very intensive and endangered its natural population (Arafin 1993). This situation has occurred in all Indonesian waters. Accordingly, the Ministry of Forestry declared a ministerial decree No. 12/KPTS–II/1987 which prohibits any form of exploitation and trade of the topshell.

In spite of the decree, illegal fishing still occurs, especially in regions where the traditional regulation 'sasi' was not strictly applied. The catches are marketed under 'miscellaneous mother-of-pearl shells'.

The need of fishermen of a valuable marine resource and of government policy to protect this species has created a conflict of interest. To solve this conflict, the Directorate General Forest Protection and Nature Conservation (PHPA) – Ministry of Forestry decreed No. 07/KPTS/DJ-VI/1988 mentioning that protected species may be exploited and traded if they are produced from culture. Correspondingly, research institutions and private enterprises are encouraged to initiate the culture of protected species.

Division of Marine Resources, R&D Center for Oceanology, Indonesian Institute of Sciences has been granted a three-year research project on the culture of the topshell (*Trochus niloticus* L.). The research was initiated in April 1994 and aims to find and promote seed production techniques.

Work on seed production has been reported previously (Heslinga 1980, 1981; Heslinga and Hillmann 1981; Heslinga et al. 1983; Nash 1985, 1988, 1993; Amos 1991; Kikutani and Patris 1991). This paper describes results in the hatchery of *T. niloticus* L. in Ambon (Eastern Indonesia).

Materials and Methods

Marine culture facility

Division of Marine Resources, R&D Center for Oceanology is situated in Inner Ambon Bay (3°34′S, 126°36′E), Ambon Island, eastern Indonesia. The Experimental Mariculture Laboratory consists of a

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seawater pump, three successive sand filter tanks and two reservoir tanks (35 m^3 and 50 m^3). Due to limited depth, the pump can be operated only during high tides. The culture tanks consist of six 2.25 m^3 rectangular fibreglass tanks, five 3 m^3 cylindrical fibreglass tanks, five 3 m^3 cylindro-conical fibreglass tanks and two concrete rectangular tanks of 5 m^3 and 10 m^3. Annual variation of seawater salinity ranged 30–34 ppt, while water temperature in tanks ranged 28–30°C.

Broodstock collection

The broodstocks were collected by skindiving or scubadiving from various locations where the natural population is abundant: Kei Islands, Rhun and Hatta islands (Banda islands), Nolloth and Sirisori Islam (Saparua Island). The minimal basal shell diameter of the broodstock is 60 mm. The broodstock were tagged with numbered PVC rings and released on reef flats in Sirisori Islam, Morella and Liliboi (Ambon Island). These seashores are the natural habitat of the topshell. A total of 979 individuals was tagged and used for spawning experiments.

Induced spawning techniques

In each spawning experiment, the broodstock were brought into the laboratory. They were cleaned of epiphytic biota by brushing the shells vigorously. Induced spawning was conducted by putting the animals in small plastic jars containing filtered UV-treated seawater which was aerated strongly for 6–8 hours. In the evenings, the broodstock were moved into 2.25 m^3 fibreglass tanks containing filtered UV-treated seawater. The animals were left without aeration in the tank for spawning in darkness. If spawning was not obtained, the aeration treatment was repeated the following day or two days later.

Larval and juvenile rearing

Spawned eggs were left for at least 10 minutes in the spawning tank to permit fertilisation to take place. The eggs were then collected in a sieve of 60 μm mesh size, then rinsed by flowing filtered seawater. During the collection and rinsing the eggs, the bottom of the sieve was kept in a shallow plastic container to prevent physical damage to the eggs. The eggs were then moved into rearing tanks containing cultured sessile diatom prepared previously or hatching tanks filled with filtered seawater. In the latter, swimming pediveliger were usually found 40–48 hours after fertilisation. To induce the settlement of these pediveligers, they were moved to another tank containing sessile diatoms. The number of eggs was about 200 000 eggs/m^3 seawater.

Depending on the number of tanks available, the stocking density in larval rearing might vary from 25 000 to 100 000 pediveligers/ m^3 seawater. The density was lowered if food was lacking. Since sessile diatom medium was used in the trochus culture, replacement of seawater was done only when the water quality was very poor, otherwise siphoning was performed regularly. The duration of rearing was 5–9 months, depending on the availability of ocean nursery facilities.

Food supply

Sessile diatom of the genus *Navicular* was grown in culture medium prepared by mixing the following chemicals in 1 ton of filtered seawater: 100 g ammonium chloride, 90 mL sodium silicate, 15 g sodium phosphate, 15 g calcium carbonate and 1 L of metal mixture. The mixture consisted of 1.93 g iron chloride, 3.88 g manganese chloride, 3.16 g ammonium molybdate, 0.83 g zinc chloride, 0.09 g cobalt chloride, 12.35 g boric acid and 27.74 g ethylene diamine tetra-acetic acid (EDTA) dissolved in 1 L distilled water. Semi-pure culture of *Navicula* spp. was then added to the culture medium. The sessile diatoms are dense enough to be used 7–10 days later. During the rainy season from April to September, diatom growth was promoted by putting two 40W white tubular lamps about 20 cm above the water surface in the tank.

This method was used mainly during the first weeks of larval and juvenile rearing. When the juveniles attained approximately 5 mm, the tanks were equipped with coral rubble to increase surface area and provide shelter for the juveniles.

Ocean nursery

Ocean nursery experiment of trochus juveniles was conducted in cages 2 m × 2 m × 0.2 m and several 10 m × 1 m × 0.2 m (L × W × H) built on reef flat at Morella, Ambon Island. The cages were made of fishing net of 2 mm mesh. Predators were excluded by covering the cages with similar net and cementing the lower part of the sides to the bottom. Twice a week the fishing net was cleaned using a plastic brush. This method proved adequate during the calm season (eastern monsoon). In the rough season (western monsoon) which occurs September–October to February–March, wave action tore off the netting from the bottom. Improvement was made using concrete bricks of similar size for the cage wall. To increase the surface area, some hollow concrete bricks were put in the cage.

Juveniles measuring 8–15 mm were released in these cages for several months. The stocking density was 50 individuals/m^2.

Results

Spawning

Spawning usually occurred at night from 2100–2400 hours, most in the fourth quarter of the lunar cycle (5–7 days before new moon).

A total of 10 synchronous spawnings regrouped in seven different batches (Tn001–Tn007) was obtained from 31 August 1994 to 21 December 1995 (Table 1). High spawning rates occurred mainly in September 1994 and January, March and December 1995 (35%, 38%, 44% and 46% respectively). Female individuals showed high spawning in March and December (11% and 17%), while in males it occurred in March 1995 (33%). In September 1994, January and December 1995 the spawning proportion of males was similar, i.e. 29% of total broodstock induced.

Hatching rate

The hatching rates were recorded for three batches (Table 2), and ranged 57.25%–73.47%. For Tn006, the average number of eggs spawned by female individuals was higher but hatching rate was low compared to other batches. Spawning experiment data revealed that these female individuals spawned 10 days after the beginning of induced spawning. The frequent treatment to induce spawning may cause exhaustion of the animals which consequently spawned poor eggs.

On the other hand, Tn002 and Tn007 were spawned on the first and second days of the spawning experiment. These fit females produced good quality eggs with high hatching rates.

Table 2. Hatching rate of topshell (*T. niloticus*) obtained for three different spawning batches.

Batch code	Tn002	Tn006	Tn007
No. of females spawned	5.00	4.00	12.00
No. of eggs	400 000.00	1 616 000.00	2 149 000.00
Hatching rate (%)	73.47	57.25	70.23

Growth rate

The growth obtained for different batches during larval and juvenile rearing is presented in Table 3, which shows that growth rate is not consistent from one batch to another.

For the first three months (12 weeks), Tn002 attained the highest growth rate compared to other batches, but decreased afterwards. Oppositely, Tn006 showed slow growth during the first months and reached the greatest size (13.03 ± 2.27 mm) at 6 months old. This inconsistent growth rate (and survival rate) was due mainly to the shortage of food supply observed in the rearing tanks. In fact, 20 000 juveniles of 4–7 mm can graze out the cultured sessile diatom on the surface of the tank (6.5 m^2) within a week or so.

Table 1. Result of spawning experiment on the topshell (*T. niloticus* L.) at Experimental Mariculture Laboratory, R&D Center for Oceanology, Ambon.

No.	Batch code	No. of broodstock	Date of spawning	Spawning		
				Male	Female	Total
1	Tn001	67	31/08/1994	13	1	14
2	Tn001	55	05/09/1994	16	3	19
3	Tn002	58	31/01/1995	17	5	22
4	Tn003	99	22/03/1995	33	11	44
5	Tn003	148	28/03/1995	4	3	7
6	Tn004	101	03/04/1995	2	1	3
7	Tn004	101	04/04/1995	2	2	4
8	Tn005	155	30/04/1995	5	3	8
9	Tn006	55	28/10/1995	6	4	10
10	Tn007	69	21/12/1995	20	12	32

Table 3. Growth of basal diameter of *T. niloticus* obtained for different batches during larval and juvenile rearing phase.

Age (weeks)	Batch code			
	Tn001	Tn002	Tn006	Tn007
0	0.28 (0.02)	0.24 (0.01)	–	–
2	0.33 (0.02)	0.52 (0.08)	0.38 (0.08)	0.31 (0.06)
4	0.78 (0.07)	1.32 (0.10)	1.05 (0.12)	0.79 (0.09)
6	–	2.08 (0.25)	1.23 (0.21)	1.10 (0.14)
8	1.09 (0.11)	2.90 (0.47)	1.75 (0.21)	1.52 (0.23)
10	–	4.00 (0.64)	1.96 (0.31)	1.81 (0.29)
12	5.07 (0.73)	5.94 (0.96)	2.40 (0.87)	2.26 (0.56)
14	–	6.16 (0.99)	4.21 (1.09)	2.82 (0.72)
16	7.06 (0.83)	6.36 (1.05)	6.43 (1.22)	4.94 (1.40)
18	6.67 (0.68)	7.68 (1.16)	8.56 (1.80)	6.21 (1.14)
20	–	7.19 (1.26)	10.24 (2.67)	7.13 (1.41)
22	–	8.11 (1.54)	12.51 (2.78)	
24	8.27 (1.22)	11.27 (1.72)	13.03 (2.27)	
26	–	12.66 (2.13)		
27	12.57 (1.63)	–		
28	–	13.53 (2.53)		
31	20.35 (2.90)			
36	24.55 (4.54)			

Value in parenthesis is standard deviation from measurement of 100 individuals sampled randomly.

When food shortage occurred, the juveniles began to creep the wall of the tank beyond the water level. These juveniles were not capable of returning to the water and tended to die the following day. This phenomenon was observed more frequently in tanks containing bigger juveniles.

Ocean nursery

Data from the growth study in the ocean nursery are not yet available. However, the growth of tagged individuals grown under laboratory conditions and those held in cages is presented in Table 4, which shows that the growth of cage individuals is very high (20.73 mm diameter and 17.53 mm height) in comparison to those grown under laboratory conditions (8.63 mm and 5.24 mm respectively). Furthermore, the lower standard deviation of individuals from laboratory tanks may show limitation of food in the laboratory so that individuals were more homogenous.

Discussion

In order for successful activity in aquaculture, there is basic information to be studied, including the availability of broodstock and mature eggs. Due to intensive fishing during past decades, the broodstock of trochus was found only in some regions in eastern Indonesia (Arafin 1993).

Table 4. Growth of young trochus (*T. niloticus* L.) grown for 37 weeks under laboratory conditions and in ocean nursery cages.

Rearing type	Laboratory tank	Ocean nursery cage
Density (individuals/m^2)	Unknown	48
No. individuals	36	34
Initial diameter (mm)	18.77 (2.48)	23.17 (2.41)
Initial height (mm)	14.86 (1.49)	17.69 (1.85)
Final diameter (mm)	27.40 (2.16)	43.91 (8.85)
Final height (mm)	20.10 (1.80)	35.22 (9.84)
Growth		
Diameter (mm)	8.63	20.73
Height (mm)	5.24	17.53

Values in brackets are the standard deviation.

In eastern Indonesia a great number of eggs was obtained in January, March, September and December. Spawning could also be obtained in April, August and September, but the quality of the eggs was not as good as during the main spawning season. In Okinawa (26.5° N, 128° E), the spawning of trochus was possible only from late May to mid-October (Murakoshi 1991). In New Caledonia, the spawning of trochus occurred from December to April which coincided with the warmest months (Anon. 1989).

The next important aspect is information on induced spawning and rearing techniques. The present study shows that strong aeration followed by the use of UV-treated seawater is effective to induce spawning of the topshell *T. niloticus* L. Induced spawning was conducted successfully 5–7 days before new moon.

Larval and juvenile rearing techniques were shown to be applicable. Meanwhile, there are some inconsistencies in hatching and survival rates, and growth rates among batches. Hatching rate in the trochus hatchery may be related to the improper timing and technique of induced spawning which resulted in poor egg quality. Differences in survival and growth rates are due mainly to food shortage. In order to increase the production of natural food, corrugated PVC sheets can be used in the rearing tanks. In Okinawa, a 2.75 m^3 tank can produce about 48 600 seeds of 3–5 mm (35.4–45.5% survival rate) in 3–3.5 months (Murakoshi 1991). Limited food may cause slower growth and a more homogenous size of juveniles reared.

This main problem of food supply in the hatchery is due partly to the lack of appropriate facilities. When juveniles grow bigger, the feeding rate increases accordingly and food shortage invariably occurs. An ocean nursery seems to be a suitable solution. In Japan, juveniles were transferred into concrete tanks built in the reef flat as soon as they reached 3–5 mm. Since no report is available of this culture stage, it is difficult to assess the success of this method, but it seems that these young juveniles are still too vulnerable.

In the present study, low concrete cages were used to rear juveniles of 23 mm basal diameter. An ocean nursery study using juveniles of different size classes is underway. The preliminary result shows that the size of juveniles to be reared in this system could be as small as 10 mm, a size reached by 20–24-week-old juveniles. Since aquaculture activity is expensive, this fact is useful because it can be used to reduce operating costs. Furthermore, by moving the juveniles into cages on the reef flat, the hatchery facility could be used for another batch.

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Current Status of Topshell *Trochus niloticus* Hatcheries in Australia, Indonesia and the Pacific — A Review

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Abstract

Five institutional and one private hatcheries for trochus, *Trochus niloticus,* are currently in operation. The research hatcheries are at the Northern Territory University (NTU), Darwin, Australia; the Indonesian Institute of Science (LIPI), Ambon, Eastern Indonesia; Barrany Lompo Island belonging to the University Hasanuddin in Ujong Pandang, Eastern Indonesia; the Fisheries Department, Port Vila, Republic of Vanuatu; and Sopu field station at Tonga. The sole private hatchery is on Seram Island, Eastern Indonesia. The oldest hatchery currently in operation is at Port Vila, in operation since the late 1980s. The other hatcheries were established in the early to mid-1990s. The hatchery at NTU is unique and uses saline bore water for its water supply; all other hatcheries pump seawater directly from the sea. It is also the only hatchery currently in operation using a closed recirculating hatchery system. The other hatcheries use flow-through or semi-flow-through systems for producing juveniles. All the hatcheries have successfully produced juveniles in the last few years. It is encouraging that a private trochus hatchery is currently in operation in Indonesia. It occupies a floor space of 600 m^2 and is the largest trochus hatchery currently in operation in the world.

SINCE the first successful spawning and juvenile production of trochus *T. niloticus* was reported in Pulau by Heslinga (1980) and Heslinga and Hillmann (1981), considerable interest has been generated in the possibilities of (a) aquaculture of the topshell and (b) using hatchery-produced juveniles to reseed depleted reefs or translocate juveniles to virgin reefs in other Pacific countries. Trochus mariculture is ideally suited to many countries in the Indo-Pacific region. It is a very 'forgiving' species with a relatively simple lifecycle. Other attributes that make it such a suitable species for mariculture are:

- broodstock are readily collected and maintained in the hatchery;
- trochus are fecund and spawn the year round;
- the eggs are lecithotropic and hatch into planktonic larvae that do not need to be fed during the larval phase;
- the larval cycle is short and metamorphosis and settlement as benthic postlarvae (P/L) are completed within four to five days;
- once settled, the P/L feed readily on benthic diatoms and other associated benthic algae and microbenthic invertebrates growing on the substrates;
- no known diseases of juvenile and adult trochus have been reported;
- hatchery technology for producing juveniles is simple and improved methods of inducing the trochus to spawn are continually being developed;
- trochus are hardy and are readily transported during all stages of the lifecycle.

Consequently, beginning in the mid-1980s and early 1990s, many countries in the Pacific were interested in establishing hatcheries to produce trochus juveniles for culture, or for reseeding to enhance catchable wild fisheries, or to establish new fisheries.

This paper reports on the early attempts to establish hatcheries in some Pacific countries and on those currently in operation in Australia, Indonesia and the Pacific.

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Development of Trochus Hatcheries in the Tropical Indo-Pacific

The Pacific

Not all countries in the Pacific were interested in or have the technical and financial capabilities to develop trochus hatcheries and produce juveniles for reseeding as a management option for fisheries.

Palau

The first successful trochus hatchery in the Pacific was established in the Micronesian Mariculture Demonstration Center (MMDC) on Palau (Heslinga 1980). Using a flow-through system in 5000 L circular concrete tanks, several hundred thousand small juveniles were successfully produced. Since the early production, the MMDC hatchery has been upgraded using equipment from the Japanese International Cooperation Agency (JICA) and FAO–SPADP projects (Kikutani and Patris, 1991). However, recently the hatchery work has slowly wound down due to lack of operational funds (Kikutani 1991).

Fiji

There was no dedicated trochus hatchery in Fiji. However, the Makogai Mariculture Station in Fiji carried out two spawning of trochus in 1989 within its hatchery complex. Its purpose was to produce enough trochus juveniles to graze and clean the giant clam tanks within the hatchery complex. Since that time no serious attempts have been made to establish a trochus hatchery in the station or elsewhere in the country.

Western Samoa

Trochus does not occur naturally in Western Samoa. In 1991, a shipment of adults was translocated from Fiji to Western Samoa (Henrietta 1991) and some spawned while being held in the Fisheries Department aquaculture ponds. Unfortunately no juveniles were produced and no further attempt to develop a trochus hatchery was made.

Tonga

Trochus does not occur naturally in Tonga. A trochus hatchery was set up at Sopu by JICA as part of Japanese Government aid to the Ministry of Fisheries hatchery in Sopu, Tonga. No further details on the hatchery are available but Kikutani (pers. comm.) has indicated that the hatchery is running well in conjunction with the green snail *Turbo marmoratus*.

Republic of Vanuatu

Vanuatu is the only country in the Pacific to operate a trochus hatchery continuously and successfully since the latter part of the 1980s. The hatchery was provided by the South Pacific Aquaculture Development Project of FAO (FAO–SPADP) and was upgraded in 1989 and 1992. The hatchery currently houses two 5000 L rectangular concrete raceways, one 4000 L fibreglass tank, two 1000 L, two 500 L and three 100 L polycarbonate tanks. A new extension, partly funded by the Australian Centre for International Agriculture Research (ACIAR) trochus reseeding research project implemented in 1995, covering a floor area of about 100 m^2 is under construction. The hatchery extension is due for completion in December 1996.

The hatchery in Vanuatu is supplied with seawater pumped directly from the harbour of Port Vila into the hatchery tanks. Where required, the water may be filtered through a 1–5 μm filter bag which is tied to the mouth of the tap with a strong rubber band. Spawnings are usually carried out in the 100 L polycarbonate tanks although 500 L tanks are used when more broodstock are available for spawning.

Spawnings are usually carried out during new or full moon. Selected spawners are crowded in the spawning tank and during the day at least one change of water is carried out. Soon after sunset, the water is changed again and spawning normally occurs soon after. Under the ACIAR reseeding project, spawning has been improved and made more predictable by using heaters to elevate the water temperature of the spawning tank. A 1–3°C increase in water temperature is usually adequate to trigger the spawning.

After spawning, the fertilised eggs are rinsed in clean filtered seawater and released into the 5000 L concrete tanks for hatching and larval growth. During this period, gentle aeration is provided and no water change is needed. After 4–6 days, the larvae metamorphose into postlarvae and settle on the concrete walls and substrates left in the tank. A week later, water change commences.

In January 1996, three successful spawnings were carried out and a total production of over two million eggs was obtained. The juveniles from this spawning will be used for the ACIAR-funded Trochus Reseeding Research Project involving Australia, Indonesia and Vanuatu.

Federated states of Micronesia

There is no dedicated trochus hatchery but some juvenile production took place under the direction of Stephen Lindsay at the Kosrae hatchery in 1993–94. The trochus juveniles were used in polyculture with giant clam juveniles.

Australia

Queensland Fisheries hatchery

The first trochus 'hatchery' in Australia was established 1982–1984 (Nash 1985) at the Queensland Fisheries Department field station on Green Island. The tanks in the field station were set up for holding fish but were successfully converted on an ad hoc basis for rearing trochus juveniles. The trochus were allowed to spawn in glass aquarium tanks supplied with a continuous flow of seawater. After spawning, the fertilised eggs were transferred to either 100 L glass tanks or to 4700 L fibreglass tanks for ongrowing without special care being provided. Sufficient juveniles were produced using this system for growth rate study. After 1984, the system ceased to be used for trochus work and reverted to its original function.

Orpheus Island hatchery, Queensland

The second trochus hatchery in Australia was at the field station on Orpheus Island and was run by the James Cook University. It was set up in 1988–89 under the ACIAR–JCU giant clam project. Trochus juveniles were produced for stocking in polyculture with giant clams. The hatchery was discontinued at the end of the clam project but revitalised for spawning trochus in 1992–93 as a JCU PhD student research project (Laura Castell, pers. comm.). The research station has an aquarium system consisting of glass tanks and large 2500-L fibreglass tanks. Some of these tanks were used for spawning trochus and the 2500 L tanks were used for rearing juveniles on a continuous-flow basis. Over two years some 5000–6000 juveniles were produced for research. As for the Green Island trochus facility, the trochus hatchery function was abandoned when the PhD study was completed in 1995.

Northern Territory University hatchery

The only dedicated and specialised trochus hatchery currently in operation in Australia was developed and established in 1993 at the Northern Territory University (NTU), Darwin under a research grant provided by the Department of Employment, Education and Training (DEET) under its Targeted Institutional Links (TIL) Program. The hatchery is land-based and obtains its seawater supply from a saline bore. An efficient and simple closed recirculating system for rearing trochus juveniles was developed; a full description of the hatchery, its management and running is provided by Lee (these Proceedings). Currently, the NTU hatchery consists of ten 250 L glass tanks, seven 3000 L circular fibreglass tanks and ten 6000 L rectangular fibreglass tanks. It is the most functional and efficient state-of-the-art trochus hatchery in operation in any part of the world.

The NTU hatchery also pioneered and simplified the spawning of trochus by using aquarium heaters to elevate the water temperature by 1–3°C to induce the trochus to spawn. Elevation of water temperature coupled with 2–3 changes of water is sufficient to bring about spawning regardless of the phase of the moon or the time of the year. In addition to spawning, the postlarvae (P/L) rearing method has also been improved at NTU. Until recently, all trochus hatcheries in the Indo-Pacific and Japan used fibreglass plates to provide additional surface area for food production, i.e. benthic diatoms. It was discovered at NTU that this was undesirable as P/L and juveniles grown on fibreglass plates do not recognise coral rubble as their natural habitat when released. Consequently, a new protocol for juvenile production was put in place at NTU. This involved setting up a system where coral pieces and rubbles were introduced into the larval tank, fertilised and reseeded with benthic diatoms such as *Nitschia* sp. Within 7–10 days, the corals were colonised by the benthic diatoms, and P/L released into the tank grew rapidly. For more details on the NTU hatchery, see Lee (these Proceedings).

During its three years, the NTU hatchery produced a few hundred thousand juveniles varying 1–25 mm in size (basal diameter) for growth rate and nutrition studies. In early 1996, the F_1 that were spawned in the NTU hatchery in late 1993 were successfully induced to reproduce, thereby closing its lifecycle in a land-based hatchery using saline bore water. The NTU trochus project was boosted by ACIAR in July 1995 when it agreed to expand the DEET project into a further phase on reef reseeding research involving Australia, Indonesia and Vanuatu. Consequently, the NTU trochus hatchery has been expanded and will be in full operation until 1998. Research will continue to improve the hatchery system between now and 1998. Long before the end of this period, it is hoped that Aboriginal interest groups will set up their own hatcheries in King Sound, Western Australia. NTU has agreed to transfer the hatchery and grow-out technologies to the Aboriginal communities in King Sound.

Indonesia

Ambon hatchery

The first research hatchery for trochus in the country was established in 1994 by the Indonesian Institute of Sciences (LIPI) at its regional station in Ambon, Eastern Indonesia. The hatchery consists of four 3000 L rectangular fibreglass tanks, an irregular-shaped concrete tank of about 6000 L capacity and some smaller fibreglass tanks. Recently, another large concrete tank of 24 000 L was added. The method of spawning trochus in LIPI was based on the method described by Kikutani and Patris (1991). Since commencement, the hatchery has produced many thousand juveniles for reseeding research and related studies. LIPI is now linked to the ACIAR-funded trochus reef reseeding research and its hatchery operation will be supported by the project till 1998.

Ujong Pandang hatchery

The second research hatchery for trochus in Indonesia was set up recently by the University Hasanuddin (UNHAS) at its field station on Barrany Lompo Island (BLI) which is situated about 10 km off the coastal city of Ujong Pandang in South Sulawesi. The field station of UNHAS was built in 1992–93 under the Marine Science Education Project (MSEP) funded by the Asian Development Bank.

The MSEP field station on BLI is supplied with seawater via a twin 90 mm high density polyethylene pipe with the intakes sited some 100 m offshore from the station. After filtration, the seawater is pumped to a reservoir which supplies all the tanks within the field station complex. In 1994, part of the field station facility was adopted for spawning trochus.

The facility allocated for trochus consists of a 9000 L rectangular broodstock-holding tank (7.5 m × 1.5 m × 0.8 m high) and a large rectangular larval rearing tank of 12 500 L (7.8 m × 1.8 m × 0.9 m high). The water in the trochus tanks was changed continuously and estimated to be at a daily rate of about 80%. Trochus were successfully spawned on 10 January 1996 and some 80 000 small (1–3 mm) juveniles are now growing in the hatchery using the continuous seawater flow-through system. To encourage algae growth and increase food production for the young P/L, fertilisers in the form of urea and ammonium sulfate are also added regularly to the rearing tank.

Seram Island hatchery

The first private purpose-built commercial trochus hatchery in the world was established in early 1996 at Pohon Batu on the island of Seram, Eastern Indonesia. Details of the hatchery are summarised below.

Size of hatchery building	12 m wide × 50 m long
Water source	Intake pipe of 90 mm OD black polyethylene pipe placed some 6 m below sea level with a sub-sand slotted intake end
Water supply	Seawater pumped and stored in a 50-ton concrete reservoir tank; reservoir supplies seawater to the hatchery by gravity
Laboratory	A small algae and preparation room measuring 5 m × 8 m is situated at one end of the hatchery
Hatchery	10 × 1000 L circular fibreglass spawning tanks, 12 × 3400 L larvae rearing tanks and 10 × 15 000 L juvenile tanks
Hatchery system	Flow-through system
Juvenile production	One million 5–8-month-old juveniles annually (8–10 spawnings/year)
Grow-out system	A combination of intertidal pond, hanging baskets and ocean nursery
Production target	100–300 tonnes/year

The company has been very progressive in its planning and development by employing suitably qualified consultants and seeking advice from research scientists involved in this field.

The commercial hatchery was completed in April 1996 and the first spawning achieved in the same month.

Discussion and Conclusions

Currently seven trochus hatcheries are in operation in the Indo-Pacific region. There is one in Australia, three in Indonesia and three in the Pacific.

The purpose-built trochus hatchery in Australia is based in the Aquaculture complex of NTU. This state-of-the-art hatchery has been in operation since 1993. The hatchery system used for producing trochus juveniles is highly efficient in water utilisation and labour input. Estimated production cost for 1–3 mm size class juveniles varies from <1.0 to 3.3 cents/juvenile. By operating the hatchery tanks on constant recirculating mode, a minimal amount of water is used for each hatchery cycle. In addition, the use of saline bore water to supply the hatchery eliminates the need for filtration of incoming water; the hatchery is also free of fouling organisms and other water quality problems associated with using raw seawater supply. The hatchery has improved the method of (a) spawning by using heaters and (b) production of juveniles by using coral rubble inoculated with benthic diatoms to enhance food production in the hatchery tanks.

The three Indonesian hatcheries include a newly established private hatchery. It is pleasing to see that private sector investors in aquaculture have taken up the challenge of establishing a commercial hatchery at Seram Island, Indonesia. Based on information available to the authors, the newly established private hatchery is well-planned, guided by qualified staff and with a high chance of achieving its set target of producing a million 5–8-month-old juveniles annually to reseed the reefs under the company's control. The other two government-funded hatcheries produce juveniles for research only.

There are three trochus hatcheries in the Pacific and among them, the hatchery at Port Vila, Vanuatu is perhaps the most active and is undergoing expansion in 1996. The Port Vila hatchery will continue to produce juveniles for the ACIAR-funded reseeding research project; surplus juveniles from the hatchery will be released in selected reefs scattered across the country.

Acknowledgments

We wish to thank R. Braley for supplying the latest information on the trochus hatcheries on Seram Island and on Barrany Lompo Island, Ujong Pandang. Ms Laura Castell provided information on the hatchery on Orpheus Island.

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A Simplified Method of Transporting *Trochus niloticus* Broodstock over Long Distances for Spawning

C. L. Lee* and C. Ostle†

Abstract

Mature trochus broodstocks 65–120 mm in size were successfully transported over long distances from King Sound, Broome, Western Australia to the trochus hatchery at the Northern Territory University (NTU), Darwin, Northern Territory. The transport involved travelling 250 km of mostly unsealed road, overnight in Broome and jet the following morning to NTU, about 1000 km east of Broome. Using the method recommended, survival rates on arrival at NTU were in excess of 95% and many broodstocks spawned naturally in the hatchery tanks at NTU. Induced spawnings on the translocated broodstocks were also successfully carried out at NTU.

IN July 1993, the Northern Territory University (NTU), Darwin and the University Nusa Cendana (UNDANA), Kupang, eastern Indonesia were funded by the Targeted Institutional Links (TIL) Program, Department of Employment, Education and Training (DEET) to carry out research work on the biology and hatchery requirements of the topshell *Trochus niloticus* (Mollusca: Gastropoda). Preliminary studies indicated that no trochus broodstock were found within easy access of NTU. The nearest trochus fishery was in King Sound, some 250 km by road, mostly dirt track, from Broome, Western Australia, about 1000 km by air west of Darwin.

Over the past 50 years, live trochus broodstocks have been extensively translocated across long distances over waters in the Pacific (Asano 1938; Van Pel 1957; Parkinson 1984; Sims 1985; Gillett 1986 1989; Izumi 1987). Numerous methods have been employed by these workers to transport their trochus, e.g. by airfreight in damp sacks placed in wooden crates, commonly referred to as the 'dry' method; in flooded skiff and boxes; in circulating tanks on ships; in plastic bags filled with water and oxygen. A sum mary of the results obtained by these authors is shown in Table 1.

The current study was initiated with the view to developing a consistent and efficient method of transporting trochus broodstocks over long distances by road and air to NTU, Darwin.

Materials and Methods

General procedures for transporting trochus from King Sound to NTU

Trochus broodstocks for the project were collected by the Aboriginal communities in King Sound and transported by road to Broome. On arrival in Broome, they were released into a large black plastic mesh cage, suspended in seawater and held overnight (usually). The broodstocks were repacked in oxygen-filled bags (without seawater) and placed in styrofoam boxes for transportation by jet to Darwin the following morning. On arrival at NTU, the broodstocks were released in tanks filled with fresh, clean seawater and used for spawning in the hatchery. Over the last two years, beginning in September 1993, 15 shipments

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involving 1001 broodstocks were transported to NTU with a high degree of success.

Collection and handling in King Sound

Trochus broodstocks were collected with the help of the Aboriginal communities from the intertidal areas of King Sound Peninsula or from the surrounding islands.

Aboriginal fishermen collecting broodstock were asked to follow a simple set of guidelines during collections to minimise stress on the animals:

- collect all the broodstock at one time;
- transfer the trochus to a damp and clean sack as soon as possible after collection;
- keep the trochus in the damp sack in the bottom of the boat; keep the bag shaded from direct sun, and away from dirty and oily water at the bottom of the boat;
- send trochus to the delivery point as soon as possible after collection.

On arrival at the collection point, the shells were kept either in damp sacks or in black mesh cages (BMC) with mesh size measuring 10 mm × 20 mm and held under water on a sand-free bottom for up to 24 hours before transport to Broome. Road transport by airconditioned 4-wheel-drive vehicle from King Sound to Broome took about 3 hours for a distance of 230 km. Most was over corrugated dirt track.

Three methods of treating and preparing the shells for transportation by road to Broome were evaluated. These are summarised below.

1. Adult trochus were removed from the water, drained and individually wrapped in dry newspaper. They were then neatly arranged in layers in a rectangular plastic basin or 'Nelly' bin measuring 60 cm long × 40 cm wide × 35 cm high; each basin may take up to three layers of trochus placed with the larger diameter of the shell facing downward, and up to 80 shells can be transported in one basin. Prior to placing the shells in the basin, a damp sack or layers of newspaper were placed on the bottom to absorb excess moisture and road vibrations. The basins were kept shaded from direct sunlight during the road journey to Broome.
2. Adult trochus were placed in the basin as in method 1 but without being wrapped individually. To reduce desiccation, after placing the final layer of shells they were covered with a damp sack or layers of damp newspaper. The basins were similarly kept shaded from direct sunlight during the road journey to Broome.

Table 1. Summary of translocation of trochus — methods and mortality.

Author	Transport conditions and duration	Mortality
McGowan 1958	Copra bags?; dry for up to 36 hours	Nil
Asano 1938	Circulating water tank on ship	
	2 days	Nil
	10 days	21%
Van Pel 1957	Wooden crates; dry transport	Some;
		No figures given
Sim 1985	Recirculating water tank on ship;	38%
	wet sacks	100%
Fagolimul and Price 1987	Recirculating water tank on ship	Varies 0–36%;
		Mean of 8%
Parkinson 1984	Recirculating water tank on ship; up to 5 days	22%
Gillett 1986	Flooded skiff and boxes with 6 hours dry time	2.2%
	Dry in trays; up to 133 hours out of water	100%
	Plastic bags with water/oxygen; 3 hours out of water	Nil
Gillett 1989	Dry in bags and crates with final release using parachutes; up to 48 hours out of water	Low mortality; no figures given

3. Adult trochus were removed from the water and transferred to damp sacks, each containing up to 50 shells. The sacks were placed in rectangular plastic basins as described previously, each basin holding up to two bags of shells. The basins were kept shaded from direct sunlight during the road journey to Broome.

Handling of shells in Broome

On arrival in Broome, due mainly to the difficulty in getting a same-day air-connection to Darwin, the shells were unpacked and transferred into a BMC, each carrying up to 160 animals. The BMC were left suspended in at least 2 m of water alongside a boat or the jetty in the Broome Harbour and held overnight.

The following morning the shells were removed from the BMC and transferred into double-layer plastic bags lined with newspaper. Depending on the size of the trochus, 20–40 shells with newspaper between them were put into each bag which was then charged with industrial oxygen. Two such bags could be transported in a foam box 60 cm long × 40 cm wide × 25 cm high.

Transport time and handling on arrival at NTU

The removal of the shells from Broome Harbour and packing and preparation for air transport from Broome to Darwin required up to 2.5 hours for about 100 shells. Flying time from Broome to Darwin is about two hours and up to 3.5 hours if the plane stops over in Derby on the way. On arrival, airport clearance may take up to an hour. The shells were therefore out of water for a maximum of about seven hours from Broome to the NTU hatchery.

On arrival at NTU, the shells were unpacked and placed into holding tanks for quarantine purposes. Healthy shells were selected and cleaned with a stiff brush to remove all encrusting fouling organisms before transfer to the spawning tanks. The spawners were closely observed overnight and all mortalities recorded.

Results and Recommendations

It was found that the preparation time for method 1 involving wrapping shells individually in dry newspaper for transport by road was too time-consuming and appears to produce no distinct advantage. Consequently, methods 2 and 3 were adopted for all subsequent road transport of broodstocks.

The results of trials using methods 2 and 3 are shown in Table 2. During the first eight shipments where the shells were wrapped individually, no mortality was recorded when the total exposure time to air was less than eight hours prior to transport by air to NTU. Similar results were obtained for unwrapped shells (shipments 9 to 15) when the total exposure time was less than eight hours. In contrast, when the exposure to air was more than 16 hours, high mortality was recorded. The total transport time, up to 96 hours, apparently has no effect on survival as long as the shells are not out of water for a continuous eight hours at any one time.

Table 2. Transport of trochus broodstock from King Sound to NTU.

Trip number	No. of animals	Out of water (hours)	Transport time (hours)	Transport method	Survival (%)
1	200	6	48	a,b	100
2	72	16	48	a,b	69.4
3	145	5	48	a,b	87.3*
4	18	5	72	a,b	100
5	24	5	96	a,b	100
6	22	16	24	a,b	86.4
7	41	24	24	a,b	73.2
8	90	5	21	c,d	100
9	27	4	48	c,d	100
10	80	5	28	c,d	100
11	46	5	48	c,d	95.6
12	52	5	24	c,d	100
13	94	4	48	c,d	100
14	70	4	48	c,d	100
15	59	4	48	c,d	86.4*

[a] Animals individually wrapped with paper for shipment from King Sound to Broome.

[b] Animals individually wrapped with paper and packed into oxygen-filled bags for shipment by air from Broome to NTU.

[c] Animals held in a group in a damped sugarbag for shipment from King Sound to Broome.

[d] Animals held in a group and packed into oxygen-filled bags for shipment by air from Broome to NTU.

*Some of the animals were collected at an air temperature of 40°C.

On arrival at NTU, many of the shells were successfully subjected to induced spawning experiments. Details of spawning, juvenile production and the postlarval rearing system used at NTU are given in Lee (these Proceedings).

Based on the results it would appear that the trochus broodstocks should not be out of water for more than eight hours during extended transport over long distances. If the transport time is longer, the broodstocks must be submerged in seawater for time to recover before being taken out of water for packing and subsequent shipment. It is therefore recommended that the following procedures be followed for shipping broodstocks from King Sound to NTU.

Road transport from King Sound to Broome

About 80 shells may be shipped in a rectangular plastic basin measuring 60 × 40 × 35 cm; three layers of shells may be placed in the basin with the larger diameter face down. Alternately, up to 40 shells may be placed into damp sacks; two bags may be placed into each basin.

Each basin must be lined with layers of newspaper or damp sack to reduce road vibrations; after packing, the top of the basin is similarly covered.

The shells must be released into seawater within eight hours of packing for road transport in King Sound.

Shipping from Broome to NTU

Depending on the size, between 40 and 80 shells can be shipped in a foam box measuring 60 cm long × 40 cm wide × 25 cm high. Each box contains two oxygen-filled plastic bags; depending on the size of the broodstocks, each bag can carry between 20 and 40 animals.

The plastic bags used for packing the shell must be at least 50 µm thick. A double-bag system with a layer of newspaper between the bags must be used to prevent the bags being punctured by the pointed apex of the trochus shells.

A layer of newspaper is placed on the bottom of the box prior to packing the bags.

The shells must be released into seawater at NTU within eight hours of packing and shipping from Broome.

Acknowledgments

We wish to thank the Aboriginal Communities in King Sound for their support in providing broodstocks for this work and Kathy Kellam for looking after the animals in the hatchery, NTU. This project is conducted with funds provided under the Targeted Institutional Links Program, Department of Employment, Education and Training, Canberra.

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Reproductive Biology of *Trochus niloticus* L. from Maluku, Eastern Indonesia

Pradina, S. A. P. Dwiono, P. E. Makatipu and Z. Arafin*

Abstract

Observation of *Trochus niloticus* was carried out in Maluku waters to obtain reproductive information. About 10–20 individuals were collected every full moon from December 1992 to December 1993 in Rhun Island (Banda islands), and about 15–25 individuals from Nolloth (Saparua Island) every three days between June and July 1993. Gonad index and oocytes diameter were analysed from three parts of each gonad.

The change of mean gonad index and oocytes diameter showed that the main spawning season of trochus in Rhun Island occurred from March to June. Those from Nolloth indicated that spawning period was around full moon. Some observations of the spawning of trochus in the laboratory are discussed here for valuable information.

Further histological examination of female gonads (ovaries) demonstrated some interesting phenomena. Gonad condition is more or less similar in every part of each ovary. Mature oocytes were released through the opening located at the periphery of the conical area, near the connective tissue separating the digestive gland and the ovary. Oocytes might be spawned even from an immature gonad, suggesting that trochus spawn partially.

Variation in gonad development among ovaries might indicate that female trochus spawn unsynchronously. Mature ovaries were found mostly in individuals collected from Rhun, while developing stages were dominant in trochus from Nolloth.

MUCH information on trochus has been published, including that in Pacific, Japanese and Australian waters. The animal became very popular as the fashion industry uses its beautiful shell for buttons. Its natural population required extra attention due to some reports on its depletion (McGowan 1957; Heslinga and Hillmann 1981; Wells 1981; Arafin 1993; Hahn 1988).

Some laboratories, namely Palau, Vanuatu and Okinawa, have been able to produce young trochus. Still, many reproductive phenomena of trochus have not yet been uncovered, such as how gametes are released, how long one reproductive cycle is, what the most important factors affecting the release of the gametes are and what factors cause the variation of spawning period among different populations.

Reports of reproductive studies are based mainly on biostatistics (morphometry) and gonad histology (Rao 1937; Hahn 1993) or through observation of spawning experiments in the laboratory (Nash 1985). Combining that information is useful to obtain more comprehensive information on the reproductive behaviour of trochus.

Maluku waters apply a customary law called 'sasi' (prohibition), which regulates opening and closing seasons of fishing, and minimum legal sizes of particular biota. This law was based mainly on socio-economics. Therefore a decline in trochus catch in some regions is still reported while the sasi is still strictly followed. This study might enforce traditional regulation by supplying reproductive biology information of the species.

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Material and Methods

In order to determine the spawning season, monthly samples of 10–20 trochus with basal shell diameter 60 mm or more (from a depth of 1–5 m) were collected from Rhun Island, the Banda Islands (Fig. 1). Sampling was conducted every full moon from December 1992 to December 1993.

Previous reports stated that the trochus reproductive cycle follows the lunar cycle, yet there is variation in the period in many waters. Therefore another study was made on trochus from Nolloth, Saparua Island. Samples of 15–25 individuals of 60 mm or more in basal shell diameter were collected every three days from June to July 1993.

Gonad index estimation

A conical area (consisting of gonad and digestive gland) of each individual was cut into three sections. All sections were preserved in Bouin solution for further analysis. Gonad area from three parts of each gonad was drawn on millimetre block paper using camera lucida. Gonad index (GI) was estimated following Komatsu's procedure (Komatsu 1992):

$$GI = \frac{\text{Gonad area}}{\text{Total conical area}} \times 100\%$$

The mean GI of both sexes was plotted against sampling date to estimate the change of the parameter.

Oocytes diameter analysis

Oocyte diameter was obtained from histological slides. Three parts of each gonad were sectioned as thick as 7 micron using a standard paraffin method and stained with Ehrlich's haematoxylin-eosin. A total number of 100 oocytes (jelly coat excluded) from each female gonad (ovary) was measured twice perpendicularly and only those cut through the nucleus were measured.

The mean oocytes diameter was plotted against sampling date to estimate the variation of this index. Gonad in spent condition was scored zero without considering the remaining oocytes.

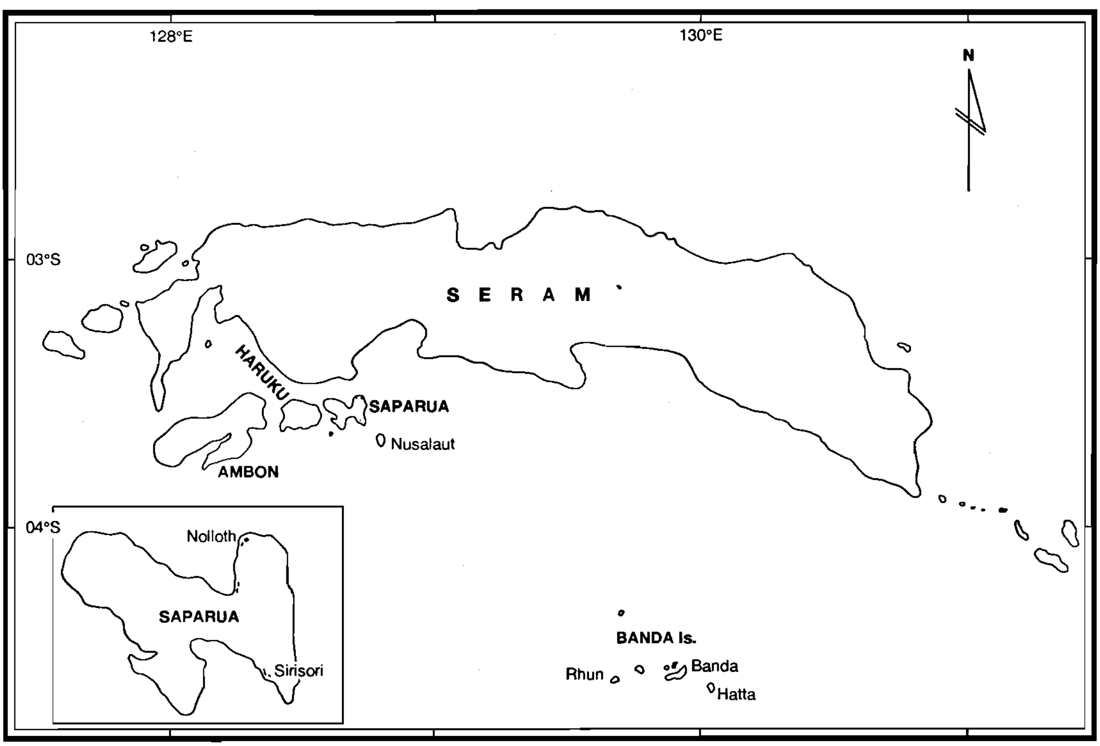

Figure 1. Map of sampling location of *T. niloticus* in Central Maluku, eastern Indonesia.

Gonad development

This study was done on the same histological slides used for oocytes diameter analysis. The stage of gonad development was determined for each ovary. The reproductive cycle was separated into five development stages (Pradina and Dwiono 1994):

a. proliferation: gonad has only small oocytes, from low density to high density;

b. early developed: oocytes increase in diameter and some have a 'jelly coat' (pitted membrane) around each oocyte (the jelly coat is an indicator of maturing oocytes as reported by previous authors); small or young oocytes are dominant;

c. late developed: this stage is marked by the increase in the number of oocytes with the jelly coat; gradation of oocyte size is obvious;

d. mature: mature oocytes dominate the ovary; at this stage, small oocytes are often found;

e. after-spawn stage: the gonad cavity becomes smaller, trabeculae indefinite with some young oocytes attached, occasionally some mature oocytes remain. This category includes spent and partial spawning. Partial spawning refers to individuals showing spawning signs such as well-determined 'flow' of mature oocytes into the opening and loose appearance caused by numerous empty spaces in the gonad.

Results

All individuals sampled possessed gonad tissue and sex could be recognised easily. A total of 292 individuals collected from Rhun Island consisted of 148 female (50.7%) and 144 male (49.3%), a ratio close to 1:1. The sample from Nolloth consisted of 107 female and 85 male individuals. This sample might not represent the actual population sex ratio since in each sampling trochus collection stopped when 10 female individuals were obtained.

The fluctuation of mean gonad index (MGI) from individuals collected from Rhun was similar for both sexes (Fig. 2). This value began to decrease in March, then declined sharply from April until June and started to increase slowly afterward. The highest value of MGI occurred in March (62.86% for male individuals and 72.40% for female), while the lowest value was found in June (22.91% male and 19.64% female).

The mean oocyte diameter (MOD) showed a slight drop in January, a decline from March to June and continued to decrease until August (Fig. 3). From September, the mean oocyte diameter fluctuated. The highest value of MOD occurred in February (118.11 µm), the lowest was in August (42.70 µm). When the MOD is compared to female MGI, some interesting phenomena can be pointed out: there was a time lag between the decrease of MOD and that of female MGI, and the decrease of oocyte size (MOD) was not always followed by a decline of MGI. The latter was found mainly between June and August when MGI reaches minimum values. Histological slides revealed that the gonads have empty spaces left by spawned oocytes or caused by reabsorption of some unspawned oocytes. Therefore the MGI was still high while MOD had already diminished.

The continuous decrease of both MOD and MGI from March to June indicate the main spawning season of *Trochus niloticus* in Rhun Island, while the slight decline in October, December and January could have been due to minor spawnings of the species.

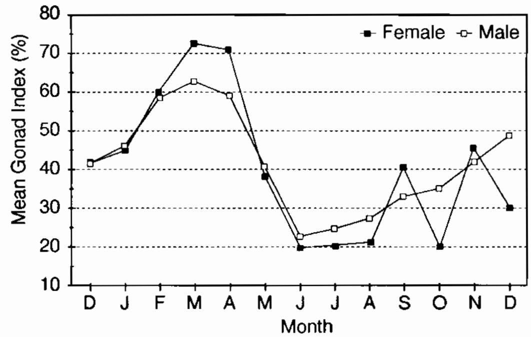

Figure 2. Female and male mean gonad index of *Trochus niloticus* L. sampled December 1992 to December 1993, Rhun Island (Banda Islands).

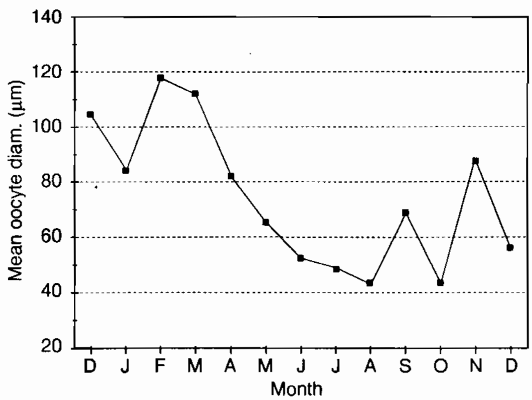

Figure 3. Mean oocyte diameter of *Trochus niloticus* L. sampled from December 1992 to December 1993 in Rhun Island (Banda Islands).

Individuals collected from Nolloth every three days showed a fluctuation in MGI for both sexes (Fig. 4). A sharp decline of MGI occurred between the 6th and 12th day of the lunar cycle in male and between the 9th and 12th day in female. The highest value of male MGI occurred on the 6th day (42.69%) while in females it was found on the 24th day of the lunar cycle (43.83%). The lowest male MGI was found on the 29th day of the lunar cycle and on the 12th day in the female. Mean oocyte diameter varied in a different pattern from the female MGI and showed a time lag (Fig. 5). The highest MOD occurred on the 9th day (83.37 µm), while the lowest was obtained on the 15th day (53.05 µm) of the lunar cycle. Both female MGI and MOD showed a sharp decrease between the 9th and 12th days of the lunar cycle. On day 12 of the lunar cycle, the female MGI started to increase, which may indicate the end of the spawning period. Mean oocyte diameter continued to decrease until the 15th day. Microscopic observation revealed that the gonads sampled on the 15th day of the lunar cycle were mostly in a proliferating stage with a relatively large gonad cavity. Unspawned oocytes were present and some of them were beginning to be reabsorbed.

A spent gonad was found on the 18th day of the lunar cycle. The gonad cavity was reduced, no mature oocytes remained, trabeculae were indefinite and very few small oocytes were present.

Some gonads showed a complex feature such as: having two groups of size without gradation; presence of spawning signs in developing gonad; presence of viable mature oocytes in proliferation stage gonad; and presence of necrotic (degenerating) oocytes in the mature gonad. The releasing signs of mature oocytes were observed near the extremity (periphery) of the connective tissue which separates the ovary from digestive glands; these mature oocytes flowed out through the opening formed by the connective tissue.

Discussion

The main spawning season, indicated by sharp decline of the MGI and MOD, occurred from March to June. These months coincided with the season change from west to east monsoon when the Banda Sea is cooler and has its highest waves. Fluctuation in female MGI and MOD between September and December suggests that minor spawnings occurred in that period. Unless individuals (mature female) could keep their mature oocytes until the main spawning season, the presence of mature gonad in trochus samples from Rhun suggests that minor spawning might occur throughout the year, as mentioned by previous workers.

Spawning experiments trialed from August 1994 to December 1995 showed that the spawning rate was higher when conducted from late December to May. The fact that no spawning was obtained from late May 1995 to August 1995 (Pradina et al. 1996; Anon. 1996) corroborated the MGI and MOD curves which reached minimum values between June and August.

An interesting phenomenon relating observations of gonads and oocytes with the spawning experiment emerged in March. In this month, while MGI and MOD reached maximum values, the spawning experiment resulted in 'epidemic spawning' where nearly half (48%) of the broodstock released its gametes.

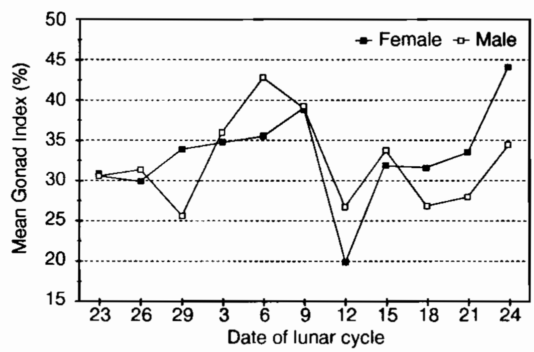

Figure 4. Female and male mean gonad index of *Trochus niloticus* L. sampled from June 14 to July 14 1993 (23rd day to the 24th day of following lunar cycle) in Nolloth, Saparua Island.

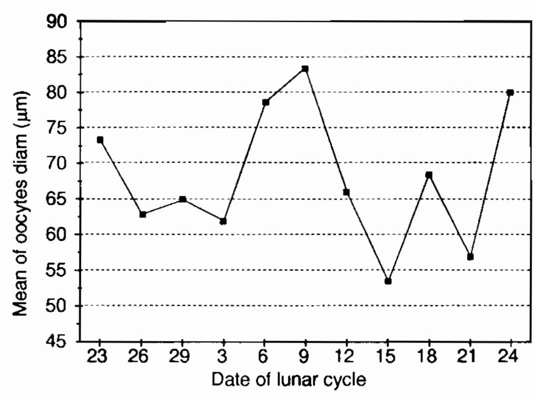

Figure 5. Mean oocyte diameter of *Trochus niloticus* L. sampled from June 14 to July 14 1993 (23rd day to 24th day of the following lunar cycle) in Nolloth, Saparua Island.

Gonad condition of trochus from Nolloth indicates that the main spawning period occurs between the 9th and 12th days of the lunar cycle. Unfortunately, only one spawning experiment was available on trochus from Nolloth to confirm this finding. Spawning in the laboratory occurred on the 24th day of the lunar cycle (31 August 1994).

The closest geographical distance from Nolloth where trochus can be found is Sirisori Islam (Saparua Island). On this population, some spawning experiments were made from January to December 1995 except in November where no spawning experiment was attempted, and spawnings were obtained from the third week of January until the beginning of May and from the third week of August to the end of October and in December. No spawning was obtained in June, July and August. It seems that the spawning season of trochus in Nolloth is similar to that of Rhun Island (Banda Islands). In these experiments, the spawning occurred mainly from six days before to four days after new moon (Pradina et al. 1996; Anon. 1996). Heslinga and Hillmann (1981) mentioned that the spawning of trochus in Palau occurs mostly a few days after new moon. Nash (1985) found that spawning of trochus generally occurred within 2–4 days of either new moon or full moon, while Hahn (1993) suggests that natural spawning of trochus occurs primarily at the new lunar phase (the day of the new moon and three following days).

If the spawning season of trochus in Nolloth is similar to that in Rhun Island and Sirisori Islam, it means that June is out of season. Therefore the data obtained for Nolloth to predict the spawning period are not valid, since the prediction will be biased. This indication is supported by histological examination which shows that most individuals from Nolloth are then in the developing stage.

The variability in oocyte diameter and development stage within a given gonad as observed on histological slides shows the complexity of the pattern of reproductive biology of the animal. It seems that the animal is capable of spawning whole mature oocytes completely or partially during a spawning period. Furthermore, the variation of gonad development among females caused unsynchronous spawning to occur frequently. Therefore it is difficult to estimate the duration of a reproductive cycle of trochus from the present study. For this species, a new method taking into consideration interindividual variation is needed. Hahn (1993), based on statistical methods suggested that there were eight different subpopulations of female, and he concluded that the spawning cycle of trochus is eight months. Nash (1985) found that the female reproductive cycle of trochus on the Great Barrier Reef ranged 2–4 months.

Acknowledgments

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The Reproductive Cycle of *Trochus niloticus* in King Sound, Western Australia

R. Gimin and C. L. Lee*

Abstract

Adult trochus (48–100) with basal shell diameter greater than 60 mm were collected at approximately monthly intervals between May 1995 and April 1996 from King Sound (KS), Western Australia. The reproductive cycle of trochus from KS was established using gonad indices (gonad index and gonad bulk index) and histological examinations. Fluctuations in gonad indices showed that trochus from KS exhibits a seasonal changes in its reproduction; this is well supported by histological data. There are two distinct spawning periods. The first occurred in July–August and the second in February through to April; minor spawning episodes occur throughout the year. This pattern of spawning is in contrast to the trochus from New Caledonia where the reproductive cycle shows a resting quiescent period. The sex ratio and size at first maturity were determined. Male and female were present in an approximately 1:1 ratio (n = 411 for males, 394 for females; $\chi^2 = 0.359$; $P>0.05$). Individual sexes could be distinguished for all trochus bigger than 50.8 mm in basal shell diameter.

TRADITIONALLY the trochus fishery played a significant role in the economy of coastal fishermen in many Indo-Pacific nations (McGowan 1967; Fagolimul 1988; Arafin 1992) including the Aboriginal communities living along the northern Western Australian coast. As part of the study of the hatchery production and nutrition of trochus from King Sound, Western Australia, its reproductive cycle was investigated.

The biology and reproduction of trochus from many countries have been studied, e.g. Andaman Sea, India (Rao 1937), Banda Islands, Indonesia (Arafin and Purwati 1993), Palau (Heslinga and Hillmann 1981), New Caledonia (Bour 1989) and more recently in French Polynesia (Hahn 1993). In Australia, aspects of trochus reproduction have been reported from Low-Isle, Queensland (Moorhouse 1932) and the Great Barrier Reef (Nash 1985, 1993). Based on the findings of these authors it can be concluded that the reproductive cycle of trochus within its natural distributional range can be divided into two broad categories — seasonal and continuous spawners.

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In the first category, trochus from parts of Queensland, Australia (Moorehouse 1932) and New Caledonia (Bour 1989) were reported to be seasonal spawners with spawning extending over several months of the year. In the case of trochus from Low-Isle Reef, spawnings were concentrated during the winter months, while in New Caledonia, during the summer months. In contrast, populations of trochus from the second category i.e. trochus from Banda Islands, Palau, French Polynesia and the Great Barrier Reef were believed to spawn continuously throughout the year. These differences in duration and frequency of spawning of trochus from different regions within its natural distributional range are possibly in response to changes in environmental factors such as temperature (Moorhouse 1932; Bour 1989), precipitation or water turbulence during the wet season (Arafin and Purwati 1993) and lunar cycle (Hahn 1993).

The present study was undertaken with the aims of (a) determining the reproductive cycle of trochus from a dry monsoonal region of northern Australia, (b) adding to knowledge of the reproductive cycle of trochus from another region of the Indo-Pacific, and (c) determining the size at which spawning first takes place. This is achieved by using gonadal indices and histological data and comparing results of this study with those studies conducted in Indonesia and the Pacific.

Material and Methods

Study site

Broodstock for the study was collected from Mermaid Island and Sunday Island in King Sound, WA (Fig. 1). Samples of 48–100 mature broodstock with minimum basal shell diameter > 60 mm were collected monthly for 12 months commencing in May 1995.

The shells were shipped alive by air to the Northern Territory University (NTU), Darwin some 1000 km east of the collecting sites. The method of transporting the trochus is described by Lee and Ostle (these Proceedings). On arrival at NTU, sex was determined according to the method of Dobson and Lee (1996).

Sex ratio

After the sex of the trochus was determined, the total number of males and females collected each month was subjected to a χ^2 goodness-of-fit test to test the null hypothesis that the male to female sex ratio for trochus in King Sound is 1:1. At the end of 12 months sampling, data for all the broodstocks were pooled, divided into different size classes and subjected to a similar χ^2 goodness-of-fit test.

Reproductive cycle

Gonad indices

Every month, 15 females and 10 males were randomly selected and the apex of the shell cut with an electric diamond cutter to expose the gonad-digestive gland complex (sometimes referred to as the visceral coil or tail). The visceral coils were dissected out and preserved in AFA fixative (a mixture of 70% ethanol, concentrated formaldehyde and glacial acetic acid in the ratio of 1.0:0.2:0.1) to harden the gonad (Hahn 1993).

Each preserved visceral coil was sectioned at three predetermined positions located near: the tip, midportion and the base of the coil. Based on preliminary gross observations, the tip was determined to be fully occupied by gonad tissue; the base was predominantly digestive gland tissue; the mid-portion contained both gonad and digestive gland tissues. The mid-portion was therefore selected as the area best suited for measuring and determining the gonad indices and the subsequent histological study.

Gonad indices were determined by first tracing the outlines of the gonad and digestive gland on a clear plastic sheet. The areas occupied by the gonad and digestive gland were measured by superimposing the tracings on a Gormack graph paper (1 mm × 1 mm) using the method of Young and DeMartini (1970). The gonad index (GI) and the gonad bulk index (GBI) for each specimen were computed by using the method developed by Young and DeMartini (1970), Wilson and Schiel (1995) and Hooker and Creese (1995) for abalone.

Gonad Index = (gonad area × 100)/(maximum shell length)

Gonad Bulk Index = (gonad area × 100)/(total cross-sectional area)

Gonad histology

For histological examination, portions of the gonad-digestive gland complex were dehydrated in a series of four increasing ethanol concentrations (70%, 80%, 90% and 100%). Following dehydration, the tissue was cleared with histolene and impregnated with paraffin wax. Dehydration and wax impregnation were done in a Lx120 Tissue Processor. The wax-impregnated tissue was subsequently embedded with paraffin using a Tissue-Tek II embedding centre. The resulting block was sectioned at 5 µm, and the sections stained in an automatic stainer with Harris Hematoxylin and Eosin.

Gonad sections were examined under the microscope and the gonad classified into different reproductive stages according to the criteria of Bour (1989), Wells and Keesing (1989) and Hahn (1993). Depending on the gametogenic activity and quantity of gametocytes present, six developmental stages were identified: early active (EA), active (A), late active (LA), ripe (R), partly spawn (PS) and spent (S).

One hundred oocytes or ova with visible nucleus were measured. Stalked oocytes were measured on their lesser diameter; polygonal or round oocytes were measured on shorter as well as longer diameter and the mean diameter computed (Hahn 1993). To obtain representative samples of ova for measurement, 3–5 radial axes perpendicular to and radiating out from the digestive gland side of the ovarian wall were drawn on the slides. Starting from the ovarian wall towards the digestive gland, measurements of all ova that fell along these axis were recorded. For spent individuals no measurements were taken, but they were included in statistical analysis by assigning them a nominal oocyte size of zero. The oocyte sizes from each monthly sample were grouped into 10 size classes of 23 µm incre-

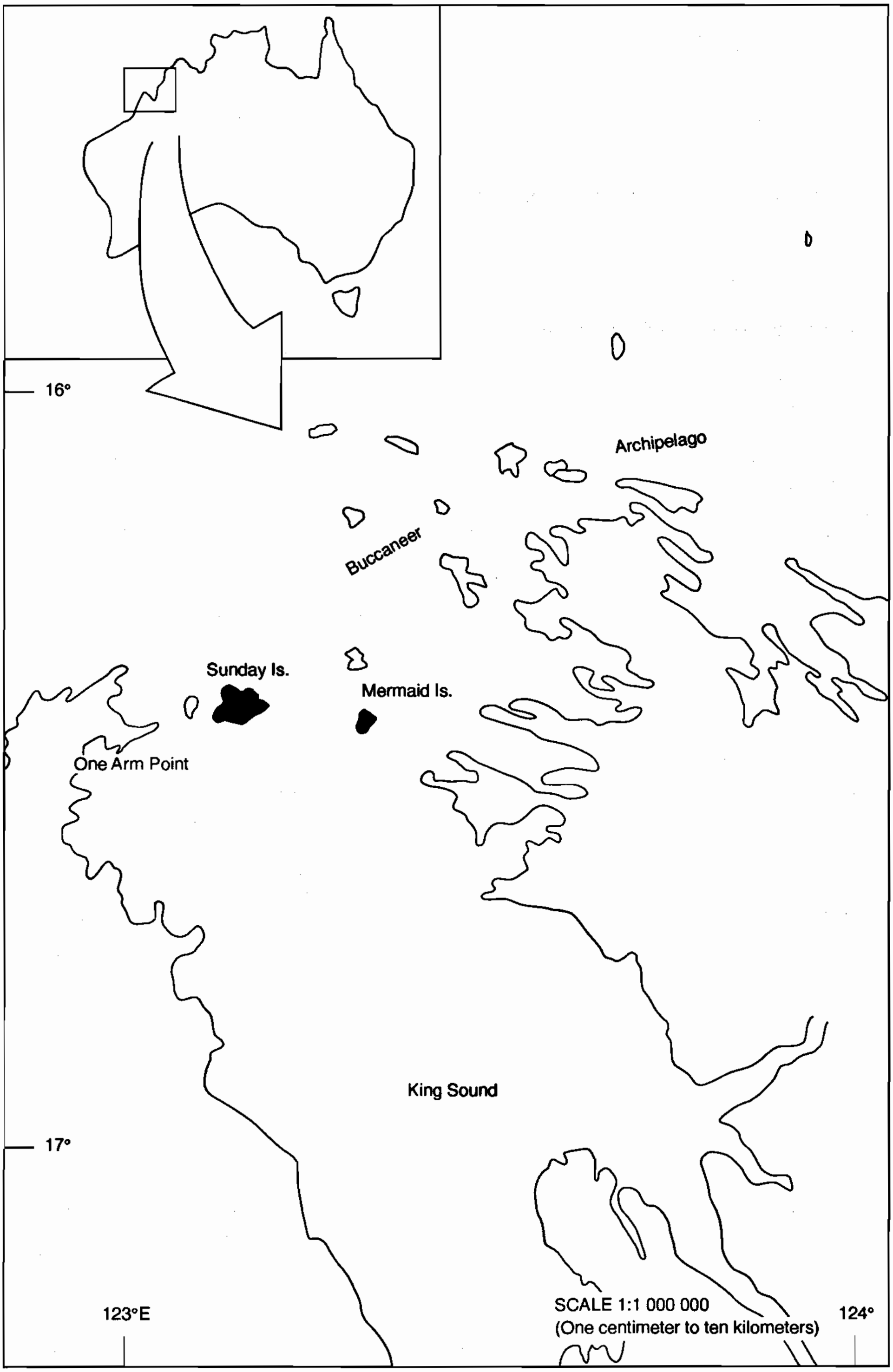

Figure 1. Map showing the location of King Sound, Western Australia.

ments and a polygon size–frequency distribution over the 12 months sampling was constructed.

Size at sexual maturity

The size at sexual maturity was determined from samples collected in January and April 1996. Thirty-four specimens (maximum diameter < 61 mm) were examined histologically for the presence of gametes.

Results

Sex ratio and size–frequency distribution

During the 12-month study, some 805 adult trochus were sampled and their sexes determined. The χ^2 goodness-of-fit test and the χ^2 value for the monthly and pooled samples are given in Table 1. Since the calculated χ^2 did not significantly deviate from the male to female sex ratio of 1:1, the hypothesis that the population from King Sound consists of equal numbers of male and female individuals was therefore accepted.

Table 1. Numbers of male and female trochus collected per month and the χ^2 value for differences from an expected ratio of 1 male:1 female (χ^2 p.05; 1 DF = 3.841).

Month sampled	Males	Females	Total	χ^2
May 1995	49	41	90	0.71
June 1995	55	43	98	1.47
July 1995	25	23	48	0.08
August 1995	29	26	55	0.16
September 1995	24	28	52	0.31
October 1995	40	43	83	0.11
November 1995	34	28	62	0.58
December 1995	32	36	68	0.24
January 1996	27	23	50	0.32
February 1996	23	26	49	0.18
March 1996	39	48	87	0.93
April 1996	34	29	63	0.40
Total	411	394	805	0.36

To establish whether the sex ratio of trochus changes with size (and age), the trochus were divided into 10 size classes. The calculated partial χ^2 showed that except in one class size (63.0–69.9 mm size class with χ^2 = 7.08 and males outnumbered females), there were no significant differences in the proportion of males and females (Table 2) within the different adult size classes of the population from King Sound. This confirmed the report of Rao (1937) and other authors that *T. niloticus* was gonochoristic.

Table 2. Numbers of males and females of class size and the χ^2 value from an expected ratio of 1 male:1 female (χ^2 p.05; 1 DF = 3.841).

Class size	Males	Females	χ^2
56.0–62.9	0	1	1.00
63.0–69.9	35	16	7.07*
70.0–76.9	89	72	1.80
77.0–83.9	119	123	0.07
84.0–90.9	76	88	0.88
91.0–97.9	52	65	1.44
98.0–104.9	25	25	0.00
105.0–111.9	10	3	3.77
112.0–118.9	4	1	1.80
119.0–125.9	1	0	1.00

Asterisk signifies a significant difference from the expected ratio.

The reproductive cycle

Gonad indices (GI and GBI)

The GI and GBI for both males and females using box-and-whisker plots are shown in Figure 2. Generally the gonad indices showed similar patterns throughout the year indicating strong correlation between the two indices. A Spearman correlation coefficient of 0.837 was calculated for males ($z = 9.132$, $p<0.01$) and of 0.653 for females ($z = 10.579$, $p<0.01$). Hence the hypothesis that the two indices were independent was rejected.

Variation in frequency of gonad developmental stages

In females, peak of sexual maturity occurred in July when the ovaries were predominated by ova that were R (87%) or in late LA (13%) stages. S, PS and EA ovaries were found in many individuals in the August samples indicating that spawning had occurred in the preceding month. The high GBI and the presence of PS, EA oocytes recorded throughout September to November indicated that some spawning occurred throughout this period. From November to February 1996, 60–100% of the individuals have gonads with high R and LA ova. Spawning was recorded throughout February to April. A similar trend in testis condition was observed in males.

The annual reproductive cycle therefore can best be described as continuous minor spawning throughout the year with major seasonal peaks in July–August and February–April.

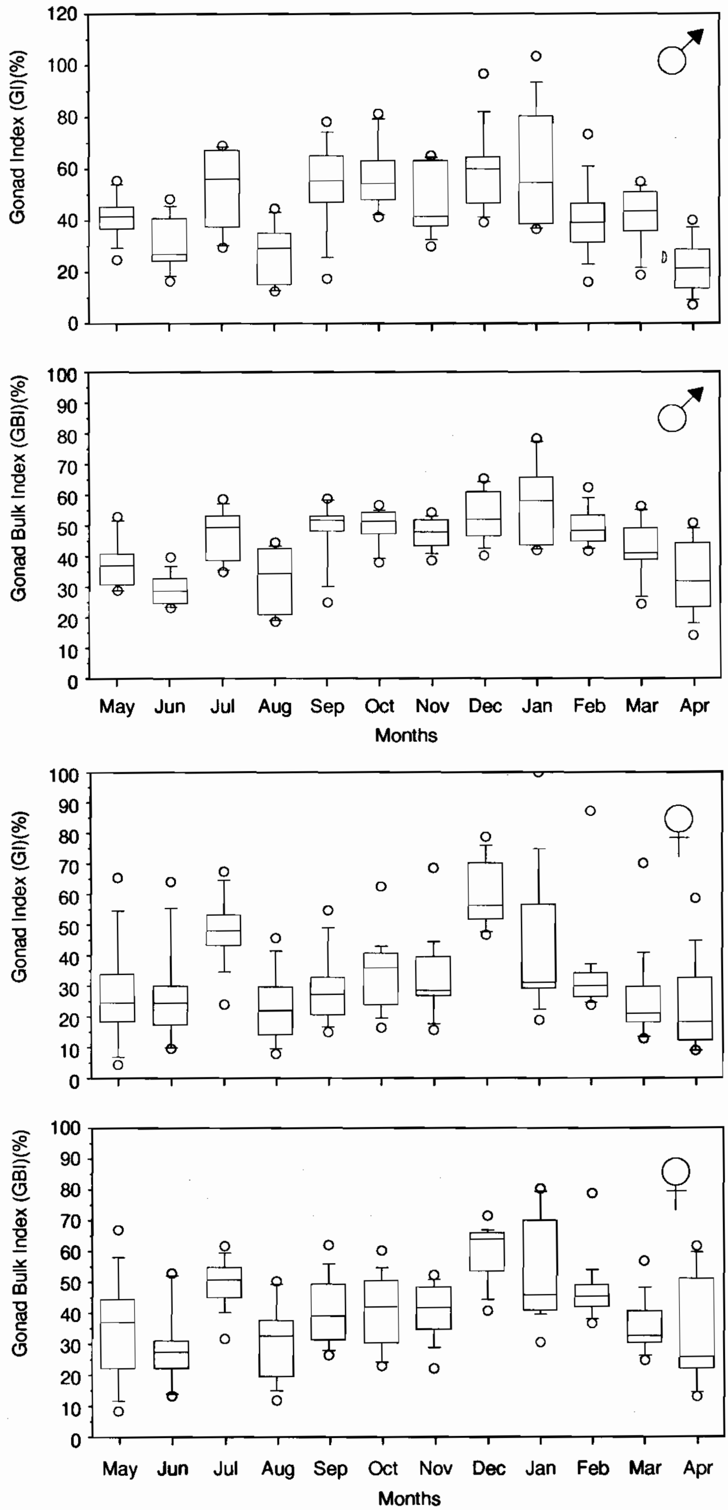

Figure 2. Box-and-whisker plots of GI and GBI over 12-month sampling.

Oocyte size–frequency distribution

A wide range of variation in oocyte sizes was observed between samples and within the same individual. Oocyte size–frequency distribution for 1500 oocytes/ova/month for 12 months is shown in Figure 3. Within samples, the oocyte ranged in size from 30 to 230 μm, with some spent specimens assigned a nominal oocyte size of zero.

Large oocytes/ova (>138 μm) predominant throughout the year suggest that mature individuals were present year-round. A decrease in proportion of large oocytes (115–230 μm size group) coupled with the presence of a greater number of small oocytes (0–92 μm size group) in August indicated that spawning had occurred in the previous month. Between September and November, there was a progressive increase in the size of the oocytes measured, i.e. an increase from 138–160.9 μm size class in September to 161–183.9 μm size class in November, indicating growth and maturation of the oocytes. A greater abundance of large oocytes (138–230 μm size class) reappeared in December 1995 and this condition remained till January 1996. In the following months, there was again a marked increase of small oocytes (<92 μm), and by April 1996 a significant number of size class 0–23 μm was recorded. The presence of the small-size oocytes provide conclusive evidence that spawning occurred during the period. The results provide further support to the contention that the trochus in King Sound exhibit continuous minor spawning throughout the year with major spawning peaks in July–August and February–April.

Size at sexual maturity

In order to determine size at sexual maturity, 34 young trochus 40–61 mm in basal diameter were arranged into 10 size classes at 2 mm intervals. Of the number, 13 small specimens were immature and the sexes were undetermined, 12 specimens were recognised as males and 9 were females. The minimum size at commencement of sexual maturity, as indicated by the presence of recognisable gametocytes, was similar for both sexes. In the case of males it ranges in size 50.5–60.2 mm; the testes contained active to ripe sperms. In the case of females, mature oocytes were recorded in individuals with basal diameters of 50.9–60.6 mm. No gametogenic activity nor sex differentiation were recognised on male and female specimens below 40.8 and 48.2 mm respectively.

Discussion

Changes to the sex ratio of a mature population of different size classes can be used to check whether one sex is more predominant in certain size classes, reflecting hermaphroditism, or in the case of a gonochoristic species, to check whether there is differential growth or mortality rate between the two sexes (Franke 1986; Hughes 1986; Wells and Keesing, 1989). In the present study, no difference or changes in the male to female ratio were detected in the different size classes of trochus investigated. This confirms that trochus is gonochoristic (Rao 1937).

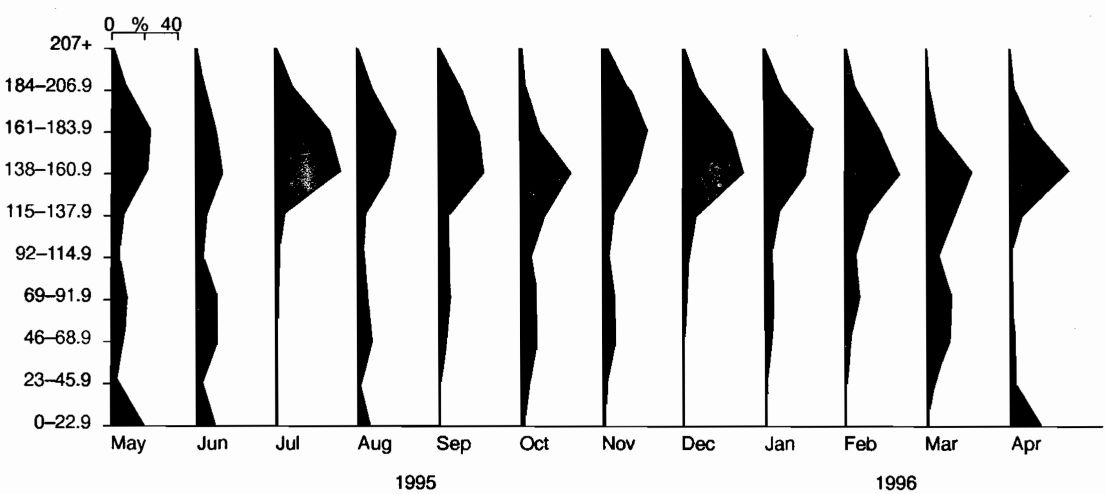

Figure 3. Percentage frequency distribution (abscissa) for different oocytes/ova size class (ordinate) over 12-month sampling.

Gonad indices such as GBI are a good means of following the reproductive cycle of trochus. This is confirmed by histological examinations where the pattern of changes of the gonadal cycle follows closely that of the GBI. In males, high GBI value coincided with the occurrence of greater numbers of spermatids in LA and R stages in the testes, and in the case of females, large-size mature ova in the gonads. Similarly, low GBI indicated that the population consisted of individuals with EA, PS or S gonads. It is therefore concluded that GBI can be reliably used to follow the reproductive cycle of *T. niloticus*. Since GBI measurement is a simple procedure and does not required specialised laboratory equipment, this method is recommended.

Combining the information from GBI, composition of gonadal stages and oocytes/ova size–frequency distribution, it is concluded that the population of trochus in King Sound spawn year round, with two seasonal peaks — a short period in the months of July and August, and a prolonged spawning phase taking place in December through to April. The two spawning peaks are interspersed by at least a few individuals spawning throughout the rest of the year. This pattern of spawning is in contrast to the trochus from New Caledonia where a resting quiescent period, as indicated by prolonged existence of a large proportion of immature stages during the colder months May–October was reported by Bour (1989). When temperatures rose in summer (November–April), rapid maturation followed by spawning events took place. This difference in the reproductive cycle of trochus from KS and New Caledonia could be accounted for by the seasonal climatic conditions in the two locations. KS has a very short and mild winter similar in climatic conditions to that in eastern Indonesia and Vanuatu. In contrast, New Caledonia has a more distinctive but mild winter spread over many months.

Present study indicates that all individuals from KS within the 50.8–52.7 mm size class or above can have sex determined. Hahn (1993) predicted that the trochus from French Polynesia required 7–8 months to complete its reproductive cycle. If Hahn's hypothesis is correct, under favourable conditions, it can be deduced that trochus reach sexual maturity and began spawning 7–8 months after sexual differentiation was first recorded. By this time, the basal diameter has reached 55–60 mm. Heslinga (1981) reported that under favourable laboratory conditions, sexual maturity and spawning can be achieved within a year for hatchery-reared trochus at size 50–54 mm. In comparison, trochus reared in NTU hatchery spawn at age 2.5 years when they are about 55 mm in basal diameter. Moorhouse (1932) reported spawning in a female measuring 44.0 mm; he believed that this specimen was a stunted individual. Nash (1985) reported that sexual maturity of trochus from the Great Barrier Reef was in animals measuring 55–65 mm. It can therefore be concluded that trochus begin to spawn when they reach a size range of 55–65 mm.

Acknowledgments

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Preliminary Studies of the Induced Spawning of *Trochus niloticus* (Linnaeus) Using Artificial Stimuli

G. Dobson*

Abstract

Exposing the gonads of *Trochus niloticus* to the external environment by removing a small section of shell over the gonad significantly increases the spawning response to stimuli. Direct physical stimulation by passing a water current over the exposed gonad further increases the spawning rate. Female trochus may respond in one of two ways to direct stimulation of the ovary — passive release of non-viable, negatively buoyant ova or viable, neutrally buoyant ova actively released into the water column.

THE marine gastropod *Trochus niloticus* is recognised as providing an ideal fishery for small oceanic nations as it can be exploited with minimal access to both financial and technical resources. Recent increases in fishing effort have led to overfishing of natural stocks, which in turn has led to a need for regulation of the fishery and a need to restock areas where the natural trochus populations have been observed to be depleted (Bour 1990). An integral part of any restocking program is the development of appropriate hatchery techniques, including a suitable means to induce spawning.

When trialing methods to induce spawning at the Northern Territory University (NTU) Aquaculture Unit, it was noted that trochus which had had their shells accidentally cut through while being sexed (Dobson and Lee 1996) seemed to respond more readily to stimuli than trochus with an intact shell.

This paper compares the spawning reactions of cut and uncut trochus, The aim was to make a preliminary investigation of the value of this technique as a method of inducing spawning in this species.

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Materials and Methods

Mature trochus ranging 60–110 mm basal diameter were transported to the NTU hatchery from King Sound, Western Australia by road and air according to the method of Lee and Ostle (these Proceedings). On arrival they were placed in a holding tank to recover before being sexed using the 'window' method (Dobson and Lee, 1996). Trochus were then placed, in groups of either 1 female and 1 male or 2 females and 2 males, into glass aquaria measuring 200 mm × 320 mm × 280 mm and filled to a depth of 80 mm (approximately 5 L of water).

The water used in all trials was aged in holding tanks containing broodstock. It was lightly aerated, maintained at a temperature of 30°C ± 2°C and, except where hypersalinity was used as a stimulus, at a salinity of 35 ppt. The water was changed regularly by syphoning out all water and waste before refilling rapidly from a bucket.

The trials were conducted over nine months from September to May. This period includes the monsoon season in northern Australia which is reported (anecdotal) to be the major spawning season for trochus.

Trochus display a lunar spawning periodicity (G. A. Heslinga, pers. comm.) so trials were begun one or two days before and terminated two days after a full or new moon.

Due to their limited availability, most unspawned trochus were used in more than one trial and discarded only after they had spawned actively. All trochus were rested for at least two months between trials.

Trials were conducted in two stages:

Stage I — response to non-physical stimuli

A total of 447 trochus (225 males and 222 females) were used for trials. From these a group comprising 85 males and 82 females each had a section of shell measuring approximately 5 mm × 5 mm removed to expose the gonad (henceforth referred to as 'cut' trochus). The rest of the group were left with shells intact (henceforth referred to as 'uncut' trochus).

Groups comprising cut and uncut individuals were subjected to one of the following potential stimuli to induce spawning: exposure to UV radiation; desiccation; exposure to H_2O_2; exposure to hypersalinity.

Stage II — Response to physical stimuli.

A group of 158 mature trochus (75 males and 83 females) each had a hole measuring approximately 5 mm × 5 mm cut through the shell to expose the gonad. A control group (11 males and 11 females) was left uncut but exposed to conditions identical to those of the cut group.

After being placed in aquaria, 59 males and 67 females were 'massaged' by using a 10-mL plastic pipette to pump water over the exposed gonad in pulses one second apart. 'Massage' sessions were of either 30 or 60 seconds duration and were repeated up to eight times.

A third group (16 males and 16 females) were cut but not massaged. In all other respects they were exposed to identical conditions.

Results

During all trials the females displayed two distinct forms of spawning.

(a) passive—the female remains passively on the bottom with no part of the body extended beyond the shell. Ova fall from within the mantle cavity and spread across the bottom of the tank. The ova are often clumped and negatively buoyant. Numbers released vary from less than 100 to about 5000 and are highly variable in their development. Passive spawning can be regarded only as aborted ova and is useful only to obtain undamaged ova to determine female ripeness. No ova released passively proved viable. In instances where the initial release was well developed there was often a subsequent active spawn of fully developed ova.

(b) active—the female either remains on the bottom or climbs toward the surface and releases a continuous stream of neutrally buoyant and separate ova through an extended, loosely cupped syphuncle. The number released is in the order of 100000–500000 and the majority are usually viable.

Males, whether cut or uncut, displayed only one spawning behaviour.

The male extends its tightly rolled syphuncle upward and releases a continuous stream of sperm, punctuated by contractions, into the water column. The release is copious and can last up to 3/4 hour or can be repeated in shorter sessions up to three times.

In Stage I trials, cut male trochus showed an increased spawning response to all non-physical stimuli except H_2O_2 when compared to uncut males (Table 1). In cut females the rates increased with the use of UV light and hypersalinity, but not with desiccation or H_2O_2 (Table 2). Low numbers of trochus were available for trials with both these stimuli so these results cannot be considered conclusive and are

Table 1. Comparison between spawning behaviour of male Trochus with shells cut and uncut under non-physical stimuli.

Stimulus	Cut			Uncut		
	No.	No. spawn	Spawn (%)	No.	No. spawn	Spawn (%)
UV light	19	6	31.6	22	3	13.6
Desiccation	8	3	37.5	20	1	5
H_2O_2	10	1	10	34	1	2.9
Salinity	20	7	35	20	3	15
Controls	28	5	17.8	44	2	4.5
Total	85	22	25.9	140	10	7.1

included only for the overall comparison of cut and uncut responses.

The overall increase was from 7.1% in uncut males to 25.9% in cut males (Table 1) and from zero to 4.9% active spawn in females (Table 2).

When the gonad was massaged (Stage II) the spawning rates increased from zero for cut but not massaged females to 29% active spawn for those massaged for 30 seconds (Table 3). In males the increase was from 18% for those not massaged to 82% for those massaged for 30 seconds (Table 4).

Of the 86 males and 94 females used in Stage II, a total of 43 males (50%) and 13 females (13.8%) spawned actively. Seven of the females produced viable ova. There were 93 passive spawnings; however, some of these were females which spawned repeatedly.

It was noted that 30-second massage sessions appeared to have no adverse effects on the trochus, even when repeated up to eight times. Repeated 60-second sessions appeared to place considerable stress on the animal resulting in strong contraction movements within the shell, lethargy and some mortality. Altogether 32 trochus were subjected to more than two 60-second sessions. After three sessions one died and after five sessions four females and one male (12%) had died.

The added stress of 60-second sessions markedly reduced the spawning in both males and females (Table 5, Chi square — $\chi^2 = 16.2$ (df = 1; a = 0.1). Passive spawning in females remained the same.

Table 2. Comparison between spawning behaviour of female trochus with shells cut and uncut under non-physical stimuli.

	Cut No.	Cut Active No.	Cut Active (%)	Cut Passive No.	Cut Passive (%)	Uncut No.	Uncut Active No.	Uncut Active (%)	Uncut Passive No.	Uncut Passive (%)
UV	17	1	5.9	2	11.8	21	0	0	0	0
Desiccate.	8	0	0	4	50	20	0	0	0	0
H_2O_2	10	0	0	1	10	32	0	0	0	0
Salinity	20	2	10	10	50	20	0	0	1	5
Controls	27	1	3.7	3	11.1	47	0	0	1	2.1
Total	82	4	4.9	20	24.4	140	0	0	2	1.4

Table 3. Spawning response in female trochus to direct physical gonad massage with sea water.

Massage regime	No.	Passive No.	Passive (%)	Active No.	Active (%)	Fertile No.	Fertile (%)
30 seconds	34	29	85	10	29	6	17
60 seconds	33	28	84	3	9	1	3
Nil	16	5	31	0	–	0	–
Uncut	11	0	–	0	–	0	–
Total	94	–	–	–	–	–	–

Table 4. Spawning response in male trochus to direct physical gonad massage with sea water.

Massage regime	No.	No.	(%)
30 seconds	34	28	82
60 seconds	25	9	36
Nil	16	2	12
Uncut	11	2	18
Total	86	–	–

Table 5. Comparison of spawning response in male and female trochus to differing duration of massage sessions.

Massage regime	Males			Females				
	No.			No.	Passive		Active	
		No.	%		No.	%	No.	%
60 seconds	25	9	36	33	28	84	3	9
30 seconds	34	28	82	34	29	85	10	29

Discussion

Massaging creates a fluctuating pressure on the gonad. A similar effect can be obtained by draining and then rapidly refilling the tank. Of the trochus that spawned actively, 100% of females and 95% of males spawned soon after there was a water change. Passive spawning in females was evenly distributed before and after water changes. It is conjectured that both gonad massage and, to a lesser extent, rapid changes of water simulate, in an exaggerated form, the effects of wave and storm action on the animal. This conjecture is in agreement with anecdotal evidence and with studies by Gimmin (pers. comm.) placing peak spawning periods before the start and at the end of the monsoon, the stormiest periods of the year in a monsoonal climate, and supports Grange (1976) who concluded that rough water is a spawning stimulant for some species of Trochid and Turbinid gastropods.

It seems likely that the modes of spawning seen here are atypical behaviour brought about by cutting the shell. An uncut female spawns by forcibly ejecting the ova into the water column or, if she has climbed high enough, through the air. The hole in the shell makes it impossible for her to build the pressure inside the shell to eject the ova forcibly.

There is no noticeable damage to the trochus caused by removing a section of shell provided that the area removed is no more than 5 mm × 5 mm. Within a week a tough membrane forms over the gonad, after three weeks the membrane has begun to calcify and after six weeks the hole is sealed by new hard shell. If the hole is larger than 5 mm × 5 mm the gonad and digestive gland are occasionally forced out. In most instances the organs were retracted and the animal recovered, but in some cases the organs were lacerated by sharp edges to the cuts and the animals died.

This study shows that directly stimulating the gonad can be an effective method of inducing spawning in *Trochus niloticus*, especially in males. Cutting the shell leads to some modification of the spawning behaviour in females, but this has not been seen to be detrimental to either the ova or the female. This is a preliminary investigation, and the results suggest that the method described warrants further investigation.

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Early Development and Growth of Juvenile *Trochus niloticus* Reared Under Different Salinities

R. Gimin and C. L. Lee*

Abstract

Fertilised trochus eggs were tested for their ability to complete their embryonic development, growth and survival when maintained at salinities of 25, 30, 35, 40 and 45 parts per thousand (ppt). The results indicated that it was possible successfully to rear hatchery-produced trochus to two-month-old juveniles between the salinity range of 30 and 40 ppt. However, the survival rates changed drastically as the salinities dropped to 30ppt (survival, 44.67%) or increased to 40 ppt (survival, 42.67%) when compared to those maintained at 35 ppt (75%). Embryonic development failed to be completed at salinities of 25 and 45 ppt. Growth rate of postlarvae (P/L) to juveniles over two months was highest at 30 ppt (3.34 mm), slightly lower for those maintained at 35 ppt (3.25 mm) and significantly slower for the P/L maintained at 40 ppt (2.25 mm) ($P<0.05$). Similarly, P/L survival rates were different in the three salinities tested. Best survival rate (48.5%) was also recorded from 30 ppt followed by 37% for those at 35 ppt. The lowest survival rate (32.5%) was recorded at 40 ppt. The survival rate of P/L at 40 ppt was significantly different from those maintained at either 30 or 35 ppt ($P<0.05$).

A variety of physical environmental parameters is known to influence, enhance or depress survival, hatching, larval development and settlement and metamorphosis of marine gastropods. Such factors include salinity, light quality and intensity, photoperiod, temperature, substrates and diets (Heslinga 1981; Hughes 1986).

Salinity is an important ecological factor determining the survival of many stenohaline marine gastropods like trochus, *Trochus niloticus*. An optimum salinity level for example is necessary for egg incubation; sudden rapid changes in salinity may cause premature bursting. The effect of salinity on the physiological processes of marine invertebrates has been reviewed extensively by Kinne (1971) and other related studies on the effect of salinity in combination with such factors as temperature or food on early larval development of invertebrates like crustacean and molluscs were reported by Loosanoff and Davis (1963), Hartnoll (1978), Hrs-Brenko (1978) and Lau et al. (1994). More recently, Yi and Lee (these Proceedings) have looked into the effects of salinity and temperature on the oxygen consumption and survival of juvenile trochus reared in the hatchery. There has been, however, relatively little information available of the effect of salinities on early development and growth of the juvenile. This paper reports the effects of salinities on (a) the embryonic development, hatching rate and metamorphosis to postlarvae (P/L) and (b) growth of P/L to two-month-old juveniles.

Materials and Methods

Egg production

Mature broodstocks were collected from King Sound, Western Australia and transported by road and air to the Northern Territory University (NTU) hatchery according to the method described by Lee and Ostle (these Proceedings) and the sex determined according to the 'window' method (Dobson and Lee 1996). After the sexes were determined, the brood-

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stocks were induced to spawn and the fertilised eggs were collected and washed with fresh seawater (35 ppt) before transferring into experimental units.

Salinity and postlarvae production

The fertilised eggs were subjected to hatching at five different salinities i.e. 25, 30, 35, 40 and 45 ppt with the 35 ppt acting as the control. Test salinities, with five replicates per salinity, were prepared by mixing treated bore water at 50 ppt with the required amount of tap water. Each experimental container (EC) used for the experiment was derived from a 800-mL plastic bowl filled with 600 mL of test solution; stocking density was maintained at 2 eggs/10 mL. A lid was also loosely placed on the top of each EC to reduce evaporation. The salinity in all ECs was checked twice a day and adjusted accordingly. Dissolved oxygen in the bowls was supplied via perforated air tubing that provided gentle aeration to the water during the experimental period.

All 25 ECs were fixed to three styrofoam boxes each measuring 550 mm × 380 mm × 250 mm; each box can carry up to 10 EC. The boxes were placed in the hatchery shed away from direct sunlight. This arrangement provided adequate insulation and reduced temperature fluctuation to the ECs to an acceptable level during the experimental period. About 30 hours after hatching, diatom-coated shellgrits were put carefully into the bowls to cover the bottom. This provided the substrate for the larvae to settle into postlarvae (P/L).

During the experiments, all ECs were checked every 15 minutes for the first 16 hours. However, for determining the embryonic development and stages a sample of 10–15 eggs/larvae from each bowl was removed and observed under a dissecting microscope at set intervals (Table 1). Due to time constraints, after 16 hours observations were limited to the most prominent stages of the egg/larval development, i.e. hatching, trochophore and veliger larvae, and metamorphosis to postlarvae. During the experiment, it was observed that all eggs subjected to salinities of 25 and 45 ppt failed to develop after 20 hours and were therefore discontinued. Comparison of eggs/larval quality among the different salinities treatments were conducted by counting proportion (%) of the eggs or larvae reaching a particular stage at the time of sampling. The results were subjected to Cohran's test prior to using one-way analysis of variance (ANOVA) to examine the effects of salinity on the larvae reaching a particular developmental stage. Where required, the data were subjected to arcsine transformation prior to analyses using the statistical software package Statview 5+.

Salinity and juvenile growth

Nine days P/L were used for the growth rate experiments using different salinities. The 800-mL plastic bowls described previously were used and into each bowl seasoned shellgrits with benthic diatom *Nitzchia sp.* growing on their surfaces were introduced together with 40 P/L. Salinities tested were 30, 35 and 40 ppt. During the experiment, adequate food was maintained by replacing old shellgrits with fresh diatoms and algal-coated ones every three days. Seventy-five per cent of the water in the bowls was also replaced daily and during this time the salinities were adjusted accordingly.

Growth rates of P/L were determined by measuring the maximum basal diameter of 5–10 P/L using an ocular micrometer inserted into a stereo dissecting microscope at about fortnightly intervals. During the experimental periods, five measurements were carried out. At the end of the experiments, the results were subjected to one-way analysis of variance (ANOVA).

Table 1. Survival through embryonic development (mean % ± SD) of eggs maintained at different salinities.

Stage	Time after fertilisation	Salinities 25 ppt	30 ppt	35 ppt	40 ppt	45 ppt
Cleavage	20 minutes	69.556±9.481^{a}	88.640± 7.340^{b}	91.140± 5.291^{b,c}	85.862± 8.481^{b}	78.278±11.677^{c,a,b}
Gastrulation	7 hours	–	44.978± 9.193^{a}	78.000±11.646^{b}	46.360±10.451^{a}	–
Hatching	12 hours	–	46.000±12.575^{a}	90.666± 9.250^{b}	86.278± 9.529^{b}	–
Veliger	20 hours	–	29.522±16.680^{a}	89.240±15.363^{b}	79.646±17.168^{b}	–
Metamorphosis	6 days	–	44.670±10.436^{a}	75.000±16.583^{b}	42.670±10.111^{a}	–

Values which are followed by different superscript(s) are significantly different ($P<0.05$)
N = 5 for each salinity tested

Results

Salinity and postlarvae production

Effects of different salinities on embryonic development of fertilised eggs to newly metamorphosed P/L are summarised in Table 1. It can be seen that fertilisation and cleavage can occur in a wide range of salinities. Twenty minutes after fertilisation, the eggs undergo two divisions. The number of eggs (by per cent) completing the first two divisions are different under different salinity regimes and ranges from 69.56% to 91.14%. ANOVA shows significant difference in the number of eggs completing the first two cell divisions at the salinity range of 30 to 40 ppt in comparison to those maintained at 25 ppt (P<0.05). In fact, all samples, except for one sample at the lowest and one sample at the highest salinities (25 and 45 ppt), did not reach the gastrula stage and the eggs failed to develop further.

Gastrulation took place about 7 hours post-fertilisation and was successfully completed by many of the eggs maintained at 30, 35 and 40 ppt. Among them, the highest gastrulation rate (78%) was recorded at 35 ppt and it was significantly different (P<0.05) from those maintained at 30 and 40 ppt (44.98% and 96.36%, respectively).

At about 12 hours post-fertilisation, the eggs hatched into free-swimming trochophore larvae. Hatching rates of eggs incubated in 35 and 40 ppt were similar (86.28% and 90.67%, respectively) and were significantly different (P<0.05) from those maintained at 30 ppt, which showed a hatch rate of only 46%.

The beginning of the veliger stage was indicated by the completion of the larval shell and the animal being able to withdraw its body into the shell. The proportion of trochophore larvae reaching veliger stage at 20 hours was the highest at 35 ppt (89.24%) but this was not significantly different from those maintained at 40 ppt (79.65%). The lowest percentage of trochophore reaching the veliger stage was observed at 30 ppt (29.52%) and this was significantly different (P<0.05) from those maintained at 35 and 40 ppt. Many of the veligers maintained at 30 ppt were also observed to have elongated shells and spent most of the time lying on their side.

Metamorphosis was recognised when the crawling larvae lost their vela. The highest rate (75%) of metamorphosis was recorded at a salinity of 35 ppt; this was significantly different (P<0.05) from those maintained at 30 and 40 ppt which gave 44.67% and 42.67% metamorphosing rates, respectively.

Salinity and juvenile growth

Table 2 and Figure 1 summarise the results of the growth and survival rates of juveniles reared under three different salinity regimes over a two-month period. Optimal salinity for early juvenile growth appeared to be from 30 to 35 ppt. Growth rate of juvenile was higher at 30 ppt but this was not significantly different from those maintained at 35 ppt. After two months, the mean shell diameter of juveniles maintained at 30 and 35 ppt were 3.34 mm and 3.25 mm respectively. Significantly poorer growth was shown by juveniles maintained at 40 ppt, and at the end of the experimental period the mean shell diameter recorded was only 2.245 mm; this was significantly different (P<0.05) from those juveniles maintained at 30 and 35 ppt.

Survival rates after the two-month rearing period were also different between the three salinities tested. The best survival rate (48.5%) was attained by the juveniles at 30 ppt and was not significantly different from those maintained at 35 ppt (37%). Survival rate of juveniles at 40 ppt was significantly different (P<0.05) than those maintained at 30 ppt only.

Table 2. Growth of trochus from metamorphosing stages till two-month-old juveniles.

Salinity	Shell growth at date				Survival rate
	22/11/95	23/12/95	07/01/96	21/01/96	(%)
30 ppt	0.38 ± 0.02^{a}	1.27 ± 0.2^{a}	2.34 ± 0.44^{a}	3.34 ± 0.30^{a}	48.50 ± 13.87^{a}
35 ppt	0.39 ± 0.01^{a}	1.23 ± 0.10^{b}	2.42 ± 0.23^{a}	3.25 ± 0.32^{a}	$37.00 \pm 9.86^{a,b}$
40 ppt	0.36 ± 0.01^{b}	1.02 ± 0.128^{b}	1.53 ± 0.27^{b}	2.24 ± 0.40^{b}	32.50 ± 10.31^{b}

Values which are followed by different superscript(s) are significantly different (P<0.05).
Values are means of five replicates ± SD.

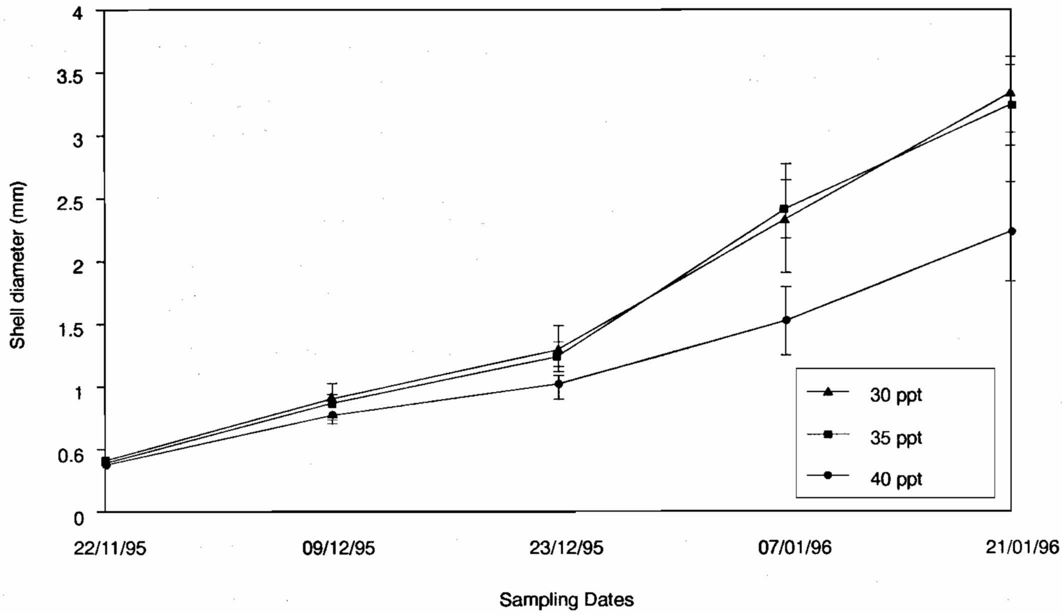

Figure 1. Growth rate (mean ± SD) of trochus juveniles at different salinities.

Discussion

The present study indicates that the newly fertilised eggs are highly sensitive to very low (25 ppt) and very high (45 ppt) salinities during their early embryonic development. Newly hatched trochophore and veliger larvae are similarly stenohaline although the larvae are able to survive and metamorphose successfully to P/L at a salinity range of between 30 and 40 ppt. This salinity tolerance apparently remains relatively unchanged after settlement. The ability of marine larval molluscs to develop normally under a range of salinities has been documented on bivalves like oyster and clams. Loosanoff and Davis (1963) reported that oyster *Crassostrea virginica* acclimatised to low salinity (27 ppt) were able to mature and spawn, and the larvae grew successfully in salinities of 15–35 ppt. Whether these larvae and newly metamorphosed P/L develop normally or show greater occurrence of abnormality is not known.

Optimum salinity for metamorphosis to P/L was recorded at 35 ppt and decreased significantly ($P<0.05$) as the salinities increased to 40 ppt or decreased to 30 ppt. After settlement, the P/L appeared to grow slightly better at 30 ppt than at 35 ppt; growth and survival rates were significantly better at 30 and 35 ppt than those maintained at 40 ppt. The salinity tolerance for larvae and P/L recorded in the experiment may provide some explanation of the reproductive cycle of trochus from the King Sound Peninsula, Western Australia.

Trochus are believed to have an asynchronous reproductive cycle (Hahn 1993) and at least a percentage of females is known to spawn during the new and full moon of each calendar month. This is possibly the case for trochus in many parts of their natural distributional ranges. However, in King Sound where the trochus broodstock were obtained, the situation is complicated by the presence of heavy monsoonal rain concentrated during the months of December and April each year. During this time, intertidal invertebrates and their larvae may be exposed to low salinity for a few hours or for days at a time (Yi and Lee, these Proceedings). During this period of heavy rain, spawnings are unlikely to lead to successful recruitment as the fertilised eggs have a much lower chance of completing their embryonic development. Therefore it could be hypothesised that trochus in King Sound have evolved a reproductive strategy that involves concentration of spawning at the beginning (September to November) and at the end (March and April) of the wet season when monsoonal rains are less frequent and the dilution effect of rainwater on the intertidal zone less marked, in order to optimise reproductive efficiency and to ensure high survival for offspring. If the hypothesis is true, then the present study coupled with an investigation of the gonadal cycle of the trochus in King Sound would provide further insight into the production of trochus in the King Sound region. This aspect is currently being investigated by NTU staff.

Acknowledgments

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Effects of Temperature and Salinity on the Oxygen Consumption and Survival of Hatchery-reared Juvenile Topshell *Trochus niloticus* (Mollusca: Gastropoda)

S. K. Yi* and C. L. Lee†

Abstract

Juvenile *Trochus niloticus* were successfully produced in closed recirculating hatchery tanks supplied with saline bore water. Juveniles (Js), after reaching 3 mm in shell diameter, undergo rapid growth where the relationships between the shell width (W) and total weight (Tw) and dry weight of soft body (Dw) are represented by the equations $Tw = 0.305W^{2.787}$ ($r^2 = 0.995$) and $Dw = 0.0542W^{o2.172}$($r^2 = 0.976$). During this period of active growth, the oxygen consumption rate (Ocr) can be presented by the equation $Ocr = 1131.655/W^{1.139}$ ($r^2 = 0.933$). The maximum oxygen consumption of 264 µL O_2/h/gTw was recorded at 31°C and 35 parts per thousand (‰); oxygen consumption is reduced and stress sets in when the temperature exceeds 34°C. Js are less affected by salinity at low temperature than at higher temperature (24–TL_{50}). At 31°C, the salinity tolerance is between 23 and 51‰. This is well within the operating temperature of a trochus hatchery situated in the tropics. For optimal growth it is recommended that a trochus hatchery in the tropics should maintain a water temperature of 30–31°C and a salinity of 31–37‰.

THE coral reef snail commonly known as the topshell, *Trochus niloticus* Linnaeus, 1767 is one of the most important gastropods in the Indo-West Pacific due to demand for its nacreous shell and meat. Its natural range extends from Sri Lanka and the Andaman Sea in the west to Samoa Islands in the East, from Ryuku Islands in the north and to New Caledonia in the south (Hedley 1917; Rao 1937). Since 1927, trochus has been successfully translocated to many areas (Hoffschir et al. 1989; Kubo 1991; Gillett 1993) resulting in its distributional range extending to most of the Indo-West Pacific islands.

Commercial trochus fishery began about 100 years ago, and in recent years interest in the shell has accelerated as a result of increasing demand for it to make buttons for high-priced garments, inlays for furniture, bracelets, tourist souvenirs, etc. This has resulted in overexploitation of natural trochus populations which in many areas have suffered serious decline despite efforts to manage the fishery in some countries (Bouchet and Bour, 1980; Heslinga and Hillman, 1981; Nash 1985). Consequently, there has been considerable interest in trochus aquaculture and in stock enhancement of trochus populations in coral reefs with hatchery-produced juveniles (Nash 1985; Hahn 1989; Kubo et al. 1989; Hoffschir 1990; Castell 1993).

As part of the research into the hatchery management, nutrition and growth of trochus juveniles (Js) in the Northern Territory University (NTU), their physiology was studied in order to develop a protocol for managing them through a better understanding of oxygen consumption, water temperature and salinity requirements.

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Materials and Methods

Juvenile production and water supply

Trochus broodstock were collected from King Sound, Western Australia and transported by air to the NTU, Darwin, Australia some 1000 km east of the collection site. The method of transportation of broodstock, inducing spawning, hatchery system and methods of producing juveniles at NTU for the experiments have been reported by Lee (these Proceedings) and Lee and Ostle (these Proceedings).

Experimental animals: measurements and treatments

Beginning in December 1993, Js of up to 10 mm in basal shell diameter were harvested periodically and held in glass aquaria (each measuring 30 cm wide × 60 cm long × 40 cm high) for 24 hours (water temperature 28–30°C) to evacuate their gut contents before being used for experiments. The Js were divided into seven different size groups by basal shell width (Table 1) and for each size group, 8–30 animals which fell into a standard deviation range for the group were selected and used for each size-dependent treatment. For oxygen consumption and tolerance experiments related to salinity and temperature, Js belonging to the 5 mm size group were used. At the end of each experiment, the basal shell width (W) was measured to the nearest 0.01 mm, the total wet weight of the animals (Tw), wet weight of soft body (Sw) and dry weight of the soft body (Dw) were determined to the nearest 0.01 mg; the dry weight was obtained after drying the soft body of the animals in an oven at 50°C for 96 hours.

Oxygen consumption

The water used in the oxygen consumption study was passed through a 1-µm filter bag and boiled to kill micro-organisms. After cooling, the required salinity was attained by mixing with distilled water. The water was again passed through a filter paper of 1 µm, exposed to ultraviolet irradiation for a further two hours and saturated with air at room temperature with gentle aeration before using.

All experiments were conducted in 300-mL oxygen bottles placed in water baths, and unless otherwise stated, placed under continuous illumination. After the Js were placed into an oxygen bottle, the airtight lid was sealed and the bottle incubated for 4–6 hours at a given temperature. A blank (without animal) with a similar size oxygen bottle was also maintained to correct for microbial respiration. All experiments were conducted in duplicate. Oxygen consumption was measured using a Yellow Springs Instruments oxygen meter (YSI-57).

Salinity and temperature tolerance

Tolerance tests (24 h-TL_{50}) were conducted by exposing young trochus to 14 different temperatures (between 12° and 37°C) and 28 different salinities (between 7 and 61‰) regimes. Test containers of known salinities were submerged two-thirds of their height in a water bath with a pre-set temperature. Once a constant temperature was reached, 10 young trochus of similar 5 mm size were introduced into the test containers without temperature acclimatisation. At the end of 24 hours, the operculum of each test animal was gently touched with a fine needle and the response was recorded. Animals that reacted readily

Table 1. Number of juvenile trochus and their size ranges used for each set of experiments.

Size group (mm)	No. animals	Shell width (mm)	Total weight of whole animal (mg)	Dry weight of soft body
1	30	1.34 ± 0.15	0.68 ± 0.089	0.07
2	30	2.25 ± 0.15	3.55 ± 0.240	0.34
3	30	3.31 ± 0.16	7.86 ± 1.010	0.61
4	23	4.17 ± 0.16	15.04 ± 2.630	0.74
5	20	4.91 ± 0.27	24.07 ± 3.820	1.21
7	12	7.06 ± 0.48	61.10 ± 13.40	2.29
10	8	10.08	232.26	8.20

to touching were classified as unaffected; animals that failed to respond to touch were transferred to a recovery tank maintained at 35‰ and 24–26°C. After three hours in the tank, those Js that recovered were classified as being comatose and those that did not recover were classified as dead. The upper and lower incipient lethal temperatures and salinities (24 h - TL50) were determined using a computer model based on Litchfield and Wilcoxon (1949).

Results

Size–weight relationships of hatchery juveniles

The relationships between shell width and weight of the young Js used in this study were exponential (Fig. 1). These relationships are described by the equations:

$Tw = 0.305W^{2.787}$ $(R^2 = 0.995)$
$Dw = 0.054W^{2.172}$ $(R^2 = 0.976)$.

Oxygen consumption

Under a salinity regime of 35‰ at 30°C, the respiration rate of the young Js decreased with increasing size and body weight (Fig. 2). The best-fit lines for the oxygen consumption rate (Ocr) for light and dark conditions are described by the two equations:

$Ocr\ (Light) = 1181.030/W^{1.108}$ $(R^2 = 0.929)$
$Ocr\ (Dark) = 1471.938/W^{1.182}$ $(R^2 = 0.949)$

where Ocr is in µL O_2/h/gTw.

Since no clear difference was found in the Ocr conducted under dark and light conditions, the two equations could be combined to give a general Ocr equation where

$Ocr = 1131.655 / W^{1.139}$ $(R^2 = 0.933)$

From Figure 2, it can be noted also that the Ocr of the young Js sharply decreased with increasing body size after 3 mm. With W at about 4 mm, the Ocr was close to 220 µL O_2/h/gTw. Further growth of Js did not markedly change the Ocr.

At 35‰, changes in water temperature greatly affected the Ocr; the rate increased in a step-wise manner with increasing temperature of up to 31°C and decreased thereafter (Fig. 3). There was a sharp increase of Ocr between 12° and 18°C followed by a static period when the Ocr remained at about 120 µL O_2/h/gTw (between 18° and 26°C). The maximum Ocr of 264 µL O_2/h/gTw was recorded at a water temperature of 31°C.

In contrast to the effect of water temperature, salinity has a less marked effect on the Ocr of Js. Between 27 and 43‰ the Ocr were recorded within a range 90–146 µL O_2/h/gTw; the maximum Ocr of 146 µL O_2/h/gTw was recorded at 39‰ and above 43‰, the Ocr decreased sharply with increasing salinity (Fig. 4).

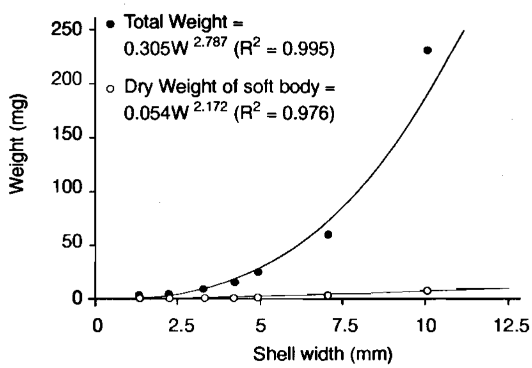

Figure 1. Size–weight relationships of juvenile trochus.

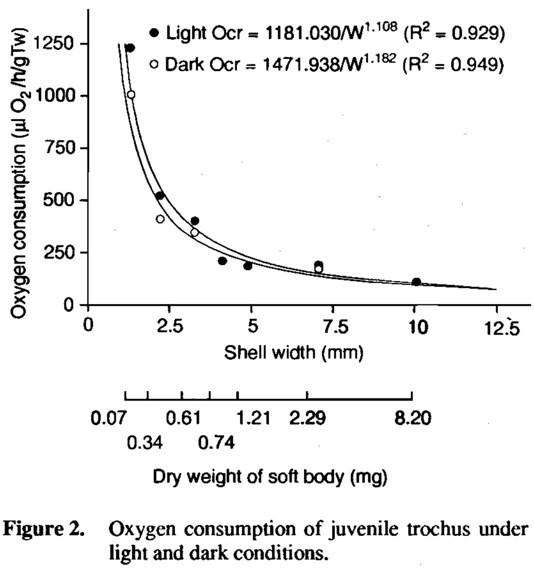

Figure 2. Oxygen consumption of juvenile trochus under light and dark conditions.

The relationship between Ocr, temperature and salinity is shown three-dimensionally in Figure 5. Generally, the Ocr increases with increasing water temperature and salinity.

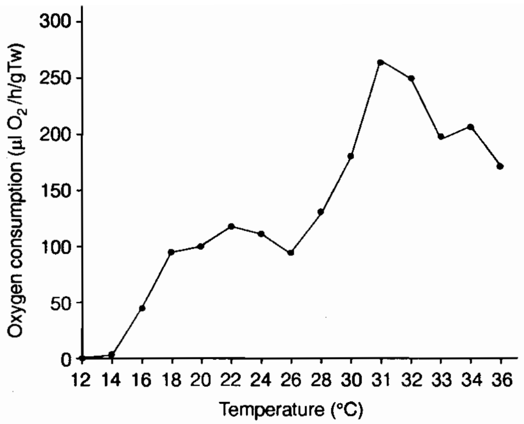

Figure 3. Oxygen consumption rate of juvenile trochus at 35‰ under different temperature regimes.

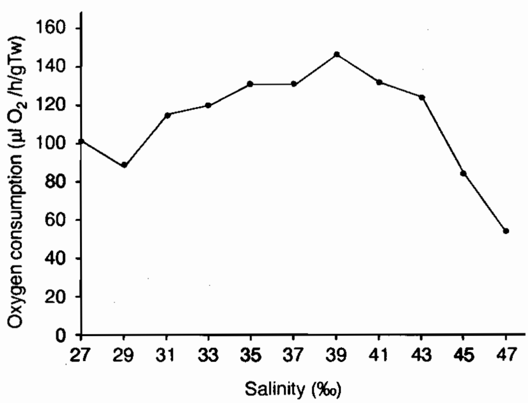

Figure 4. Oxygen consumption of juvenile trochus at 28°C under different salinity regimes.

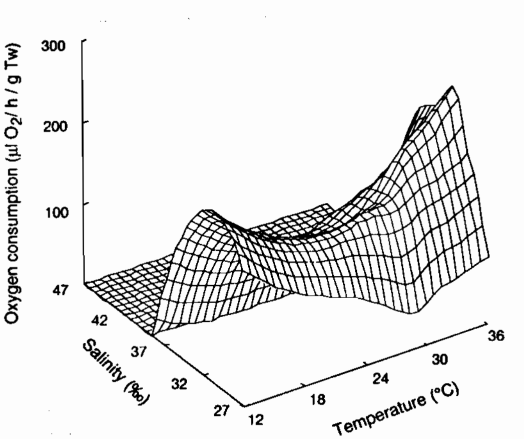

Figure 5. Oxygen consumption (µl O_2/h/gTw) of juvenile trochus as a function of temperature and salinity.

Tolerance to temperature and salinity

The upper and lower tolerance limits, as 24h-TL_{50} against salinity and temperature of Js are shown in Figure 6. Generally, there is a tendency for (a) the upper tolerance limits to salinity to decrease with increasing temperature and, (b) the lower tolerance limits to increase with increasing temperature. This has resulted in a wider range of tolerance to salinity at low temperatures (14.8–56.4‰ at 20°C) and a much narrower tolerance to salinity at higher temperatures (24.9–51.3‰ at 34°C). The ranges of tolerance to salinity at 20° and 34°C are reduced to 17.0–51.8‰ and 26.6–50.3‰ respectively when comatose individuals are considered to be dead.

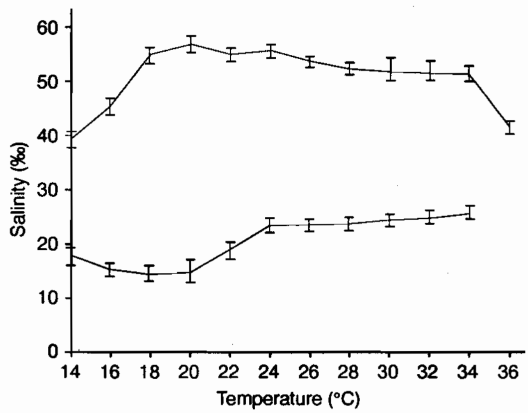

Figure 6. Upper and lower tolerance limits of juvenile trochus against different salinity and temperature. Bars represent the 95% confidence intervals.

Discussion

Size–weight relationships

A literature review has indicated that little is known about the size–weight relationships of trochus postlarvae up to Js 15 mm in size. Heslinga (1981) carried out some 12-month studies on the size–weight relationships of hatchery-reared trochus on Palau, Caroline Islands. He believed that in the first four months of their life, Js showed slow growth which was designated as the lag phase of the juvenile growth cycle. Unpublished data from the NTU studies indicate that the Js had a much shorter lag phase, e.g. about eight weeks when the Js reached 2–3 mm in diameter, before the growth rate took off exponentially. During the phase of rapid growth, a huge demand is placed on the hatchery to produce sufficient quantities of benthic algae and diatoms to feed the Js; this has become a major constraint in developing a mass hatchery production system. In terms of economics, it is believed that the 2–3 mm size class is the better group for harvesting for aquaculture or for release into reefs for stock enhancement programs and research.

Oxygen consumption

The relationship between shell size and Ocr recorded in the present study is very interesting. It has been reported that smaller marine gastropods have higher basal metabolic rates (BMR) than larger ones (Hughes 1986). This is obviously the case with Js smaller than 5 mm (Fig. 2). The high Ocr recorded for juveniles in the present study may be accounted for by their feeding behaviour pattern in the hatchery tanks. In the hatchery, Js of this size group can be observed to graze actively and crawl along the substrate and wall of the tanks at all hours. In contrast, as they grow larger, the feeding activities are reduced during daylight hours. Since the Ocr were recorded over 24 hours, the lower Ocr recorded for the larger Js may partly reflect reduced feeding activities and perhaps lower BMR.

Figure 3 shows that the highest Ocr was observed at water temperatures around 31°C and a sharp decrease above this temperature seemed to be caused by heat stress to the experimental animals. Sandison (1967) found that the maximum respiratory rate of intertidal gastropods was recorded at a water temperature a few degrees below coma temperature; on entering or exceeding coma temperature, the Ocr fell sharply and this reduction in oxygen consumption continued until heat death. The relatively constant Ocr observed between 18° and 26°C indicates that the Js have some compensatory mechanisms to regulate and maintain a 'normal' Ocr which is about 110 µL O_2/h/gTw. As the water temperature dropped below the lower optimal range, the Js closed their opercula, reduced mobility and decreased the rate of metabolism. In contrast, when the water temperature exceeded the upper optimal range, the rapid increase in the Ocr indicated an active attempt at regulation of body functions. Above 32°C, stress gradually set in and Js slowly enter comatose conditions.

Intertidal animals show a certain degree of response to tidal rhythms in their behaviour (Palmer 1974; Taylor and Naylor, 1977) and a circatidal rhythm of oxygen consumption in molluscs has been observed by a number of researchers (Zann 1973; Marsh and Branch, 1979; Kim 1994). Further experiments are needed to study the circatidal and circadian effects on oxygen consumption and Ocr of intertidal animal such as trochus Js.

Salinity and temperature tolerance

It has been reported that for many tropical lower and subtidal gastropods, the upper lethal water temperature was above 42°C (Sandison 1967; Markel 1971; Stirling 1982) and this is normally far above any temperature which might reasonably be expected to occur in the field (Kinne 1970). The thermal tolerance of a marine invertebrate species is a sophisticated function of its previous thermal history and other environmental factors. Within a species, this tolerance often varies in accordance with lifecycle stages and habitat conditions. In general, animals inhabiting the higher littoral zone and lower latitude show higher thermal tolerance than those in the lower littoral zone and higher latitude respectively (Ritz and Foster, 1968; Foster 1969; Kenny 1969; Kinne 1970). The present study provided additional data which confirm this hypothesis.

Wallis (1976) found that high salinities aid thermal resistance of a tropical mussel *Trichomya hirsuta*. However, Brenko and Calabrese (1969) noted that the effects of salinity and temperature were significantly related only as the limits of tolerance of either factor were achieved. In the present study, it is believed that the salinity was less likely to have an impact on the survival of trochus Js, and cooler temperature seemed to enhance their salinity tolerance. This could be explained in terms of their adaptation to the environmental conditions existing in King Sound. King Sound experienced somewhat lower water temperature compared to many other tropical habitats. The water temperature recorded in a pearl farm at King Sound (at 2 m depth) was 24–31.4°C. However, the intertidal water temperature during winter is likely to fall below 20°C. Furthermore, heavy monsoonal rains are common during the wet season between December and March each year. During heavy thunderstorms, the salinity may drop drastically for a few hours before tidal movements help to reduce the effect of the salinity stress on the intertidal Js. Since tropical thunderstorms are accompanied by rapid cooling and lowering of water temperatures, the effect of salinity stress on the Js during the summer months is also reduced, as shown by the results of the present study.

During the course of this study, no Js smaller than 35 mm in basal diameter were collected from the study site in King Sound. The habitat of Js under 5 mm is unknown, but it was reported that Js about 10 mm can be found on coral rubble in the lower intertidal reef areas (Shokita et al. 1991) while bigger trochus of about 25 mm can be found in shallow subtidal reef flats (Nash 1985) where the water temperature is cooler. The thermal tolerance of juvenile trochus in the wild is unknown, but based on the present laboratory study, Js are likely to be found beneath coral rubble among coralline algae beds during daylight hours at low tide where the temperature is likely to be cooler. At dusk they may emerge to graze on the surface of the coral rubble. This should be verified in future studies to be conducted in King Sound.

The results of the present study indicated that a temperature of 30–33°C and a salinity of 31–37‰ should be maintained during the culture period in the hatchery. To ensure an adequate supply of dissolved oxygen, the tanks must also be well aerated during the culture period. The results also showed that the water temperature for Js production is not limiting on hatcheries operating within the tropics where water temperatures normally fluctuate 28–32°C.

Acknowledgments

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Effects of Different Substrata on the Growth Rate of Early Juvenile *Trochus niloticus* (Mollusca: Gastropoda)

R. Gimin and C. L. Lee*

Abstract

Juvenile trochus, *Trochus niloticus,* were reared for 72 days in three different substrata (fibreglass plate, shell grit and pieces of coral) which were prepared in a similar way and inoculated with the same algal species *Nitzschia* sp. Feeding ad libitum, the best growth rate (101.603 ± 9.851 μm/day) was recorded for juveniles growing on shell grits (SG), slightly lower growth (99.949 ± 6.702 μm/day) was recorded for those growing on coral pieces (CP) and significantly poorer growth (81.489 ± 10.646 μm/day) was noted on juveniles reared on fibreglass plates (FRP). The growth rate of juveniles using FRP as substrata was significantly lower ($P<0.05$) from those juveniles grown on either SG or CP. Differences were also recorded in the survival rates of juveniles grown on the three different substrata. Survival rate was highest (66 ± 7.416%) for the juveniles grown on SG, slightly lower (65 ± 6.124%) for those grown on CP and significantly lower (45 ± 17.678%) for those grown on FRP substrate. The survival rate of juveniles grown on FRP as substrata was significantly lower ($P<0.05$) from those juveniles grown on SG and CP. Based on the results of this study, the hatchery system developed at the Northern Territory University has chosen an appropriate substrate for the biological filter of the closed recirculating system.

MOST intertidal benthic invertebrates use the substrata as a source of food, for attachment and for shelter, as well as protection either from predators or adverse environmental conditions, e.g. dislodgment by wave actions, tidal currents or prolonged exposure to desiccation. The effects of substrata on different groups of marine invertebrates have been reviewed by Newell (1979), Bacescu (1985), Crisp and Bourget (1985) and Dall et al. (1990). The most common substratum used for increasing the surface area for growing benthic algae in trochus hatcheries is corrugated PVC plates (Shokita et al. 1991) or fibreglass plates (C. Lee, pers. comm.).

In the Northern Territory University (NTU) trochus hatchery, newly settled postlarvae were allowed to grow and graze on benthic algae growing on surfaces of shell grits (SG) that made up the filter materials of the biological filter bed of the closed recirculating system. However, a few weeks after metamorphosing to postlarvae, the young juveniles overgrazed the benthic algae on the SG and mass mortality from starvation occurred unless additional food grown on suitable substrata was provided.

This paper reports a series of experiments carried out to determine the effects of growth and survival of hatchery-grown juvenile trochus *Trochus niloticus* using fibreglass plates (FRP), shell grit (SG) and coral pieces (CP) as substrata. They were conducted to test the hypothesis that when food (benthic algae) is provided ad libitum, the nature and type of substrata would have no effect on the survival and growth rates of juveniles maintained in the hatchery.

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Materials and Methods

Experimental animals and conditions

Five hundred one-month-old juveniles growing in the closed recirculating system of NTU hatchery (Lee, these Proceedings) were removed on 23/12/95 for experimentation. The juveniles were transferred into a clean glass aquarium 35 cm × 20 cm × 20 cm where they were held for two days without food. After that period, they were counted and 20 juveniles were transferred into individual experimental unit (EU) and reared for 72 days. During the experimental period, the growth rates were determined by measuring the maximum width of the basal diameter of up to 50% of the juveniles found in each EU. As they grew bigger, all the juveniles in the EUs were measured fortnightly and the mortality recorded.

Each EU was made of a 10 L plastic round pail with the top half cut off leaving a volume of about 5 L. The bottom of the pail had an area of 220 cm^2. During the experiment, each EU was supplied with 4 L of saline bore water at 35 ppt which was pretreated according to the method of Lee (these Proceedings). The EUs were placed within a large water-filled shallow circular tank (diameter 1.1 m) which was placed outdoors under a waterproof shadehouse (Fig. 1). This arrangement produced less violent changes in the water temperature of the EUs, which fluctuated between 28 and 33°C during the experiment.

During the period of study, selected diatom-coated substrata (SG, CP and FRP) up to 10 mm diameter were provided and spread evenly on the bottom of each EU. The water in the EUs was aerated gently and changed every two days. At each change of water, fresh substrata covered with diatoms were introduced. The types and preparation of the three substrata (FRP, SG and CP) for diatom growth before being used for the experiments are given below.

Substrata

FRP — this substratum was made by cutting 1-mm new corrugated roofing fibreglass sheet into rectangular pieces measuring 30 mm long × 15 mm wide (Fig. 1). This was approximately similar in size to the shell grits used in the present study. Prior to cutting the sheet to size, the surface of the sheet was roughened with sandpaper to provide better algal attachment. It was then washed, rinsed with fresh water and spread across the bottom of a 35-cm diameter plastic basin (algae food production basin) and covered with seawater at 35 ppt.

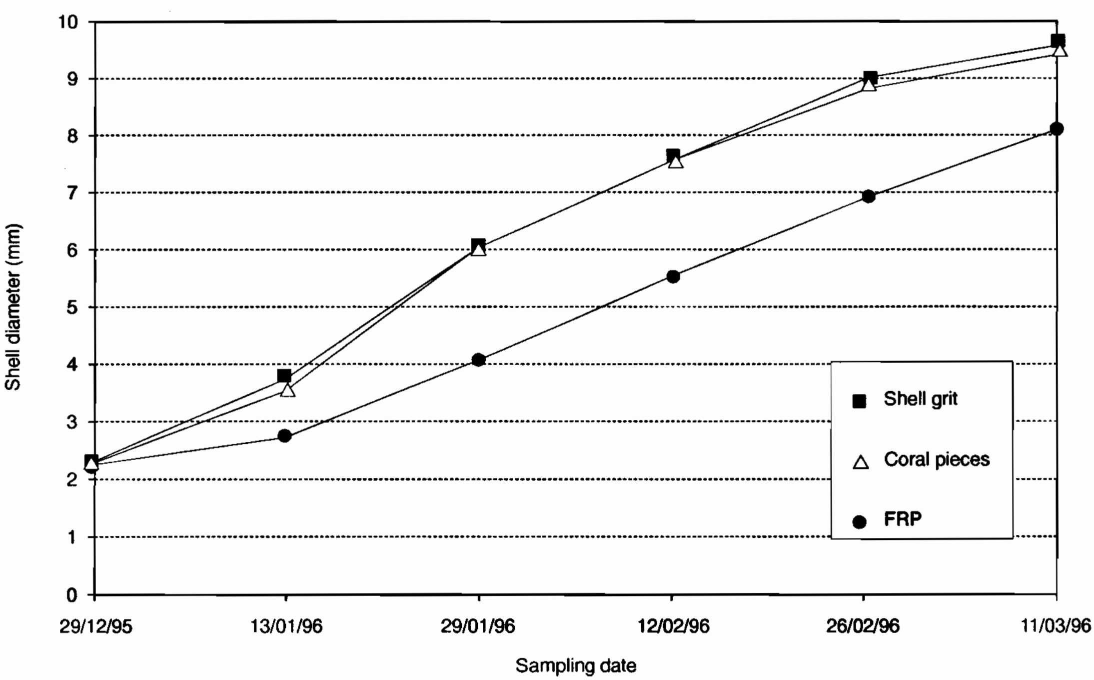

Figure 1. Growth rate of juvenile trochus grown on different substrata (shell growth is given as mean ± SD).

SG — shells consisting mainly of mixed marine bivalves were collected from Darwin Harbour and sun-dried for two weeks. This treatment ensured that no food organisms were present on the substratum. After washing with fresh water, they were redried for two days before being spread across the bottom of a 35-cm diameter algae food production basin and covered with seawater at 35 ppt.

CP — mixed dried coral pieces were collected from the beach off Lee Point Reserve in Darwin, washed with fresh water and left to dry in the sun for three weeks. Selected pieces were broken with a hammer to get small blocks of about 25 mm × 25 mm × 10 mm and washed again before being spread across the bottom of a 35-cm food production basin and covered with seawater at 35 ppt.

Algae production on the substrata

Each algae food production basin containing different substrata was filled with 30 L of saline bore water maintained at 35 ppt. Gentle aeration was provided with the help of an airstone connected to all the basins. The sessile diatom *Nitzschia* sp. was initially cultured in f/2 medium before been inoculated on the substrata in the basin. To promote the diatom growth, a commercially marketed complex fertiliser Aquasol™ was added at the rate of 5 ppm. After one week, a thin brownish layer of diatom mat covered the substrata; these substrata were used for feeding juvenile trochus by transferring to the EUs when required. During the experimental period, production of the diatom-coated substrata was maintained by regularly introducing new substrata and inoculating the medium with fresh diatoms.

Experiment design and statistical analysis

The experiment was designed to test the effect of growth rates and survival of juvenile trochus grown on three different substrata (treatments) when fed ad libitum; each treatment had five replicates. The experiment was carried out from 29 December 1995 for 72 days. Growth rate of juveniles was recorded by measuring the basal shell diameter (µm/day). Survival rate was counted at the end of the experiment.

Data on growth rate were subjected to log-transformation and compared by one-way ANOVA followed by Fisher's PLSD at P=0.05. Survival rate between substrata was initially arc-sine transformed before applying the same analysis. All statistical tests were done using the Statview5+ package.

Results

The daily growth (in µm/day) and survival (in %) rates of juveniles grown on the various substrata during the experimental period are summarised in Table 1. Fortnightly shell growth using the three substrata over the experimental period is illustrated graphically in Figure 1.

The analyses of variances indicated that there were differences in the growth rate of juveniles between substrata. The highest daily growth was observed in juveniles reared on SG (101.603 ± 9.851 µm/day) although this was not significantly different (P>0.05) with the growth rate of juveniles (99.947 ± 6.702 µm/day) reared in CP. Significantly poorer growth rate was achieved by juveniles grown on FRP (81.489 ± 10.646 µm/day) (P<0.05).

There were also differences in survival rates between substrata. SG gave the highest survival rate (66 ± 7.416%). This was followed by the juveniles grown on CP which showed similar survival rates (65 ± 6.124%). The lowest survival rate was recorded by juveniles grown on FRB plates (45 ± 17.678%); the result was significantly lower (P<0.05) than those recorded for SG and CP substrata.

Table 1. Growth and survival of trochus juveniles reared for 72 days on different substrata.

Substrata	Mean shell diameter (mm)		Shell growth mm/day	Survival rate (%)
	29/12/95	11/03/96		
Fibreglass plate	2.237 ± 0.104	8.105 ± 0.770	81.49 ± 10.65[a]	45 ± 17.68[a]
Shell grit	2.273 ± 0.029	9.588 ± 0.685	101.60 ± 9.85[b]	66 ± 7.42[b]
Coral pieces	2.277 ± 0.079	9.473 ± 0.549	99.95 ± 6.70[b]	65 ± 6.12[b]

Values followed by different superscripts are significantly different (P<0.05).

Discussion

The results of the current study indicated that under hatchery conditions and in the presence of suitable food provided ad libitum, the type and nature of the substratum can have measurable effects on the growth rates and survival of early juveniles.

A substrate may influence the trochus juveniles in two different ways. It may affect the juveniles (a) directly, i.e. as a substrate for attachment or as a source of food or shelter, or (b) indirectly through regulating the type and abundance of food types growing on its surfaces.

The direct effects of a substratum on the growth and survival of trochus juveniles could be manifested in numerous ways. It could be possible that juvenile *T. niloticus* obtained part of its nourishment in the form of inorganic substances while scraping the rocks and coral rubble on which it lived (Rao 1937). If this is true, then the CP used in the present study would be the most suitable substratum since it is more easily subjected to the effect of erosion by scraping than the harder and inert SG. As the growth of juveniles using CP is not significantly different from those grown on SG ($P>0.05$), it would appear that the substratum itself does not contribute directly to the nutrition of the animal.

It is more likely that the nature of the substratum may influence juvenile growth and survival directly via the provision of shelter and longer feeding opportunities. Trochus is a nocturnal animal and feeds during night hours; its activities and feeding are inhibited by sunlight (Heslinga 1981). Due to its homogenous thickness, FRP tend to pack together and do not provide hiding spaces to the juveniles. By necessity, the juveniles are therefore limited to the upper surface of the substratum where high light intensity may cause reduction of feeding activities and increased stress or the animals. On the other hand, the uneven nature of SG and CP provides ideal surfaces for shelters and feeding at most hours of the day. If this is true, it is possible that the significantly higher growth and survival rates recorded ($P<0.05$) for juveniles held in SG and CP in comparison with those provided with FRP as a substratum are due to their living in a lower-stress environment where activities and feeding are less impeded for longer hours each day.

Indirectly, a substrate may influence the juveniles through regulating the type and abundance of food types growing on its surfaces. The nature and composition of the substratum, for example, may influence and determine the type of algae attached to it (Wood 1985; Hartog 1985). In the present study, the three substrata were inoculated with similar kinds of algae and *Nitzschia* sp. was observed to colonise the surfaces as the dominant species. Therefore it is unlikely that there are marked differences in the composition of the microalgae food found growing on surfaces of the substrata. In any case, food is not a limiting factor in the experiment as the animals were fed ad libitum. In addition, the lower survival rate of juveniles maintained on FRP would result in a greater abundance of food available to the surviving juveniles. Since the survivors do not show faster growth rate, it would be sensible to deduce that factors other than food must be affecting the growth and survival of test animals growing on FRP.

The present study confirms the suitability of natural substrata over artificial ones as better for maintaining juveniles. This generally supports the preliminary study of Dobson (1994) who reported that juveniles growing on shell grits and coral pieces as substrata produced a higher growth rate when compared to juveniles grown with FRB as substratum.

Acknowledgments

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A Report on Collections of Benthic Marine Algae from Darwin, Northern Australia

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Abstract

Benthic marine algae were collected in Darwin Harbour and environs, Northern Territory, Australia, in connection with the Trochus Research Program at the Northern Territory University. A total of 100 taxa has been identified and classified according to division, order, family, genus, and species. Some collections have been tentatively identified only to the generic level. These taxa represent 62 Rhodophyta (Red Algae), 16 Phaeophyta (Brown Algae), and 22 Chlorophyta (Green Algae).

THIS report on the identification of benthic marine algae from Darwin Harbour on the north coast of Australia is part of the nutrition studies in the *Trochus* Research Program at the Northern Territory University (NTU). Marine macroalgae are the principal food source of a large number of marine vertebrate and invertebrate herbivores (Hawkins and Hartnoll, 1983). This is also the case for the herbivore *Trochus niloticus* Linn. Their guts were found to contain a large quantity of Cyanophyceae and brown algae, and, to a lesser extent, other small red and green algae (Asano 1944, cited by Bour 1990). Although there have been studies on the use and dietary value of macroalgae for abalone and other gastropods (Hahn 1989), little information is available on the topic regarding trochus. The purpose of this work was to take stock of what benthic marine algae are available within Darwin Harbour (12°26′S, 130°51′E) and make the information available for the trochus nutrition section of the project. The collections in this report have been deposited in the Herbarium of the University of Michigan (MICH), Ann Arbor, and also in the personal herbarium of the second author.

Studies of algae of the northern coast of Australia date back to Sonder (1871) and include such reports as Womersley (1958) on Arnhem Land. Yet compared with the present status of our knowledge of benthic marine algae of eastern, southern and western Australia, knowledge of algae occurring on the coast of northern Australia lags relatively far behind. Lewis (1984, 1985, 1987) has compiled inventories of benthic marine algae for northern Australia, and these are very useful lists of species and literature from scattered sources. Recent publications include collections from northern Australia such as of *Gracilaria* (Withell et al. 1994) and mangrove algae (King and Puttock 1989, 1994a,b).

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List of Taxa

RHODOPHYTA	
NEMALIALES	
Galaxauraceae	
Galaxaura rugosa (Ellis & Solander) Lamouroux	Colls.: NTU 97; NTU 99
Gloiophloea sp.	Coll.: NTU 100
Tricleocarpa fragilis (Linn.) Huisman & Townsend	Colls.: NTU 44; NTU 98 (cystocarpic); NTU 163 (cystocarpic); NTU 168 (cystocarpic)
GELIDIALES	
Gelidiaceae	
Pterocladia caerulescens (Kütz.) Santelices	Coll.: NTU 101
Gelidiellaceae	
Gelidiella acerosa (Forsskål) Feldm. & Hamel	Coll.: NTU 143 (tetrasporic)
Gelidiella myrioclada (Børges.) Feldm. & Hamel	Coll.: NTU 201
CORALLINALES	
Corallinaceae	
Amphiroa crassa Lamouroux	Colls.: NTU 60; NTU 165
Amphiroa foliacea Lamouroux	Colls.: NTU 102; NTU 204
Amphiroa fragilissima (Linn.) Lamouroux	Colls.: NTU 60; NTU 70
Jania adhaerens Lamouroux	Colls.: NTU 103; NTU 165-B
GIGARTINALES	
Rhizophyllidaceae	
Portieria hornemannii (Lyngbye) P. C. Silva	Colls: NTU 2; NTU 43 (cystocarpic); NTU 54; NTU 175-C
Peyssonneliaceae	
Peyssonnelia inamoena Pilger	Coll.: NTU-11
Caulacanthaceae	
Catenella nipae Zanardini	Colls: NTU 104 (tetrasporic); NTU 105; NTU 166 (male and cystocarpic)
Hypneaceae	
Hypnea boergesenii Tak. Tanaka	Coll.: NTU 167 (tetrasporic)
Hypnea pannosa J. Agardh	Coll.: NTU 106 (cystocarpic)
Hypnea spinella (C. Agardh) J. Agardh	Colls.: NTU 27 (tetrasporic); NTU 28 (cystocarpic and tetrasporic); NTU 79; NTU 107 (tetrasporic); NTU 180
Hypnea valentiae (Turn.) Mont.	Colls: NTU 6 (tetrasporic); NTU 108
Solieriaceae	
Eucheuma denticulatum (N. L. Burman) Collins et Hervey	Coll.: NTU 83
Solieria robusta (Grev.) Kylin	Colls.: NTU 21 (tetrasporic); NTU 49 (cystocarpic); NTU 72 (cystocarpic)
Halymeniaceae	
Halymenenia floresii (Clemente) C.Agardh	Coll.: NTU 13
Halymenia porphyraeformis Parkinson	Coll.: NTU 7 (cystocarpic)

Corynomorphaceae	
Corynomorpha prismatica (J. Agardh) J. Agardh	Coll.: NTU 200
GRACILARIALES	
Gracilariaceae	
Gracilaria arcuata Zanardini	Coll.: NTU 109
Gracilaria canaliculata Sond.	Coll.: NTU 10; NTU 110

Xia (1986) proposed to merge this species within *Gracilaria salicornia* (C. Agardh) Dawson. Withell et al. (1994) provisionally accepted Xia's taxonomic judgment, but at the same time that also recognised 'a *Gracilaria canaliculata* Sonder form of *Gracilaria salicornia*' as occurring in northern and western Australia as well as New Caledonia, Sri Lanka, and India. Wynne (1995) recognised two distinct species in material occurring in the Seychelles Islands: plants of *G. canaliculata* have non-constricted or barely constricted axes, and plants of *G. salicornia* have strongly and regularly constricted axes.

Gracilaria eucheumoides Harvey	Coll.: NTU 82
Gracilaria preissiana (Sond.) Womersley & Min-Thien	Coll.: NTU 30
Gracilaria salicornia (C. Agardh) Dawson	Coll.: NTU 89 (tetrasporic); NTU 164 (cystocarpic)
Gracilaria sp. A	Coll.: NTU 68
Gracilaria sp. B	Coll.: NTU 87
Gracilaria sp. C	NTU 4
RHODYMENIALES	
Rhodymeniaceae	
Botryocladia leptopoda (J. Agardh) Kylin	Coll.: NTU 9
Ceratodictyon spongiosum Zanardini	Coll.: NTU 153
Gelidiopsis intricata (C. Agardh) Vickers	Coll.: NTU 111 (tetrasporic)
Champiaceae	
Champia indica Børgesen prox.	Coll.: NTU 5 (tetrasporic)
Champia parvula (C. Agardh) Harv.	Colls.: NTU 112; NTU 165-A (cystocarpic)
CERAMIALES	
Ceramiaceae	
Centroceras clavulatum (C. Agardh) Mont.	Coll.: NTU 34; NTU 39
Ceramium clarionense Setch. & N. L. Gardn.	Coll.: NTU 174; NTU 175-B
Ceramium sp. A	Coll.: NTU 205
Ceramium sp. B	Coll.: NTU 164-A
Spyridea filamentosa (Wulfen) Harv.	Coll.: NTU 80 (cystocarpic)
Wrangelia tayloriana Tseng	Coll.: NTU 32 (tetrasporic)
Delesseriaceae	
Caloglossa monosticha Kamiya (Kamiya et al., 1997)	Coll.: NTU 143-A
Hypoglossum simulans M. J. Wynne, I. Price & D. L. Ballant.	Coll.: NTU 114 (tetrasporic)
Dasyaceae	
Heterosiphonia crassipes (Harv.) Falkenberg	Coll.: NTU 25 (immature tetrasporangial stichidia)
Rhodomelaceae	
Acanthophora muscoides (Linn.) Bory	Coll.: NTU 42
Acanthophora spicifera (Vahl) Børgesen	Coll.: NTU 115

Rhodomelaceae (cont'd)

Bostrychia tenella (Lamouroux) J. Agardh var. *tenella*	Coll.: NTU 116
Chondria riparia (J. Agardh) DeToni	Coll.: NTU 117

Chondria riparia was distributed by Harvey as No. 16 in Friendly Island Alg. Exsicc. The taxon was validated by J. Agardh (1852) under the name *Chondriopsis riparia* and later transferred to *Chondria* by DeToni (1903). Grunow (1874) stated that the species is distinguished by its dark colour and its small cortical cells, which are seldom up to twice as long as broad. The Darwin collection was compared with a syntype of this species in MICH and observed to be in good agreement.

Chondria sp. A	Coll.: NTU 118
Chondria sp. B	Coll.: NTU 206
Chondria sp. C	Coll.: NTU 155
Chondria sp. D	Coll.: NTU 160
Herposiphonia secunda (C. Agardh) Ambronn f. *secunda*	Coll.: NTU 165-C (tetrasporic)
Laurencia majuscula (Harv.) Lucas	Colls.: NTU 51 (cystocarpic); NTU 119; NTU 120; NTU 182

This species is related to *Laurencia obtusa* (Huds.) Lamouroux, but the surface cells near branch apices are convex and projecting. This material is in agreement with Saito's (1969) characterisation of this species. Saito and Womersley (1974) stated that *L. majuscula* has a broad distribution in tropical, subtropical and temperate Pacific and Indian Ocean waters.

Laurencia glandulifera (Kütz.) Kütz.	Colls.: NTU 29; NTU 90

Silva et al. (1996) have proposed that the name *Laurencia glandulifera* (Kütz.) Kütz. has priority over the two taxonomic synonyms L. *paniculata* (C. Agardh) J. Agardh and *L. patentiramea* (Mont.) Kütz. Earlier, Silva et al. (1987) accepted Ardissone's (1883) treatment of *L. paniculata* as a taxonomic synonym of *L. patentiramea*. This species seems to have a broad distribution in temperate and tropical waters (Yamada 1931; Tseng 1943; both as *L. paniculata*). Saito and Womersley (1974, as *L. paniculata*) described the occurrence of this species in southern Australia.

Laurencia sp. A	Coll.: NTU 33 (tetrasporic)
Laurencia sp. B	Coll.: NTU 157
Leveillea jungermanioides (Hering & Martens) Harv.	Colls.: NTU 121; NTU 175-A
Polysiphonia sp. A	Coll.: NTU 24
Polysiphonia sp. B	Coll.: NTU 207
Tolypiocladia calodictyon (Harv. ex Kütz.) P. C. Silva	Coll.: NTU 3 (cystocarpic and tetrasporic); NTU 208
Tolypiocladia glomerulata (C.Agardh) Schmitz	Coll.: NTU 175

PHAEOPHYTA

ECTOCARPALES

Ectocarpaceae

Feldmannia indica (Sond.) Womersley & A. Bailey	Coll.: NTU 203

DICTYOTALES

Dictyotaceae

Dictyopteris polypodioides (DeCandolle) Lamouroux	Coll.: NTU 122; NTU 185
Dictyopteris woodwardia (Turner) Schmidt	Coll.: NTU 123

Dictyotaceae (cont'd)

The margins of the blades in this species are beset with minute teeth. This species was described by Turner (1811) from the north coast of Australia. Its range includes northern and eastern Australia (Lewis 1985) and also India (Børgesen 1937) and Sri Lanka (Durairatnam 1961). Wynne (1995) pointed out that 'woodwardia' rather than 'woodwardii' is the correct epithet in that Turner (1811) explicitly employed the former to commemorate both T. J. Woodward and the fern genus, which this brown alga resembles.

Dictyota cervicornis Kütz.	Coll.: NTU 84

Although not previously reported from northern Australia (Lewis 1985), the range of this species, which was first described from Florida (Kützing, 1859), apparently includes the Indo-Pacific: Fiji Islands (Kapraun and Bowden 1978; Ajisaka and Enomoto 1985), and Singapore (Wei and Chin 1983).

Dictyota ciliolata Kütz. var. *lata* Womersley	Coll.: NTU 85

Womersley (1985) based his account of this variety on a collection from Arnhem Land.

Dictyota Sp. A (possibly a juvenile stage of *D. dichotoma*).	Coll.: NTU 18
Dictyota Sp. B (with alternate branching)	Colls.: NTU 124
Padina australis Hauck	Colls: NTU 20; NTU 125; NTU 126
Padina boryana Thivy in W. R. Taylor	Colls.: NTU 88; NTU 183

Although the name *Padina tenuis* Bory has been applied to this species (Womersley and Bailey 1970; Allender and Kraft 1983; Lewis 1985), Papenfuss (1977) has presented reasons for the retention of the name *P. boryana*. Another name that has been used for this alga is *Padina commersonnii* Bory, which Papenfuss (1977) asserted to be a superfluous and thus illegitimate name. Tanaka and Nozawa (1962, as *P. commersonii*) depicted plants from southwestern Japan, illustrating the rhizomatous bases that were also present in the Darwin material.

Padina tetrastromatica Hauck	Colls.: NTU 41; NTU 127
Padina sp. A	NTU 158
Spatoglossum asperum J. Agardh	Coll.: NTU 128; NTU 186
SCYTOSIPHONALES	
Scytosiphoaceae	
Hydroclathrus clathratus (Bory) Howe	Coll.: NTU 129
Rosenvingea nhatragensis Dawson	Coll.: NTU 95
FUCALES	
Sargassaceae	
Sargassum decurrens (Turner) C. Agardh	Coll.: NTU 187
Cystoseiraceae	
Cystoseira trinodis (Forsskål) C. Agardh	Coll.: NTU 209
CHLOROPHYTA	
ULVALES	
Ulvaceae	
Enteromorpha flexuosa (Wulfen ex Roth) J. Agardh	Coll.: NTU 21
Enteromorpha sp. A. (simple)	Coll.: NTU 159
Enteromorpha sp. B (branched)	Coll.: NTU 156
CLADOPHORALES	
Anadyomenaceae	
Anadyomene brownii (J. E. Gray) J. Agardh	Coll.: NTU 1; NTU 61
Cladophoraceae	
Rhizoclonium riparium (Roth) Harv.	Coll.: NTU 91

CAULERPALES	
Bryopsidaceae	
Bryopsis sp. A	Coll.: NTU 210
Caulerpaceae	
Caulerpa lentillifera J. Agardh	Colls.: NTU 45; NTU 86; NTU 131; NTU 149; NTU 171; NTU 178
Caulerpa manorensis Nizamuddin	Colls: NTU 15; NTU 148
Caulerpa racemosa (Forsskål) J. Agardh var. *laetevirens* (Mont.) Weber-van Bosse	Colls.: NTU 132; NTU 172
Caulerpa racemosa (Forrsk.) J. Agardh var. *lamourouxii* (Turn.) Weber Bosse	Coll.: NTU 177
Caulerpa serrulata (Forsskål) J. Agardh var. *serrulata* f. *lata* (Weber-van Bosse) Tseng	Colls: NTU 133; NTU 162; NTU 179
Caulerpa sertularioides (S. G. Gmelin) Howe	Colls: NTU 134; NTU 135; NTU 162
Caulerpa vesiculifera Harv.	Coll.: NTU 14
Udoteaceae	
Avrainvillea lacerata J. Agardh	Coll.: NTU 49
Halimeda borneensis W. R. Taylor	Colls: NTU 21; NTU 136; NTU 137; NTU 152
Halimeda macroloba Decaisne	Coll.: NTU 211
Halimeda opuntia (Linn.) Lamouroux	Colls: NTU 17; NTU 138
Halimeda taenicola W. R. Taylor	Coll.: NTU 212
Halimeda tuna (Ellis & Solander) Lamouroux	Coll.: NTU 16; NTU 139; NTU 140; NTU 141; NTU 142
SIPHONOCLADALES	
Siphonocladaceae	
Boergesenia forbesii (Harv.) Feldm.	Coll.: NTU 169
Valonia aegagropila C. Agardh	Colls: NTU 47; NTU 144
DASYCLADALES	
Dasycladaceae	
Neomeris van-bosseae Howe	Coll.: NTU 170

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Chemical Composition of Algae for Use in *Trochus niloticus* Studies

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Abstract

The proximate biochemical composition (carbohydrate, lipid and protein) of 12 species of brown, green and red benthic macroalgae that could be useful in the aquaculture of *Trochus niloticus* were investigated to assess their nutritional value. Five samples of naturally occurring mixed benthic microalgae from NTU Aquaculture Centre and the dominant diatom, *Navicula* sp., were included for comparison. The lipids were further analysed for fatty acid composition to determine levels of long-chain polyunsaturated fatty acids (PUFAs), including eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), which have been reported as essential to the survival and growth of juvenile stages of many molluscs.

The proximate analysis of macroalgae found carbohydrate as the major biochemical component (8.3–22.1% dry weight) followed by protein (1.2–9.0%) and lipid 1.8–7.8%). The mixed benthic macroalgae were found to have higher percentages of lipid (10.1–21.2% dry weight) and protein (19.5–48%) than seaweeds, and lower levels of carbohydrate (9.1–16%). The nutritive value of green, blue-green and red macroalgae may be influenced by high levels of inorganic matter (ash) within the range 61–75% of total dry weight. Brown macroalgae had lower levels of inorganic matter (31–47%) while lowest amounts were found in benthic microalgae (12–36%). The results are comparable with literature values for macroalgae and benthic chrysophytes.

High amounts of EPA (22.9% of total fatty acids) were found in the red macroalgae *Tolypiocladia glomerulata* and the benthic diatom *Navicula* sp. (14.2%). None of the macroalgae examined contained amounts of DHA higher than 0.9%, while the mixed microalgae ranged from 0.4–1.8%.

Overall, all algae species studied contained adequate levels of protein, lipid and carbohydrate for gastropod nutrition, but benthic microalgae had superior amounts of protein and lipid and the essential fatty acids EPA and DHA.

THIS is a report of chemical analyses made in support of nutrition studies in the Trochus Research Program at the Northern Territory University (NTU).

A major limiting factor in the cultivation of gastropods such as abalone and topshell under aquaculture is the availability of suitable algal rations. Many studies have reported on the use and dietary value of macroalgae for abalone, *Haliotis* spp. and other herbivorous gastropods (Uki et al. 1986; Hahn 1989; Mercer et al. 1993), but little has been reported of the nutritional value and chemical composition of algae that may be useful in topshell *Trochus niloticus* culture.

The early literature reported that the gut contents of a large number of *T. niloticus* contained approximately 10% macroalgae and microalgae together with 90% inorganic debris including crushed coral, sand, fragments of foraminifera, sponges, hydroids, crustaceans and molluscs (Rao 1937; Ansano 1944). Later studies confirmed that *T. niloticus* consumes large quantities of sand and sediment, together with minor amounts of low, filamentous brown and green macroalgae, coralline algae and microalgae growing on the surface of rocks and dead corals (McGowan 1956; Heslinga and Hillman, 1981; Nash 1985; Hahn 1989; Isa 1991).

The optimum nutritional requirements of *T niloticus* are not known, but the closely related gas-

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tropod *Haliotis* spp. (abalone) requires dietary protein within the range 15–28% dry weight, dietary lipid 3–5% dry weight and carbohydrate 20–30% (Uki and Watanabe, 1992; Mercer et al. 1993). These requirements were based on maximum growth rate, muscle yield and feed conversion efficiency. Many tropical macroalgae and microalgae have levels of protein, lipid and carbohydrate within these ranges and were included in this study.

The juvenile stages of many marine organisms, including larval and juvenile stages of bivalves (Langdon and Waldock, 1981), molluscs (Uki et al. 1986) and prawn larvae (Kanazawa et al. 1977), require one or both of the polyunsaturated fatty acids (PUFAs) eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) for survival and growth.

For this reason, the fatty acid composition of each alga was investigated.

The aims of this study were:

(i) to determine the proximate chemical composition, including ash, carbohydrate, lipid and protein, of 12 species of macroalgae and three benthic microalgae used in nutrition studies in the Trochus Research Program at the NTU, and

(ii) to investigate levels of long-chain polyunsaturated fatty acids, including eicosapentaenoic acid and docosahexaenoic acid, in the 15 species.

Materials and Methods

Macroalgal collection

Seaweeds were collected from the surface rocks and coral reefs in King Sound, Western Australia, where *Trochus niloticus* occurs naturally, and from rocks and coral reefs of the intertidal zone off East Point and Nightcliff Beach, Darwin Harbour, Northern Territory. The seaweeds included macroalgae of the divisions Chlorophyta (green algae), Phaeophyta (brown algae) and Rhodophyta (red algae) (Table 1). Samples were collected into plastic bags, stored on ice, frozen within two hours and transported to NTU. In the laboratory, seaweeds were washed with seawater to remove sand and surface debris, rinsed with 0.5 M ammonium formate to remove sea salt, freeze-dried, ground and stored at –75°C prior to chemical analysis.

Source of microalgae

Naturally occurring benthic microalgae, including *Navicula* sp. and two prymnesiophytes, were isolated from the surface of tanks at NTU Aquaculture Centre and duplicate cultures of each species grown in 1.5 L of medium f/2 (Guillard and Ryther, 1962) in 3 L conical flasks under standard NTU conditions (pH 8.3±0.2, temperature 25±1°C, photon flux density 80±2 μmol/m^2/s and salinity 25±1 ppt). Cultures were harvested by centrifugation in late logarithmic phase of growth, rinsed with 0.5 M ammonium formate, freeze-dried and stored at –75°C prior to chemical analysis.

Analytical methods

For each species, duplicate samples were analysed for total carbohydrate, total protein, total lipid, total ash (inorganic matter) and fatty acid composition.

Total carbohydrates were determined by the colorimetric method of Dubois et al. (1956), after extraction with 0.5 M H_2SO_4. Total lipid was analysed gravimetrically after extraction with chloroform-methanol (2:1) by the method of Bligh and Dyer (1959). Total inorganic matter (ash) was determined gravimetrically after heating at 550°C in a Heraeus Thermicon muffle furnace. (For further details of these methods see Renaud et al. 1991).

Total nitrogen in seaweeds from Darwin Harbour was determined by the Kjeldahl semi micro-digestion method with potentiometric endpoint detection following digestion with concentrated H_2SO_4 and $CuSO_4$ catalyst at 390°C (Windrift block digester) for two hours (for details see Lambrinidis 1994). The total nitrogen content of seaweeds collected from King Sound and of microalgae was determined by Flow Injection Analysis (Lachat 8000). For all samples, total protein was calculated from total Kjeldahl nitrogen (%) × 6.25.

The extracted lipids from each species were converted to fatty acid methyl esters (FAME) by digestion with 14% BF_3-methanol complex under high purity nitrogen at 60°C for two hours. FAME samples were extracted into hexane, reduced to 1 mL under a stream of nitrogen and analysed immediately or stored at –75°C. Fatty acid compositions investigated were analysed using a Varian Vista 6000 gas chromatograph with split-splitless injector (temperature 250°C; split ratio 1:60) and flame ionisation detector (temperature 300°C), using a BP225, polar capillary column (50% cyanopropyl 50% phenyldimethylsiloxane, 25 m × 0.2 mm internal diameter (id)). Data were collected and processed using a Varian Star chromatography system. Individual fatty acids were identified by comparing the retention times with commercially available external standards (Sigma, USA). FAME were then further analysed by GC-MS (Varian Saturn 3) with split-splitless injector and mass selective detector, using a non-polar column (DB-5MS, 30 m × 0.25 mm id) to confirm the identification of fatty acids. The shorthand notation used in fatty acid nomenclature is $L{:}B(n{-}x)$ where L = chain length, B = number of double bond and $(n{-}x)$ = position of the ultimate double bond from the terminal methyl group. More details of these methods are given in Renaud et al. (1994).

Results and Discussion

Proximate chemical analysis

The proximate analysis of seaweeds found carbohydrate the major biochemical component (range 7.5–25% dry weight), followed by protein (1.2–9.0%) and lipid (1.8–7.8%) (Table 1). The highest amount of protein was found in the brown macroalga (Phaeophyta) *Padina boryana* collected from both King

Table 1. Proximate chemical composition (ash content, total carbohydrate, total lipd and total protein) of macroalgae collected from King Sound, Western Australia, and/or Darwin Harbour, Northern Territory, and of benthic microalgae isolated from NTU Aquaculture Centre.

Code	Location	Proximate analysis (% dry weight)	Ash	CHO	Lipid	Protein
MACROALGAE						
Chlorophyta (green)						
Boodlea sp.	KS5	WA	61.1	8.3	4.0	3.3
Halimeda borneensis	GL11	NT	75.3	17.5	4.1	2.7
Cyanophyta (blue-green)						
Symploca sp.	GL12	NT	70.4	15.2	4.2	3.1
Phaeophyta (brown)						
Dictyota ciliolata	GL8	NT	47.2	15.2	7.8	4.1
Pachydicyons sp.	KS6	WA	36.6	10.5	2.1	6.2
Padina australis	GL13	NT	47.2	20.5	4.7	6.1
P. boryana	KS1	WA	34.6	9.7	7.4	9.0
P. boryana	GL3	NT	36.8	21.4	5.5	8.1
Rosenvingea nhatrangensis	GL14	NT	45.4	12.1	3.2	1.2
Sargassum sp.	KS2	WA	31.4	8.5	5.9	4.1
Sargassum sp.	GL2	NT	45.2	12.6	2.6	3.4
Rhodophyta (red)						
Acanthopora muscoides	GL6	NT	65.5	19.4	1.8	3.6
Hypnea sp.	GL10	NT	62.6	17.2	5.8	6.4
Tolypiocladia glomerulata	GL7	NT	64.4	22.1	2.1	4.2
MICROALGAE						
Chryosphyta						
Bacillariophycae (diatom)						
Navicula sp.	NT13	CAS	12.1	16.0	19.0	48.0
Mixed benthic microalgae						
Sample 1	CAS1	CAS	21.7	12.0	21.0	41.0
Sample 2	CAS2	CAS	34.4	11.4	21.2	21.5
Sample 3	CAS3	CAS	33.5	12.1	10.1	24.4
Sample 4	CAS4	CAS	36.2	9.1	17.9	19.5

WA=King Sound, Western Australia. NT=Darwin Harbour, Northern Territory. CAS= NTU Aquaculture Centre.

Sound and Darwin Harbour (9.0 and 8.1% dry weight, respectively). Lowest percentages of protein (2.7–3.3%) were found in green algae (Chlorophyta) and blue-green algae (Cyanophyta). These results are slightly higher than the range of protein contents from 0.5 to 5.8% reported for 21 species of tropical macroalgae (Kumar 1993). *Dictyota ciliolata* contained the highest percentage of lipids (7.8%), closely followed by *P. boryana* collected in Darwin Harbour (7.4%). For comparison, samples of *P. boryana* collected in King Sound had a slightly lower percentage of lipids (5.5%). Total lipid levels found in this study were comparable with or slightly higher than previous reports for tropical green macroalgae (range 0.5–8.4% dry weight), red macroalgae (1.2–3.8%) and brown macroalgae algae (0.5–5.5%) (Banaimoon 1992; Mercer et al. 1993; Mishra et al. 1993). The highest percentage of carbohydrate was found in the red algae *Tolypiocladia glomerulata* (22.1%) but there was a broad range within each macroalgal class (green algae range 8.3–17.5%; brown algae range 8.5–21.4%; red algae range 17.2–19.4%). These percentages are within the ranges reported for the carbohydrate content of tropical brown, green and red macroalga (Kumar 1993; Mercer et al. 1993). These authors noted seasonal variations in carbohydrate content with increases of between 4% and 11% dry weight, depending on the macroalgal species.

The mixed benthic microalgae isolated from NTU Aquaculture Centre were found to have higher percentages of lipid (18.5–19.7% dry weight) and protein (31.5–48.0%) than the seaweeds discussed above, and lower levels of carbohydrate (5.5–13.9%) (Table 1). Highest levels of lipid (19.7% dry weight) and protein (48.0%) were found in the diatom *Navicula* sp. (Phaeophyta). This was the dominant species in the mixed microalgae that grew naturally on corrugated fibreglass sheets suspended in tanks at NTU Aquaculture Centre (Figure 1).

The nutritive value of these components is influenced by higher levels of inorganic matter (ash) in macroalgae (31–75% of total dry weight) than in diatoms (14–26%). In particular, brown, green and red algae were notable for their high ash contents (61.1–75.3% dry weight). The literature does not include ash content of macroalgae with proximate analysis data, but calculations of ash as (100 minus carbohydrate minus lipid minus protein) from published data on tropical species suggest ash contents ranging 47–80% dry weight (from the data of Kumar 1993). The high percentage of inorganic matter could influence the nutritive value of the macroalgal species, as a higher mass of algae must be consumed to give feeding herbivores adequate carbohydrate, lipid and protein.

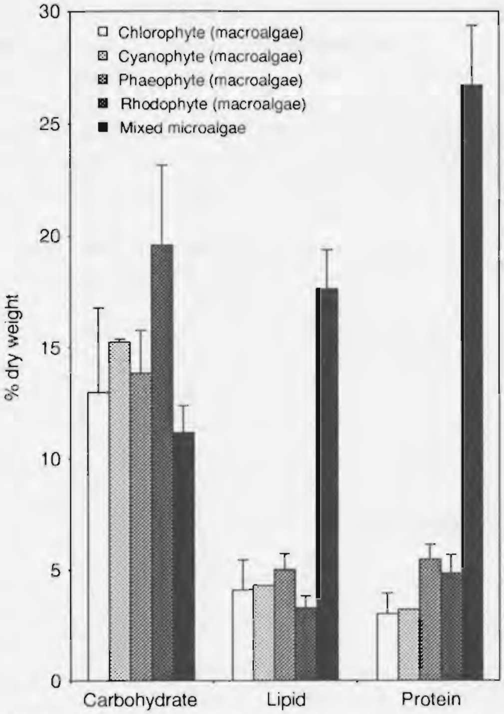

Figure 1. Proximate chemical composition of macroalgae and mixed microalgae.

Fatty acid composition

Major fatty acids

All macroalgae examined had 16:0 as the major fatty acid. Amongst the brown macroalgae, the percentage of 16:0 was within the range 25.8–41.6% of total fatty acids. This fatty acid was found together with moderate amounts of 18:1(n–9) (maximum 20.6%), 18:4(n–3) (maximum 12.6%) and the polyunsaturated fatty acid 20:4(n–6) (maximum 12.6%) (Table 2). Green macroalgae contained 16:0 within the range 33–36.1% and moderate amounts of 14:0, 16:1(n–7) and 18:2(n–6) (maximum amounts 10.9%, 7.7% and 9.4% respectively) (Table 3). Amongst the red macroalgae, the fatty acid 16:0 (range 34.9–38.1%) was present together with relatively high levels of the essential fatty acid 20:5(n–3) (range 8.3–22.9%) and moderate amounts of 14:0 (maximum 16.5%) (Table 3). Only one blue-green macroalga was examined and found to have 16:0 as its principal fatty acid (24.9%) and moderate amounts of 14:0, 16:1(n–7) and 18:1(n–7) (11.2%, 7.9% and 7.1% respectively) (Table 3).

Table 2. Fatty acid composition[a] (% total fatty acids) of benthic macroalgae, Class Phaeophyta (brown algae) collected at two locations. Data given as mean of duplicate analyses; King Sound n=1; Darwin Harbour n-3.

Fatty acid	Phaeophyta							
	King Sound			Darwin Harbour				
	Pachydicticyon sp. KS6	*Padina boryana* KS1	*Sargassum* sp. KS2	*Dictyota ciliolata* GL8	*Padina australis* GL13	*Padina boryana* GL3	*R. nhatragensis* GL14	*Sargassum* sp. GL2
Saturates								
12:0	2.2	1.3	2.7	–[b]	–	–	–	–
14:0	5.7	6.2	5.9	10.4	5.7	6.2	8.0	4.4
16:0	36.9	38.7	41.6	25.8	26.9	32.5	37.5	30.7
18:0	3.6	3.9	5.0	1.8	2.1	1.5	5.2	1.3
Sum %	48.4	50.1	55.2	38.0	34.7	40.2	50.7	36.4
Mono-unsaturates								
16:1	7.1	5.9	10.1	6.5	10.4	5.0	4.7	10.9
18:1 (n–9)	12.5	20.6	9.5	12.9	10.2	12.4	13.3	10.2
18:1 (n–7)	2.6	–	2.7	1.1	0.4	–	–	–
Sum %	22.2	26.5	22.3	20.2	21.0	17.4	18.0	21.1
Polyunsaturates								
16:2	0.4	0.7	0.3	1.7	1.0	1.2	2.9	2.1
16:3	0.5	0.8	0.5	1.3	1.0	0.4	3.1	0.5
18:2 (n–6)	3.8	3.8	3.9	5.2	4.6	4.8	2.6	5.9
18:3 (n–6)	0.8	0.6	0.5	2.3	1.5	1.4	3.0	0.9
18:3 (n–3)	4.4	2.8	2.4	2.3	5.8	6.2	1.7	7.4
18:4 (n–3)	4.4	3.7	1.8	10.6	12.6	12.5	2.2	4.9
20:2	–	–	–	1.0	–	–	–	–
20:4 (n–6)[c]	8.4	5.7	6.6	10.5	8.8	8.2	4.2	13.3
20:5 (n–3)[d]	2.9	–	3.0	3.5	2.5	0.6	3.7	3.2
22:6 (n–3)[e]	0.1	–	0.3	–	–	–	0.5	–
Sum %	25.7	18.1	19.3	38.5	37.8	35.3	24.0	34.0

[a] fatty acids less than 1% of total fatty acids in all species not included: 14:1, 15:0, 17:0, 18:5, 20:0.
[b] less than 0.1%.
[c] arachidonic acid (AA).
[d] eicosapentaenoic acid (EPA).
[e] docosahexaenoic acid (DHA).

Comparing fatty acid compositions of the two macroalgae species *P. boryana* and *Sargassum* sp. collected at both King Sound, WA, and Darwin Harbour, NT, the percentage of the major fatty acid 16:0 was slightly lower for both species collected at King Sound. The percentages of 18:1(n–9) were similar from both locations, while the percentages of 18:4(n–3) and 20:4(n–6) were higher for both species collected in Darwin Harbour (Table 2).

The fatty acid compositions for macroalgae reported in this study were comparable with previous reports for phaeophytes (Johns et al. 1979; Shameel et al. 1991), chlorophytes (Shameel and Khan, 1991; Banaimoon 1992) and rhodophytes (Afaq-Hussain et al. 1992).

The major fatty acid of the benthic microalga *Navicula* sp. was 16:1(n–7) (28.6% total fatty acids) (Table 4). This species contained moderate amounts of 16:0 (20.1%), 20:5(n–3) (14.2%) and 14:0 (10.0%). This fatty acid profile is consistent with previous reports for this species (Al-Hasan et al. 1990). The mixed microalgae were found to have either 16:0 (range 15.4–27.6% total fatty acids) or 16:1(n–7) (range 13.0–26.7%) as the major fatty acid. The percentage of 20:5 (n–3) varied from collection to collection within the range 2.1–7.8% (Table 4 and Figure 2).

Polyunsaturated fatty acids

Considering all macroalgal species, highest levels of polyunsaturated fatty acids (PUFAs) were found in the green alga *Halimeda bornensis* (42.5% of total fatty acids) and the three brown algae *Dictyota* sp., *Padina* spp. and *Sargassum* sp. (37–40% of total fatty acids) (Table 2). High amounts of the essential long-chain PUFA 20:5(n–3) were found in the red algae *Tolypiocladia glomerulata* (22.9%) and *Acanthopora muscoides* (13.4%) (Table 3).

None of the macroalgae had levels of'22:6 (n–3) higher than 1%.

The diatom *Navicula* sp. had a high percentage of PUFA (36.9% total fatty acids) and a high amount of 20:5 (n–3) (14.2%) and a small amount of 22:6 (n–3) (0.4%) (Table 4). The mixed benthic micro-algae contained 20:5 (n–3) within the range 11–17% and were the best source of 22:6 (n–3) (range 0.7–1.2%) (Table 4).

Table 3. Fatty acid composition[a] (% total fatty acids) of benthic macroalgae, Divisions Chlorophyta, Cyanophyta, Rhodophyta (green, blue-green and red algae) collected in Darwin Harbour. Data given as mean of duplicate analyses; Darwin Harbour n-3.

Fatty acid	Chlorophyta		Cyanophyta	Rhodophyta		
	Boodlea sp.	*Halimeda bornensis*	*Symploca* sp.	*Acanthophora muscoides*	*Hypnea* sp.	*Tolypiocladia glomerulata*
	KS5	GL11	GL12	GL6	GL10	GL7
Saturates						
12:0	3.1	–[b]	4.7	1.1	–	1.1
14:0	10.9	6.9	11.2	6.7	16.5	9.3
16:0	36.1	33.0	24.9	38.1	34.9	37.0
18:0	4.1	2.4	1.6	2.0	2.2	1.4
Sum %	54.2	42.3	42.4	47.9	53.6	48.8
Mono-unsaturates						
16:1 (n–7)	7.4	7.7	7.9	4.7	9.8	5.4
18:1 (n–9)	–	3.3	7.1	2.7	7.6	4.3
18:1 (n–7)	10.7	3.1	8.9	10.2	2.8	2.8
Sum %	18.1	14.1	23.9	17.6	20.2	12.5
Polyunsaturates						
16:2	1.5	6.0	6.6	2.6	1.6	0.7
16:3	0.8	0.3	3.6	1.5	1.7	0.6
16:4	4.2	8.3	2.2	–	–	–
18:2 (n–6)	8.5	9.4	1.9	0.9	2.3	1.8
18:3 (n–6)	4.8	1.9	2.6	2.4	1.2	1.1
18:3 (n–3)	0.5	12.9	0.9	1.1	0.8	0.7
18:4 (n–3)	0.5	2.2	2.7	1.5	1.5	1.0
18:5 (n–3)	–	–	1.5	–	–	–
20:4 (n–6)	0.7	0.5	6.8	7.8	4.6	3.7
20:5 (n–3)	0.3	0.5	2.1	13.4	8.3	22.9
22:6 (n–3)	0.9	0.5	–	–	–	–
Sum %	22.7	42.5	30.9	31.2	22.0	32.5

[a] fatty acids less than 1% of total fatty acids in all species not included: 14:1, 15:0, 17:0, 20:0, 20:2.
[b] less than 0.1%.

Table 4. Fatty acid composition[a] (% total fatty acids) of benthic macroalgae isolated from NTU Aquaculture Centre. Data given as mean of duplicate analyses; Darwin Harbour n=1.

Fatty acid	Dominant diatom *Navicula* sp. NT13	Mixed benthic microalgae GL June	CAS 1 August	CAS 2 October	CAS 3 January	CAS 4 March
Saturates						
12:0	–	–	–	–	–	2.7
14:0	10.0	5.4	7.4	19.0	15.9	6.1
16:0	20.1	24.8	15.4	12.8	19.4	27.6
18:0	1.8	2.7	1.6	0.4	1.6	2.4
Sum %	31.9	32.9	24.4	32.2	36.9	38.8
Mono–unsaturates						
16:1(n–7)	28.6	26.7	18.2	21.2	13.0	22.1
18:1(n–9)	0.5	2.2	1.6	0.1	2.3	2.7
18:1(n–7)	0.3	1.7	1.9	1.1	1.7	1.9
Sum %	29.4	30.6	21.7	22.4	17.0	26.7
Polyunsaturates						
16:2(n–7)	6.9	2.6	7.9	4.1	2.9	3.1
16:3(n–4)	–	–	–	2.5	–	2.9
16:4(n–3)	–	–	–	8.3	–	1.0
16:4(n–1)	1.1	3.1	12.6	4.4	6.8	1.8
18:2(n–6)	3.5	2.1	1.5	0.9	4.1	3.2
18:3(n–6)	2.7	1.5	0.7	0.8	1.7	0.9
18:3(n–3)	0.6	2.4	3.8	1.1	3.9	3.3
18:4(n–3)	2.5	2.7	3.0	–	3.1	1.9
20:4(n–6)	5.0	2.9	2.1	4.1	7.8	3.5
20:5(n–3)	14.2	13.0	17.0	15.3	14.0	10.7
22:6(n–3)	0.4	1.8	0.8	0.7	0.8	1.2
Sum %	36.9	32.1	49.4	42.2	45.1	33.5

[a] fatty acids less than 1% of total fatty acids in all species not included: 14:1, 15:0, 17:0, 18:5, 20:0, 20:2.

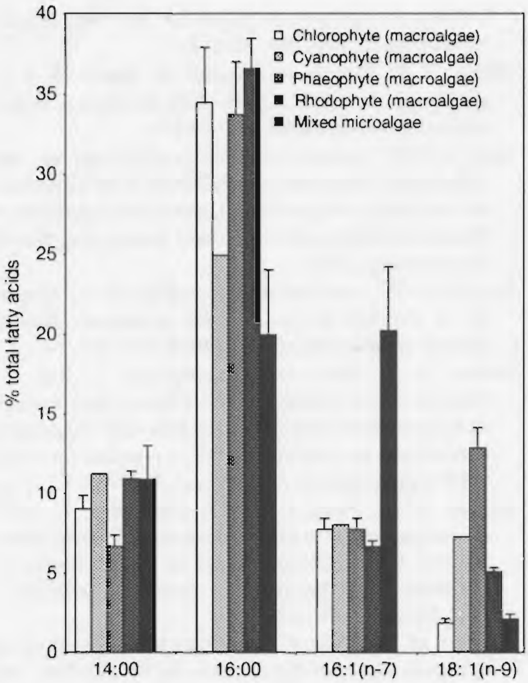

Figure 2. Major C14, C16 and C18 fatty acids of macroalgae and mixed microalgae

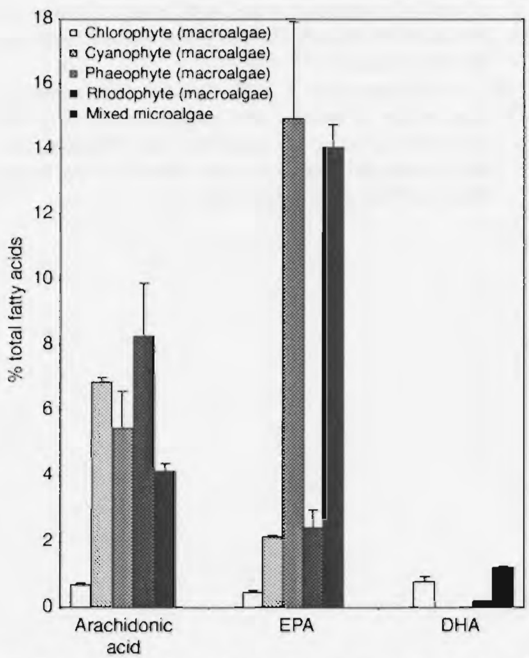

Figure 3. Major long-chain polyunsaturated fatty acids of macroalgae and mixed microalgae

Conclusions

(i) All macroalgae examined contained adequate levels of protein, lipid and carbohydrate. A mixture of red, green and brown algae is needed to ensure adequate levels of the essential PUFAs EPA and DHA.

(ii) Comparing macroalgal species collected at King Sound and Darwin Harbour, there was little difference in proximate chemical composition, but Darwin Harbour samples had slightly higher levels of essential PUFAs.

(iii) The NTU mixed microalgae is of high nutritional quality, containing higher levels of protein, carbohydrate, lipid and essential PUFAs than macroalgae.

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Fatty Acid Composition and Proximate Biochemical Composition of Wild and Hatchery-held Broodstock of the Marine Topshell, *Trochus niloticus* (Mollusca: Gastropoda)

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Abstract

The fatty acid composition and proximate biochemical composition (carbohydrate, lipid, protein and ash) of foot tissue of adult *Trochus niloticus* (Gastropoda: Archaeogastropoda) collected from King Sound, north-west Australia (Group 1), were determined. Results were compared with those for animals from the same collection maintained for a year in the hatchery at Northern Territory University Aquaculture Centre (Group 2). The animals were mature broodstock and there was no significant difference in the total live weight of Group 1 and Group 2 at harvest.

Foot tissue of Group 1 was composed of carbohydrate (3.8% dry weight), lipid (6.1%), protein (77.7%) and ash (5.4%). Animals of Group 2 had ash and protein contents similar to those of Group 1, but there was significantly lower lipid (5.3% compared to 6.1%; $p < 0.05$) and higher total carbohydrate (5.0% compared to 3.9%; $p < 0.05$). There was no significant difference in proximate composition between male and female for either group.

The principal fatty acids (greater than 5% total fatty acids) were palmitic acid (16:0), stearic acid (18:0), oleic acid (18:1 (n–9)) and arachidonic acid (20:4 (n–6)). Percentages of eicosapentaenoic acid (20:5 (n–3)) and docosapentaenoic acid (22:5 (n–3)) in Group 1 animals were lower than percentages reported for other archaeogastropods. There were changes in the fatty acid composition of Group 2 animals which included significant increases in the percentage of polyunsaturated fatty acids 16:3 (n–6), 18:2 (n–6), 20:5 (n–3), 22:5 (n–3) and 22:6 (n–3) and significant decreases in the percentage of saturated fatty acids 16:0 and 18:0. The results for 16:0, 18:0, 16:3 (n–4) and 20:5 (n–3) reflected the fatty acid composition of the hatchery mixed microalgae feed.

THE marine topshell, *Trochus niloticus* (Linnaeus) (Gastropoda: Archaeogastropoda) has been harvested for over a century for the production of mother-of-pearl buttons, furniture inlays, costume jewellery and other products. This small fishery is of economic importance to many countries in the Pacific, Papua New Guinea, Indonesia and Australia (McGowan 1956; Heslinga 1981; Bour et al. 1990). In recent years, price increases for shells of *T. niloticus* have resulted in overfishing and overexploitation of the stocks in many countries in its distributional ranges (Heslinga and Hillmann, 1981; Nash 1985; Bour 1990; Nash 1993) and there are fears that they are endangered in most of their natural fishing grounds. Increasingly, the aquaculture potential of this animal is being investigated.

A review of the literature of the feeding habits of *T. niloticus* indicated that the animal grazes the substrate with a rasp-like radula, consuming large quantities of sand, crushed coral and sediment, together with minor portions of low, filamentous brown and

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green macroalgae (Rao 1937; Ansano 1944; McGowan 1956; Heslinga and Hillman 1981; Isa 1991), microalgae (Rao 1937; McGowan 1956; Heslinga and Hillmann 1981; Isa 1991), coralline algae (Heslinga and Hillmann 1981; Nash 1985; Hahn 1989) and bacteria (Heslinga and Hillmann 1981).

Few, if any, biochemical studies have been reported on the members of the family Trochidae and almost nothing is known about the chemical composition, nutritional value and fatty acid composition of juvenile and adult *T. niloticus*. There are reports of the fatty acid compositions of a large number of marine molluscs including gastropods such as the commercially important abalone (see review by Joseph, 1989) but few reports on the proximate chemical composition of other marine archaeogastropods (Giese 1966; Webber 1970). The principal fatty acids of abalone (family Haliotidae), (Bannatyne and Thomas, 1969; Kochi 1975; Yamada and Hayashi, 1975) and of members of families Patellidae (Gardner and Riley, 1972; Johns et al. 1980) and Trochidea (Johns et al. 1980) are palmitic acid (16:0), oleic acid (18:1(n–9)), arachidonic acid (20:4(n–6)), eicosapentaenoic acid (20:5(n–3)) and docosapentaenoic acid (22:5(n–3)).

As part of the research project on the induced spawning, hatchery production and nutrition of trochus at the Northern Territory University (NTU), this study was conducted to establish the baseline proximate biochemical composition (carbohydrate, lipid, protein and ash) and fatty acid composition of the adult *T. niloticus* collected from the wild, and to compare the results with similar animals maintained for a year in the hatchery. The fatty acid compositions of marine macroalgae collected from the coral reefs where *T. niloticus* occur naturally and of the mixed microalgal diet at NTU were investigated to determine whether the dietary intake was related to the fatty acid composition of *T. niloticus*. The results of this study will provide information on nutritional requirements and feeding conditions necessary for maintaining mature *T. niloticus* broodstock for spawning in the hatchery.

Materials and Methods

Sample source

Fifty individual mature adult *T. niloticus* were collected from King Sound, north-west Australia (17°0′S, 12°30′E) and transported to the NTU Aquaculture Centre. Ten individuals, five males and five females, were weighed (total live weight = W_T; wet weight of meat without shell = W_M), and dissected upon arrival in the laboratory, ground to a fine powder (Retsch automatic grinder), and stored at –75°C until chemical analysis (Group 1). Samples of low, filamentous algae from King Sound were collected into plastic bags from the surface of rocks and dead coral close to browsing *T. niloticus*, stored on ice and frozen within two hours, transported to NTU, freeze-dried, ground and stored at –75°C.

A similar number of *T. niloticus*, five males and five females, were fed on naturally grown mixed microalgae and maintained in a 3000 L aquaculture tank for one year before being harvested, weighed, dissected, ground and stored at –80°C prior to chemical analysis (Group 2). The mixed microalgae, together with minor amounts of microscopic benthic invertebrates, grew naturally on 30 cm × 60 cm corrugated fibreglass sheets suspended in the tanks. The main microalgae species were *Navicula* sp., a benthic diatom, and benthic prymnesiophytes, although the species composition of diatoms changed during the year. Subsamples of mixed microalgae were collected from the plates on different months, freeze-dried and stored at –75°C.

Analytical methods

Duplicate samples of foot tissue of *T. niloticus* were analysed for total ash (inorganic matter), total carbohydrate, total protein, total lipid and fatty acid composition. The ground foot tissue was very hygroscopic and samples were re-freezedried prior to lipid and protein analysis. Algal samples were analysed for fatty acid composition only.

Ash was determined gravimetrically after heating at 550°C, total carbohydrates were analysed by the method of Dubois et al. (1956) and total lipid was determined gravimetrically by the method of Bligh and Dyer (1959). (For more details of these methods see Renaud et al. 1991). Total protein was calculated from total Kjeldahl nitrogen (× 6.25) determined by Flow Injection Analysis (Lachat 8000) following digestion of samples of ground *T. niloticus* (20 mg), with concentrated sulfuric acid and $CuSO_4$ catalyst at 390°C (Windrift block digester) for two hours.

Fatty acid methyl esters (FAME) were prepared by direct transesterification of lipid extracts with 14% BF_3-methanol complex at 60°C for two hours. FAME samples were dried under a stream of nitrogen and redissolved in a known amount of hexane.

The gas chromatographic analysis of the methylated fatty acids was performed on a Varian Vista 6000 GC equipped with a BP225, polar capillary column (50% cyanopropyl 50% phenyl dimethylsiloxane, 25 m × 0.2 mm internal diameter (id)), and an FID detector. The individual fatty acids were identified by comparing the retention times with commercially available external standards (Sigma, USA). FAME were then analysed by GC-MS (Varian Saturn 3) with split-splitless injector and mass selective detector, using a non–polar column (DB-5MS, 30 m × 0.25 mm id) to confirm the identification of fatty acids. More details of these methods are given in Renaud et al. (1994). Data were processed by a Varian Star chromatography system. Fatty acid structure was represented as L:B(n–x) where L=chain length, B=number of double bond and (n–x) = position of double bond closest to the terminal methyl group.

Statistical analysis

Data were compared by one-way analysis of variance (ANOVA) with group or sex as the independent variable and dependent variables W_T, W_M, protein, carbohydrate, lipid and individual fatty acids.

Results and Discussion

Body weight

Body weights of *T. niloticus* broodstock at harvest are shown in Table 1. There was no significant difference between groups in the total live weight, W_T (Group 1 mean 149 g; Group 2 mean 164 g) or in the wet weight of meat without shell, W_M (mean 25.6 g and 29.5 g respectively). There was no significant difference between male and female of either group.

Proximate chemical analysis

The proximate analysis of foot samples of Group 1 (Table 1) found ash 5.4% of dry weight, total carbohydrate 3.8%, total lipid 6.1% and total protein 77.7% (Table 1). *T. niloticus* of Group 2 had a small but significant decrease of 0.8% in total lipid content ($p < 0.05$) and a small increase of 1.3% in total carbohydrate ($p < 0.05$), when compared with Group 1 (Table 1). These were only minor changes and may indicate that the gross chemical composition of the hatchery diet was comparable with the diet available to animals at King Sound. There was no significant difference between male and female for any of the analyses in either group.

Comparing the results for Group 1 *T. niloticus* with those for muscle of other gastropods, the total carbohydrate was slightly lower than the reported range of 5–23% dry weight in the abalone, *Haliotis* sp. (Webber 1970), the total lipid content was within the range reported for foot tissue of *Haliotis* spp. (4–8% dry weight: Giese 1966; Webber 1970), and the total protein content was higher than the range reported by those authors (46–60% dry weight).

Table 1. *Trochus niloticus* from King Sound, northwest Australia: W_T, W_M and proximate chemical composition of foot (ash content, total carbohydrate, total lipid and total protein; % dry weight).

Organism	Sex	W_M[a] (g)	W_M[a] (g)	Proximate analysis (% dry weight) Ash	CHO[b]	Lipid	Protein
Group 1							
	F[c]	153 (19.0)	27.3 (4.3)	5.3 (0.5)	3.8 (0.5)	6.3 (0.9)	76.6 (2.2)
	M[d]	144 (16.0)	24.3 (2.9)	5.5 (0.6)	3.8 (1.3)	6.0 (1.0)	78.8 (1.7)
	Combined	149 (17.2)	25.9 (3.8)	5.4 (0.6)	3.8 (0.9)*	6.1 (0.9)*	77.7 (2.2)
Group 2							
	F[c]	163 (39.6)	30.7 (8.1)	4.6 (1.1)	4.5 (0.6)	5.4 (0.7)	78.2 (3.5)
	M[d]	164 (80.8)	28.3 (11.3)	6.0 (0.4)	5.8 (0.4)	5.3 (0.5)	79.5 (2.5)
	Combined	164 (60.0)	29.5 (9.3)	5.3 (1.1)	5.1 (0.8)*	5.3 (0.6)*	78.9 (3.0)

[a] (Mean (standard deviation), n=5)
[b] Carbohydrate
[c] Female
[d] Male
* Significant difference (p<0.05) between combined group 1 and group 2.

Fatty acid composition

The principal fatty acids of foot tissue of *T. niloticus* collected from the wild (Table 2, Group 1) were 16:0 (34.9% total fatty acids), 18:0 (8.4%), 18:1(n–9) (5.5%) and 20:4(n–6) (12.7%). The percentage of 16:0 was higher than levels reported in foot tissue of two other archaeogastropod species, *Haliotis discus* (Kochi 1975) and *Patella peroni* (Johns et al. 1980) (21.9% and 9.1% respectively), while the percentages of the polyunsaturated fatty acids (PUFAs) 20:5(n–3) (2.3%) and 22:5(n–3) (3.4%) were 3–6% lower than previously reported levels for *H. discus* and *P. peroni* (Kochi 1975; Johns et al. 1980).

The fatty acid composition changed after one year under aquaculture. *T. niloticus* broodstock held in aquaculture tanks showed significantly lower values of the saturated fatty acids 16:0 ($p<0.05$), and 18:0 ($p=0.0001$) (Table 2; Group 2). This contributed to lower (39.1% versus 49.6%) total saturated fatty acid content. There were significant increases in the polyunsaturated fatty acids 20:5 (n–3) ($p<0.05$), 22:4 (n–6) ($p=0.0001$), 22:5 (n–3) ($p<0.01$) and 22:6 (n–3) ($p=0.0001$) for Group 2 animals.

Table 2. *Trochus niloticus* foot from King Sound, northwest Australia: fatty acid composition of Group 1 and Group 2 broodstock. Data given as mean (standard deviation); n=5.

Fatty acid	Group 1			Group 2		
	Female	Male	Combined	Female	Male	Combined
Saturates						
14:0	2.5 (0.3)	2.2 (0.4)	2.4 (0.4)	2.1 (0.3)	2.2 (0.3)	2.1 (0.3)
16:0	35.9 (2.0)	33.9 (5.8)	34.9 (4.2)[b]	32.1 (2.8)	31.1 (1.5)	31.6 (2.2)[b]
18:0	9.8 (1.2)[c]	7.0 (1.9)[c]	8.4 (2.1)[b]	3.3 (0.3)	3.5 (0.1)	3.4 (0.2)[b]
20:0	1.7 (0.8)	0.8 (0.5)	1.2 (0.8)	2.2 (0.4)	2.0 (1.2)	2.1 (0.3)
Sum %	49.9	43.9	46.9	39.7	38.8	39.1
Mono-unsaturates						
14:1	1.3 (0.1)	1.3 (0.2)	1.3 (0.2)	1.3 (0.1)	1.2 (0.2)	1.3 (0.1)
16:1	0.6 (0.2)	1.8 (0.8)	0.8 (0.6)	0.9 (0.2)	0.8 (0.1)	0.8 (0.2)
18:1(n–9)	5.8 (0.3)	5.2 (0.7)	5.5 (0.6)[b]	4.6 (0.7)	4.0 (0.3)	4.3 (0.6)[b]
18:1(n–7)	3.1 (0.4)	2.9 (0.2)	3.0 (0.4)	2.9 (0.3)	2.7 (0.3)	2.6 (0.3)
20:1	1.8 (0.9)	2.9 (0.5)	2.4 (0.9)	3.1 (0.7)	3.4 (0.2)	3.3 (0.5)
Sum %	12.6	14.1	13.0	12.8	12.1	12.3
Polyunsaturates						
16:2(n–7)	1.6 (0.2)	1.5 (0.3)	1.5 (0.2)[b]	2.1 (0.1)	2.0 (0.1)	2.0 (0.1)[b]
16:2(n–4)	4.2 (1.0)	2.6 (0.5)	3.4 (1.5)	3.3 (0.7)	3.7 (0.6)	3.5 (0.7)
16:3(n–4)	1.9 (0.8)	1.9 (0.8)	1.9 (0.8)[b]	5.3 (0.7)	5.4 (0.5)	5.4 (0.5)[b]
18:2(n–6)	3.1 (0.5)	3.5 (1.6)	3.3 (0.1)[b]	4.2 (1.0)	5.1 (0.7)	5.0 (0.8)[b]
18:3(n–6)	0.4 (0.8)	[a]	0.2 (0.6)[b]	1.9 (0.4)	1.7 (0.2)	1.8 (0.3)[b]
18:3(n–3)	2.2 (0.8)	2.1 (0.7)	2.1 (0.7)	1.7 (0.6)	1.4 (0.3)	1.6 (0.5)
20:4(n–6)	11.5 (1.9)	13.8 (3.1)	12.7 (2.7)	13.2 (2.4)	3.7 (0.5)	13.5 (1.7)
20:5(n–3)	1.9 (0.3)[c]	2.7 (0.5)[c]	2.3 (0.6)[b]	2.7 (0.2)	2.9 (0.2)	2.8 (0.2)[b]
22:4(n–6)	3.1 (0.3)	4.2 (1.3)	3.7 (1.1)[b]	1.2 (0.1)	1.3 (0.2)	1.3 (0.1)[b]
22:5(n–3)	2.7 (0.4)	4.1 (0.7)	3.4 (1.4)[b]	5.0 (1.6)	5.2 (0.6)	5.1 (1.1)[b]
22:6(n–3)	2.2 (0.3)[c]	3.6 (1.0)[c]	2.9 (1.0)[b]	5.1 (0.5)	5.6 (0.7)	5.3 (0.6)[b]
Sum %	34.8	40.0	37.4	45.7	48.0	47.3

[a] Less than 0.1%.
[b] Significant difference ($p<0.05$) between combined group 1 and group 2.
[c] Significant difference ($p<0.05$) between female and male.

This contributed to the 10% increase in total polyunsaturated fatty acid content of Group 2 animals which were held in NTU for one year (47.3% versus 37.4%) (Table 2).

Dietary fatty acids

There are many reports of the relationship between the fatty acid composition of the diet with that of lipids of marine invertebrates, including molluscs (see review by Joseph 1989). The relationship between the fatty acid composition in algal diet species and the fatty acids in gastropods that feed on the algae is well established (Bannatyne and Thomas, 1969; Kochi 1975; Johns et al. 1980). Kochi (1975) investigated both muscle and viscera of *H. discus* and related the high levels of the fatty acids 20:4(n–6) and 20:5(n–3), especially in the viscera, to the diet of red and brown algae. Johns et al. (1980) studied the fatty acid composition of the limpet, *Cellana tramoserica*, collected from two sites, one with a predominance of green algal feed species and the other with mixed red and brown algae. These workers reported higher levels of 14:0, 16:0, 18:1(n–9) and 18:1(n–7) in *C. tramoserica* (whole animal only) feeding on green algae, and higher levels of 20:4(n–6) and 20:5(n–3) in those feeding on red and brown algae.

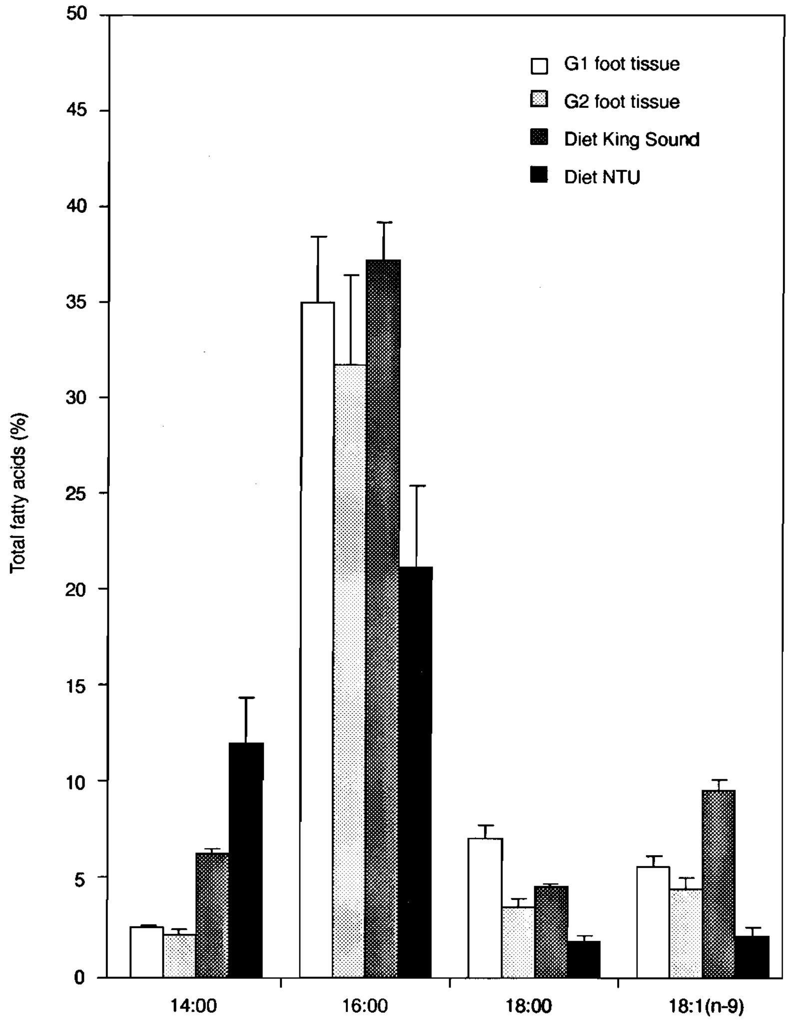

Figure 1. Comparison of selected saturated and monounsaturated fatty acids in foot tissue of *T. niloticus* (n = 10) and of the mixed algal diets (n = 5) consumed by groups 1 and 2. Error bars show standard deviation from the mean.

In a comparison of the major fatty acids in the mixed macroalgae from King Sound with those of the mixed microalgal diet at NTU, the present study found significantly higher levels of the saturated fatty acids 16:0 and 18:0 in the King Sound macroalgae ($p < 0.01$ in each case). The high levels of these fatty acids in Group 1 *T. niloticus* could could be attributed to these high levels in the diet (Fig. 1). On the other hand, the NTU hatchery feed was dominated by mixed diatoms (brown algae) with lower levels of saturated fatty acids and significantly higher levels of polyunsaturated fatty acids 16:3(n–4), 20:5(n–3) and 22:6(n–3) ($p = 0.04$, 0.001 and 0.028 respectively). Again, the significantly higher levels of these PUFAs in Group 2 animals could be related to higher levels in the diet (Fig. 2). However, there was no consistent relationship between percentages of other fatty acids in *T. niloticus* foot tissue and percentages in the microalgal feed. The overall increase in the total PUFA content in Group 2 animals is important as recent studies have established that a higher level of some PUFAs is essential for maturation in females, for the production of high-quality eggs and for survival and growth of early stages of most marine species (Ackman 1967; Watanabe 1982; Sargent et al. 1989).

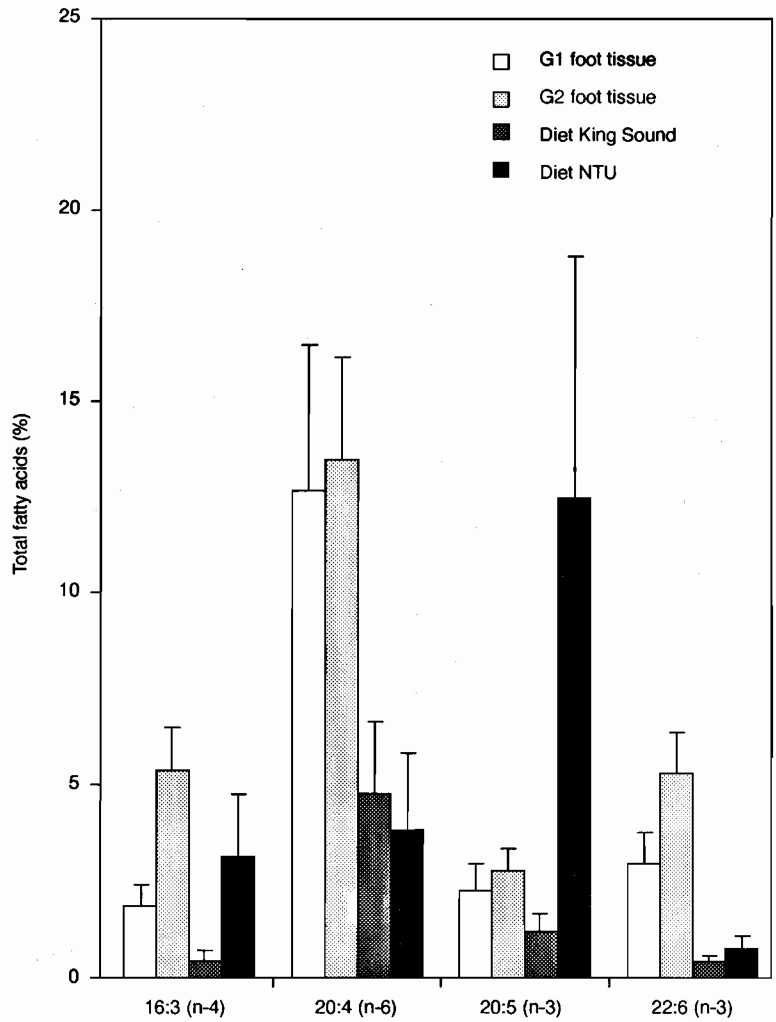

Figure 2. Comparison of selected polyunsaturated fatty acids in foot tisssue of *T. niloticus* (n = 10) and of the mixed algal diets (n = 5) consumed by groups 1 and 2. Error bars show standard deviation from mean.

The results of the present study indicate that *T. niloticus* broodstock can be maintained successfully under hatchery conditions using mixed microalgae growing on corrugated fibreglass sheets. The growth of hatchery-produced juvenile trochus and the changes in their biochemical composition as they mature are under further investigation at NTU.

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Food Preference of *Trochus niloticus* Fed Algae from Darwin Harbour

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Abstract

Food preference of adult trochus, *Trochus niloticus* L., was investigated by estimating the feeding index and food consumption for 10 species of seaweeds (*Halimeda borneensis, Symploca* sp., *Dictyota ciliolata, Padina australis, Padina boryana, Rosenvingea nhatrangensis, Sargassum* sp., *Acanthophora muscoides, Tolypiocladia glomerulata* and *Hypnea* sp.) and a mixed microalgal diet.

It was found that trochus ate larger amounts of the soft filamentous forms (e g. *Symploca* sp., *Hypnea* sp., and *Tolypiocladia glomerulata* and the corticated *Acanthophora muscoides* than the leathery brown algae (*Rosenvingea nhatrangensis, Sargassum* sp. and *Padina* spp.) and the calcareous green algae (*Halimeda borneensis*). When food preference was expressed as feeding indices, it was found that there was no correlation between structural characteristics of the diets and feeding index. The mixed microalgal diet scored the highest index.

THE tropical topshell *Trochus niloticus* is a large gastropod mollusc belonging to the family Trochidae. It is an Indo-Pacific species and is endemic to the waters of the Indo-Australian Archipelago (McGowan 1970). In Australia its range is restricted to northern Australia and extends from the Great Barrier Reef (GBR) to the waters of north western Australia (Moorehouse 1932). The animals are large and slow moving, and are therefore easily surveyed by walking along the reefs at low tide or by diving using scuba.

Trochus animals seem to favour the windward zone of coral reefs (Shokita et al. 1991; Nash 1993) and actively move out over the substratum in search of food at night and hide in crevices during the day or when exposed by low tides (Moorehouse 1932).

Trochus is an Archaeogastropod (Bour and Gohin 1985; Nash 1993). It eats mainly filamentous and microscopic algal forms because of the structure and function of the radulae (Steneck and Waitling 1982). Despite this, large seaweeds of the orders Chlorophyceae, Phaeophyceae and Rhodophyceae have been found in the stomach contents of 20 adult trochus (Nagao Asano 1944). However, there are no experimental data to confirm that trochus feeds on both microalgae and macroalgae.

This paper studies the feeding of trochus using 10 macroalgae and a mixed microalgal diet by measuring feed consumptions and feeding indices.

Methods and Materials

All trochus animals were supplied by Dr Chan Lee, School of Biological Sciences of the Northern Territory University (NTU). Ten species of seaweeds were collected at low tide from the intertidal areas off Fannie Bay, East Point and Nightcliff beaches within the Darwin Harbour (Table 1).

The seaweed samples were carefully detached from the substrates with their holdfast intact and placed in sealable plastic bags with a little seawater and transported to the aquaculture complex within 1–2 hours of collection. At the complex the algae were washed with clean 30 ppt salinity borewater to remove sand and debris and then placed into plastic

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colanders (30-cm diameter) which were floated on the surface of the water in a 1500-L fibreglass tank in the shadehouse. The algae were kept in the tanks up to 23 days (except *Sargassum* sp., 79 days) before being used.

Table 1. Algal species used for the feed preference experiments.

Species	Collection site
Bacillariophyceae	
Diatoms	NTU trochus holding tanks
Chlorophyceae	
H. borneensis	East Point
Cyanophyceae	
Symploca sp.	East Point
Phaeophyceae	
D. ciliolata	East Point
P. australis	East Point, Nightcliff
P. boryana	Fannie Bay, Nightcliff
R. nhatrangensis	Nightcliff
Sargassum sp.	East Point
Rhodophyceae	
A. muscoides	Fannie Bay, East Point, Nightcliff
T. glomerulata	East Point
Hypnea sp.	East Point

Experimental design

The consumption rates of algae by adult *T. niloticus* were studied by feeding experiments carried out in 60-L aquaria filled with 35 L of filtered (through 45μm bag) bore water fitted with a 2-L shell grit biofilter with airlift (Fig. 1). Four adult trochus (60–110 mm maximum basal diameter) were randomly selected, numbered with a permanent ink marker (Artline 700) and starved for two days, before they were placed in each experimental tank with a known amount (wet weight) of seaweed in the centre. To monitor the algal growth and/or degradation during the experimentation period, a control for each diet was set up in the same conditions, but without the animal. Four replicates for each alga (except *P. boryana, n*=2) were set up. The food preference experiments using seaweeds were compared with the normal diet of the benthic microalgae. For this experiment, the microalgal food was available on the surface of fibreglass plates (25 cm × 10 cm dimension). The food was placed in tanks using random numbers.

The wet weight of the algae was measured at the beginning and the end of the experiment to estimate the amount of seaweeds being consumed by the animals. The difference in wet weights, corrected for the control value, indicates the amount of algae consumed by the trochus animals in two days of feeding. The amount of microalgae was estimated similarly; however three plates (25 cm × 10 cm) were cut from existing plates and used for each tank and were replaced after the first day of feeding.

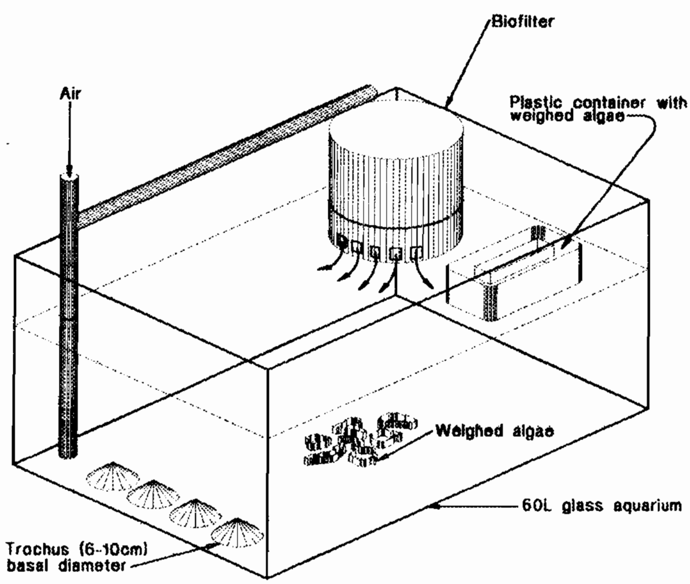

Figure 1. Diagram of the 60-L aquarium used for the food preference experiment.

The feeding on seaweed by the animals was observed during the night since trochus are nocturnal animals (Heslinga 1981a,b). Observations were made every half-hour from 7:30 p.m. to 2:00 a.m. the next morning for two consecutive days. The score was recorded as positive when the trochus mouth was extended fully onto the seaweeds or microalgal plate The feeding index was calculated as follows:

$$\text{Feeding index } (\%) = \frac{\text{Number of positive scores}}{\text{Total number of observations}} \times 100$$

The data were analysed using ANOVA. Wherever required, the dataset was arcsine square root transformed to meet ANOVA assumption of normality. Data ranking was done using Tukey HSD multiple comparisons.

Results

Algal consumption

All diets offered to adult trochus were consumed, except *Sargassum* sp. (Fig. 2). Diets can be grouped into three groups, depending on the level of consumption. Thus *Symploca* sp., *Hypnea* sp., *Rosenvingea nhatrangensis* and *Acanthophora muscoides* were in the highly consumed group; *Tolypiocladia glomerulata, Padina australis* and *Dictyota ciliolata* were the moderately consumed group; and mixed microalgae, *Halimeda borneensis* and *Padina boryana*, the least consumed group.

There is a significant difference between the mean weight of each algal species consumed as determined by a simple one-factor ANOVA ($P = 0.006$). A Tukey HSD multiple comparisons analysis shows that this difference is between the most consumed (first of the pair) and the least consumed (second of the pair) algal species as indicated below:

* *Symploca* sp. and *Sargassum* sp. ($P = 0.019$)
* *Symploca* sp. and *H. borneensis* ($P = 0.043$)

The brown alga *P. boryana* was not included in this analysis as there were only two experiments using this species (due to availability). The amount of *P. boryana* consumed is less than *H. borneensis* and the diatoms, and it therefore most likely to be significantly different to the *Symploca*.

Feeding frequency

Since there was no difference between the feeding index of day 1 and day 2 (data not shown), the results were pooled for further analysis. It was found that all diets were eaten by the trochus (Fig. 3). The diets can again be grouped into three groups based on feeding index. Those with highest feeding indices were mixed microalgae and *P. boryana*. Those with moderate feeding indices were *Symploca* sp., *Hypnea* sp., *T. glomerulata* and *P. australis*. Those with low feeding indices were *D. ciliolata, A. muscoides, R. nhatrangensis, H. borneensis* and *Sargassum* sp. (Fig. 3).

Discussion

The results confirm the observations by Asano (1944) that trochus animals feed on both seaweeds and microalgae. Like other gastropods (Steneck and Waitling 1982), trochus animals feed mostly on diets with soft thalli and have difficulty consuming those having structural toughness.

All filamentous algae used in this work (Table 2) were consumed in greatest quantities, the exception being *R. nhatrangensis*, a leathery macrophyte which was consumed comparably with the filamentous algae. However, this algae exhibited one of the lowest feeding frequencies. This algae has a hollow structure which is able to retain water if damage occurs to its thalli. Large weight losses of this species of up to 20% were also observed in the control. Therefore, to obtain more accurate data, the duration of the experiment could have been extended with more replicates to obtain a better estimate of consumption. This was not possible due to availability of the algae. All other leathery brown macrophytes were not consumed as much as the filamentous forms, which is consistent with the model.

P. boryana was not readily consumed even though trochus were frequently observed on this algae. This may be due to the animals inability physically to remove the tissue from the tough thallus (Table 1). It was noted, however, that the trochus scraped off material from the growing apices of the thalli. This is consistent with the findings of Cronin and Hay (1996) which suggest that this is in response to 'higher levels of chemical defences in older tissues, and not tissue toughness or nutrient value', although the effect of algal toughness has been negatively correlated with the food preferences of abalone. However, Leighton (1966) noted that several species of herbivorous invertebrates consumed tougher algal species (*Eisenia* sp., *Laminaria* sp., *Pterogyphora* sp. and *Cystoseira* sp.) more than the relatively tender algae (*Macrocystis* sp. and *Eregia* sp.) offered to them. It was suggested that chemical perception may have been an important factor affecting selection.

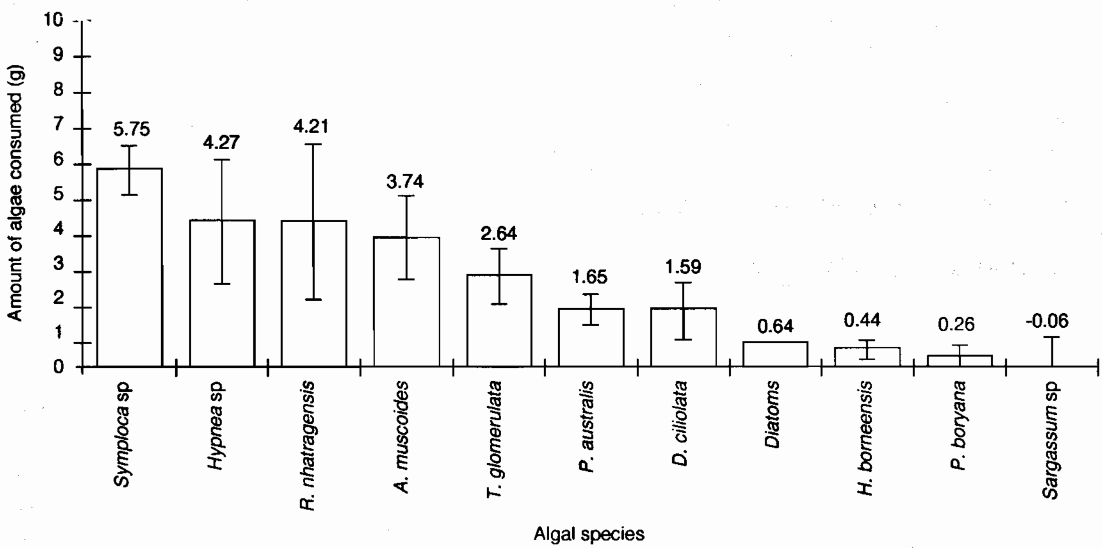

Figure 2. Weight of algae (g) consumed by adult *T. niloticus*, expressed as mean ± SE (n = 4), except *P. boryana*, mean ± range (n = 2). Average tank water temperatures ranged 23.5–29.5°C at 7:00 p.m. and 21.7–28.1°C at 12:00 a.m. The pH of the water in the experimental aquaria ranged 7.7–8.5 at 7:00 p.m. and 12:00 a.m. The values on top of the bars represent the means.

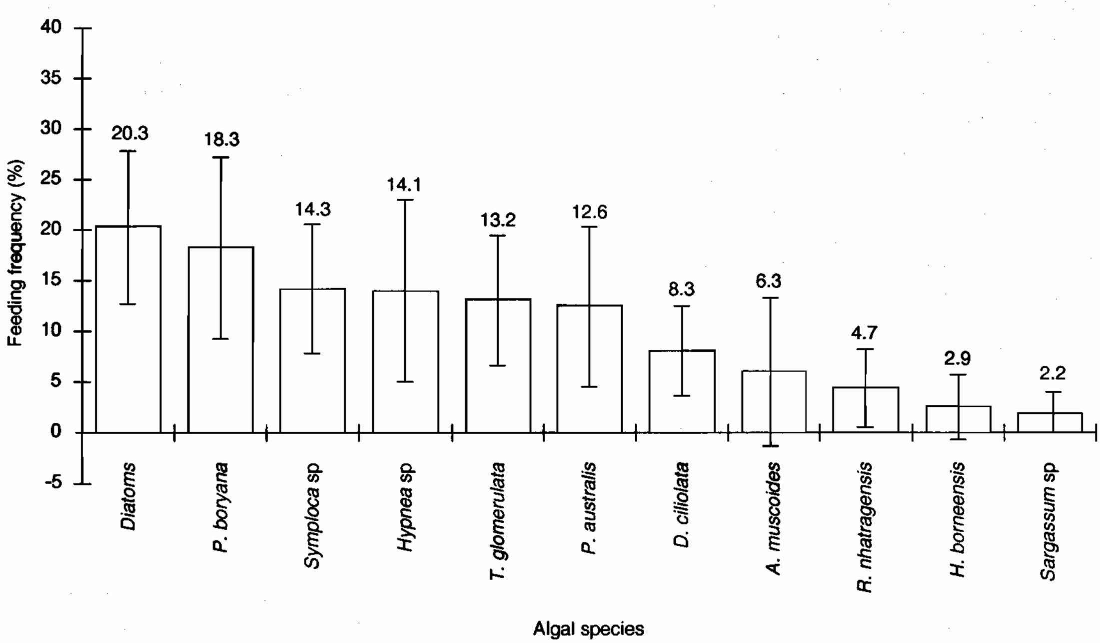

Figure 3. Mean feeding index (%) of adult *T. niloticus* fed 11 algal diets, n=4, except *P. boryana*, n=2. Bars indicate standard error except for *P. boryana* (range). The values on top of the bars represent the means.

Table 2. Algal morphology, after Steneck and Waitling (1982). Algae are ranked by increasing grazing difficulty.

Species of algae	Functional form group
Benthic diatoms	Microalgae
Symploca sp.	Soft filamentous (blue-green algae)
Hypnea sp.	Filamentous algae
T. glomerulata	Filamentous algae (spongy)
A. muscoides	Corticated macrophyte
P. boryana	Leathery macrophyte
P. australis	Leathery macrophyte
D. ciliolata	Leathery macrophyte
R. nhatrangensis	Leathery macrophyte
Sargassum sp.	Leathery macrophyte
H. borneensis	Articulated corticated macrophyte

The blue-green alga *Symploca* sp. has very soft filamentous thalli. The trochus were highly attracted to this algae, and it was the most eaten of all the algae (Fig. 2). Trends that appear in both the consumption and the feeding index experiments are summarised below:

Hypnea sp. > *T. glomerulata* > *P. australis* > *D. ciliolata* > *H. borneensis* >*Sargassum* sp.

The feeding index data are for the untransformed results, resulting in very large standard deviations. Figure 3 shows the feeding index (%) for all the algae in descending order. The animals prefer the diatoms, which were available evenly and are easily grazed algal forms. The attractiveness of the red algae may be due to their relatively tender tissues compared to the brown algae and the calcareous green algae.

Conclusion

The highest feeding indices were recorded for the benthic diatoms, the soft filamentous forms (*Hypnea* sp., *T. glomerulata* and *Symploca* sp.) as well as the corticated red algae *A. muscoides* and the leathery brown algae (*P. boryana*). There was no correlation between feeding indices and amount of algae consumed. For example, the leathery brown seaweed *P. boryana* gave a high feeding index but the amount consumed was low.

Acknowledgments

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Preliminary Investigation of an Artificial Diet for the Marine Topshell, *Trochus niloticus* (Mollusca: Gastropoda)

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Abstract

Seven artificial diets containing differing combinations of casein, fishmeal, soybean meal, semolina, cornstarch or whole-wheat flour were prepared with formulations based on published artificial diets for abalone. Diets were tested for salt-water stability using hourly measurements of turbidance for six hours and after 18 hours. The artificial Diet E containing fishmeal (20% dry weight), soybean meal (30%) gelatine (15%), whole-meal wheat flour (30%) and agar (5%) was found to have the lowest 16-hour water turbidance.

Diet E was further modified to include one of the attractants black pepper (Diet E1), cinnamon (Diet E2), powdered *Spirulina* sp. (Diet E3) or peanut butter (Diet E4). The feeding attraction of these diets to juvenile *Trochus niloticus* was tested over the 10-hour afternoon and evening period of maximum feeding activity. Hourly observations were made of the location of each animal. It was found that Diet E3 had the highest positive response index (65% of observations found animals close to the diet). Lower positive responses were recorded to Diet E1 (53%), Diet E2 (51%), Diet E4 (46%) and the control (41%). The feeding attraction of Diet E3 to mature *T. niloticus* was tested over a 10-hour afternoon and evening period with hourly observations of animal location. Animals with access to the experimental diet E3 spent 80% of their time close to or on top of the diet. Control animals, with no access to the experimental diet, spent 40–45% of their time at either end of the tank and 15% of time in the centre. The gross chemical composition of the most attractive diet, Diet E3, was carbohydrate (33.0% dry weight), lipid (10.2%), protein (42.1%) and ash (2.5%).

Few, if any, research groups have investigated artificial diets for the topshell *Trochus niloticus*, so the extensive literature on artificial diets for abalone has been used as a starting point for research. Japanese and Chinese researchers have been investigating artificial feeds for abalone for many years (Ogina and Ohta, 1963; Uki et al. 1986) and both Japan and China now produce high quality commercial diets for use in abalone aquaculture (Fleming et al. 1995). In recent years, production of artificial feeds for abalone has commenced in Australia (Gorfine 1991; Hanna 1992), South Africa (Britz 1993) and New Zealand (Flemming et al. 1995), and research into formulations is in progress in Korea, France and Mexico (see review by Flemming et al. 1992).

In a review of the development of artificial diets for abalone, Flemming et al. (1995) found that the most common protein sources in abalone diets were fishmeal, defatted soy meal and casein. A few diets included supplements of synthetic amino acids lysine and arginine. The lipid content was usually 3–5% (dry weight) of diet, and was supplied as fish oil or vegetable oil. The requirement of abalone for highly unsaturated fatty acids of the (n–3) and (n–6) families was found to be 1% of a diet containing 5% lipid (Uki

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and Watanabe 1992). Energy and carbohydrate made up 30–60% of artificial diets and was provided in the form of cheap cereals such as soybean meal, wheat or corn flour and maize or rice starch. Starch could be both an energy source and a binder. Other binders that have been used are dextrin, sodium alginate and gelatin, but may authors keep details of binders and feed attractants confidential. Little information is available about levels of vitamins and minerals. Most follow the Uki test diet with 1.5% vitamins and 4% minerals, and many include Vitamin E as antioxidant (Uki and Watanabe 1992).

A successful artificial diet requires a water-stable binding agent. In a study of seven binding agents, five alginates in combination with agar and/or gelatine, on 24-hour water stability of artificial weaning diet, Knauer et al. (1993) found the best binder to be 1:3 agar/gelatine. This binder was used in all Northern Territory University (NTU) test diets.

Harada and Kawasaki (1982) investigated the feed stimulating effect of 32 species of green, brown and red seaweeds to abalone *Haliotis discus* and found that only certain algae provoked feeding behaviour — three out of six green algae tested, four out of 12 brown and two of 14 red algae. In a further study of the feeding attraction of *Haliotis discus* to 18 fragrant and pungent spices Harada (1992) reported the strongest attractants were black pepper, anise, basil and cinnamon.

The aims of this short preliminary study were as follows:

(i) to test the water stability of artificial diets formulated with different sources of protein, lipid and energy,

(ii) to investigate the feeding attraction of *T niloticus* to diets which included successful abalone attractants, and

(iii) to establish the chemical composition of the formulation found to be most attractive to *T. niloticus*.

Table 1. Formulations of artificial diets used in 16-hour water stability trial.

Component	Diet A	Diet B	Diet C (% dry weight)	Diet D	Diet E
Protein source					
Casein	15	–	–	–	–
Fishmeal	15	50	15	15	20
Soybean meal	25	–	35	20	30
Spirulina sp.	–	15	–	–	–
Gelatine (binder)	15	15	15	15	15
Energy source					
Semolina (wheat)	25	15	30	–	–
Corn starch	–	–	–	25	–
Wheat flour, wholemeal	–	–	–	–	30
Lipid source					
Cod liver oil	0.5	0.5	0.5	0.5	0.5
Binder					
Agar	5	5	5	5	5
Sodium alginate	–	–	–	20	–
Vitamins					
Vitamin E	0.1	0.1	0.1	0.1	0.1
Vitamin C	0.1	0.1	0.1	0.1	0.1
Reference	Modified Australian (FRDC–CRC)	Modified South African (Rhodes Uni.)	Composite	Modified Japanese (commercial)	Modified Australian (commercial)

Materials and Methods

Preparation of artificial diets

Seven artificial diets were prepared with formulations based on published artificial diets for abalone (Table 1). Protein sources used included casein (BDH), fishmeal (Delaneys), soybean meal (Roberts), *Spirulina* sp. (Lifestream Research International) and gelatine (Davis). Energy sources included semolina (Sungrain Products), corn starch (BDH) or wholemeal wheat flour (Grain Meal). Lipid was supplied as cod-liver oil (Amcal) and lipids in the fishmeal. Binders used were agar (BDH) or sodium alginate (Ajax). Vitamins included Vitamin E (Gold Cross) and Vitamin C (Cenovis). Feeding attractants were black pepper *Piper nigrum*, cinnamon *Cinnamomum zeylandicum* (both Masterfoods), green algae *Spirulina* sp. (Lifestream Research International) or peanut butter paste (ETA).

All cereal ingredients, agar and gelatine were mixed to a paste with cold water and brought to the boil with stirring. The mixture was allowed to cool to 55°C and the remaining ingredients added with vigorous stirring. When the temperature reached 45°C the formulation was plated onto a substrate made of 10 x 20 cm plastic oyster mesh (1-mm grid size). The diets were cooled for two hours at 4°C for a minimum of two hours before further testing.

Water stability test

Diets A, B, C, D and E were tested for water stability. Duplicate samples of each diet plated onto 10 x 20 cm plastic oyster mesh were immersed in rectangular glass tanks containing 20 L salt water. Measurements of turbidity were made hourly for six hours and after 16 hours (Beckman Spectrophotometer). The experimental design was two tanks per diet, and two control tanks with mesh but no diet.

Feeding attraction to *T. niloticus*

Diet E was further modified to include one of the attractants black pepper (Diet E1), cinnamon (Diet E2), powdered *Spirulina* sp. (Diet E3) or peanut butter (Diet E4). The feeding attraction of these diets to juvenile *Trochus niloticus* was tested over the 10-hour afternoon and evening period of maximum feeding activity. Hourly observations were made of the location of each animal. Experimental design: 10 juveniles of diameter within the range 0.8–1 cm per 20 L tank; two tanks per diet; two controls with no food; all animals starved for 24 hours prior to testing.

The feeding attraction of Diet E3 to mature *T. niloticus* was tested over a similar 10-hour afternoon and evening period with hourly observations of animal location. Experimental design: one individual animal of diameter 7–8 cm per 20 L tank; eight tanks with Diet E3; two controls with no food; all animals starved for 24 hours prior to testing.

Chemical analysis

Duplicate samples of Diet E3 were analysed for total ash (inorganic matter), total carbohydrate, total protein and total lipid.

Ash was determined gravimetrically after heating at 550°C, total carbohydrates were analysed by the method of Dubois et al. (1956) and total lipid was determined gravimetrically by the method of Bligh and Dyer (1959). Total protein was calculated from total Kjeldahl nitrogen (x 6.25) determined by Flow Injection Analysis (Lachat 8000) following digestion of ground samples with concentrated sulfuric acid and $CuSO_4$ catalyst.

Fatty acid methyl esters (FAME) were prepared by direct transesterification of lipid extracts with 14% BF_3-methanol complex and analysed by gas chromatography (Varian Vista 6000). The individual fatty acids were identified by comparing the retention times with commercially available external standards (Sigma, USA). Fatty acid structure was represented as L:B(n–x) where L=chain length, B=number of double bond and (n–x)= position of double bond closest to the terminal methyl group. More details of these methods are given in Renaud et al. (1994).

Results and Discussion

It was found that the preparation of each artificial diet required 30 minutes preparation time. It was then necessary to store prepared plates in the refrigerator for at least two hours before use, to ensure that the gelling process was complete.

Water stability

The artificial Diet E containing fishmeal (20% dry weight), soybean meal (30%) gelatine (15%), wholemeal wheat flour (30%) and agar (5%) was found to have the lowest 16-hour turbidity reading, the least smell and the lowest estimate of bacteria (Table 2).

Turbidity measurements at 16 hours were very low, and the water in all diet tanks had values within the range 0.024–0.026 on a scale of 0 to 1. All diets used 1:3 agar/gelatine as binder. This was the best binder reported in a study of seven binding agents including five alginates in combination with agar and/or gelatine (Knauer et al. 1993).

Feeding attraction to modified diet

It was found that Diet E3 containing *Spirulina* sp. had the highest positive attraction for both juvenile (Table 3) and mature (Table 4) *T. niloticus*.

For the experiment on juveniles, 65% of observervations found animals close to the diet (Table 3).

Table 2. Estimation of turbidity, smell and bacterial contamination of aquarium water after 16-hour water turbidity test on five artificial diets.

	Control	Diet A	Diet B	Diet C	Diet D	Diet E
Turbidity	0	0.025	0.025	0.026	0.025	0.024
Poor smell	Nil	+++	++	++	+	+
Bacteria present	Nil	+++	++	++	+	+

Table 3. Attraction of juvenile *T. niloticus* to artificial diets containing one of the attractants black pepper (E1), cinnamon (E2), *Spirulina* sp. (E3) or peanut butter (E4).

Diet	Attractant	Time spent in tank sector (%)								
		Close to diet			Tank centre			Far end from diet		
		1	2	Mean	1	2	Mean	1	2	Mean
Control	Nil	44	15	41	18	64	30	39	16	29
Diet E1	Black pepper	48	57	53	44	19	32	11	25	15
Diet E2	Cinnamon	49	53	51	37	9	23	12	35	26
Diet E3	*Spirulina* sp.	39	91	65	17	6	12	44	10	23
Diet E4	Peanut butter	40	51	46	40	21	31	16	28	23

Table 4. Attraction of mature *T. niloticus* to artificial diet E3 containing *Spirulina* sp.

	Replicate	Time spent in tank sector (%)		
		Close to diet	Tank centre	Far end from diet
Control	1	50	0	50
	2	30	30	40
	$\bar{X}$	40	15	45
Diet E3	1	60	40	0
	2	90	10	0
	3	100	0	0
	4	60	20	20
	5	100	0	0
	6	100	0	0
	7	60	20	20
	8	70	30	0
	$\bar{X}$	80	16	4

Lower positive attraction was recorded for Diet E1 containing black pepper (53%), Diet E2 containing cinnamon (51%), Diet E4 containing peanut butter (46%) and the control (41%).

The gross chemical composition of Diet E3 was made up of 33.0% (dry weight) carbohydrate, 10.2% lipid, 42.1% protein and 25% ash.

Mature *T. niloticus* with access to the experimental diet E3 containing *Spirulina* sp. spent 80% of their time close to or on top of the diet. Control animals spent 40–45% of their time at either end of the tank and 15% of time in the middle.

Conclusion

It was found that the preparation of each artificial diet was relatively labour-intensive and diets required refrigeration before use.

Diet E containing fishmeal (20% dry weight), soybean meal (30%), gelatine (15%), wholemeal wheat flour (30%) and agar (5%) had the lowest 16-hour water turbidance, the least smell and the lowest estimate of bacteria.

Modified Diet E3 containing powdered *Spirulina* sp. had the highest positive attraction for both juvenile and mature *T. niloticus.*

The gross chemical composition of Diet E3 was carbohydrate (33.0% dry weight), lipid (10.2%), protein (42.1%) and ash (25%).

Acknowledgments

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Fatty Acid Composition Characteristic of *Trochus niloticus* (Mollusca: Gastropoda) Fed on Naturally Growing Microalgae in an Aquaculture System

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Abstract

The fatty acid composition of juvenile *Trochus niloticus* (three, 12 and 18 months old) artificially spawned and reared at Northern Territory University (NTU) Aquaculture Centre was investigated. The difference in fatty acid composition of two size groups (10–25 mm diameter) and 26–40 mm diameter) of 12-month-old animals was also investigated.

The principal fatty acids of three-month-old whole animals were palmitic acid (16:0), stearic acid (18:0), oleic acid (18:1(n–9)), γ-linolenic acid (18:3(n–6)) and arachidonic acid (20:4(n–6)). There were significant changes in fatty acid composition of foot tissue of *T. niloticus* with age. These changes included increases in the percentage of the long-chain polyunsaturated fatty acids (PUFAs) 20:4(n–6), 20:5(n–3), 22:4(n–6), 22:5(n–3) and 22:6(n–3), and decreases in the percentage of shorter-chain saturated and monounsaturated fatty acids, including 14:0, 18:0, 14:1, 16:1, 18:1(n–9) and 18:1(n–7). There was very little difference between the fatty acid composition of the two size groups of 12-month-old *T. niloticus*, except for the percentage of 18:3(n–6) which was 4% lower in the 10–25 mm size group.

The percentage of the principal PUFA, 20:4(n–6), increased from 6.4–8.0% of total fatty acids in three-month-old whole juveniles to 14.2–20.8% in foot tissue of 18-month-old juveniles. The percentage of 20:5(n–3) increased from 1.6–2.0% at three months to 2.2–5.0% at 18 months, while the percentage of 22:6(n–3) increased from 0.6–1.0% to 3.2–5.4.

THERE is an extensive literature of the fatty acid compositions of marine molluscs with contributions dating from the early 1970s (Joseph 1989). The literature includes reports of the fatty acid composition of Archaeogastropoda including families (Haliotidae (abalone) (Bannatyne and Thomas, 1969; Kochi 1975; Yamada and Hayashi, 1975), Patellidae (Gardner and Riley, 1972; Johns et al. 1980) and Trochidae (*Austrocochlea constricta*) (Johns et al. 1980) but no reports of the fatty acid composition of the topshell *Trochus niloticus*.

Our earlier paper presented data on the fatty acid composition of mature *T. niloticus* recently collected from the wild and of animals from the same collection subsequently maintained for one year at NTU hatchery (Rehbung et al., these Proceedings). This paper reports a baseline study of changes in fatty acids composition of juvenile *T. niloticus* bred at NTU Aquaculture Centre and fed on naturally growing microalgae in the aquaculture system over an 18-month period. The study includes a comparison of the fatty acid compositions of small and large animals at age 12 months.

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Materials and Methods

Sample Source

Larval *T. niloticus* bred from wild broodstock from King Sound, north-west Australia, were hatched at NTU Aquaculture Centre in September 1993. Larval and juvenile *T. niloticus* were grown-out in 3000 L aquaculture tanks fed on mixed microalgae naturally grown on 30 cm × 60 cm corrugated perspex sheets suspended in the tanks. Juveniles were sampled after 3, 12 or 18 months. Ten samples of three-month-old juveniles (approximately 1300 individuals per sample, whole animal including shell) were cleaned with distilled water, dried with tissue paper, freeze-dried, ground to a fine powder and stored at –80°C prior to chemical analysis. The 12-month-old juveniles were divided into two groups according to size of shell diameter (10–25 mm and 26–40 mm groups).

Ten samples of the smaller group (8–10 individuals per sample, foot only) and 10 samples of the larger group (2–3 individuals per sample, foot only) were prepared. Eighteen-month-old juveniles were analysed individually with a total of 10 samples each consisting of the foot of a single animal. For 12- and 18-month-old juveniles foot tissue was dissected, rinsed with distilled water, freeze-dried, ground and stored at –80°C prior to chemical analysis.

Analytical methods

Duplicate samples of ground *T. niloticus* (whole organism or foot) were analysed for fatty acid composition. Samples (0.3g) were extracted by a modified Bligh and Dyer (1959) method, with 5 mL of chloroform-methanol (2:1 v/v) containing 0.005% BHT (butylated hydroxytoluene antioxidant) using sonication for 30 minutes. Distilled water (1 mL) was added and then samples were vortex-mixed for 1 minute and centrifuged at 1000×g (2500 rpm) for 15 minutes. The chloroform layers were removed and the debris extracted twice more with 5 mL of chloroform-methanol solution as in the initial extraction but without sonication. The pooled extracts were washed with KCl (0.8%) followed by methanol:NaCl (0.85%) (1:1, v/v), then dehydrated with anhydrous sodium sulfate and filtered through Whatman filter No. 40. The extracts were evaporated to dryness at 30°C under a stream of nitrogen and then diluted to a known concentration with hexane for subsequent fatty acid analysis. Fatty acid methyl esters (FAME) were prepared by direct transesterification of lipid extracts with 14% BF_3-methanol complex as described previously (Renaud et al. 1994).

Statistical analysis

Data were compared by one-way analysis of variance (ANOVA) with age as the independent variable and dependent variables the individual fatty acids. Post hoc multi-comparisons were made using the Tukey Test.

Results and Discussion

Fatty acid composition

The major fatty acids (greater than 5% of total fatty acids) of 3-month-old animals were palmitic acid (16:0) 29.3% of total fatty acids, stearic acid (18:0) 9.5%, oleic acid (18:1(n–9)) 6.5%, γ-linolenic acid (18:3(n–6)) 5.4% and arachidonic acid (20:4(n–6)) 7.2% (Table 1).

Changes in fatty acid composition over the 18-month study period are shown in Table 1.

There was an overall increase in the percentage of polyunsaturated fatty acids (sum %) in the lipids of older animals, which increased from 30.4% of total fatty acids for 3-month-old juveniles to 54.8% for 18-month-old animals. This trend was influenced by increases of 3.0% or more in the amounts of long-chain PUFAs 20:4(n–6), docosatetraenoic acid (22:4(n–6)), docosapentaenoic acid (22:5(n–3)) and docosahexaenoic acid (22:6(n–3)) (10%, 3.0%, 3.0% and 3.4% increases respectively). There was an overall decrease in the percentage of monounsaturated fatty acids, which was influenced by decreasing percentages of 16:1 and 18:1 fatty acids in older animals (Table 1). There was no clear trend in the change in saturated fatty acids with animal age. There were small decreases in the percentages of the saturated fatty acids 14:0 and 18:0 in older animals, and a peak in the percentage of 16.0 in 12-month-old animals (Table 1).

Studies of fatty acid biosynthetic pathways in marine animals have shown that 18:2(n–6) is converted to 18:3(n–6) which is then chain-elongated to 20:3 (n–6) and then converted to 20:4(n–6) (Sargent et al. 1993). Crustaceans (Kanazawa et al. 1977), bivalves (Langdon and Waldoch, 1981) and gastropods (Uki et al. 1986) have only a limited ability to elongate and desaturate 18-carbon PUFAs of the n–3 or n–6 series to longer chain (PUFAs). The nutritional requirements of juvenile Trochidae are poorly understood but data are available for abalone (Haliotidae) (Uki et al. 1986). These researchers demonstrated that the two PUFAs 20:5(n–3) and 22:6(n–3) are essential dietary fatty acids for the growth of abalone because these animals have very low desaturase activity to convert dietary 18:3(n–6) and 18:3(n–3) to higher unsaturated fatty acids. In the present study, the increase in long-chain C20 and C22 PUFAs in 12- and 18-month-old *T. niloticus* demonstrates that the NTU mixed microalgal diet which contained 10.7–17.0% of the fatty acid 20:5(n–3) and 0.4–1.8% of 22:6(n–3) (Renaud et al., these Proceedings) was an adequate source of essential PUFAs to support continued long-term growth of this species.

Comparison of fatty acid composition of two size-classes of 12-month-old animals

When comparing the two sizes of 12-month-old *T. niloticus* there were small differences in the percentages of several fatty acids (Table 1). The percentages of 14:1 and 18:3(n–6) were significantly higher in the 25–40 mm size group (1% and 4.2% higher, respectively; p=0.0001 in both cases), while the percentages of 16:1(n–7) and 20:5(n–3) were significantly lower (0.5%, 0.8% and 0.9% lower respectively; p<0.02 in each case). The higher level of 18:3(n–6) was interesting. The mixed microalgal diet was available to all 12-month-old *T. niloticus* and contained small but adequate amounts of 18:3(n–6)

Table 1. Fatty acid composition of juvenile *T. niloticus* aged 3, 12 and 18 months fed on naturally growing mixed microalgae. (Data given as % total fatty acids (mean ± standard deviation); n=10.)

Fatty acid	3 months (whole)	12 months (foot)		18 months (foot)
		10–25 mm	26–40 mm	
Saturates				
14:0	4.0±0.5^{a}	2.4±0.4^{b}	2.2±0.3^{b}	1.4±0.4^{c}
16:0	23.9±1.8^{a}	34.3±4.9^{b}	32.1±2.4^{b}	28.8±2.0^{c}
18:0	7.4±1.8^{a}	8.5±1.0^{b}	8.2±1.2^{b}	5.0±0.4^{c}
20:0	2.7±1.0^{a}	1.0±0.6^{b}	2.4±0.9^{a}	3.3±0.5^{c}
Sum %	38.0	46.2	44.9	38.3
Mono-unsaturates				
14:1	1.8±0.2^{a}	1.3±0.2^{a}	2.3±0.3^{b}	1.4±0.4^{a}
16:1	4.0±0.2^{a}	1.2±0.2^{b}	0.8±0.2^{c}	0.5±0.2^{d}
18:1(n–9)	6.5±0.8^{a}	6.0±0.7^{a}	5.5±0.6^{a}	3.8±0.3^{b}
18:1(n–7)	4.4±0.3^{a}	2.8±0.3^{b}	2.5±0.3^{b}	2.5±0.7^{b}
20:1	1.4±0.3^{a}	3.3±0.5^{b}	2.5±0.3^{c}	0.2±0.4^{d}
Sum %	18.1	14.6	13.6	8.3
Polyunsaturates				
16:2(n–7)	2.7±0.6^{a}	1.9±0.2^{a}	1.7±0.4^{a}	3.6±0.4^{b}
16.2(n–4)	1.5±0.5^{a}	4.2±0.7^{b}	3.4±0.5^{b}	2.3±0.7^{c}
16:3(n–6)	3.0±0.1^{a}	2.5±0.6^{a}	2.5±0.6^{a}	5.5±0.6^{b}
18:2(n–6)	3.3±1.0^{a}	2.6±0.8^{a}	2.8±0.3^{a}	3.8±1.1^{a}
18:3(n–6)	3.9±1.9^{a}	2.1±1.1^{b}	6.3±0.7^{c}	4.1±0.5^{a}
18:3(n–3)	2.4±0.3^{a}	2.6±1.2^{b}	1.8±0.2^{b}	1.3±0.3^{c}
20:4(n–6)	7.2±0.7^{a}	10.0±1.3^{b}	9.8±1.2^{b}	17.2±3.6^{c}
20:5(n–3)	1.8±0.2^{a}	2.8±0.3^{b}	1.7±0.2^{a}	3.6±1.4^{b}
22:4(n–6)	2.5±0.9^{a}	3.8±0.6^{b}	3.4±0.5^{b}	5.5±0.6^{c}
22:5(n–3)	1.2±0.3^{a}	2.6±0.4^{b}	2.5±0.4^{b}	4.2±0.9^{c}
22:6(n–3)	0.9±0.3^{a}	3.4±0.6^{b}	2.8±0.4^{b}	4.3±1.1^{c}
Sum %	30.4	38.5	38.7	54.8

[a,b,c,d] Values in a row with unlike superscripts are significantly different at 95% level.

(0.8–2.7/5 total fatty acids). This suggests that the larger 12-month-old *T. niloticus* ingested more microalgae and thus had greater supplies of the fatty acid 18:3(n–6) available for biosynthesis to 20:4(n–6) or that the smaller *T. niloticus* may not have consumed enough microalgae to meet their normal growth requirements for long-chain PUFAs.

Conclusions

The naturally growing mixed microalgal diet at NTU Aquaculture Centre was an adequate source of fatty acids, including essential PUFAs, to support continued long-term growth of *T. niloticus*.

To establish which long-chain PUFAs are essential for this species further study is needed on starved and carbohydrate-fed juvenile *T. niloticus* over the first 14–21 days post-hatching. Another study could use artificial feeds containing individual PUFAs to investigate the effect of these on the growth rate of juvenile *T. niloticus*.

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The Growth of Juvenile *Trochus niloticus* Fed on Algae

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Abstract

The growth of juvenile *Trochus niloticus* (15.5 mm maximum basal diameter) fed on three species of algae (*Acanthophora muscoides, Padina boryana and P. australis*) was investigated. It was found that the animals fed *A. muscoides* had the highest daily growth rates, giving 0.022 mm increase in diameter and 6 mg increase in wet weight per day, followed by *P. australis* (0.028 mm, 5 mg per day). Animals fed *P. boryana* had a smaller increase in weight (3 mg per day), while the unfed control exhibited a decline in weight (–125 micrograms per day). However, there was no difference in diameter for animals fed *P. boryana* compared to the starved animals.

RESEARCHERS have estimated the growth of the trochus utilising mark–recapture experiments of released juveniles or wild animals with growth estimated using the von Bertalanffy growth function (VBGF) (Smith 1987, Hoffshir et al. 1990). A summary of the results of growth studies utilising VBGF calculated the average trochus size (maximum basal diameter) for the first year as 30 mm, 60 mm after two years, and about 80 mm by the third year (Nash 1993). This represents an annual growth increment of about 25 mm. Hoffshir and coworkers introduced artificially spawned juvenile trochus with an average diameter of 19 mm to Lifou reefs, New Caledonia. After 11 months the mean growth (increase in diameter) was 45 ± 4 mm. This is on average 12 mm higher (33 ± 3 mm) than that of the juveniles kept in their original aquaculture breeding ponds for the same period (Hoffshir et al. 1990). Heslinga (1981a,b) reared trochus animals, which were fed ad libitum to a mean shell diameter of 7.8 mm after 4 months and 62 mm after one year. These results show that the animals can grow faster in tanks than in their natural environment. This growth in turn is limited by algal production in the tanks. It is, however, not economically possible to culture trochus to commercial size in tanks due to the large tanks required to culture food for the animals (Nash 1993). Trochus under culture have been fed benthic diatoms and other unidentified filamentous and benthic microalgae (Heslinga 1981a; Heslinga and Hillman, 1981). In Okinawa, Japan, the sessile diatom *Navicula ramosissima* is the selected food for juvenile trochus (Isa 1991). Although adult trochus has been shown to consume macroalgae (Lambrinidis et al., these Proceedings) there are no such data on juvenile trochus animals.

This study looks at the suitability of three intertidal macroalgae, namely *Acanthophora muscoides, Padina boryana and P. australis* as suitable food sources for juvenile trochus.

Materials and Methods

Three species of macroalgae *A. muscoides, P. boryana* and *P. australis* were collected fortnightly from Nightcliff, Darwin Harbour between June and September, as described previously (Lambrinidis 1994).

Juvenile *Trochus niloticus* (287 days old, average maximum basal diameter 15.5 mm) were supplied by

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Dr Chan Lee, NTU Aquaculture Centre. For each algal species and control, 10 juvenile trochus were selected at random, numbered with a permanent ink marker (Artline 700) and then placed into 60 L glass aquaria, each containing 35 L of sterilised treated bore water (in duplicate). Groups of 10 juvenile trochus of the same age were randomly assigned to different numbered tanks into which the algae were added. Two types of control were used: one had no added food, the other contained a benthic algal diet grown on fibreglass plates. The live food of macroalgae and benthic microalgae were offered to the animals ad libitum. To ensure this about 200°g of macroalgae were supplied every fortnight and the benthic microalgal plates every two days or whenever necessary. For growth measurements each numbered animal was measured and weighed as described by Lambrinidis (1994). The pH and temperature were recorded between 9:30 and 10:00 a.m., unless otherwise stated, the tankwater constantly maintained and cleaned to ensure the highest possible water quality.

Linear least-squares regressions were fitted to all the growth curves. The regression equations characterised a linear relationship of the form $y=a+bx$, where y is either the increase in maximum basal diameter (mm) or total fresh weight (g), b is the regression coefficient (which is a measurement of growth rate), and a is the intercept of the y axis.

Daily growth rate G was determined for each animal from day zero to day 79, according to the formula: $G = (W_1-W_0)/(t_1-t_0)$, where W_1= weight at harvest time (t_1), W_0= weight at time 0 (t_0). The growth rates were then analysed statistically by one-way analysis of variance (ANOVA), with growth as the source of variance. Pairwise comparisons after ANOVA were made utilising Scheffe's F-test (confidence level is 95% in all cases). The data for maximum basal diameter were treated the same way, with D_1 = diameter at harvest time (t_1) and D_0= diameter at time 0 (t_0). $G = (D_1-D_0)/(t_1-t_0)$, The growth of animals that died or fell out of tanks within the first 28 days were not included as these underestimated growth rates. Only one replicate tank for the animals fed *P. australis* was used since seven of the animals died of unknown causes, and it was assumed that the three remaining animals had been affected.

Results

On the basis of fresh weight and diameter dimensions, juvenile trochus grew best when fed with benthic microalgae (Fig. 1). Juvenile trochus grew reasonably well when fed *Padina australis* and *Acanthophora muscoides*. Surprisingly, *P. boryana* did not show to be a suitable food for juvenile trochus.

Linear least-squares regression for measurements of maximum basal diameter were calculated for the data collected after day 28 to eliminate the effects of the lag phase. The values of the coefficients a and b, the coefficients of determination (r^2) for the regression analysis, are given in Table 1. For both the measurements of fresh weight and maximum basal diameter, animals fed on the benthic microalgae showed the highest growth rates.

Animals fed on *P. australis* and *A. muscoides* had an equal rate of growth but both were lower than those fed the microalgae. The animals fed *P. boryana* showed little growth, while the animals in the unfed control exhibited a reduction in weight and diameter.

The means and standard deviations of the daily growth rates are given in Table 2. The daily growth rates show the same trend as those calculated with the method of least-squares regression, although estimates for growth in terms of increase in diameter are a little lower since the lag phase has not been taken into account. The mean growth (fresh weight and maximum diameter) for the animals fed the three seaweeds and the two controls was significantly different ($P = 0.0001$ for both), as determined by ANOVA (with P, the probability of the null hypothesis that the mean growth of the animals is equal, irrespective of diet, rejected and accepting the alternative hypothesis).

The results of the Scheffe's F-test (Table 3) confirm that the growth (fresh weight and diameter) of the animals fed *P. australis* was not significantly different to the growth of animals fed *A. muscoides* at the 95% confidence level. The effect of diet was significant for all other treatments. There was no significant difference between the mean growth of animals fed *P. boryana* and the unfed control for diameter only.

The growth rates at the 95% confidence level for the animals can therefore be ranked according to diet for (a) change in weight and (b) change in maximum basal diameter, as follows:

(i) benthic diatoms > *A. muscoides* = *P. australis* > *P. boryana* > control.

(ii) benthic diatoms > *P. australis* = *A. muscoides* > *P. boryana* = control.

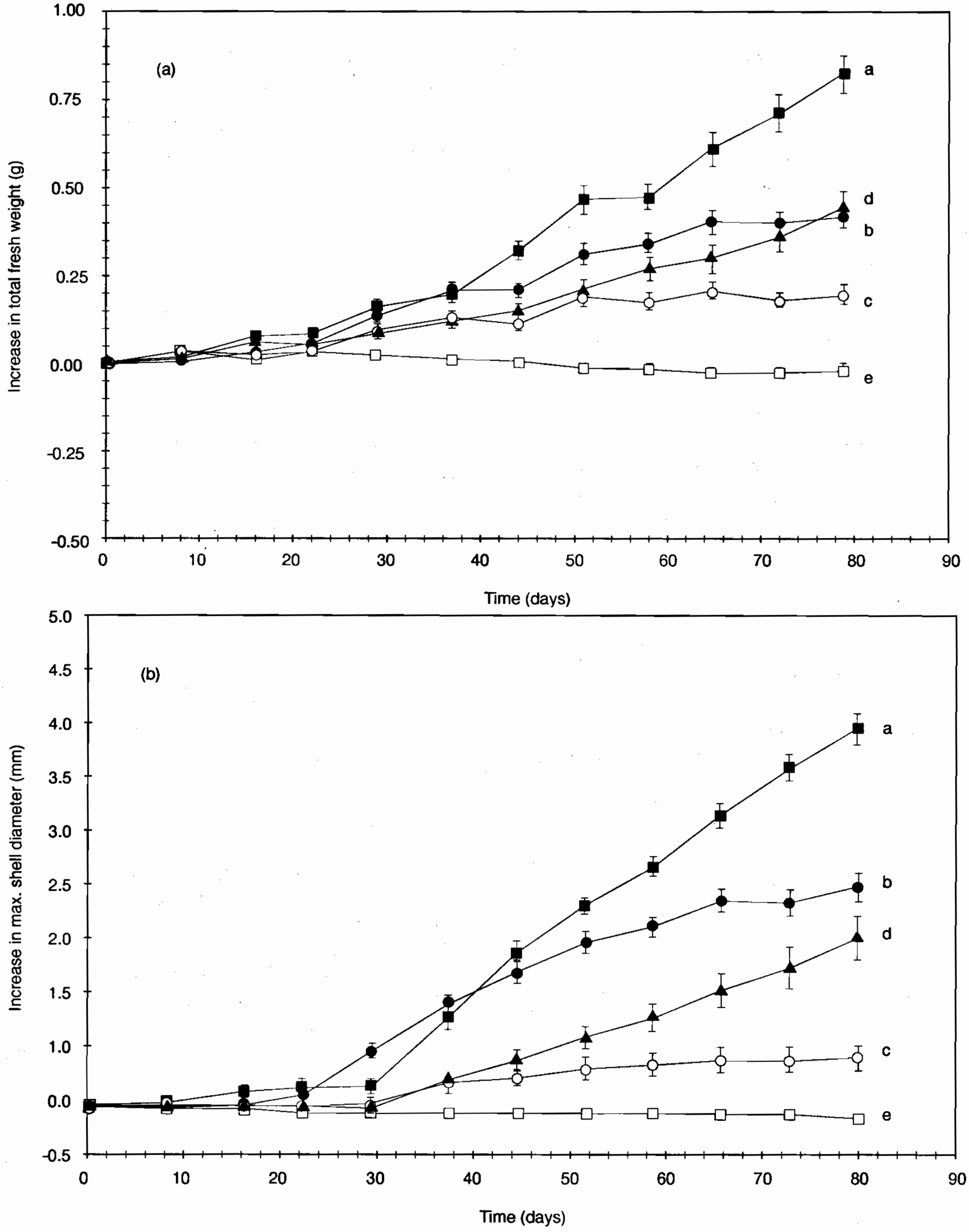

Figure 1. Growth of juvenile *T. niloticus* fed on three seaweeds and two controls. Results are for (a) increase in total fresh weight and (b) increase in maximum basal diameter. Each point is the mean of at least nine measurements with error bar (SE) on each side of the mean: a, diatoms (control 1); b, *P. australis*; c, *P. boryana*; d, *A. muscoides*; e, no food (control 2).

Table 1. Values of the coefficients *a* and *b* of the equation *y=a+bx*, calculated by the method of least-squares regression, with r^2, the coefficient of determination for the growth of all juvenile *T. niloticus* fed three algal diets and two controls for (a) increase in maximum basal diameter from days 29 to 79 and (b) increase in fresh weight from day zero to day 79.

Algal species fed	a	b	r^2
(a)			
P. boryana	– 0.111	0.008	0.091
P. australis	– 0.215	0.033	0.767
A. muscoides	– 1.035	0.034	0.422
Mixed microalgae	– 1.467	0.073	0.817
Unfed (control 2)	– 0.029	– 0.001	0.042
(b)			
P. boryana	0.002	0.003	0.493
P. australis	– 0.040	0.006	0.863
A. muscoides	– 0.044	0.006	0.610
Mixed microalgae	– 0.087	0.013	0.800
Unfed (control 2)	0.029	– 0.001	0.150

Discussion

The results clearly show that the three seaweeds *P. australis*, *A. muscoides* and *P. boryana* available in the intertidal zone of Darwin Harbour during the months of June to September are suitable as a live food source for young trochus in the laboratory. However, the growth rates of animals fed on seaweeds were significantly lower than those of animals fed on the benthic diatom control. This could be due to the inability of the young animals to graze on tough seaweeds, resulting in less-than-optimal food intake.

Growth of hatchery-reared trochus was investigated by Heslinga and Hillmann (1981) in 5000 L and 500 000 L tanks at 27–30°C. They reported that animals in the larger tanks reached a mean size of 2.1 mm, 2.2 months post-fertilisation, while animals in the 5000 L tank reached a mean shell size of about 5 mm in the same time. The higher growth in the larger tank was accredited to a higher algal production in the tank. In that study animals reached a mean shell diameter of 43 mm at age 10 months and increased in mean shell diameter by 19 mm in the following two months. The animals spawned by Heslinga and Hillmann (1981) had higher rates of growth than animals from the wild. By comparison, in the current study one-year-old animals grew 5 mm or less during the following two months. This result is similar to growth rates of juvenile trochus cultured in baskets hung in the sea (Isa 1991).

Animals fed *P. boryana* showed only a small increase in weight and diameter. The increase in weight was significantly different to that of the control but there was no difference in the shell diameter increases. This suggests that the animals were able to obtain some nutrition but not enough for significant shell growth. It is possible that the animals cannot obtain nutrients from the algae because the cell walls of the algae are too tough, or because the mouth structure of the juveniles is not adapted to feeding on that type of algae. Instead the animals may be surviving on the small amounts of bacteria found on the surface of the algal thallus. Feeding on attached microalgae has been observed in juvenile abalone in southern California (Tutschulte and Connell, 1988), although adults fed on the macrophytes.

Animals of the unfed control did not die but showed slight decreases in weight and diameter. The weight loss is to be expected since the animals used reserved materials during the period of starvation.

It is apparent from Figures 1a and 1b that there is a significant lag phase before shell growth occurs, which is consistent for all animals on different diets. Heslinga (1981a) noted a significant four-month lag phase in the first four months of growth of larval trochus; after this, growth was rapid. However, the lag phase in this study may be due to a number of other factors. Handling stress is prominent in delicate animals. The animals may suffer enough trauma from being removed from their surroundings that they require a period of acclimatisation before they become

Table 2. Mean growth rate of juvenile *T. niloticus* fed four algal diets and no food for 79 days for (a) maximum basal diameter and (b) total fresh weight.

Diet	n	Mean growth rate (mm per day)	Standard deviation
(a)			
P. boryana	18	0.006	0.006
P. australis	9	0.028	0.005
A. muscoides	18	0.022	0.012
Mixed microalgae	20	0.055	0.009
Control	18	–0.001	0.001
(b)		(g per day)	
P. boryana	18	0.003	0.001
P. australis	9	0.005	0.001
A. muscoides	18	0.006	0.002
Mixed microalgae	20	0.013	0.003
Control	18	–0.000125	0.001

Table 3. Results of Scheffe's pairwise comparisons of means after ANOVA for values with a difference in growth rates for (a) total fresh weight and (b) maximum basal diameter of juvenile *T. niloticus* fed different algal diets and no food.

	Total fresh weight (g)	Max diameter (mm)
Algal diets compared	95%	95%
P. boryana vs control	*	NS
P. australis vs *A. muscoides*	NS	NS

* = significant at that confidence level; NS = not significant at that confidence level.

used to their new environment and begin to grow. Another factor would be temperature. Over the course of the growth experiment, maximum air temperatures ranged 28.8–31.9°C. and minimum air temperatures increased from 15.4 to 24.7°C, with 9:30 a.m. tank water temperatures closely following minimum air temperatures. Therefore days became consistently warmer, favouring growth, since metabolism increases with increasing temperature (Spotte 1992). However, more experimental work must be carried out to confirm this, as Smith (1987) observed no seasonal variation in growth of trochus in the field.

Conclusion

Significant increases in daily growth rates of juvenile *T. niloticus* fed the three species of seaweeds were found, when compared to the unfed control. However the animals fed mixed diatoms in the second control tank had higher daily growth rates compared to animals fed seaweeds.

The growth rates for the animals can be ranked for change in weight according to diet as follows:

- benthic diatoms > *A. muscoides* = *P. australis* > *P. boryana* > control.

The growth rates for animals can also be ranked for change in maximum basal diameter according to diet as follows:

- benthic diatoms > *P. australis* = *A. muscoides* > *P. boryana* = control.

Acknowledgments

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A Study of Density, Abundance and Distribution of Juvenile Trochus and Associated Small Molluscs in Kei Besar Island, Indonesia

J. C. Dangeubun and S. Haumahu*

Abstract

Juveniles of topshells, *Trochus niloticus,* occurred in 20% of observations on the intertidal zone of the east coast of Kei Besar Island, having density and abundance of 0.55 snails/m^2 and 1.83 snails/m^2, respectively. Juvenile trochus were found mostly underneath rocks or rubble along the coast on the low-tide exposed area of the intertidal zone. There are 37 species of juvenile mollusc (33 species gastropods and four species bivalves) occupying the same habitat as juvenile trochus. The associated species occurring in highest densities in decreasing order, are *Cellana radiata, Rissonia spirata, Natica sertata, Mitra* sp. and *Rhinoclavis* sp., while the associated species of highest abundance in decreasing order, are *Mitra* sp., *N. sertata, C. radiata, Rhinoclavis* sp., *Notonister* sp., *R. spirata,* and *Pyramidella terebelloides.* Species found at the highest frequency of occurrence, in decreasing order, are *C. radiata* (85%), *R. spirata* (40%) *Epitonium lamelosa* (35%), and *Nerita albicila* (35%).

ONE factor considered very important in seeding juvenile trochus is seeding density (Castell 1995). It is likely that dense patches of juveniles become the focus of attention for predators (Boulding and Hay, 1984; Moran 1985) or may suffer lower predation rates, alternatively, compared to randomly distributed individuals (Ray and, Stoner 1994). However, very little is written on the quantitative study of natural juvenile trochus, except Smith (1987) and Castell (1995), or on any juveniles of other molluscs.

The biological structure of any community shows significant relationships among quantitative values of associate species (Odum 1971). The stability of any community is believed to be determined by the density and the distribution of the species in, and their interaction with, its environment. Dynamic relations within the species in their environment form distinctive types of arrangements and programmed activities known as community structure. Hutchinson (1953) used the term 'pattern' to describe such structure and identified many different arrangements and programmed activities in communities such as: stratification patterns (vertical layering); zonation patterns (horizontal segregation); activity patterns (periodicity); food-web patterns (network organisations in food chains); reproductive patterns (parent–offspring associations); coactive patterns (resulting from competition, antibiosis, mutualism, etc.); and stochastic patterns (resulting from random forces).

Kei Besar is an eastern Indonesian island whose inhabitants live mainly along the coast. Most income is derived directly from the utilisation of coastal and marine resources. One such resource is the trochus population. As everywhere, the natural stock of trochus in Kei Besar has declined and is limited. This condition creates a potential site for reseeding juvenile trochus. Until the present, little attempt has been made to study any aspect of trochus populations on this island, in spite of its significant economic impact on local fishermen.

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This research attempts to describe some of the biotic structures that characterised the community where juvenile trochus were found. Those structures are the species composition of small molluscs and their frequency of distribution, density and abundance. It is expected that the result of this study will provide some information about associated small molluscs in the intertidal zone of Kei Besar Island.

Materials and Methods

Two sites, Mastel and Ded Hanoat, of Sather intertidal zone of Kei Besar (Fig. 1) were surveyed during November–December 1995 for the presence of juvenile trochus. At each study site, two transect lines 100 m apart were placed at the dry area horizontal to the coastal line, during low and high tides (the chosen dry area is based on local knowledge about where trochus juveniles could be observed). Along the lines, a 1 × 1 m quadrat was placed in the dry area at intervals of 10 m. In each quadrat, all substrate (mostly large rocks) was carefully observed for the presence of juvenile trochus. All small (up to 20 mm) molluscs were collected from the quadrats where juvenile trochus were found. A single-factor ANOVA was performed to test for significant differences in the number of juvenile trochus observed during high and low tide sampling. The collected specimens were then sorted according to species and counted. Plotting these data using bar graphs demonstrates the frequency of occurrence of species associated with juvenile trochus. Data were further analysed using formulas described by Odum (1971) to determine species density and abundance.

Results

Juvenile *T. niloticus*

Juvenile trochus were found only at one site of the study area, Ded Hanoat. At this site, juvenile trochus were present at 20% of observations: 6% at low tide observation and 14% at high tide observation. From the entire observation of juvenile trochus, this study found that the calculated density and abundance of juveniles of this species are 0.55 snails/m^2 and 2.5 snails/m^2, respectively. The size of the observed juvenile trochus ranged 4.2–12.1 mm with an average of 6.92 mm (SD=1.61). Table 1 shows the calculated values of frequency of occurrence, density, and abundance of juvenile trochus for each sampling period.

A single-factor ANOVA revealed no significant difference in the total number of juvenile trochus found during low and high tides ($F=0.056 < F_{.005\,(1),1,98}$).

Associated small molluscs

Thirty-seven species from 31 families of mollusc, size ranges 2.00–12.10 mm, were found occupying the same habitat as juvenile trochus. Of those species, 33 were gastropods and four were bivalves.

Figures 2, 3, and 4 show the frequency of occurrence, density, and abundance of these associated species. In the highest frequency of occurrence, in decreasing order, were *Cellana radiata* (85%), *Rissonia spirata* (40%), *Epitonium lamelosa* (35%) and *Nerita albicila* (35%). The highest densities of associated species, in decreasing order, were *C. radiata, R. spirata, Natica sertata, Mitra* sp. and *Notosinister* sp., while the highest abundance, in decreasing order, were *Mitra* sp., *N. sertata, C. radiata, Notosinister* sp., *Rhinoclavis* sp. and *R. spirata.*

Discussion

Two sites (Mastel and Ded Hanoat) in Sather were observed for the presence of juvenile trochus in this study. Both sites are considered the ideal location or fishing ground for adult trochus by the local fishermen in Sather, Kei Besar. This was proved during the survey, when adult trochus were found at high occurrence. In this study, however, juvenile trochus were found at one site only, Ded Hanoat. This result indicates that juvenile trochus cannot be found in all the adult fishing grounds, and that its distribution is limited to a particular habitat. However, in this study, there was no attempt to study in-depth the physical characteristic of this particular habitat.

The density of 0.55 individuals/m^2 of juvenile trochus found at the study site in Ded Hanoat, Sather is considerably high compared to those found in Orpheus Island, Vanuatu (Castell 1995) or in Guam (Smith 1989). The site at Ded Hanoat is slightly cape-like, while the site at Mastel is a bay-like position (see Fig. 1). Substrate at Ded Hanoat is generally characterised by homogenous boulders and rocks throughout the entire shore, where all the observed juveniles were found underneath those boulders and rocks. At Mastel, the substrate varied with patches of sand, seagrass, boulders and rocks. At this site no juvenile trochus were found.

It is speculated that the physical characteristics of the habitat is one of the main factors that determine the

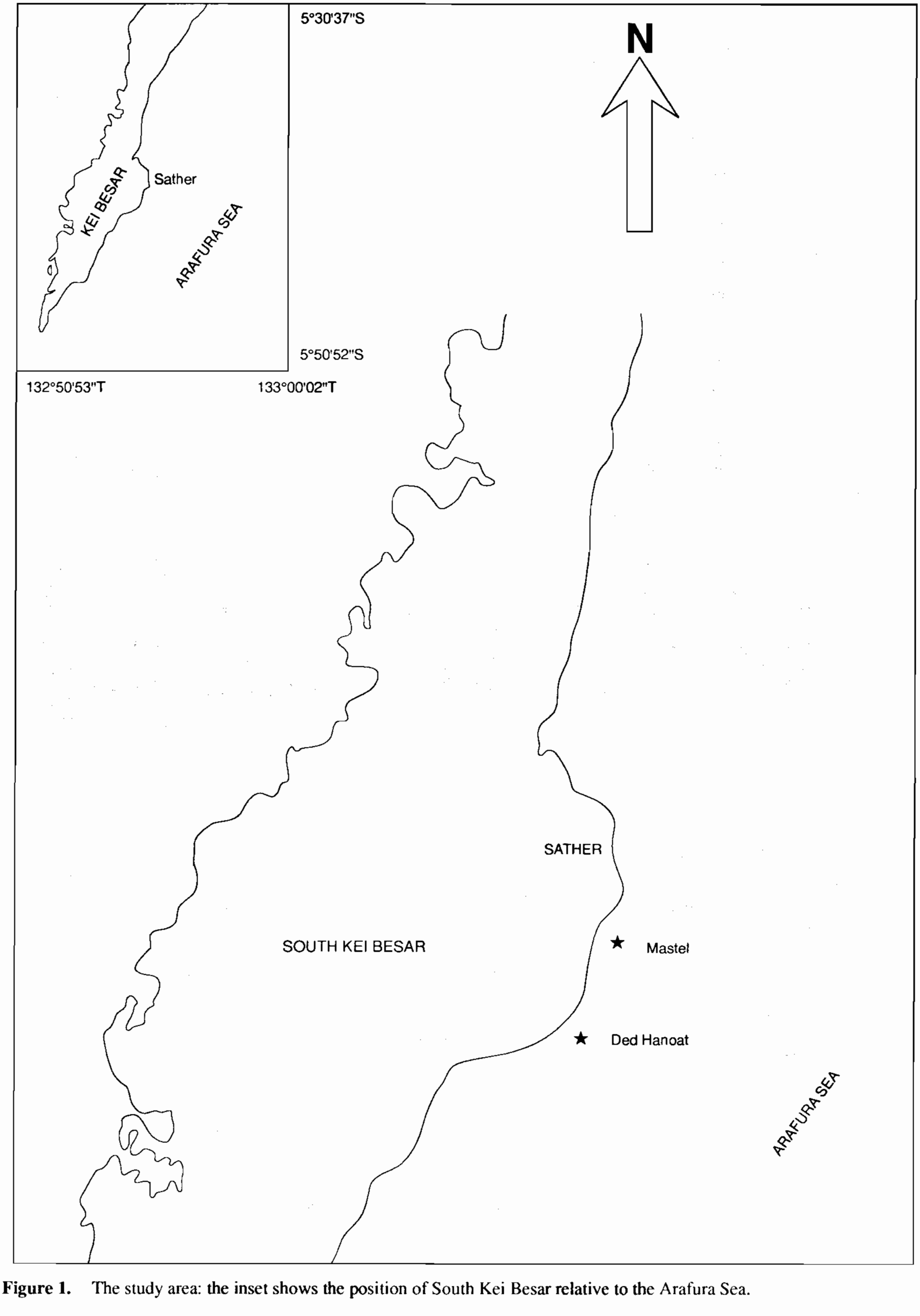

Figure 1. The study area: the inset shows the position of South Kei Besar relative to the Arafura Sea.

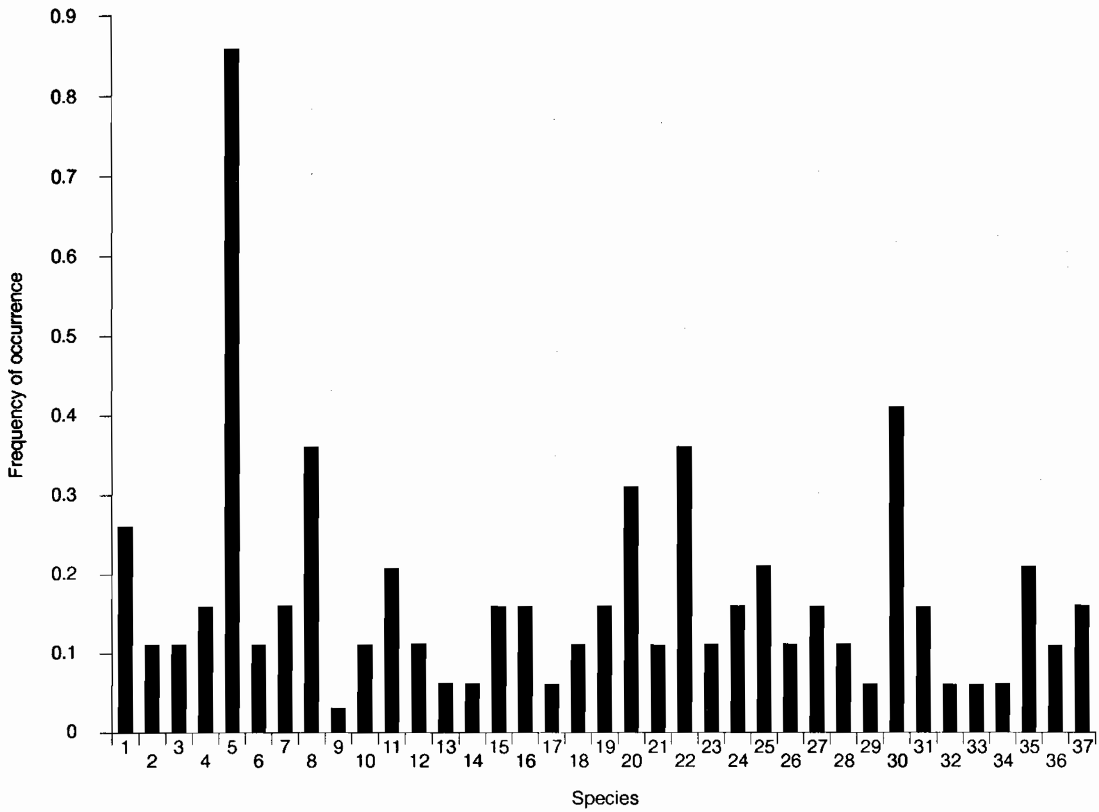

Figure 2. Frequency of occurrence of associated small molluscs found at the intertidal zone of Kei Besar.

Gastropods
1 = *Agaronia nebulosa*
2 = *Angiola* sp.
3 = *Astrea buschii*
4 = *Cellana radiata*
5 = *Conus* sp.
6 = *Conus sponsalis*
7 = *Engina zonalis*
8 = *Epitonium lamelosa*
9 = *Euchelus atratus*
10 = *Latirus* sp.
11 = *Latirus poligonus*
12 = *Liotina peronii*
13 = *Maculotriton seriale*
14 = *Marginella tricincta*
15 = *Mitra* sp.
16 = *Mitra tabanula*
17 = *Morula* sp.
18 = *Morula anaxeres*
19 = *Morula funiculus*
20 = *Natica sertata*
21 = *Nassarius coronatus*
22 = *Nerita albicila*
23 = *Nodilittorina pyramidalis*
24 = *Notosinister* sp.
25 = *Pyramidella terebelloides*
26 = *Pyrene punctata*
27 = *Rhinoclavis* sp.
28 = *Rissonia* sp.
29 = *Rissonia rosea*
30 = *Rissonia spirata*
31 = *Turbo chrysostomus*
32 = *Vexillum* sp.
33 = *Velliora corrugata*

Bivalves
34 = *Cardita variegata*
35 = *Isognomon perna*
36 = *Modiolus micropterus*
37 = *Tellina* sp.

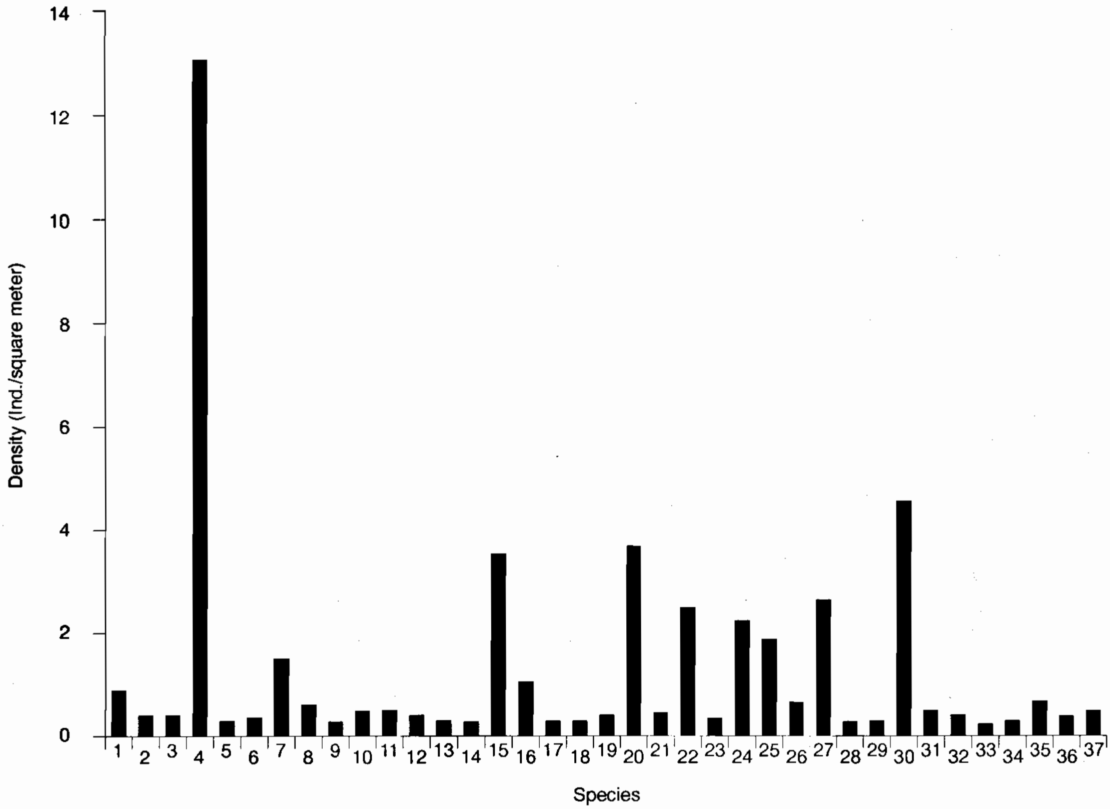

Figure 3. Density of associated small molluscs found in the intertidal zone at Kei Besar.

Gastropods
1 = *Agaronia nebulosa*
2 = *Angiola* sp.
3 = *Astrea buschii*
4 = *Cellana radiata*
5 = *Conus* sp.
6 = *Conus sponsalis*
7 = *Engina zonalis*
8 = *Epitonium lamelosa*
9 = *Euchelus atratus*
10 = *Latirus* sp.
11 = *Latirus poligonus*
12 = *Liotina peronii*
13 = *Maculotriton seriale*
14 = *Marginella tricincta*
15 = *Mitra* sp.
16 = *Mitra tabanula*
17 = *Morula* sp.
18 = *Morula anaxeres*
19 = *Morula funiculus*
20 = *Natica sertata*
21 = *Nassarius coronatus*
22 = *Nerita albicila*
23 = *Nodilittorina pyramidalis*
24 = *Notosinister* sp.
25 = *Pyramidella terebelloides*
26 = *Pyrene punctata*
27 = *Rhinoclavis* sp.
28 = *Rissonia* sp.
29 = *Rissonia rosea*
30 = *Rissonia spirata*
31 = *Turbo chrysostomus*
32 = *Vexillum* sp.
33 = *Velliora corrugata*

Bivalves
34 = *Cardita variegata*
35 = *Isognomon perna*
36 = *Modiolus micropterus*
37 = *Tellina* sp.

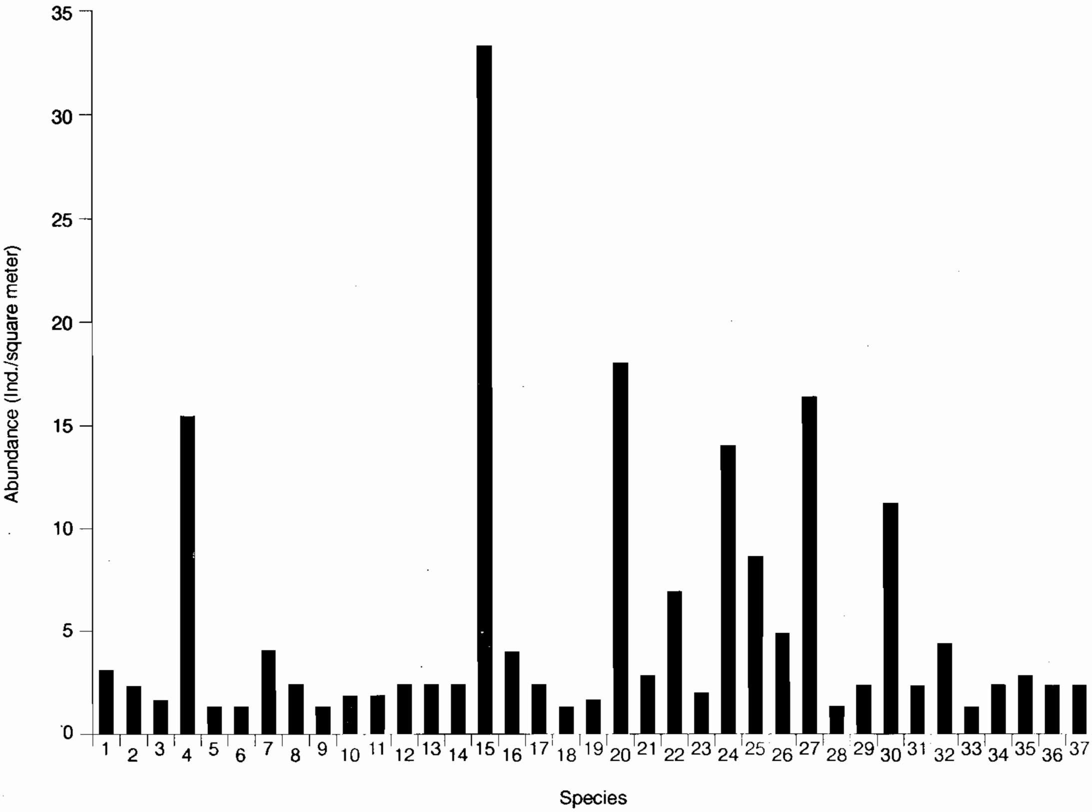

Figure 4. Abundance of associated small molluscs found in the intertidal zone at Kei Besar.

Gastropods
1 = *Agaronia nebulosa*
2 = *Angiola* sp.
3 = *Astrea buschii*
4 = *Cellana radiata*
5 = *Conus* sp.
6 = *Conus sponsalis*
7 = *Engina zonalis*
8 = *Epitonium lamelosa*
9 = *Euchelus atratus*
10 = *Latirus* sp.
11 = *Latirus poligonus*
12 = *Liotina peronii*
13 = *Maculotriton seriale*
14 = *Marginella tricincta*
15 = *Mitra* sp.
16 = *Mitra tabanula*
17 = *Morula* sp.
18 = *Morula anaxeres*
19 = *Morula funiculus*
20 = *Natica sertata*
21 = *Nassarius coronatus*
22 = *Nerita albicila*
23 = *Nodilittorina pyramidalis*
24 = *Notošinister* sp.
25 = *Pyramidella terebelloides*
26 = *Pyrene punctata*
27 = *Rhinoclavis* sp.
28 = *Rissonia* sp.
29 = *Rissonia rosea*
30 = *Rissonia spirata*
31 = *Turbo chrysostomus*
32 = *Vexillum* sp.
33 = *Velliora corrugata*

Bivalves
34 = *Cardita variegata*
35 = *Isognomon perna*
36 = *Modiolus micropterus*
37 = *Tellina* sp.

Table 1. Frequency of occurrence (FOO), density (D), and abundance (A) of juvenile *T. niloticus* in each site at each sampling period.

Site/period		NQ	NQT	NT	FOO	D	CA
Mastel							
Low tide	T1	25	0	0	0	0	0
	T2	25	0	0	0	0	0
High tide	T1	25	0	0	0	0	0
	T2	25	0	0	0	0	0
Ded Henoat							
Low tide	T1	25	8	17	0.32	0.68	2.13
	T2	25	6	12	0.24	0.48	2.00
High tide	T1	25	2	8	0.08	0.32	4.00
	T2	25	4	18	0.16	0.64	5.50

NQ = number of quadrats, NQT = number of quadrats where juveniles were found; NT = number of trochus; T = transect.

presence of juvenile trochus. For the size of juveniles found in this study (4.2–12.1 mm), it is expected that they are very vulnerable to predators, and that the absence of predators will greatly influence their density. In the case of Ded Hanoat, they are very exposed to rough wave actions that disadvantage the population of predators (mostly crustacean), whose feeding mechanisms involving holding the shells before actively breaking them. Too rough wave action prevents predators holding their prey properly before attacking it, or even from being in that area. Only organisms such as gastropods that attach to their substrate will adapt to such rough habitat. In addition, juveniles have the ability to hide underneath boulders or rocks.

As for the presence of the associated small molluscs, no comparison could be made. However, 37 associated species, as found in this study, could be considered a relatively high number. Species interaction, in relation to the availability of food and space and other coactive or biological patterns were not investigated here. However, it would be an interesting aspect and potential challenge to proceed to study those species interactions. Among the species that might be considered important in species interaction in this study site of Sather, Kei Besar are *Mitra* sp., *Cellana radiata*, *Rissonia spirata* and *Natica sertata*. *Mitra* sp. is counted for its abundance in the composition of the entire species array in this special community of juvenile trochus habitat. *C. radiata* and *R. spirata* are counted for their frequency of occurrence in the observation, while *N. sertata* may be important because of its high density.

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Catch History of Trochus in King Sound, Northwestern Australia Between 1980 and1995

K. L. Magro*

Abstract

The marine gastropods *Trochus niloticus* are taken by hand from exposed intertidal reefs in King Sound by members of the Bardi Aboriginal Community. The annual catch of trochus declined from 135 t (shell dry weight) in 1980 to 9 t in 1983. Catch increased after 1984 to 50 t and has steadily declined to 29 t in 1995. Difference in CPUE among months was significant ($F = 2.86$, $df = 11 \times 147$, $p = 0.001$). The CPUE was highest at 263 (kg per fisher month), 242 and 233 in October, June and May respectively. These months correspond with favourable seasons according to boating conditions. CPUE was lowest at 194, 183 and 181 in April, February and January. Difference in CPUE among years was significant ($F = 19.12$, $df = 14 \times 147$, $p < 0.0001$). The CPUE varied from 237 in 1981 to 174 in 1983, increasing to 391 in 1985 before steadily decreasing from 303 in 1986 to 134 in 1995. This suggests a steady decline in stock abundance. Fluctuations in the historical catch records can be attributed to economic, social or biological factors. Over-fishing and declining stocks may be a threat to the sustainability of the fishery.

TROCHUS shell (*Trochus niloticus* Linnaeus, 1767) is a marine mollusc fished throughout the Indo-Pacific for the mother-of-pearl trade. It is used primarily for button manufacture and other pearl shell products (Carleton 1984). Trochus appears to be particularly vulnerable to overexploitation. The biology of trochus makes it a relatively easy resource to exploit; it is an obvious, large, sedentary animal accessible in shallow water environments allowing collection by dry-picking the reef flat at low-tide or by diving in sub-tidal habitats (Heslinga *et. al.* 1984, Nash 1985). Exploitation of trochus resources also fits well with indigenous lifestyles. Harvesting does not require investment in expensive equipment or vessels and harvesting grounds are often close to home. The shell requires no preservation and is easily packed and stored (Glucksman and Lindholm 1982). For these reasons, trochus are easily exploited and highly vulnerable to overexploitation.

Over-fishing has reduced the abundance and size distributions of trochus stocks in many countries throughout the Indo-Pacific (Heslinga and Hillman 1981). These declines are a serious threat to the economic status of many countries and to the sustainability of the resource. The impact of fishing activity on the natural stocks can be monitored by catch statistics which describe the amount of resource being removed and the fishing effort (Gulland 1988). Monitoring catch and effort allow comparison of exploitation with the estimated surplus production (or estimated potential) of the fishery. This comparison is particularly useful in fisheries managed with quota systems, such as the Cook Islands trochus fishery. However, it is particularly useful to record the amount removed by fisheries without quotas to enable some estimation of potential yield and to observe if stocks are being over-fished. This is beneficial where no biological information is available and collection of catch data is the only means of gathering fisheries data, as in the extensive trochus fisheries in Papua New Guinea and the Solomon Islands.

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Catch is usually measured in terms of weight, such as kilograms landed, and should include all removals from the resource, including individuals discarded before landing, although this is rarely recorded. The fishing effort (also known simply as effort) is the measurement of the labour used to collect an individual catch. Effort is regulated by the technology used in fishing and how efficiently it exploits the resource (Ricker 1975, Gulland 1983). Consequently, effort can be measured by the efficiency of the fishing gear and the time spent fishing (Ricker 1975). However, the simplest measures of effort are the numbers of fishers, although this requires there are no changes in fishing practice over time (Gulland 1983). Trochus fisheries generally have simple gear technology, either dry-picking exposed reefs during low-tide periods, or using SCUBA or snorkel on subtidal reefs. Therefore, recording effort should include information on the number of fishers, time worked and the location fished.

Dividing catch by the associated effort provides an index of catch per unit effort (CPUE), a measurement of the amount fished for a particular level of effort (Ricker 1975). CPUE is believed to correspond with the abundance of a fished resource and changes in CPUE are generally used to observe changes in stock abundance (Royce 1972, Gulland 1983). This requires the catch and effort data to come from a single unit stock and that the catchability must be equal for all individuals in the stock. These assumptions may not be valid for trochus, as individuals are widely distributed among many reefs and high density areas within the stock are usually preferentially harvested. Other factors that determine if the whole stock is equally vulnerable to fishing include economic rewards or legal restrictions on the fishers, behavioural changes in the resource and responses of the stock to different population densities, and different rates of fishing or different environmental conditions (Ricker 1975, Gulland 1983). However, the most important limitation of CPUE is that it provides indication of changes in abundance *a posteriori*. Detection of stock declines after they have occurred is not efficient in easily over-exploited trochus fisheries (Sims 1985).

Despite these problems, analysis of CPUE data may still be useful. Royce (1972) suggests the use of all available data and Gulland (1983) suggested when it is "suspected that the available statistics of CPUE do not give a reliable index of abundance ... the only alternative to doing nothing is to use the CPUE data though with suitable precautions about the sort of errors in the final conclusions ... and with maximum attention being given to obtaining better estimates of effort and CPUE". The large samples obtained from catch data can provide valuable information on the dynamics of fished populations (McGowan 1958).

Catch and effort data for trochus in King Sound have been collected since 1981. The Bardi Aborigines Association Council issues trochus fishing permits to individual community members. All permit holders sell their trochus catch to the community council who maintain records of the catch. This method of collecting basic fishery data (from record of first sale) is efficient in providing regular, unmanipulated data on catches (Royce 1972) and these data have not been previously analysed. The aim of this paper is to analyse this historical catch and effort data. The annual production, numbers of fishers and CPUE from 1980 to 1996 will be documented and a general model will be applied to determine a maximum sustainable yield and maximum sustainable effort for trochus in King Sound.

Methods

The license issued to the Bardi Aborigines Association from the Fisheries Department of Western Australia require a monthly record of the catch (in kilograms of dry shell weight) for each individual permit holder. The individual month records date back to January 1981, although statistics for seven months over the 12 years of collection were not available (that is, February, March and May 1984, April 1985, February 1986 and January and June 1987). It was possible no fishing activity occurred during these months, but the records for these months are more likely to be missing. Only the total catch and number of fishers for each month in 1980 were known. The catch data was entered onto a computer database containing 3222 observations and 4 variables, which were month, year, catch (kg) and name of individual fishers. These names were coded and confidentiality of individual fishers has been maintained in all analyses. Catch and effort data are regarded as confidential because it contains information on individual fishers (Royce 1972, Gulland 1988).

The catch data were described in two ways: the total annual production of trochus at One Arm Point was calculated by summing the monthly totals for each year from 1980 to 1995; alternatively, the total catches for the 12 months of each year were presented in a series of graphs to observe any within year differences in catch, perhaps due to seasons.

The only source of fishing effort available with the present catch records was the number of permit holders reporting catches in each month. The only people that were allowed to collect trochus were those with permits. The number of permit holders reporting catches in each month is a simple measure of fishing effort, including both gear and time components. It is probably not a true estimate of fishing effort because individual fishers may not be consistent in the amount of time spent collecting or the amount of shell removed. The difference in effort between fishers was described by determining the total number of months each fisher worked and the average monthly catches for each fisher. These estimates of effort do not account for differences between fishers, and consequently must be used with caution, with consideration given to improving the methods of measuring effort. The total number of fishers for the 12 months of each year was determined. This allowed comparison of effort within years. Comparison of effort from 1980 to 1995 was made from calculating the annual average number of fishers per month.

A correlation between catch and effort, using the total annual production and the annual average number of fishers per month, was tested. A simple linear increase in catch with increasing effort can indicate a reduction in stock biomass, if the CPUE is accurately reflecting stock abundance (Ricker 1975).

A two factor ANOVA was performed on catch per fisher among years and months. The response for the analysis were values of catch per fisher, that is, each sale was used as an independent sampling unit. The analysis was unbalanced in two ways; the number of fishers varied greatly between months and seven months no data were available. An unbalanced ANOVA requires special consideration for testing the underlying assumptions and multiple comparisons tests (Day and Quinn 1989). Two forms of Cochran's test were used to test for homogeneity of variances (Day and Quinn 1989). The data closely approximated homoscedascity and appeared to be normally distributed. Violating the assumption of normally distributed data was acceptable because the total number of observations was extremely large ($n = 3222$). The ANOVA was performed using a general linear model with type III sums of squares on SAS software (SAS Institute Inc. 1987), following recommendations for unbalanced designs with missing cells by Shaw and Mitchell-Olds (1993). A Tukey-Kramer test was used for *post hoc* multiple comparisons of significant factors.

The annual CPUE was calculated according to equation 1, where C_i is the total catch for each month and E_i is the total effort for each month. Therefore, the annual CPUE is described in terms of the average catch per fisher per month with units of kg of dry shell per fisher per month. This allowed comparison of CPUE among years. It is preferable to average the monthly values when calculating the annual CPUE (Gulland 1983), although this assumes any seasonal effects are uniform among years. This is valid because the ANOVA found most of the variation in catch per fisher was attributed to years and there was no significant difference among months

$$\mathrm{CPUE} = \sum_{i=1}^{12} \frac{Ci}{Ei} \qquad (1)$$

Results

Total annual production and effort from 1980 to 1995

The annual catch of trochus has fluctuated since the fishery began in 1979 (Figure 1). The annual production was initially very high in 1980 at 135 t dry weight. The catch decreased between 1981 and 1983, with the lowest annual production of 9 t recorded in 1983. The catch increased after 1984 to 50 t and has stabilised around 50 to 70 t, until 1990 when a slight and steady decline was observed. The catch generally increases in February to May and from September to November (Table 1). This corresponds to the *Iralboo* and *Djalalayi* seasons where weather conditions are more favourable for boating activity.

Variation in the average number of fishers per month provided some indication of the causes for fluctuations in the total annual production. The average number of fishers per month was initially very high at 29 fishers per month (Figure 2). The average number of fishers per month decreased between 1981 to 1984 and was lowest at 4.4 fishers per month. The average number of fishers per month increased to 30 in 1989 and has remained relatively high to 1990. Figure 2 shows the average numbers of fishers per month increased after 1983 until 1989, except during 1988. The effort generally increases in February to May and from September to November (Table 2).

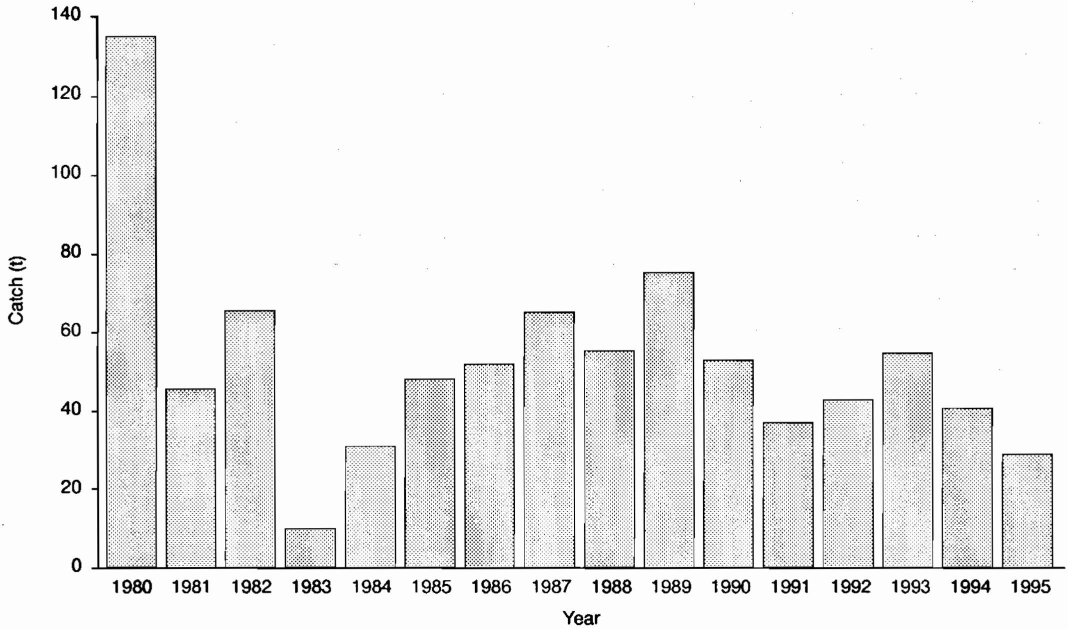

Figure 1. Total annual production of trochus at One Arm Point from 1980–1995. Catch is measured in kg dry shell weight (animal removed).

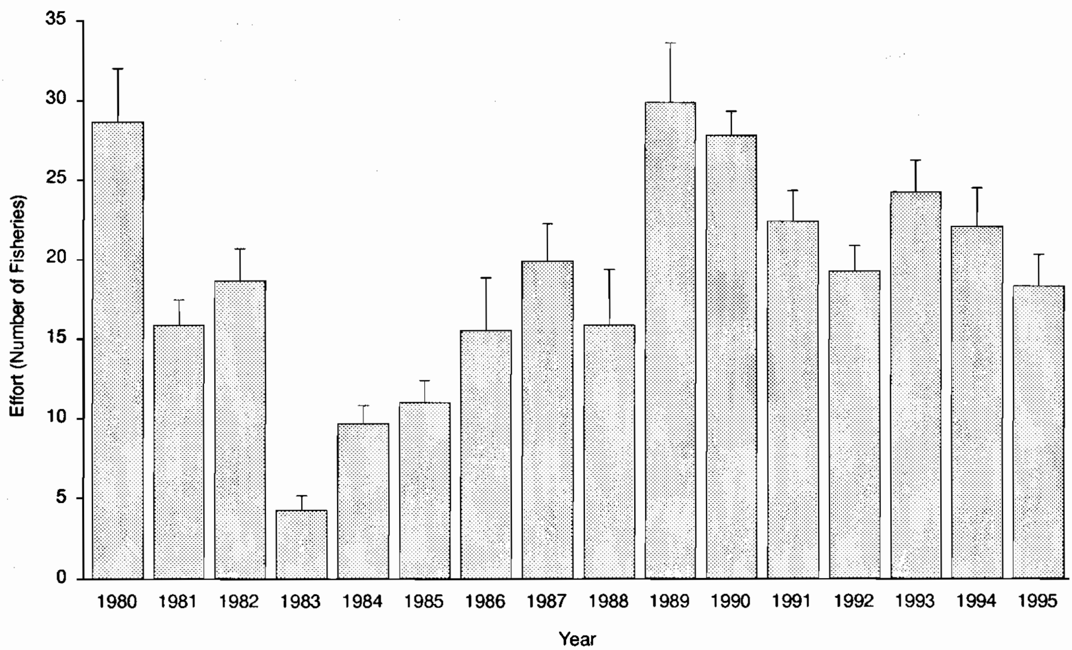

Figure 2. Annual effort (average number of fishers per month) with associated standard errors (n = 12) for the One Arm Point trochus fishery from 1980–1995.

Table 1. Summary of catch (kg dry shell weight) for each month from 1980 to 1995 (* indicates no data available).

Year	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
1980	3833	2577	7407	12878	23542	13404	9644	14154	14858	16624	11833	4468
1981	2251	2587	4055	6810	5336	4034	1903	2842	4963	6413	3416	1144
1982	2706	1511	3653	7628	6457	5784	8414	6503	7482	8113	4075	3383
1983	455	2467	1362	648	90	560	407	565	751	1332	376	239
1984	878	*	*	2823	3172	*	2799	3188	5057	6242	4960	2147
1985	3851	1564	1304	*	4396	4305	3449	7727	6574	5966	5028	3544
1986	3063	*	1958	1508	6610	4622	514	2383	5926	8583	8011	8857
1987	*	3369	6427	5550	3933	*	7011	7065	9615	12549	5316	4069
1988	2759	4333	2025	2607	3285	971	993	1413	3190	10991	14410	8723
1989	6781	4254	1963	2111	4038	2999	5269	9476	8775	12979	8340	8079
1990	4969	4119	4456	3872	5637	2204	4059	4877	6982	5501	3699	2284
1991	3707	1466	3421	2311	4050	2870	5502	3635	3017	3636	1896	1421
1992	2265	1497	2451	3686	3696	5219	3165	3824	3780	5666	4298	3821
1993	2221	2020	6678	5208	3766	6514	5253	4672	5014	5904	4842	2656
1994	3724	2048	3324	2196	5716	5268	6365	2195	4808	3807	714	686
1995	1386	1295	476	2981	3174	2083	2839	2900	3570	3920	3713	1058

Table 2. Summary of fishing effort (number of fishers per month) for each month from 1980 to 1995 (* indicates no data available).

Year	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
1980	14	14	18	32	46	29	26	34	28	50	36	21
1981	17	11	15	24	22	13	8	13	23	22	15	10
1982	12	9	11	26	24	13	26	23	30	19	19	15
1983	5	11	8	5	1	3	2	3	2	8	3	2
1984	4	*	*	8	11	*	9	6	14	15	11	10
1985	11	6	5	*	9	7	8	17	16	14	18	11
1986	11	*	6	7	11	11	4	10	18	35	36	23
1987	*	9	16	19	13	*	22	25	31	33	18	13
1988	12	16	8	12	10	4	6	6	13	32	40	33
1989	30	23	8	18	18	17	35	37	42	50	44	38
1990	33	23	27	34	31	20	27	27	37	30	26	20
1991	28	14	26	21	27	22	28	26	31	24	12	11
1992	15	10	15	20	17	25	14	21	19	24	28	24
1993	10	19	24	29	19	19	33	33	29	27	27	24
1994	27	15	22	17	31	22	31	20	34	29	9	9
1995	12	12	8	17	28	20	14	21	25	24	28	11

Relationship between catch, CPUE and effort

The total annual catch was lowest at 9 t with a corresponding effort of 4.4 fishers per month. The highest annual catch was observed at 135 t with an average annual effort of 29 fishers per month (Figure 3). The relationship between catch and effort was significant ($r = 0.666$, $p < 0.01$, $n = 16$, Figure 3) indicating a proportional increase in catch with increasing effort (i.e. each extra unit of effort results in the same increase in catch). A proportional increase in catch with effort would generally indicate a declining CPUE with increasing effort, if the abundance is decreasing with increasing CPUE. However, the relationship between annual CPUE and effort was not significant ($r = -0.208$, $p > 0.05$, $n = 16$, Figure 4) suggesting the stock abundance was not declining proportionally with increasing effort. This may reflect abundance is not related to CPUE (as discussed in the introduction i.e. stocks are separate and behaving differently), although CPUE may be behaving this way because different areas are being fished, and fishers are then moving on to new grounds. A consequence of there being no relationship is that low annual CPUE was observed across a wide range of annual efforts.

Analysis of CPUE between years and months

The ANOVA showed a significant difference in catch per fisher between months ($F = 2.86$, $df = 11 \times 147$, $p = 0.001$, Table 3). The CPUE was highest at 263 (kg per fisher month), 242 and 233 in October, June and May respectively (Figure 5). These increases occur during the months which correspond to the *Iralboo* and *Dyalalayi* seasons. The lower CPUE of 194 in April, 183 in February and 181 in January were significantly different to October, but similar to the remaining months of the year. Despite the significant difference between months, this factor accounted for only 7% of the total variation in the ANOVA. The interaction between year and month was not significant, but accounted for 34% of the variation. Differences among years account for 59% of the variation in CPUE. The ANOVA found a significant difference in catch per fisher per month among years ($F = 19.12$, $df = 14 \times 147$, $p = 0.0001$; Table 3). The annual CPUE was initially high in 1981 at 237 kg per person per month (Figure 6), but decreased to 174 kg per person per month in 1983. The annual CPUE increased between 1984 to 1985 and was highest in 1985 at 391 kg per person per month. The catch per person per month has steadily decreased from 1985 and was at an all-time low in 1995 at 134 kg per person per month. The significantly steady decline in CPUE from 1985 to 1995 suggests a long period of declining stock abundance.

Discussion

Fluctuations in the catch rates

Problems associated with analysing catch fluctuations occur in all fisheries. The annual trochus production fluctuates in most trochus fisheries (Bour 1990). The catch history trends in trochus fisheries include an initial rapid decline followed by an increase after periods of low fishing effort indicating possible recovery of stocks. This has been observed in Palau, Papua New Guinea and New Caledonia (Bour 1990). Initially high catches are made on previously unexploited stocks followed by declines in catch and CPUE.

Bour (1990) provides a review of fishing and distribution of stocks in the South Pacific, giving the total annual production for each country. The fluctuations are a reflection of trochus being an easy resource to exploit (Bour 1987), but have been attributed to wartime abstinence, alternative economic activity, decrease in market demand, declines in stock abundance, changes in fishing effort and environmental conditions (Moorhouse 1933, McGowan 1958, Nash 1985, Bour 1990). Fluctuations in catch, effort and CPUE can be generally be attributed to economic, social or biological factors. The affect of these factors on the King Sound trochus fishery is given in the following discussion.

Economic causes of catch fluctuations

The value of raw trochus shell can influence trochus catch. Approximate trochus values (per t) obtained from Kim Bridge (Aboriginal Economic Development Corporation, pers. comm.) indicate the Bardi Council received $1000 from 1980–1984, $1400 from 1985–1986, $1500 in 1987, $2200 in 1988 and $4000 in 1989. The increase in value of raw trochus in 1988 was not due to an increase on the world market, but because the Bardi Council started dealing directly with overseas buyers and eliminated the middle man. The price offered for raw shell has consistently increased since 1988 and does not appear to be correlated with trochus catch. However, fishers have become more aware of the prices offered for trochus over recent years, which could increase the influence of value on catches (Kim Bridge, Aboriginal Economic Development Corporation, pers. comm.).

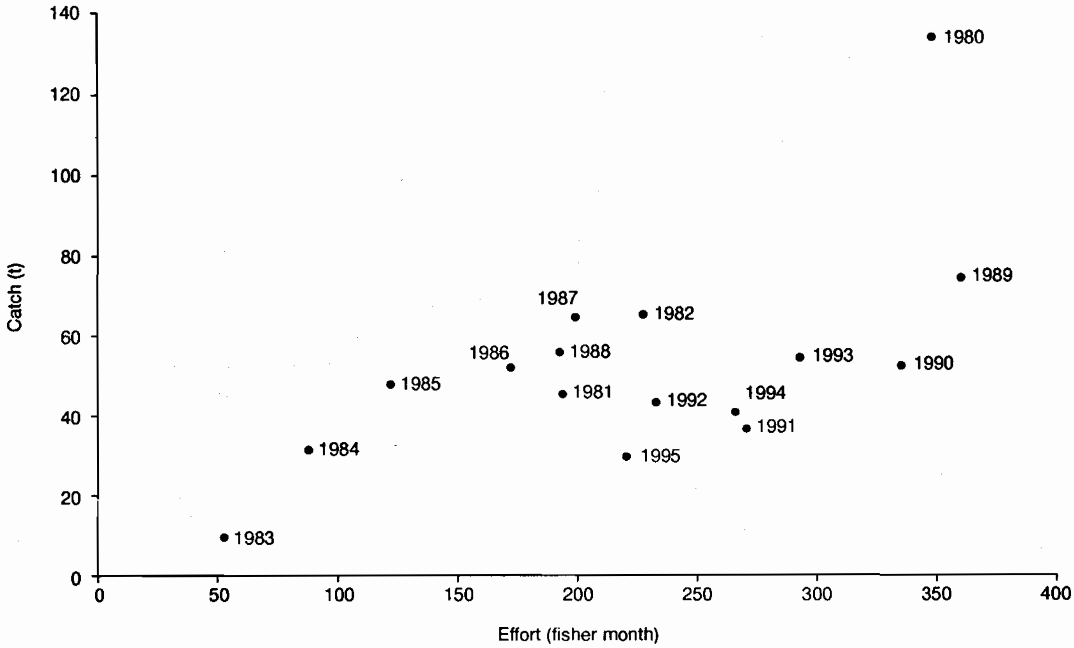

Figure 3. Total annual catch (kg dry shell weight) against effort (average number of fishers per month) for each year from 1980–1995 ($r = 0.666$, $n = 16$). The relationship is expressed by the equation: Catch = 2.74*Effort.

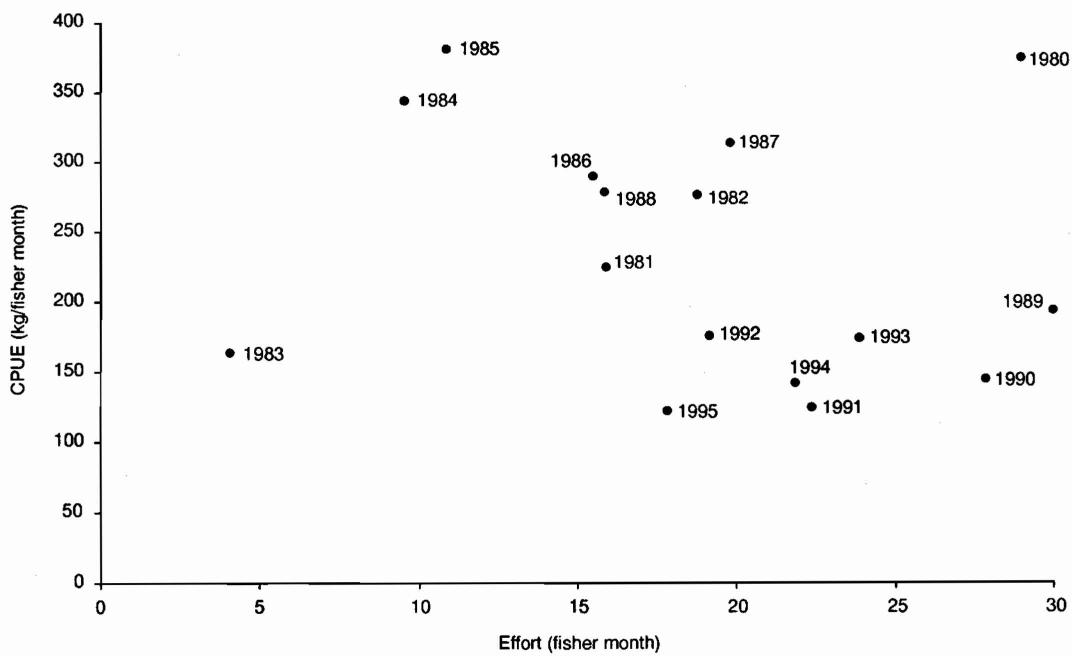

Figure 4. CPUE (kg per fisher per month) against effort (average number of fishers per month) for each year from 1980–1995 ($r = -0.208$, $n = 16$). The relationship is expressed by the equation: CPUE = –2.68*Effort + 295.76.

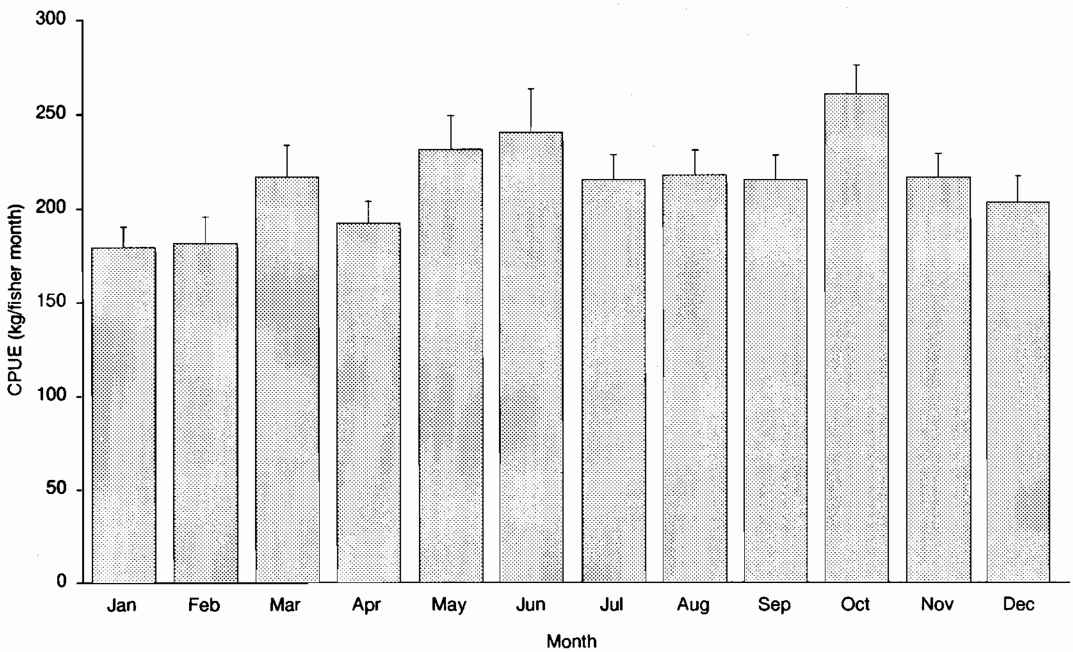

Figure 5. Average CPUE for each month with associated standard errors. CPUE is calculated from an average of the catch per fisher for each month from 1980–1995. Catch is measured in kg dry shell weight (animal removed).

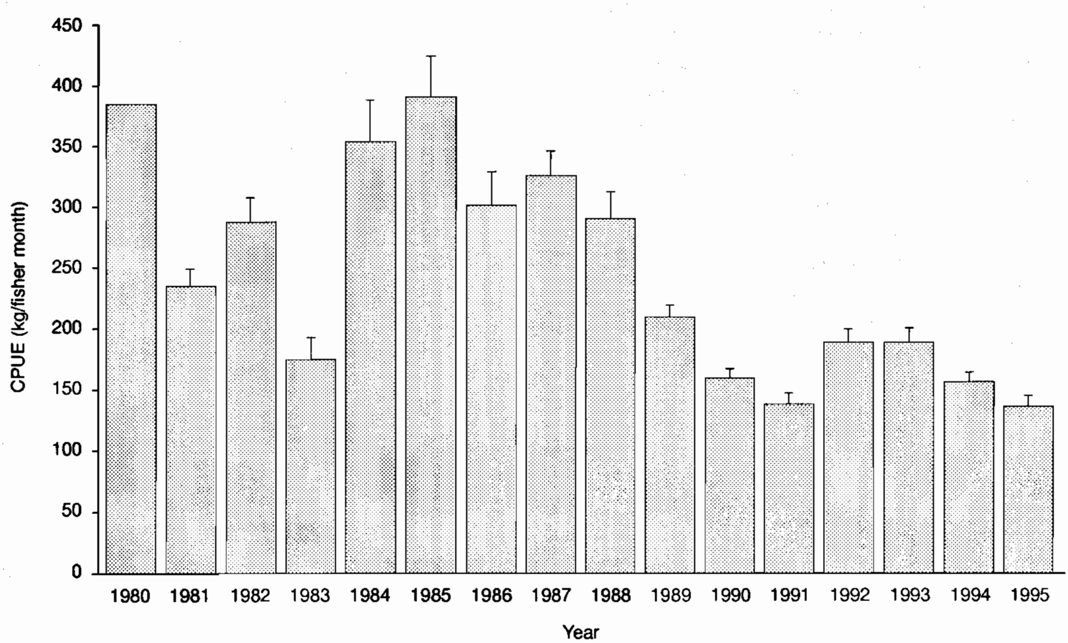

Figure 6. CPUE for each year from 1980–1995 with associated standard errors. The annual CPUE is calculated from an average of the catch per fisher for all months in the year. Catch is measured in kg dry shell weight (animal removed).

Table 3. A two factor ANOVA on catch per fisher per month by year (n = 15) and month (n = 12). Cochran's C = 0.0631; *** = highly significant and ns = non-significant.

Source	df	MS	F	p		Effect
Year	14	1 026 204	19.12	0.0001	***	59%
Month	11	153 628	2.86	0.001	**	7%
Year*Month	147	51 912	0.97	0.5959	ns	34%
Residual	3049	53 680				

Results of a Tukey-Kramer test on CPUE by year. Mean CPUE for each year given.

85 ≥	84 =	87 =	86 ≥	88 =	82 =	81 ≥	89 ≥	92 =	93 =	83 =	90 =	94 ≥	91 =	95
391	355	326	303	290	289	237	209	187	187	175	157	154	137	134

Results of a Tukey-Kramer test on CPUE by month. Mean CPUE for each month given.

Oct ≥	Jun =	May =	Aug =	Mar =	Nov =	Sep =	Jul =	Dec ≥	Apr =	Feb =	Jan
263	242	233	220	219	219	218	217	205	194	183	181

Although the value of trochus varies between countries, the catch fluctuations within countries do not appear to be correlated to price fluctuations. This has been the case on the Great Barrier Reef and the Solomon Islands (Nash 1985, Bour 1990). High prices were offered after the war at a time when stocks were abundant (Glucksman and Lindholm 1982). This makes it difficult to determine if high catches are a refection of high abundance or high economic value. Moorhouse (1933) observed a decrease in trochus production regardless of increasing value. A decline in production can further increase prices because demand for the shell increases. The Great Barrier Reef fishery revived in the 1970s when the price increased because the trochus stocks in many countries had become depleted by over-fishing (Nash 1985). The value of trochus can decline when cheaper alternatives become available on the market; the Great Barrier Reef trochus fishery collapsed in the 1950s with the development of plastic buttons (Nash 1985).

The income generated through trochus fishing is an important source of income for fishers. However, a decrease in trochus value may not affect fishers where alternative incomes can be found. For example, copra is often used as an alternative income in Papua New Guinea (Glucksman and Lindholm 1982) and Vanuatu (Amos 1991).

Social causes of catch fluctuations

The high CPUE at One Arm Point during 1981 and 1982 (compared with 1983) could be attributed to high interest in collecting trochus among community members. The CPUE was possibly low in 1983 due to involvement in a housing project at One Arm Point, and loss of initial motivation as short term money goals were achieved. The catch and effort increased in 1990 but the CPUE was very low. This could be influenced by a change in effort by fishers. However, the number of fishers had been steadily increasing from 1983 to 1989 (Figure 2). The average number of fishers in 1989 to 1991 was as high as in 1980. However, the average catch per fisher per month was much lower in the 1989 to 1991 period than in 1980.

The low CPUE may also reflect a decrease in the amount of time spent collecting. This assumes the effort is remaining consistent between years, but it is likely the fishing effort is changing over time and between fishers according to individual fisher's needs. Comparisons of effort among fishers indicate differences in both the total number of months fished and the average monthly catch. Interviews with fishers indicate the fishing effort has increased and they were more likely to spend increased time fishing during recent years. A low CPUE can be maintained when high prices enable a profit from removing small amounts of shell (Nash 1985). However, fishing activity in other countries can be reduced by lucrative alternative financial incomes, such as selling copra. Fishing activity may also be limited by the costs to fish. Glucksman and Lindholm (1982) observed "rising fuel and maintenance costs have eroded the profit" in Papua New Guinea.

Biological causes of catch fluctuations

The catches during the first few years of a developing fishery are generally high (Bour 1990). This trend was observed in trochus catches during 1980 and 1981. Stocks were abundant because they were virtually untouched commercially. Therefore, the virgin biomass was being removed. However, fisheries are often unable to maintain initial high catches. The catch declined to an all-time low from 1982 to 1984. A similar trend is observed in most trochus fisheries (Bour 1990). For example, trochus stocks in the Torres Strait appeared to be unaffected during the first 2 years of fishing, although stocks were believed to be over-fished only 3 years later (Nash 1985). Catch fluctuations resulting from biological causes can be manifest by declining stocks, decreasing sizes of shells collected and changes in the spatial distribution of fishing grounds.

Decrease in sizes when stock over-fished (Growth-over-fishing)

Size limit regulations are the only management regulation on the trochus fishery in King Sound. The minimum legal size limit was 65 mm when the fishery opened in 1979 (Commonwealth Gazette 1970). The minimum legal size limit was increased to 76 mm in 1983 (Government Gazette 1983), excluding a large proportion of the available stock. A size distribution taken in 1983 indicated very few shells below 70 mm (John Looby, Fisheries Department of Western Australia, pers. comm). The minimum legal size limit decreased from 76 mm to 65 mm in 1984 and a maximum legal size limit of 100 mm was established (Government Gazette 1984). This was likely to have made more stock available for fishing and the catch was likely to have increased. It is possible the populations in 1984, following intense fishing activity, were a different size structure from the virgin fishery which was exploited in 1980. The shells collected in 1983 were smaller than in 1980 as the average number of shells per kilogram was six and nine in 1980 and 1983 respectively (John Looby, Fisheries Department of Western Australia, pers. comm.). The catch increased between 1985 and 1989, with a slight decrease in 1988. It is possible that the population recovered under the low catch rates of 1983 and 1984, as trochus take 2 to 3 years to reach fishable size at 60 mm.

The catch and average number of fishers per month increased in 1990 but the average catch per fisher per month was very low. This most likely indicates a decline in stock levels, but it may also be influenced by a change in effort by fishers. The number of fishers working in the fishery has steadily increased from 1983 to 1989. The average number of fishers in 1989 to 1991 was as high as in 1980. However, the average catch per fisher per month was much lower in the 1989 to 1991 period, than in 1980. This could be interpreted as stocks lower in 1989 to 1990. The low average catch per fisher per month may reflect a decrease in available stock due to heavy exploitation in previous years. Interviews with fishers indicate declining catches; fishers are concerned that they only collect two bags now in locations where the shell was previously abundant. Some fishers may sell only a bucket or a quarter of a bag to the council during one month, whilst before 1987 at least 1 bag was always sold, regardless of an increase in effort. The low average catch per fisher per month, even with an increase in the average number of fishers per month and an expansion of the area fished seems to indicate a decrease in the amount of shell available. Anecdotal information from fishers also supports this.

Decrease in sizes when stock over-fished (Growth-over-fishing)

Over-fishing is initially reflected by reduction in the numbers collected, but eventually the numbers of larger individuals declines until catches are composed of higher proportions of small shells. A reduction in the size of shells collected in Micronesia and Papua New Guinea, and an increase in the number of under-sized shells confiscated from catches, lead to speculation whether the remaining stocks were sufficient to maintain population levels (McGowan 1958, Glucksman and Lindholm 1982). This is true if the stocks remaining are composed of shells below the size at sexual maturity, or have limited opportunities to breed, and the stock is reduced to the point where recruitment is restricted (Cushing 1975).

Over-fishing has certain implications for the future of the fishery, particularly if recruitment over-fishing has occurred. Firstly, the numbers required to maintain catches of smaller shells are much higher than those of larger individuals because the smaller shells weigh a fraction of the larger individuals. Ultimately the catches become difficult to maintain. The Great Barrier Reef trochus fishery removed under-size shells to maintain yearly catches during the early 1900s (Moorhouse 1933). The immediate reaction following recruitment over-fishing is to conserve the remaining stock (McGowan 1958). Unfortunately, this action is *a posteriori*, and has limited benefits to improve stock levels after declines have occurred, particularly the large declines from recruitment over-fishing.

Harvesting of close reefs by fishers

Reefs closest to human centres are generally depleted first, although this is not documented in the trochus catch data from King Sound. Information from fishers and personal observations suggest an early decline in abundance and sizes of trochus stocks closest to One Arm Point. Similar scenarios in other trochus fisheries have occurred. McGowan (1958) reported reefs closest to "relatively large human population centres" declined during the post-war boom while stocks increased on "more remote reefs". This trend is likely as small vessels are restricted to reefs closest to home (Gulland 1988) and fishing remote reefs generally requires extended periods away from home, often involving long boat trips in difficult waters (McGowan 1958). Reefs closest to human populations on the Great Barrier Reef were initially heavily fished, although catches increased during 1917 to 1927 because the fishery was continually moving to unexploited areas (Nash 1985). Similarly during the 1980s fishers moved from Cairns, because of stock declines. following reports of larger quantities in Mackay (Nash 1985). Similar patterns have also been described in abalone fisheries.

An implication of moving from exploited to unexploited fishing grounds is that it may become increasingly difficult to locate areas with harvestable levels of stock (Nash 1985) and catches can remain high over a long period of time, while the stocks are actually declining. Gulland (1983) states "failure to obtain data from all areas arises from the fact that fishers will only stay at places where catches are at least moderate and the commercial CPUE data are likely to give a biased impression of the relative density in the less productive areas". The catches will be unable to reflect the early signs of over-fishing, particularly when the fishing grounds are not indicated on the catch records. Consequently, it is important to understand not only how much shell is being removed through fishing, but from where it is being removed.

Problems associated with catch data

There are many problems associated with the use of catch and effort data from the King Sound trochus fishery and it is difficult to monitor the fishery closely with the present system. The data presented in this paper are presnted, with acknowledgment of the limitations of the results and a discussion of ways to improve the collection of future catch data. The number of fishers may not be an independent measurement as individual permit holders may work in family groups and then distribute the catch between family members or they may sell the total catch through individual permit holders. The weight value for total catch does not include under-sized and worm-encrusted shells that have been eliminated from the total catch. Month as a measure of "time spent fishing" may not account for differences relative to tides and seasonal effects. Although these differences will surface in comparisons between months, they are minimised by taking averages over year (Gulland 1983). This is indicated by the ANOVA of catch per fisher per month.

The fishery could be more closely monitoring inspection of live shells before processing to enable under- and over-sized shells to be returned to the reefs and through issuing CPUE logbooks. Gulland (1983) states "catch data should include all removals from the stock". However, the catch of under- and over-sized shells is often unknown because, while these illegal shells may be collected, they are often eliminated from the catch before being sold. Inspection of live shells would ensure shells outside the legal size limits are returned to reefs, therefore eliminating this portion of the catch which is not usually included in the catch statistics. Gauging shells as they are collected to get 65 to 100 mm shells would be the easiest way as the too small or too large shells aren't handled.

Recording catch and effort in fishers' logbooks provides a simple method of gathering data at the time when catches are being taken (Gulland 1988). The measurement of effort can be specific, for example time spent gathering in hours or more appropriate time units such as the number of tides worked (Royce 1972). It would also be appropriate for the CPUE logbooks to record the size distribution of catches and the location of reefs where shells are collected (Gulland 1983, Nash 1985). Reefs may be divided into area divisions in accordance with possible stock location (Royce 1972, Gulland 1983). Logbooks can also assist in increasing the input of fishers toward management because of the understanding generated. Problems associated with the catch data from the King Sound trochus fishery could be avoided by the use of logbooks.

Estimating MSY from catch data

The catch history presented in this paper suggests serious depletion of the trochus resource in King Sound through over-fishing. This is supported from evidence from trochus and abalone fisheries elsewhere. The fishing pressure was high in 1980 when catches exceeded 70 t and average number of fishers per month was high. Periods of low catches and low catch per fisher are shown to follow high fishing pressure regardless of the number of fishers. It seems likely the sustainable amount for King Sound is much less than 70 t. Most estimates of MSY are derived from the amount collected during maximum effort for example high initial catches. However, MSY may be more accurately derived from stable periods in the catch history. The annual catch at One Arm Point has remained within 40 to 70 t during 7 of the 12 year history. However, the catches decline despite increasing effort and expansion of the fishing grounds. Therefore, it seems the MSY of trochus in King Sound, under the present management is below 40 to 50 t.

Conclusions

The results presented in this paper suggest the trochus fishery in King Sound, like most trochus fisheries (Nash 1985, Bour 1990), is being over-fished and the annual yield is likely to be below 40–50 t, which is less than anticipated from high initial catches. The average production of trochus in King Sound from 50–70 t represents about 5% of the total world production and is much smaller than yearly productions in Fiji, New Caledonia and the Solomon Islands which have annual productions around 250, 400 and 550 t (FAO Yearbook). However, over-fishing is widespread because catch is usually greater than that which can be regularly sustained. The most successful fisheries seem to be those not exploited continuously, for example the Cook Islands. Reef closure not only reduces effort, it eliminates it and enables the stocks to recover. Other action required to compensate over-fishing includes reducing the total amount collected by decreasing fishing effort, implementing quotas or changing size limits. The catch history information presented in this paper suggest changes are required to improve management of the King Sound trochus resource.

Acknowledgments

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Estimating the Total Habitat and Biomass of Trochus in King Sound, Northwestern Australia

K. L. Magro*

Abstract

Trochus shells are fished from intertidal reefs in King Sound, northwestern Australia. The fishery currently produces approximately 50 t annually, but the biomass of trochus has never been estimated in the 16-year history of the fishery. Biomass estimates define the amount available for fishing and assist in determining fishing quotas. The total area of trochus habitat in King Sound was determined from a Landsat TM satellite image using seven spectral signals with a spatial resolution of 30 ¥ 30 m. The predictive success of spectral signals for classifying individual transects into reef edge or reef flat habitats was 96.20%. The satellite image was used to determine to total area of reef edge (9 556 ha.) and reef flat habitat (23 837 ha.) in King Sound. Extrapolation of mean trochus densities and total areas of reef edge and reef flat habitat indicated a total biomass of legal size trochus (65 to 100 mm) of approximately 70 t of trochus in King Sound. The total biomass of under size trochus (below 65 mm) was estimated to be approximately 40 t of trochus in King Sound.

A commercial fishery for trochus (*Trochus niloticus* Linnaeus, 1767) exists in King Sound, northwestern Australia. Annual catches of trochus have declined from the 1980 high of 135 t. There is no published information concerning this fishery but anecdotal accounts suggest localised depletion of trochus is occurring, especially trochus on reefs closest to access points. Although the fishery has been operational since 1979, there have been no surveys of the abundance of trochus in King Sound and the virgin biomass is unknown.

An estimate of the total number of trochus for the total area where trochus occurs is known as the standing stock. Determination of total standing stock requires extrapolating density estimates across the entire fishing region. This requires estimates of mean trochus density and the total area of trochus habitat. There have been several studies estimating the total area of trochus habitat by planimetry methods using maps and aerial photographs in the Cook Islands (Zoutendyk 1991), Palau (Ngiramolau et al. 1991) and Kosrae (Molina et al. 1991). The application of remote sensing techniques using satellite imagery has been used most recently to estimate the total area of trochus habitat, including simulated SPOT satellite imagery in New Caledonia (Bour et al. 1986); and Landsat TM satellite imagery in the Cook Islands (Nash et al. in press), Torres Strait (Long et al. 1993) and the Great Barrier Reef (Larkum 1993).

Trochus catches are recorded by the weight of shell collected. Therefore, it is often useful to discuss the amount of trochus available in the population in terms of the total wet weight. The standing stock can be converted to biomass, which is a measurement of the total wet weight of the population, if the size frequency distribution and the size weight relationship of the standing stock are known. Previous surveys indicate the relationship between size and weight for trochus in King Sound can be described by the

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following relationship (n=1730 $r^2 = 0.998$; Magro in preparation).

$$\text{Total wet weight (g)} = -8.39 * \text{Maximum basal diameter (mm)}^{3.12} \quad (1)$$

The values in this relationship can be used to estimate the biomass of trochus, according to the size distribution associated with the density estimates. Additionally, the size frequency distribution of the standing stock can be used to estimate the percentage of the total standing stock that can be exploited. For example, in the Cook Islands the mean maximum basal diameter of 10.1 cm indicated 58% of the population was within the legal size range of 8 to 11 cm (Zoutendyk 1991).

The aims of this paper are to determine if a satellite image of King Sound can detect spectral groups correlating to trochus habitat; to estimate the total area of trochus habitat; and to establish estimates of the standing stock and biomass of trochus in King Sound.

Methods

Application of satellite imagery to estimate trochus density

A Landsat Thematic Mapper (TM) satellite image of King Sound is held at the Remote Sensing Applications Centre of the Department of Land Administration. The image was recorded on 27 August 1986 at 9:40 am during a low spring tide (Australian National Tide Tables 1986). The trochus fishing area within King Sound was isolated. The image was rectified using 17 control points and enlarged to correspond with 1:50 000 navigational charts of regions within King Sound. This produces a geometrically correct image, aligned to the Australian Map Grid (AMG), direct comparison with latitude and longitude bearings or AMG co-ordinates. The mean map error of the image was 22 m, which is less than the dimensions of a single pixel.

Landsat TM images classify ground covers using seven spectral signals with a spatial resolution of 30 × 30 m (Table 1). Radiance values received by the satellite contains contributions from atmospheric scattering, surface reflectance and substrate upwelled radiance for each spectral signal. Data values correspond to radiance, a measure of reflectance, and values represent digital counts of radiance scaled between 0 to 255. Higher reflectance, usually indicates higher penetration, invokes a higher digital count. The satellite image was analysed using the International Image System (I^2S) software processing system at the Remote Sensing Applications Centre.

Table 1. Summary of spectral bands classified by Landsat TM.

Spectral band	Band number	Wavelength (μm)	Spatial resolution (m)
Blue	1	0.45 to 0.52	30 × 30
Green	2	0.52 to 0.60	30 × 30
Red	3	0.63 to 0.69	30 × 30
Infra-red	4	0.76 to 0.90	30 × 30
Mid Infra-red	5	1.55 to 1.75	30 × 30
Far Infra-red	7	2.08 to 2.34	30 × 30
Thermal	6	10.40 to 12.50	120 × 120

Estimating the total area of trochus habitat in King Sound

The total number of trochus in King Sound can be calculated if the total area of suitable habitat and a reliable estimate of density are known. The transect data provided classification of 91 known reef areas which could be classified according to actual reef site (edge or flat). Each transect corresponds to a single pixel on the satellite image. The location of each transect was placed on the satellite image using the GPS co-ordinates recorded when density estimates were made for each transect. Each pixel was visually inspected to check that it came from dry reef flat or reef edge habitat. Twelve pixels were found to lie positioned either in water or on reef flat edges producing incorrect signals or mixels. These transects were: Gregory (1), Jooloom edge (3), Jooloom flat (2), Mardaj edge (5) and Marloogoon flat (1). Consequently, these 12 pixels could not be used for analysis. Although the image was recorded during a spring low, it was not as big a spring low as when the habitat samples were taken (Australian National Tide Tables 1986; 1993), so the location of the 12 transects were under water when the image was taken. Therefore, 79 transects were used for analysis.

Multiple Discriminant Analysis (MDA) was used to determine the ability of the spectral signals to accurately classify the transects into reef edge and reef flat habitats. Discriminating variables included values for the six bands (blue, green, red, infra-red, mid infra-red and far infra-red). Additionally, ratios of the first three bands (green/red, blue/green and blue/red) were included as ratios high-light subtle differences in the satellite image. Long et al. (1993) observed that trochus habitat was best described by a ratio of green to red wavelength, with trochus being most abundant in areas with a high green to red ratio. Jupp et al. (1985) attribute high ratios of green to red to submerged coral and rubble shoal habitats.

Accurate determination of ratios required correction for atmospheric scattering. Each band scatters in the atmosphere differently and variations need to be compensated if ratios are going to be calculated. Blue wavelength has the highest contribution from atmospheric scattering. The dark pixel subtraction method was favoured to correct atmospheric back-scattering (Alex Wyllie, Remote Sensing Applications Centre, pers. comm.). The dark pixel gives the lowest reading for all bands, therefore assuming radiance equals zero and the value produced is the contribution from atmospheric back-scattering. The dark pixel for the King Sound image produced radiance values of 49, 14 and 11 for the blue, green and red bands respectively.

Multiple discriminant analysis was performed using DSCRIMINANT on SPSS/PC+. Unstandardised canonical discriminant function coefficients were calculated because variable units were similar. Wilks Lambda, converted into chi square statistics were used to test the significance of discriminant functions. Significant results from this analysis determine whether the satellite image can be classified accurately into suitable trochus habitat. Classification of suitable trochus habitat was determined for seven regions in King Sound, each corresponding to 1:50000 maps. The total area of suitable trochus habitat was determined by summing the area of suitable habitat in the seven regions.

Estimating the standing stock and biomass of trochus in King sound

The standing stock for legal size and under size trochus was determined by extrapolating the density estimates to the total area of trochus habitat in each of seven regions in King Sound. The weight for the average size of legal size and under size trochus was used to convert the standing stock to an estimate of trochus biomass. The total standing stock and biomass of trochus in King Sound were determined by summing the standing stock and biomass of trochus in the seven regions. The standard error associated with the mean density estimates was used to determine upper and lower limits for standing stock and biomass.

Results

Estimating the total area of trochus habitat in King Sound

Discriminant analysis of spectral signals according to habitat groups produced from ordination and clustering of 15 environmental variables produced an overall correct classification of 64.56% (Table 2). The spectral signals for the 6 signals and 3 ratios predicted 60.9% and 66.7% of the algal pavement and macroalgal pool habitats (combining the algal pavement and macroalgal pool habitats would improve the predictive success to > 70%). Predictive success was higher for the rubble pavement and exposed rock habitats where the spectral signals for the 6 signals and 3 ratios accurately grouped 75.0% of the transects. The first discriminant function was highly significant (Chi square = 76.467, $p < 0.001$, Table 2), explaing 77.60% of the total variation. The variables ranked highest according to unstandardised canonical discriminant function coefficients, therefore contributing the most toward the first discriminant function were (in order): blue/green ratio, green/red ratio and blue/red ratio. The values of these variables were much greater than those for the six spectral signals alone.

Discriminant analysis of spectral signals according to reef site was very effective. The spectral signals for the 6 bands and 3 ratios accurately predicted 88.9% and 100% of the 79 transect into reef edge (n=27) and reef flat (n=52) sites respectively. The overall predictive success of spectral signals produced by the satellite image into reef edge or reef flat classification was 96.20%. Only 3 of all 79 transects were inaccurately grouped (transects 2, 31 and 44).

The first discriminant function was highly significant (Chi square = 97.946, $p<0.001$, Table 3). The green/red ratio was found to have the highest unstandardised canonical discriminant function coefficients, therefore contributing the most toward the first discriminant function. Other high ranking variables included (in order): blue/green ratio, green band, blue/red ratio and red band. These results indicate the satellite image was suitable for classification into reef edge and reef flat habitat. Classification was performed for each of the seven regions in King Sound, producing an image showing the location of reef edge and reef flat habitats and the total number of pixels in reef edge and reef flat habitats within the seven regions (Table 4). The total number of pixels was converted to total area (in ha.) and this was highly variable by habitat and region. For example, 7356 ha of reef flat habitat was determined in the Sunday region while only 1896 ha on the more suitable reef edge habitat was determined (Table 4). Although the area of reef flat and reef edge habitat in the Caffarelli region was more comparable, 3179 and 3595 ha respectively, the Cafferelli region had a much larger area of the more suitable reef edge habitat than the Sunday region (Table 4).

Estimating the standing stock and biomass of trochus in King Sound

The biomass of legal size trochus was variable between the seven regions, the lowest being 2891 kg in Kimbolton and 23554 kg in Caffarelli (Table 4). The trochus biomass in the Sunday region, which is most accessible to One Arm Point, was estimated to be 15766 kg.

Table 2. Summary of Multiple Discriminant Analysis of spectral signals into habitat groups: algal pavement (n = 46); macroalgal pool (n = 21); rubble pavement (n = 4); and exposed rock (n = 8); (a) shows the unstandardised canonical discriminant function coefficients with the level of significance for univariate F-tests of each variable (df = 3,75) and the ranking of variables is given in parentheses; (b) shows statistics for the significance test; and (c) shows a summary of correct classification.

(a)

Signal	DF1		DF2		DF3		p
Blue	-0.1649	(6)	-0.6256	(6)	-0.2380	(7)	0.2866
Green	0.4438	(5)	0.6690	(5)	-1.3020	(5)	0.2358
Red	0.1397	(7)	0.3456	(7)	1.0298	(6)	0.3665
Infra-red	0.0480	(10)	0.0776	(9)	-0.0193	(10)	0.0010
Mid Infra-red	-0.0484	(9)	0.0166	(10)	0.0818	(9)	0.0020
Far Infra-red	-0.0577	(8)	-0.1803	(8)	-0.2169	(8)	0.0155
Ratio Green/Red	12.9398	(4)	16.1733	(3)	53.0094	(2)	0.5260
Ratio Blue/Green	13.4460	(3)	19.0714	(2)	26.9506	(3)	0.2031
Ratio Blue/Red	-15.4920	(2)	-9.0714	(4)	-21.3875	(4)	0.4040
Ratio Blue/Green/Red	55.3809	(1)	55.2190	(1)	-72.2323	(1)	0.1816
Constant	-18.1257		-21.7973		-24.9775		
% variability explained	77.60%		13.31%		9.08%		

(b)

Statistic	Value	Value	Value
Wilks Lambda	0.341	0.735	0.881
Chisquare	76.467	21.878	9.020
df	30	18	8
Significance	< 0.0001	0.2374	0.3406

(c)

Actual group	Predicted algal pavement	Predicted macroalgal pool	Predicted rubble pavement	Predicted exposed rock
Algal pavement	63.0%	13.0%	10.9%	13.0%
Macroalgal pool	23.8%	66.7%	9.5%	nil
Rubble pavement	nil	25.0%	75.0%	nil
Exposed rock	25.0%	nil	nil	75.0%

Overall correct classification = 65.82%

Table 3. Summary of Multiple Discriminant Analysis of spectral bands into reef site groups: edge (n=27) and flat (n=52). (a) shows the unstandardised canonical discriminant function coefficients with the level of significance for univariate F-tests of each variable (df=1×77) and the ranking of variables; (b) shows statistics for the significance test and c. shows a summary of correct classification.

(a)

Band	DF1	Ranking	p
Blue	-0.5351	7	0.0553
Green	1.841	3	0.2565
Red	-1.0732	5	0.0553
Infra-red	0.0204	10	0.0022
Mid Infra-red	-0.0023	9	<0.0001
Far Infra-red	0.3380	8	<0.0001
Ratio Green/Red	-17.9612	1	0.0147
Ratio Blue/Green	9.2616	2	0.0270
Ratio Blue/Red	1.5483	4	0.9782
Ratio Blue/Green/Red	-1.0644	6	0.7134
Constant	12.1033		

(b)

Statistic	Value
Wilks Lambda	0.257
Chi square	97.946
df	10
Significance	<0.0001

(c)

Actual Group	Predicted reef edge	Predicted reef flat
Reef edge	88.9%	11.1%
Reef flat	nil	100.0%

Overall correct classification = 96.20%

The variation in biomass between regions reflects differences in the area of suitable trochus habitat both between regions themselves and between habitat types. Trochus is more abundant in regions with larger proportions of reef edge habitat (Table 4).

The total biomass of legal size trochus for King Sound was calculated to be 71 t of trochus, with a range of 58 to 84 t (Table 5). The total biomass of under size trochus in King Sound was calculated to be 44 t, with a range of 34 to 53 t (Table 5). The total population biomass of trochus is estimated to be 115 t, and 62% of the total population weight was within the legal size limits. The difference between total biomass of under size shells and legal size shells reflects the different average size and corresponding weights of the two groups. Legal size trochus have a larger size which corresponds with a larger weight, compared to under size trochus.

Discussion

This is the first attempt to obtain an estimate of the standing stock and biomass of trochus in King Sound. This study suggests the total population of commercial trochus in King Sound was approximately 70 t. This biomass is similar to annual production between 1985 to 1992, which ranged from 40 to 70 t, approximately 60 to 100% of the current biomass estimate. Additionally, the biomass of under size trochus is estimated to be approximately 40 t, which may or may not be able to support a commercial fishery with similar harvest rates under the present management system.

Table 4. Estimates of the total area of trochus habitat and trochus biomass estimates in seven regions within King Sound. Area estimates were calculated from the number of pixels classified in the ground-truthed satellite image (1 pixel = 30 × 30 m). Standing stock was determined by extrapolating density estimates from transect sampling to total areas. Biomass was determined using a size-weight relationship for trochus (Total wet weight (g) = -8.39 × Maximum basal diameter (mm)$^{3.12}$ n = 1730, r^2= 0.998).

Region	Reef site	Number of pixels classified	Area (ha.)	Standing stock	Biomass (kg)	Total region biomass
Yampi	Edge	1185	1067	27972	6434	
	Flat	5012	4511	11537	2654	9087
Strickland	Edge	490	441	11566	2660	
	Flat	573	516	1319	303	2964
Kimbolton	Edge	371	334	8757	2014	
	Flat	1657	1491	3814	877	2891
Cascade	Edge	536	482	12652	2910	
	Flat	3312	2981	7624	1753	4664
Mermaid	Edge	1934	1741	45652	10500	
	Flat	4226	3803	9728	2237	12737
Caffarelli	Edge	3994	3595	94278	21684	
	Flat	3532	3179	8130	1870	23554
Sunday	Edge	2107	1896	49736	11439	
	Flat	8173	7356	18813	4327	15766
Total						71663

Table 5. Summary of King Sound trochus biomass estimates (kg total wet weight).

King Sound	Lower biomass	Estimated biomass	Upper biomass
Legal size shells	58641	71663	84522
Under size shells	34856	44019	53215
Total	93497	115682	137737

The application of satellite imagery for assisting the estimation of standing stock and biomass of trochus in King Sound was successful. Discriminant analysis of satellite data indicated significant differences between reef edge and flat habitats. An artificial channel created by a ratio of the green to red bands contributed the most toward significantly discriminating between reef edges and flats, supporting earlier research by Long *et al.* (1993). The area mapped as trochus habitat can affect both the accuracy and precision of the final biomass estimate (Hill and Ahmad 1992). Multiple images over time are required to remove errors associated with interpolation and extrapolation of a single image. However, satellite image processing allows mapping of extensive areas quickly, accurately and inexpensively, particularly when suitable classification has been discriminated (Bour *et al.* 1986, Long *et al.* 1993). There is some possible application for selecting areas to reseed wild populations with hatchery reared trochus juveniles based on the findings of this research.

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Establishing a Relationship Between Habitat and Abundance of Trochus in King Sound, Northwestern Australia

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Abstract

The habitat and abundance of trochus in King Sound, northwestern Australia, has never been investigated for the commercial trochus fishery established in 1979. Estimates of trochus habitat and abundance were made from strip transect sampling of reef edge and reef flat habitats. Cluster analysis of habitat data separated the transects into 4 groups: algal pavement (56 transects); macroalgal pool (22); rubble pavement (4); and exposed rock (9). The maximum basal diameter was variable between reefs, especially between fished and non-fished reefs. Although the majority of variation was explained by reef site; larger shells were found on reef edges and reef flats were dominated by smaller shells. There were very few trochus above the maximum legal size of 100 mm. The mean density of trochus within legal size limits (65 to 100 mm) on the reef edge (4.03 ± 0.630 se per 156 m^2, n=35) was significantly greater than on the reef flat (0.39 ± 0.107 se per 156 m^2, n=56). Large numbers of small shells could be attributed to over-fishing. Results from this study can be used in conjunction with satellite images to estimate the total area of habitat, standing stock and biomass of trochus in King Sound.

ABUNDANCE is a measure of the number of animals in a population. From a biological viewpoint, abundance is likely to influence the rates of growth and mortality and the recruitment of juveniles into the population (Krebs 1985; Begon and Mortimer 1986). Abundance can define the amount available for fishing and assist in the determination of suitable fishing quotas. Estimates of abundance collected over time can be used to monitor changes due to fishing and provide an indication of the success of transplants to re-seed reefs. Consequently, developing and validating methods of measuring abundance is important and requires many considerations. Many countries where trochus is harvested are conducting stock assessment, particularly pre- and postharvest surveys. There are several ways of expressing population abundance, such as density, standing stock and biomass.

Density estimates provide an estimate of absolute abundance because the total number of animals for a given spatial area is known. Density estimates for trochus can be obtained from strip transects, of variable lengths and widths. This provides an estimate of the number of trochus for a given area of reef habitat. Trochus density estimates range from a mean of 37 trochus per 100 square metres (24 transects) in Kosrae (Molina et al. 1991) to a mean of five trochus per 400 square metres (20 transects) in Palau (Ngiramolau et al. 1991). However, densities are not always uniform within habitats. Molina et al. (1991) found a peak trochus density on the upper reef slope at the 3 m contour but density decreases with increasing depth. The number and size of strip transects within the reefs need to be adjusted according to the trochus density. This will ensure that the variance associated with estimates of mean trochus density within reefs is reduced (Green 1979; Andrew and Mapstone 1987). This increases the likelihood of detecting differences in mean trochus density estimates between reefs.

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It is likely that trochus density will be variable according to the habitat, as some habitats are more suitable than others. The aims of this paper are to investigate the relationship between habitat and abundance of trochus in King Sound and to estimate trochus density.

Methods

Study sites

Most of the intertidal reefs adjacent to One Arm Point have been main trochus fishing grounds since the fishery began in 1979, and the number of shells in this region have declined following intense fishing pressure. It was necessary to discontinue fishing activity on some reefs to provide an area for trochus populations to regenerate, generating unexploited conditions with increased population levels and larger size distributions. The idea of closing (or resting) reefs is not a new one to the Bardi Community; several reefs were closed in the early 1980s by the Bardi Council to provide a rest from intense fishing activity (Eric Hunter, Bardi Aborigines Association, pers. comm.). Four reefs were closed from fishing activity for research purposes in October 1990 following consultation and discussions with the community members and elders. The reefs chosen were *Niiwardinggoon* (16°23′S and 123°06′E), *Mardaj* (16°26′S and 123°04′E), *Marloogoon* (16°25′S and 123°07′E) and *Jooloom* (16°27′S and 123°05′E) which are within 10 km of One Arm Point.

Intertidal reefs, accessible by small dinghies, occur near One Arm Point; *Bawlon* (16°26′S and 123°05′E), *Nambanan* (16°26′S and 123°10′E) and *Ngoolminjin* (16°24′S and 123°03′E) within 10 km of One Arm Point. Reefs further away from One Arm Point were also sampled; *Gararr* (16°25′S and 123°21′E and *Noonba* (16°18′S and 123°19′E) are approximately 30 km from One Arm Point across Sunday Strait. Table 1 lists the latitude and longitude of all the reefs surveyed, along with the English name for each island which the reefs are adjacent to. Reference to study sites throughout this report use the Bardi reef names.

Transect sampling

An assessment of the standing stock of trochus in King Sound was made during an extended field trip from April to May 1993. The tides and weather conditions are most suitable during this time of year for this type of work. Trochus density and reef habitat were quantitatively surveyed at a single site on five fished reefs and at two sites each on the four closed reefs (Table 1). Seven randomly placed 4.8 × 32 m strip transects were taken at each of the 13 sites. The reef habitat was recorded at 1 m intervals along the 32 m line transect in the centre of each strip transect. The strip transect was thoroughly searched for trochus, providing an estimate of trochus density (number of trochus per 153.6 m^2). The maximum basal diameter of all shells collected in each transect was measured using vernier calipers to the nearest 0.05 mm. A reading of the Australian Map Grid co-ordinates was taken at the centre of each line transect using a Magellan™ Global Positioning System (GPS). The longitude and latitudes of each transect are given in the Appendix.

Habitat description

Reefs were described according to the substrate and vertical structure of the exposed reef and the flora and fauna present. Substrate was classified as rock, rubble, sand or rock/coral pavement (comprised of either dead coral matrix or consolidated substrate). Reef height was distinguished into deep pools (< 20 cm depth), shallow pools (0 to 20 cm depth), flat, small elevation (0 to 1 m height from flat) or large elevation (> 1 m above flat). Flora and fauna were classified into small macroalgae, large macroalgae, turfing algae, green algae species (for example *Ulva* sp. and *Caulerpa* sp.), live coral (for example *Goniastrea* sp., soft coral, sponges, ascidians, barnacles, oysters, holothurians, zooanthids and anemones.

The habitat measurements taken along each transect were summed to provide a total count for each habitat variable in each transect. These totals were divided by 33 to give the percentage cover of each habitat variable in each transect. Principal Components Analysis (PCA), an ordination technique, was used to determine the major relationships between the habitat variables and to establish dominant habitat types. Cluster Analysis using UPGMA and Ward's cluster procedures were used to define the habitat groups, which were displayed on a plot of the first two principal components. Principal components with eigenvalues > 1 were deemed significant and variables with eigenvector loadings > +0.30 and < –0.30 contributed most significantly toward the principal component. PCA and Cluster Analysis were performed using PRINCOMP and CLUSTER programs respectively on SAS software (SAS Institute Inc. 1987). The dominant flora and fauna components were small macroalgae, large macroalgae, turfing algae, barnacles and oysters. The remaining flora and fauna components: including live coral; soft coral; ascidians; holothurians; zooanthids; sponges; fleshy green algae species (for example *Ulva* sp.); and other green algae (for example *Caulerpa* sp.) were present in only a few transects and were therefore excluded from analysis.

The outcome of these analyses could enable transects and the associated density estimates to be allocated into habitat types, and analyses made comparing density among these *a posteriori* groups.

Comparison of trochus size distributions between reefs

Size frequency distributions of trochus were plotted for each reef by pooling measurements for trochus collected in the seven transects on each site and allocating size measurements to 5 mm size class intervals. The total number of trochus measured varied between reef sites: Bawlon (n=6), Gararr (n=25), Nambanan (n=8), Ngoolminjin (n=58), Noonba (n=3), Jooloom edge (n=59), Jooloom flat (n=7), Mardaj edge (n=25), Mardaj flat (n=7), Marloogoon edge (n=30), Marloogoon flat (n=2), Niiwardinggoon edge (n=43) and Niiwardinggoon flat (n=25). The size measurements were useful for selecting size classes for calculating density estimates, especially as density estimates are usually only concerned with estimates of the standing stock of shells that can be legally collected within the set size limits. Consequently, trochus were placed into three groups according to their size: within the legal size range (65 to 100 mm); below the minimum legal size limit of 65 mm; and above the maximum legal size limit of 100 mm.

Estimates of trochus density

A nested analysis of variance (ANOVA) was used to compare the density of trochus between reef status (closed and fished reefs) for reef flat habitats only. The response variable for the ANOVA were measurements of the number of trochus per 153.6 m^2 from seven random replicate transects taken from eight random independent reef flat habitats. A two factor ANOVA was used to compare the density of trochus between reef flat and reef edge habitats for closed reefs only. The response variable for the ANOVA were measurements of the number of trochus per 153.6 m^2 from seven random replicate transects taken from reef edge and flat habitats from four random closed reefs. Cochran's test was used to investigate the assumption of equal variances (Day and Quinn 1989) and a log (x+1) transformation on trochus density data for both ANOVA's gave the best approximation to homoscedascity. The transformed data also gave a closer fit to normality according to the Shapiro-Wilk test using JMP software. ANOVA's were performed using a general linear model with type III sums of squares on SAS software (SAS Institute Inc. 1987). Separate analyses were performed for legal size and under size trochus.

Table 1. Summary of names and locations of the 13 reefs researched in King Sound. Longitude and latitudes were calculated from 1:50 000 maps of King Sound.

Status	Habitat	Bardi reef name	English name	Latitude	Longitude
Closed	Edge	Jooloom	Middle Island	16°27′S	123°05′E
Closed	Flat	Jooloom			
Closed	Edge	Mardaj	One Arm Point	16°26′S	123°04′E
Closed	Flat	Mardaj			
Closed	Edge	Marloogoon	Tallon Island	16°25′S	123°07′E
Closed	Flat	Marloogoon			
Closed	Edge	Niiwardinggoon	Rees Island	16° 23′S	123° 06′E
Closed	Flat	Niiwardinggoon			
Fished	Flat	Bawlon	Waterlow Island	16°26′S	123°05′E
Fished	Flat	Gararr	Mermaid Island	16°25′S	123°21′E
Fished	Flat	Nambanan	Sunday Island	16°26′S	123°10′E
Fished	Flat	Noonba	Gregory Island	16°18′S	123°19′E
Fished	Edge	Ngoolminjin	Apex Island	16°24′S	123°03′E

Results

Habitat description

The means and standard error for flora and fauna at each reef location are presented in Figure 1. Small macroalgae, large macroalgae and turfing algae are the most abundant biological feature on all reefs sampled. Reefs are generally dominated by turfing algae. The abundance of small macroalgae is greatest on the four closed reef flat sites. The most dominant faunal reef components listed in Figure 1 indicate barnacles and oysters are most abundant on reef edge sites; barnacles and oysters presented on only four reef flat sites in very small amounts.

The most common reef substrate is composed of rock/coral pavement (Figure 2). The five reef edge sites are characterised by 20% to 50% dominance of rock substrate. Sand is present in varying amounts at all 13 sites, although it is generally more common on the reef flat sites.

The means and standard error for reef height are presented in (Figure 3). Most reefs are generally flat, although elevations are present on all of the five reef edges. A very small proportion of elevations are found on only three reef flat sites. Pools occur on all reefs, however, deep pools (> 20 cm) are found most commonly and in greater proportions on reef flats.

Classification of individual transects using the habitat data (excluding large elevation) separates the transects into four groups. The same clusters appear using UPGMA and Ward's clustering techniques. The clusters presented in Figure 4 correspond with the following habitats: algal pavement (n = 56), macroalgal pool (n = 22), rubble pavement (n = 4) and exposed rock (n = 9).

Principal Components Analysis of 14 environmental variables is used to ordinate these habitat variables into significant principal components (those with eigenvalues > 1). The first four principal components have eigenvalues > 1 and are therefore considered significant. These four principal components explained 72.29% of the overall variability. The first two principal components explain 49.33% of the variability (Table 2).

Variables with eigen vector loadings > 0.30 or < 0.30 contribute significantly toward the principal component. PC1 is explained by the variables barnacles, oysters, rock and small elevations. Large macroalgae, flat, shallow and deep pools are most significant in explaining PC3. The third principal component is explained by small macroalgae, rubble, sand and pavement. The fourth significant principal component is explained by small macroalgae, turfing algae and rubble.

The separation between transects according to the principal components is evident between algal pavement and exposed rock habitats (Figure 4). The macroalgal pool habitat is also well separated from exposed rock habitat, but overlaps slightly with algal pavement. Rubble pavement shows no separation from the algal pavement habitat, but is distinguishable from macroalgal pool and exposed rock habitats.

The means and standard error for habitat components in each habitat group are shown on Figure 5. The algal pavement habitat is dominated flat, coral/rock pavement with turfing or small macroalgae. The macroalgal pool habitat is dominated by coral/rock pavement, large proportions of macroalgae, plus shallow and deep pools. Rubble pavement habitat is characterised by flat rubble substrate with turfing or small macroalgae. Finally, the exposed rock habitat is described by elevated rock substrate with barnacles and oysters. These descriptions are not totally exclusive, for example flat rock/coral pavement, which is associated with algal pavement habitat is also a major component of macroalgal pool habitat.

The transects do not necessarily group according to the reefs from where they were sampled. However, exposed rock habitats occur at Niiwardinggoon edge and Ngoolminjin. All transects in the rubble pavement habitat were sampled at Bawlon. Generally, all reef flat samples are described by both algal pavement and macroalgal pool habitats.

Comparison of trochus size distributions between reefs

The size frequency distributions from each reef are given in Figure 6. Interpretation of these distributions is hindered by the low abundance on most reefs. Distributions with higher abundances show more readily identified modes. The total abundances are generally higher at reef edge locations (including Ngoolminjin). The five fished reefs, except Ngoolminjin, have lower abundances. This is also true of the reef flat samples. Furthermore, the size distribution of fished reef and reef flat samples are dominated by shells below the 65 mm legal limit. The closed reef edge samples have the highest proportions of legal shells (between 65 to 100 mm). Size was variable between reefs, especially the closed and fished reefs. The majority of variation is explained by reef site; larger shells are found on reef edges and reef flats are dominated by smaller shells. However, there were very few trochus above the maximum legal size of 100 mm (Table 3).

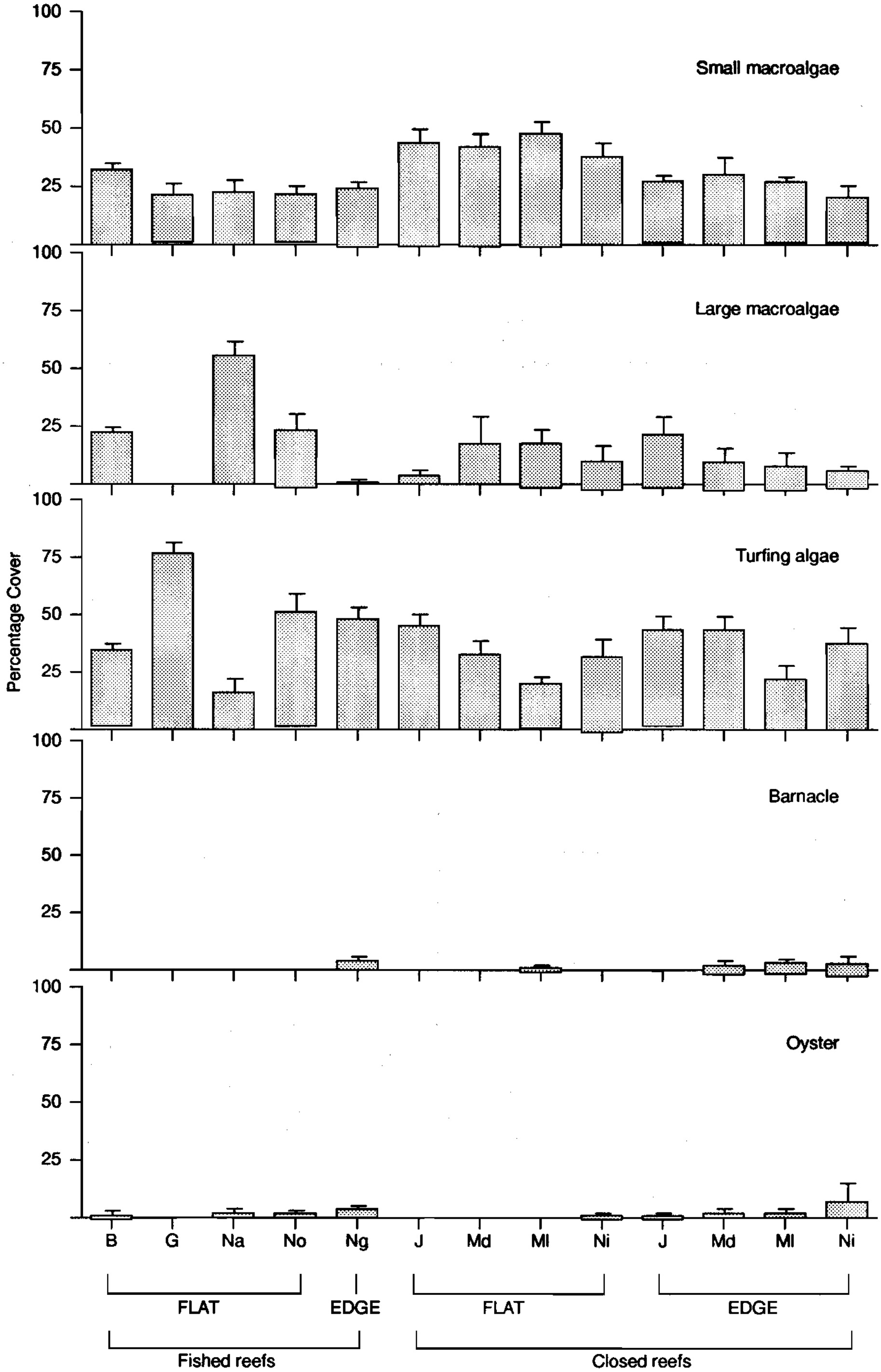

Figure 1. Mean percentage cover (and standard error of the mean, n = 7) for flora and faunal habitat components (where B = Bawlon, G = Gararr, Na = Nambanan, Ng = Ngoolminjin, No = Noonba, J = Jooloom, Md = Mardaj, Ml = Marloogoon and Ni = Niiwardinggoon).

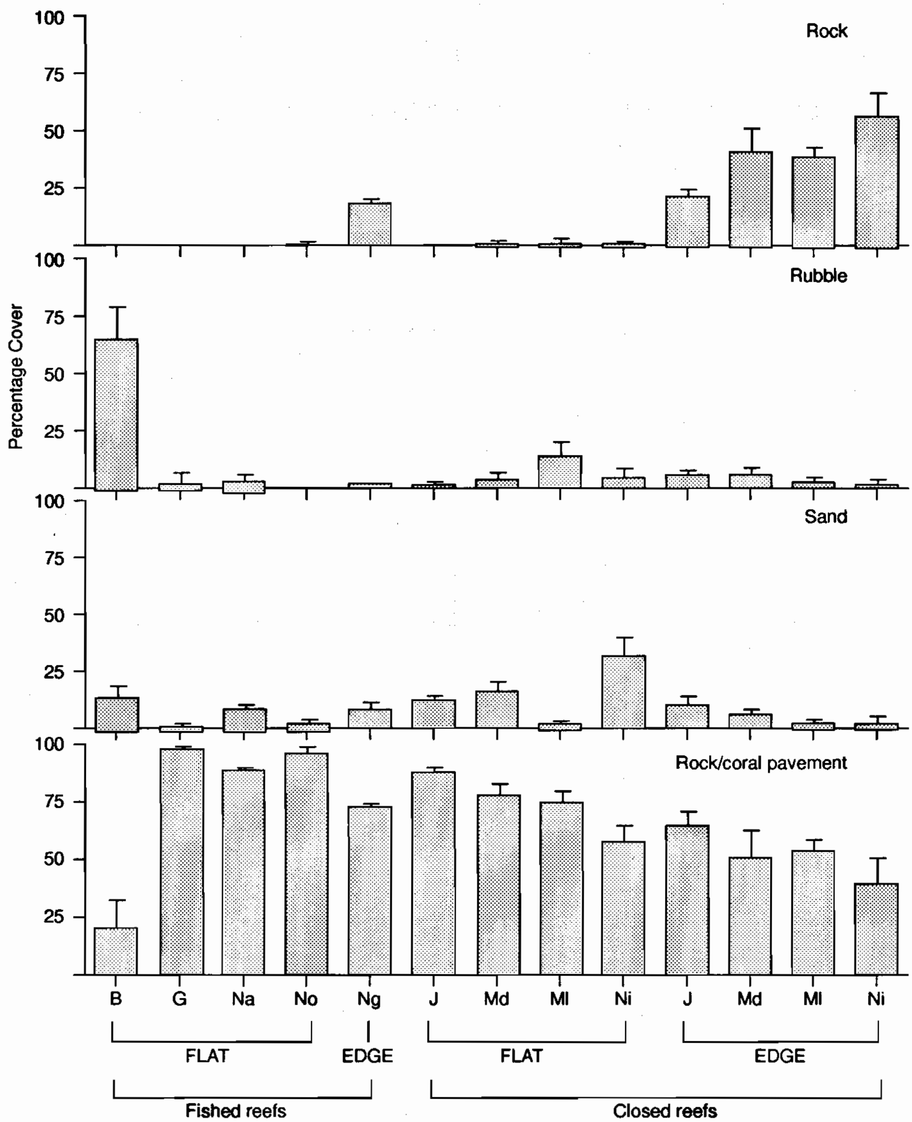

Figure 2. Mean percentage cover (and standard error of the mean, n = 7) for substrate habitat components (where B = Bawlon, G = Gararr, Na = Nambanan, Ng = Ngoolminjin, No = Noonba, J = Jooloom, Md = Mardaj, Ml = Marloogoon and Ni = Niiwardinggoon).

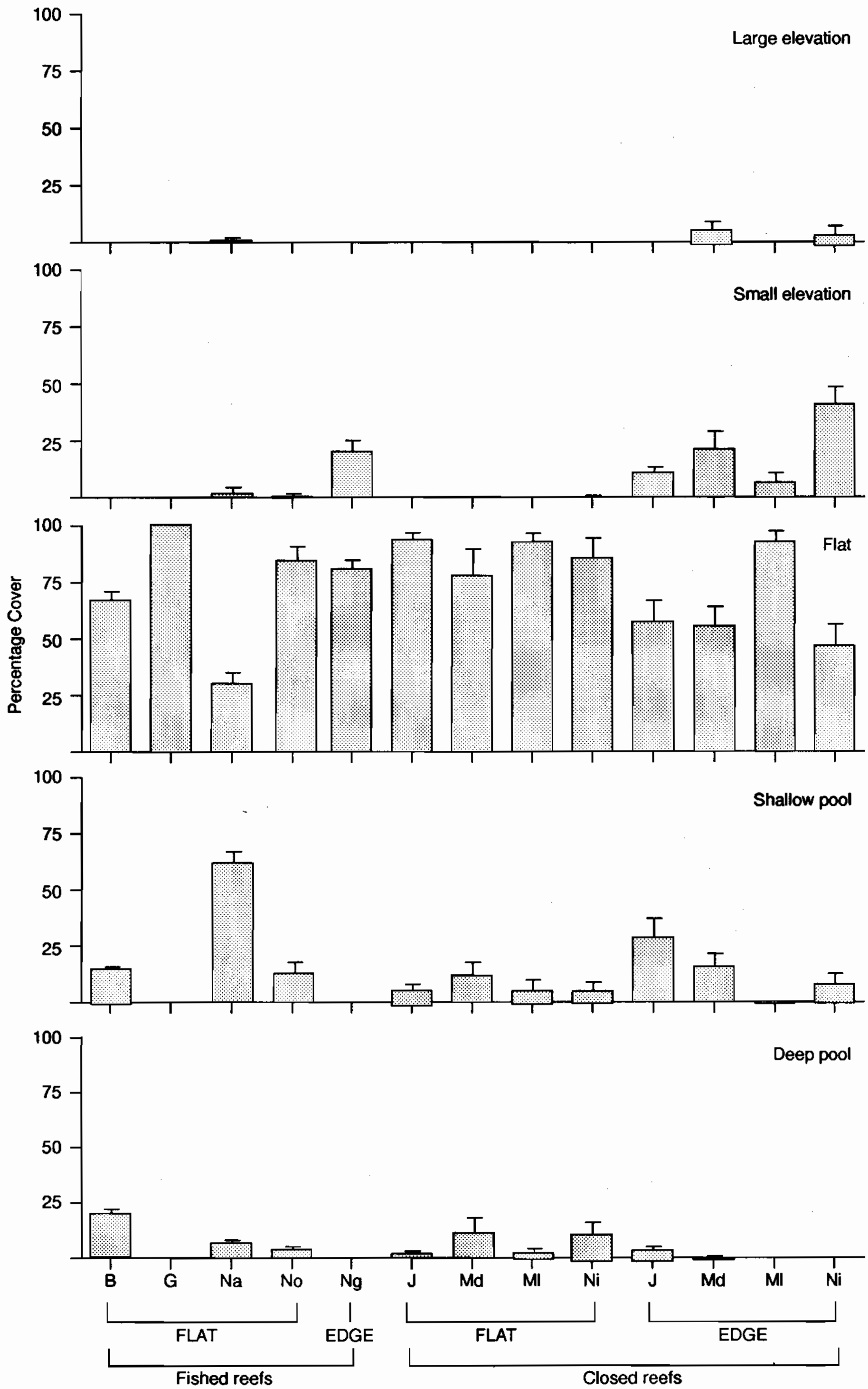

Figure 3. Mean percentage cover (and standard error of the mean, n = 7) for height habitat components (where B = Bawlon, G = Gararr, Na = Nambanan, Ng = Ngoolminjin, No = Noonba, J = Jooloom, Md = Mardaj, Ml = Marloogoon and Ni = Niiwardinggoon).

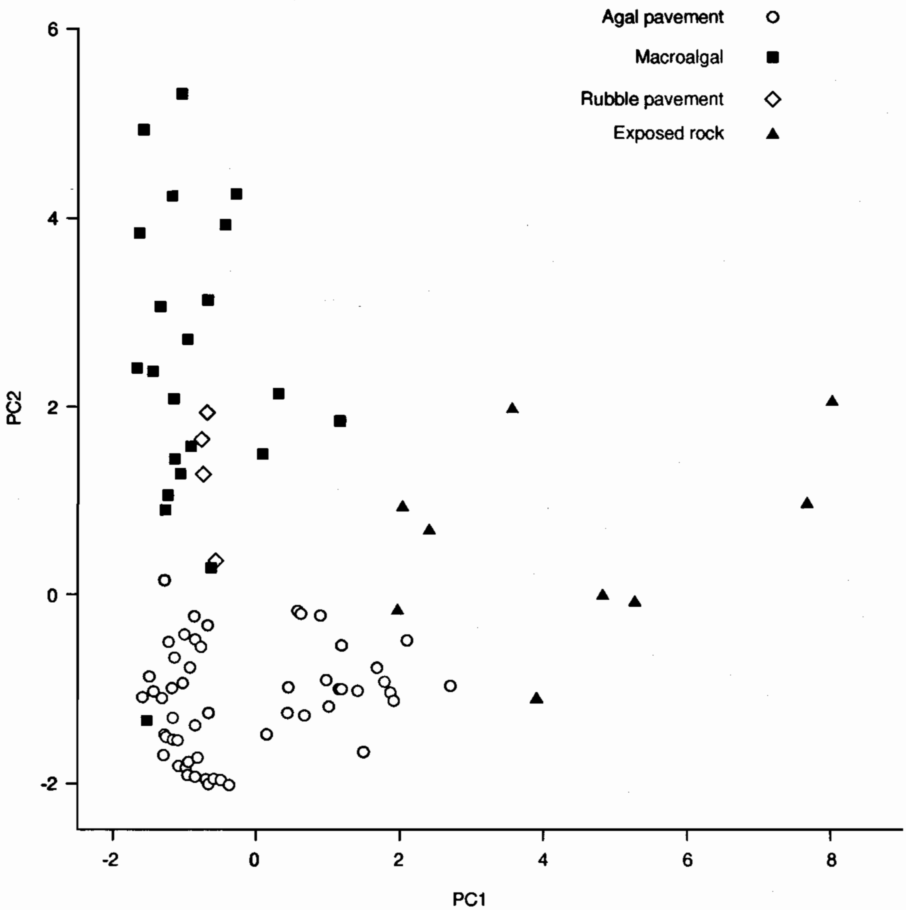

Figure 4. Principal Components Analysis of 14 habitat variables showing four habitats classified using UPGMA and Wards clustering techniques: algal pavement (n = 56); macroalgal pool (n = 22); rubble pavement (n = 4); and exposed rock (n = 9).

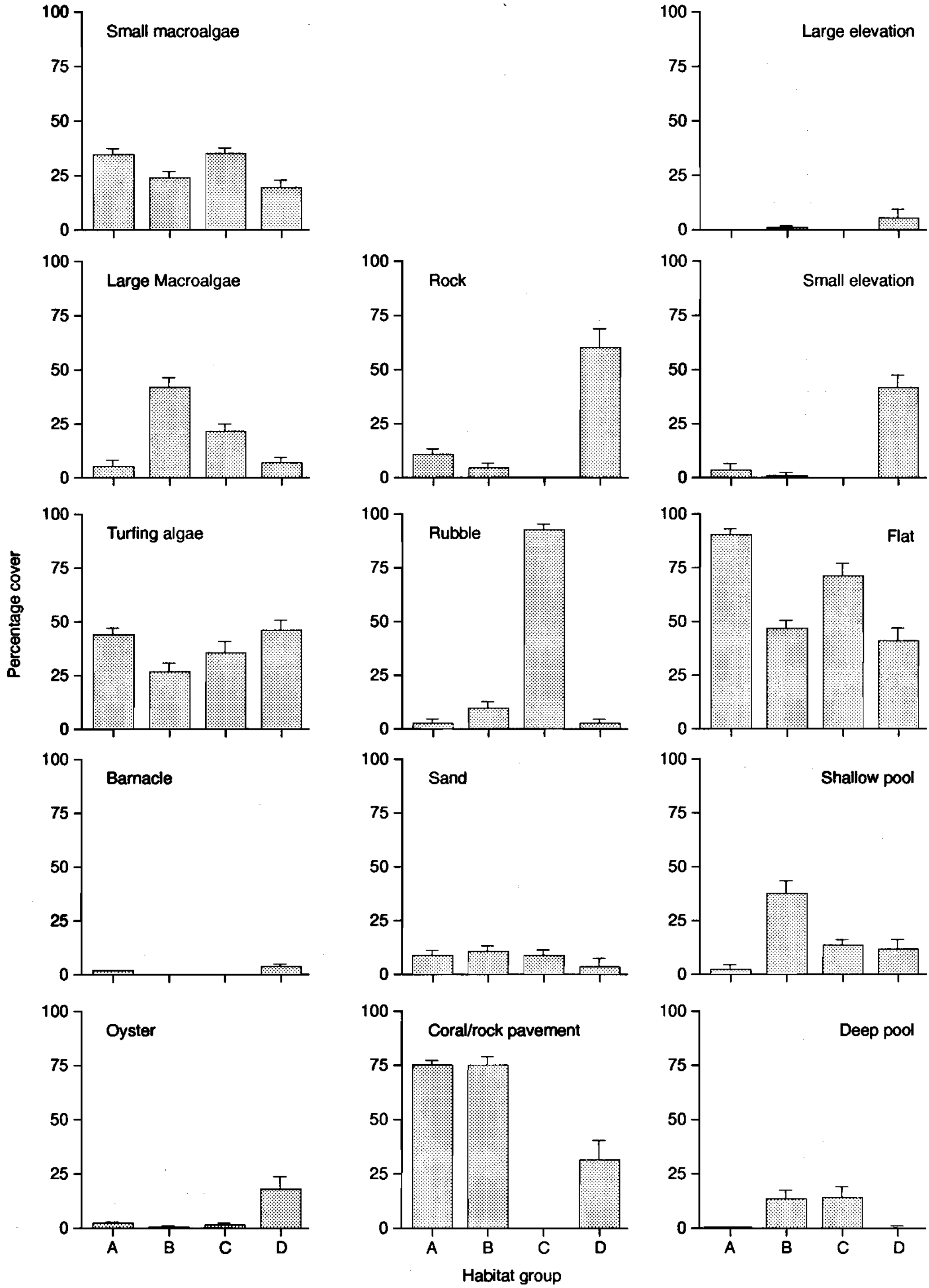

Figure 5. Mean percentage cover (and standard error) of 14 habitat variables between the four habitats separated by PCA: A = algal pavement (n = 56); B = macroalgal pool (n = 22); C = rubble pavement (n = 4); and D = exposed rock (n = 9).

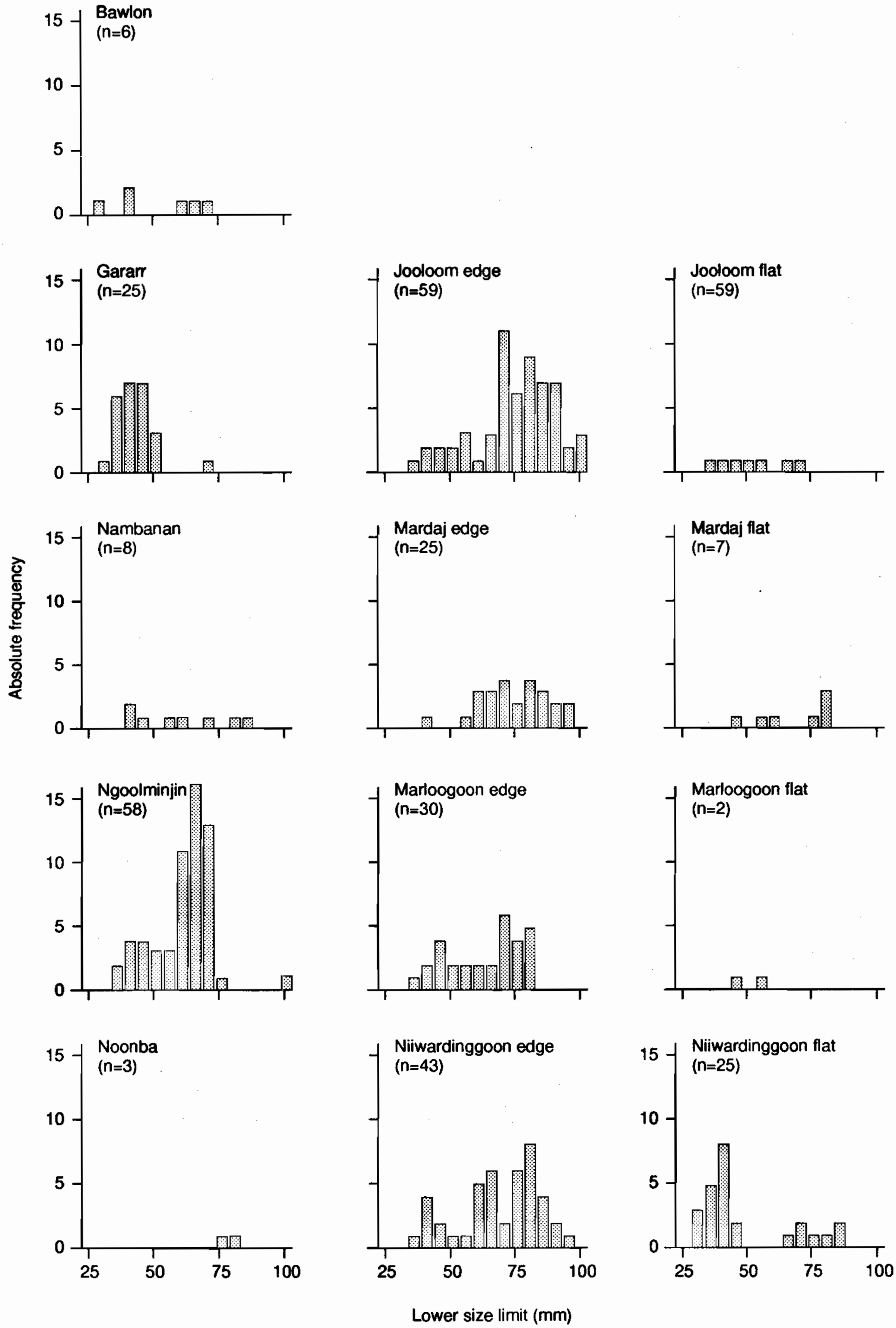

Figure 6. Comparison of size frequency distributions collected from 7 each transects at 13 reef sites in King Sound. Lower size limits of 5 mm size class limits are given.

Table 2. Summary of Principal Components Analysis of 14 environmental variables collected at 91 transects in King Sound. Significant eigenvalues for each principal component are underlined.

Variables	PC1	PC2	PC3	PC4
Small macroalgae	-0.2297	-0.2113	0.3289	0.4266
Large macroalgae	-0.1479	0.4940	-0.1377	0.0382
Turfing algae	0.0596	-0.2859	-0.2784	-0.5625
Barnacles	0.3036	-0.0795	0.0426	0.1662
Oysters	0.4073	0.0770	-0.0048	0.1362
Rock	0.4825	-0.0008	0.0735	0.0898
Rubble	-0.0687	0.1220	0.4738	-0.5027
Sand	-0.1551	0.0118	0.3869	0.2592
Pavement	-0.2559	0.0738	-0.5899	0.1485
Large elevation	0.2589	0.0468	0.0742	-0.0961
Small elevation	0.4568	0.0441	-0.0576	0.0511
Flat	-0.1760	-0.4923	0.0689	-0.0228
Shallow pool	-0.0824	0.4761	-0.1509	0.1419
Deep pool	-0.1525	0.3517	0.1724	-0.2674
Eigenvalue	3.6149	3.2908	1.9067	1.3074
% variability explained	25.82%	23.51%	13.62%	9.34%

Table 3. Summary of the total numbers of under, legal and over size trochus at each reef.

Reef	Under size < 65 mm	Legal size 65 to 100 mm	Over size > 100 mm	Total
Reef Flat				
Bawlon	4	2	0	6
Gararr	24	1	0	25
Nambanan	5	3	0	8
Noonba	0	3	0	3
Jooloom	5	2	0	7
Mardaj	3	4	0	7
Marloogoon	2	0	0	2
Niiwardinggoon	18	7	0	25
Reef Edge				
Ngoolminjin	27	30	1	58
Jooloom	11	45	3	59
Mardaj	5	20	0	25
Marloogoon	13	17	0	30
Niiwardinggoon	14	29	0	43

Estimates of trochus density

Density of trochus was not significantly different between closed and fished reefs on reef flat habitats for both under and legal size trochus (F = 0.0157, p = 0.9044 and F = 0.9169, p = 0.3753 respectively; Table 4). No significant difference in legal size trochus was observed among the 4 reefs (F = 1.1890, p = 0.3282; Table 4), although there was a significant difference in under size trochus among the eight reefs (F = 7.4112, p = 0.0001; Table 4).

There was no significant difference in the density of under size trochus between reef edge and reef flat habitats (F = 2.3283, p = 0.2245; Table 5). Mean shell density of under size trochus was no greater on the reef edge (2.00 shells per 153.6 m^2) compared with reef flats (1.08 shells per 153.6 m^2; Table 6). However, the reef edge habitats had significantly greater densities of legal size trochus than reef flat habitats (F = 17.2855, p = 0.0253; Table 5). Mean legal size trochus densities on reef edge and reef flat habitats were 4.03 and 0.39 shells per 153.6 m^2 respectively (Table 6). Additionally, there was no significant difference in the density of under size trochus among the four closed reefs (F = 2.4301, p = 0.0766; Table 5), but a significant difference among the 4 closed reefs was observed for legal size trochus (F = 3.2880, p = 0.0285; Table 5).

Table 4. A nested ~~ANOVA on density~~ (number of trochus per 156 m^2) by status (fished and closed) for reef flat habitats only. A log (x+1) transformation was used to provide the closest approximation to homoscedascity; *** = highly significant and ns = non-significant.

a. ANOVA for under size trochus (below 65 mm).

Source	df	MS	F	p	
Status	1	0.0050	0.0157	0.9044	ns
Reef (Status)	6	0.3177	7.4112	0.0001	***
Residual	48	0.0429			

b. ANOVA for legal size trochus (65 to 100 mm).

Source	df	MS	F	p	
Status	1	0.0366	0.9169	0.3753	ns
Reef (Status)	6	0.0399	1.1890	0.3282	ns
Residual	48	0.0366			

Table 5. A two factor ANOVA on density (number of trochus per 156 m^2) comparing reef edge and flat habitats for closed reefs only. A log (x+1) transformation was used to provide the closest approximation to homoscedascity; * = significant (p<0.05) and ns = not significant.

a. ANOVA for under size trochus (below 65 mm).

Source	df	MS	F	p	
Reef	3	0.1395	2.4301	0.0766	ns
Habitat	1	0.2040	2.3283	0.2245	ns
Reef * Habitat	3	0.0876	1.5260	0.2198	ns
Residual	48	0.0574			

b. ANOVA for legal size trochus (65 to 100 mm).

Source	df	MS	F	p	
Reef	3	0.2312	3.2880	0.0285	*
Habitat	1	2.7233	17.2855	0.0253	*
Reef * Habitat	3	0.1575	2.2403	0.0956	ns
Residual	48	0.0703			

Table 6. Summary of the mean density of under and legal size trochus on reef edge and reef flat habitats.

Source	n	Mean density of under size trochus (Number of shells / 156 m^2 ± se)	Mean density of legal size trochus (Number of shells / 156 m^2 ± se)
Reef Flat	56	1.1 ± 0.2283	2.0 ± 0.4140
Bawlon	7	0.6 ± 0.2973	0.3 ± 0.2857
Gararr	7	3.4 ± 0.6851	0.1 ± 0.1428
Nambanan	7	0.7 ± 0.4205	0.4 ± 0.4285
Noonba	7	0.0 ± 0.0000	0.4 ± 0.4285
Jooloom	7	0.7 ± 0.1844	0.3 ± 0.1844
Mardaj	7	0.4 ± 0.2020	0.6 ± 0.2973
Marloogoon	7	0.3 ± 0.1844	0.0 ± 0.0000
Niiwardinggoon	7	2.6 ± 1.1096	1.0 ± 0.3779
Reef Edge	35	0.4 ± 0.1071	4.0 ± 0.6304
Ngoolminjin	7	3.9 ± 1.6822	4.3 ± 1.2670
Jooloom	7	1.6 ± 0.5714	6.4 ± 1.5865
Mardaj	7	0.7 ± 0.2857	2.9 ± 1.7515
Marloogoon	7	1.9 ± 0.4040	2.4 ± 1.3067
Niiwardinggoon	7	2.0 ± 0.7559	4.1 ± 0.8571

Discussion

This research has implications for future management of the trochus resource in King Sound. Over size shells are extremely rare, indicating a reduced broodstock to reseed populations. The maximum basal diameters of trochus from all transects were low, possibly indicating growth overfishing of the trochus resource. Furthermore, the average sizes were close to the size where trochus in King Sound become sexually mature (approximately 65 mm). The problem of over-fishing may be increased if abundances are declining in addition to decreases in the average sizes collected. This is particular concern because a reduction in sexually mature shells will reduce the spawning stock biomass, resulting in recruitment overfishing.

There are a number of limitations associated with a small scale study of this nature. In particular, a large number of assumptions are involved with each analysis, e.g. the problems associated with sample size for calculating mean densities of trochus. Additional monitoring of trochus abundance in King Sound would reduce the number of assumptions associated with this research. For example, monitoring changes in abundance over time can allow determination of the impacts of fishing, if the particularly extent fishing activity is known. This is the first attempt to obtain habitat and abundance estimates for trochus in King Sound. The results from this study can be used in conjunction with satellite images to estimate the total area of habitat, standing stock and biomass of trochus in King Sound.

Acknowledgments

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Management Policy for Trochus Fishery in the Pacific

M. J. Amos*

Abstract

Commercial trochus fisheries started throughout the Pacific in the first decade of this century. The harvest of trochus has provided a significant source of revenue and employment in the Pacific region. Trochus has also played a significant role in fishery development in the Pacific. Its introduction into a number of Pacific island states represents one of the more successful facets of fishery development.

While the development of trochus fisheries has been successful in a number of areas, the effectiveness of management policies has been less clearly demonstrated. The status of trochus fisheries in some countries is not known because of inadequate or nonexistent catch statistics. Information provided by fishermen or shell buyers often provides some indication of overfishing. The vulnerability of trochus to overfishing suggests that trochus fisheries may best be conserved by implementing conservative management regimes in the early stages of fishing.

THE marine gastropod *Trochus niloticus* is coral reef-associated and has provided the basis of important inshore fisheries in many Pacific island states (Indo-Australian archipelago, Melanesia, Micronesia and Polynesia). This valuable marine resource has been harvested since early this century, primarily for the manufacture of buttons, decorative inlay work and jewellery. The flesh is edible and is typically cooked or dried.

The harvest of trochus has provided a significant source of revenue and employment in the Pacific region. It forms a substantial opportunity for social and economic development. Adams and Dalzell (1994) estimate that 80% of all harvests from inshore fisheries in the South Pacific are taken for subsistence purposes.

Fisheries for trochus throughout its range in the Pacific region declined in the 1950s when plastics replaced natural shell in button manufacture. It was not until the late 1970s trochus fisheries enjoyed a revival because the fashion houses of Europe and east Asian countries decided to use natural shell buttons on their high-quality shirts (Carleton 1984). In recent years, the demand for trochus has increased, fuelled by the current world demand for trochus shell estimated at 7000 t. The global demand for and market value of trochus shell are steadily increasing, while at the same time the price of and demand for the agricultural commodities that form an important source of income for Pacific island states (copra, cocoa and coffee) are declining. Not surprisingly, interest in harvesting trochus has intensified in recent years. Given the history of overfishing of trochus stocks in many places throughout the species' range (McGowan 1958; Heslinga and Hillmann, 1981; Nash 1985), there is concern for future management of trochus stocks in the Pacific region. The purpose of this paper is to explore past and present paradigms of trochus management with particular focus on experiences in the South Pacific region.

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Fisheries Development and Management

The vulnerability of trochus to overfishing in the South Pacific is probably due to the combination of the following factors: trochus habitat; the shell is easily found despite the inconspicuous colouration of the shell; and larval dispersal is probably limited (Nash 1993). The last means that depleted populations will take longer to regenerate, because recruitment is primarily localised. Recruitment from other reefs is likely to be only slight.

The development phase of a trochus fishery is often very short, and in the absence of proper management systems is often followed by reports of overfishing. This vulnerability to overfishing suggests that trochus fisheries in the South Pacific may best be conserved by implementing conservative management regimes in the early development stages. This section therefore outlines the principal methods by which trochus stocks may have been managed and to describe management issues and options applicable to the trochus fishery.

Management tools

Trochus fisheries in the South Pacific have been managed by a variety of methods. These include size limits, limited fishing season, total allowable catch (TAC), sanctuaries, traditionally based management systems, and stock replenishment. One or more of these management methods have been or are implemented for trochus fishing in many places throughout the species' range in the Pacific region.

Each management policy is dealt with separately in the hope that this process will lead to sensible advice for rational and effective trochus fisheries management.

Size limits

Lower size limit

This management tool (lower size limit) is widely used. Minimum size limits have ranged from 6.2 cm in Solomon Islands to 9.0 cm in Vanuatu and New Caledonia. In principle, a fishery may be managed by a minimum size limit alone, that is, if the size limit is set high enough, a sufficient proportion of the egg production may be protected by the size limit and recruitment can be sustained, regardless of the level of fishing pressure on the fraction of the population that is larger than the size limit.

The following Pacific Island countries have adopted minimum size limit policies for trochus fisheries management.

Country	Minimum size limit
PNG	2.5 inches (6.35 cm)
Solomon Islands	2.5 inches (6.35 cm)
Chuuk	3.0 inches (7.62 cm)
Kosrae	3.0 inches (7.62 cm)
Pohnpei	3.0 inches (7.62 cm)
Yap	3.0 inches (7.62 cm)
Palau	3.0 inches (7.62 cm)
Fiji	3.5 inches (8.89 cm
Cook Islands	8.0 cm
French Polynesia	8.0 cm
Vanuatu	9.0 cm
New Caledonia	9.0 cm

The minimum size limits are implemented in the form of legislations, which prohibit the harvest of shells with diameters less than the specified size limits. Any person who contravenes the size limit provisions is guilty of an offence and is liable to pay a specified fine.

A limitation of the minimum size limit only approach is that, at high levels of fishing pressure, the size limit necessary to conserve the stocks would be high, perhaps 10.0 or 11.0 cm. However, shells this size are of much lower value than smaller shells. If the price for trochus shell on the global market continues to rise, fishing pressure is likely to follow the same upward trend. Because the impact on trochus resources will continue to increase if the price continues to rise, management by size limit alone would require that the minimum size limit be uneconomically high.

Experience within the Pacific region has shown that the instigation of minimum size restriction alone for trochus fishing is not very effective. Financial constraints faced by the Fisheries Department have meant that enforcement has not been very effective.

Upper size limit

This management option (upper size limit) has been implemented together with the minimum size limit by a number of Pacific Island countries. It ranges from 10.2 cm (4 inches) in Pohnpei to 15.2 cm (6 inches) in Chuuk. Because the quality of trochus shell decreases with size due to increased deterioration by boring organisms, large shells are mostly unsuitable for button manufacture. Because fecundity increases exponentially with shell diameter, large trochus make a major contribution to the egg production of a population. Therefore protection of large shells by an upper size limit makes sense, from both marketing and biological viewpoints.

The following countries have also adopted a maximum size limit policy for trochus fisheries management.

Country	Minimum size limit
Chuuk	6.0 inches (15.24 cm)
Pohnpei	4.0 inches (10.16 cm)
Yap	4.0 inches (10.16 cm)
Cook Islands	11.0 cm
French Polynesia	11.0 cm
New Caledonia	12.0 cm

Total allowable catch

Total allowable catch (TAC) is another management measure that has been implemented in only three Pacific island countries for trochus management. One country stopped using TAC, while in the two other countries it is still implemented.

TAC can be set fora country as a whole or be assigned to individual reefs, islands, clans, families or individuals. Two most important criteria that have always been used for determining a TAC: (i) setting a quantity that appears to be historically sustainable and (ii) setting a fixed percentage of the estimated biomass of legal-sized shells. The former has been used in Vanuatu during the late 1970s to early 1980s. A TAC of 75 t per year was set for the whole of Vanuatu. This TAC was scraped out in 1983 together with the export of whole shells. This was because industrious villages and islands caught large proportions of the total quota leaving only a small amount of shells to be harvested by other less industrious islands. The villages and islands that missed out retaliated by fishing heavily the following year to ensure they gained an adequate quantity of the quota. This resulted in the annual quota being taken very early in the year, with nothing to sustain the rural dwellers or the factories for the remainder of the year. This type of competition for a share of the TAC promoted localised depletion of trochus stocks, particularly when families and clans are restricted to a small area of reef by traditional land tenure. Therefore TAC set for the entire country was not preferable in terms of biological and economical reasons.

French Polynesia implements a TAC based on the percentage of the estimated biomass of legal-sized animals, while in the Cook Islands (Aitutaki) TAC is assigned to individuals. In 1991, prior to the opening season for trochus fishing, it was decided after a trochus stock assessment survey that each individual was entitled to harvest 15 kg of trochus shells upon obtaining a licence. It was found to be an effective management tool. Success was achieved in sustaining stock levels, minimising takings of illegal-sized shells and spreading the income of earnings across the community (Tuara and Passfield, 1991). Thus assigning TACs to individuals is far preferable because it reduces between-group competition for a share of the available quota. The individual quota holders may harvest their share of the TAC at a time of their choosing.

Limited fishing season

Imposing periods of the year when trochus can be harvested has biological relevance in two situations: (i) enforcing of a TAC is impossible without it, and (ii) trochus reproduce at well-defined times of the year. It can thus be regulated such that fishing is allowed during the period when trochus are non-reproductive. This ensures that there are enough eggs to propagate the population before the trochus are harvested.

Only two Pacific island countries adopt this management policy, French Polynesia and Cook Islands. For Cook Islands, the fishing period is regulated depending on the status of trochus stocks. Time ranges from 2 to 5 days (Tuara and Passfield, 1991). Fishing is once a year.

Sanctuaries

Sanctuaries to conserve trochus stocks have been established in the following Pacific island countries: Cook Islands, Fiji, French Polynesia, Chuuk, Kosrae, Pohnpei, New Caledonia, and Palau.

The effectiveness of a trochus sanctuary depends on the direction and the strength of water currents, and the proximity of the fished stocks to the sanctuary. This is because the larval stage of trochus is no more than a few days (3–4 days) and larvae are unlikely to disperse widely. Dispersion therefore depends on the direction and how fast the current is flowing. Trochus sanctuaries are effective management tools only if they are located in optimal trochus habitat and are not fished. Heslinga et al. (1984) evaluated the effectiveness of sanctuaries in Palau and found that trochus abundance was only marginally higher within sanctuaries than outside them. The effectiveness of Cook Islands sanctuary was reduced due to illegal harvesting (poaching) within the reserve (Zoutendyk and Passfield, 1989).

Traditionally based trochus management

The sovereign rights of coastal communities with respect to the exploitation, conservation and management of the living resources in their exclusive area of reef obtained through traditional land tenure are the basis for the rational management and optimum use of these resources.

Traditionally or community-based trochus management has not been adopted for trochus management in the Pacific region. This is because reef ownership through traditional land tenure does not exist in almost all of the Pacific island countries. The only country where this type of management is adopted and successful is Vanuatu. Community-based management can be defined as sharing responsibility and authority between the government and the local community to manage a fishery or other natural resource.

While the development of the co-management is not automatic or simple, is costly to establish and requires a long-term effort, it can be effective once the resource owners are made aware of the importance of proper management of their resources.

Basically, management involves including the resource owners in managing the trochus fishery. The Fisheries Department enforces minimum size limit on the trochus shells, while the community acting on the advice of the Fisheries Department instigates such controls as: closed seasons which may be as long as five years depending on the status of their trochus stocks; limited fishing season (in most cases for a period of 3 days to 3 weeks); fishing methods (no night fishing); and fishing gear (no scuba or hooker gear).

Community-based trochus management should be conceived and understood not as a constraint upon rational exploitation but as an essential tool for the sound, sustained development of fisheries. Hence, community-based management system of trochus fisheries is an integral part of the development process. Management decisions are made on the basis of reliable data and research into the biological environment, economic and social aspects of the trochus fishery.

Stock replenishment

By translocation of adults — trochus translocation has been carried out in many parts of the Pacific region where it does not naturally occur or to places where severe depletion has occurred. It has been adopted as an important management tool. The history of successful translocation of adult trochus (Gillett 1986) suggests that it may be an effective means of accelerating the rate of recovery of a population following overfishing. This depends on the reduction or cessation of fishing.

One of the Pacific island countries in which this method have provided successful results is Cook Islands (Aitutaki). In 1957, a shipment of 300 adult trochus was first introduced from Fiji (Powell 1957). In 1981 the first trochus harvest was declared. A 15-month season was set as the fishing period, during which over 200 t of trochus shells was collected (Sims 1985). Successful translocation of trochus to other parts of the Cook Islands has also been recorded.

Some of the following Pacific island countries where trochus does not occur naturally but which through translocation of adult trochus now have trochus are Cook Islands, French Polynesia, Chuuk, Kosrae, Pohnpei, Guam, Commonwealth of the Northern Marianas, Tonga, and Yap.

By hatchery-reared juveniles — Nash (1993) proposed two possible objectives for hatchery rearing trochus: (i) seeding depleted reefs to accelerate recovery; and (ii) to help compensate in an overfishing situation in the absence of conservative fishing behaviour.

Reseeding as a management option has been explored in several Pacific island countries. It has been experimented with in Palau (Heslinga et al., 1983), Vanuatu (Amos 1995), New Caledonia and Pohnpei. This management option, unlike other options, should increase the trochus stocks beyond natural capacity, should it be successful. However, survival rates after seeding have been very discouraging. This has resulted in the discontinuation of the exploration of the option in some countries. It is thought that the limited operating funds should be directed to other realistic management approaches. For example, research in Palau during the early 1980s explored the feasibility and cost-effectiveness of trochus mariculture as a management option. Consistent production of hatchery-reared trochus was quickly attained. However, during the late 1980s the effectiveness of releases continued to elude researchers in Palau. In 1992, the Palau Marine Resource Division shifted emphasis away from the mariculture and reef augmentation approach given that research and empirical evidence indicated that 'conventional method of management was considered a more cost-effective approach to managing Palau's trochus fishery than artificial stock enhancement through mariculture'.

Conclusion

While trochus has been demonstrated to be a relatively important and equitably distributed cash crop for the Pacific island countries, there are potentially additional tendencies to be avoided — such as overcapitalisation. With the need to service large capital investments there is a tendency to fish at levels beyond those sustainable. For example, button-cutting factories require between 100 and 150 t of trochus shells per year (depending on the number of cutting machines) to be financially in operation. In some Pacific island countries there has been strong pressure to have seasons each year in settings in which these factories have been built, along with reduced minimum size regulations established. The need to recoup the investment puts pressure on the resource and is typically driving stocks to much lower densities.

Most current management tools employed appear to focus on the biological attributes of the fishery. While this has ensured the ecological sustainability of the resource it appears adjustments can be made to optimise management. Policymakers and fishery managers need to look beyond biological factors and incorporate long-term and social considerations into management regimes.

In this period of change, the challenge for the Pacific island countries is to provide a new and improved basis for the rational management and utilisation of trochus fisheries. Management policies should contribute to the betterment of the socioeconomic conditions of artisanal fishermen who are among the poorest sections of the populations. In this respect, a reassessment of strategies and policies for fisheries management and development is needed to take full account of the current and potential contributions from marine fisheries.

Proper management of trochus resources can provide a model for inshore resources. They provide a real opportunity for resource managers and governments to provide benefits for all stakeholders. Government clearly has a role to play, as does the private sector, in the management of trochus fisheries. In almost all the Pacific island countries, central authority is required for sustainable fishery management. Only in the few island countries has trochus management seriously compromised the ability of traditional systems to manage the resource effectively.

Fisheries management experts in the Pacific recognise that the underlying causes of trochus resource overexploitation and coastal environmental degradation are often of social, economic, institutional and/or political origins. The primary concerns of trochus management, therefore, should address the relationship of trochus fisheries to human welfare; and the conservation of the resources for use by future generations. That is, the main focus of trochus fisheries management in the Pacific region should be people, not trochus, per se. Policy interventions, if they are to bring about lasting solutions, must address these concerns.

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The Potential of Reseeding with Juveniles as a Tool for the Management of Trochus Fisheries

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Abstract

History has shown that populations of *Trochus niloticus* (trochus) are sensitive to intensive fishing. There is a variety of methods by which trochus resources can be managed. One option is to use hatchery-reared juveniles to reseed reefs where stocks have been depleted. This technique could form an effective part of strategies designed to manage and sustain stocks of trochus.

Over the last 13 years, reseeding of this type has been the subject of preliminary investigations in a number of countries. The success of the technique has been variable. In some cases there have been encouraging results, but hatchery-reared juveniles released into the wild have not always been recaptured in large numbers. Mortality or loss of juveniles can act to limit the impact of hatchery-reared individuals on adult populations. The preliminary nature of the studies, however, limits the scope of the conclusions that can be drawn from them.

Reseeding may be considered valuable as a means of supporting artisanal fishing communities and as a conservation and management tool. Its commercial future, however, depends on its economic viability, which in turn depends on the growth and survival of the animals (both in hatcheries and on reefs) and production costs. This paper reviews existing information on the viability of reseeding using hatchery-reared juveniles, and makes a case for a more thorough investigation of its potential.

THE marine gastropod *Trochus niloticus* is harvested commercially in Australia, Southeast Asia and the Pacific. The shells are used primarily in the manufacture of pearl buttons, and estimated global demand is considered to outweigh supply. The adult snails are quite conspicuous on many of the reefs they inhabit and provide an important source of income and protein for the predominantly artisanal fishers that harvest them. These factors combine to render stocks on accessible reefs vulnerable to overfishing (Yamaguchi 1990). Nash (1993) gives an excellent review of management tools that can be used in an effort to avoid this problem. These include the use of size-limits, limited entry, limited season, total allowable catch, sanctuaries and logbooks. Amos (1995a) has also stressed the need for modern managers to understand and cooperate with traditional systems of management. Nevertheless, many stocks, particularly those easily accessible to fishers, have been overfished. In Indonesia, for example, stocks have declined to such an extent that in 1987 a total ban was placed on collecting wild trochus (Arafin and Purwati 1993). Moratoria have also been declared periodically since 1922 for the Micronesian fishery (Clarke and Ianelli, 1995).

As stocks of trochus diminish, they are thought to be slow to recover, particularly on a local scale and on isolated reefs, owing to the short larval duration of the species (3–4 days, Heslinga 1981) and its consequently limited capacity to disperse among reefs (Yamaguchi 1990; Nash 1993). Because of the slow recovery of wild populations, one intuitively attractive management option is artificially to enhance natural stocks ('stock enhancement' or 'reseeding'). The technique has been investigated with mixed results

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for a wide range of species, including finfish (Tsukamoto et al. 1989; Svasand et al. 1990), scallops (Shumway 1991; Thomson 1992; Hatcher et al. 1993), prawns (Liu 1990; Liu et al. 1991), conches (Stoner 1994; Stoner and Davis, 1994) and abalone (Tegner and Butler, 1989; Schiel 1992 1993). This paper focuses specifically on stock enhancement of trochus. Existing research into reseeding this species and consideration of the need for further investigation of its effectiveness are reviewed.

Reseeding as a Management Tool

There has been a successful history of translocation of adult trochus among regions. Several commercial populations have been established on reefs previously uninhabited by trochus (Gillett 1991; Clarke and Ianelli, 1995) with no noticeable adverse effects (Nash 1993). There have been concerns, however, that such translocations may cause problems by inadvertently spreading unwanted organisms such as parasites and pathogens along with the trochus (Yamaguchi 1990; see also Hindar et al. 1991). This technique has not been used to replenish overfished stocks of trochus and is not favoured as a management tool for this species (Nash 1993). Attention has instead turned toward collecting local broodstock, spawning them in captivity and rearing juveniles in hatchery tanks. The aim is to release the juveniles into the wild so that they will grow up and enter the fishery. If these animals are protected from overfishing, for example by the imposition of minimum size limits, they may also reproduce before being harvested and may therefore form the basis of a self-sustaining population (Nash 1993).

Several benefits could flow from such a program. For example, it could serve to restore populations of trochus to reefs on which they have been extinguished. In addition, maintaining trochus fisheries would enable many communities to continue their traditional artisanal fishing activities, which often constitute one of their few sources of income. The value of reseeding as a management tool, however, depends on its cost-effectiveness, which depends on the cost of producing and releasing juveniles, the percentage of those juveniles that survive to enter the fishery and the market price of trochus at the time of harvest. It is not possible therefore to assess the economic viability of the technique until data exist on the hatchery costs of producing juveniles and their survival as they grow to harvestable size in the wild.

Research to Date

Since the first hatchery spawning of trochus was recorded at the beginning of the 1980s (Heslinga 1980; Heslinga and Hillman, 1981), numerous studies of induced spawning and hatchery production of juveniles have been published (Nash 1990; Amos 1991; Kikutani and Patris, 1991; Dobson and Lee, 1996; Lee, these Proceedings; Lee and Amos, these Proceedings). It is now possible to induce trochus to spawn by a variety of techniques and Lee (unpubl. data) has recently simplified considerably the method of spawning and juvenile production at the Northern Territory University (NTU).

There is no published information on the costs associated with hatchery production although preliminary data (Lee, these Proceedings) indicate that a production cost as low as 0.7 cents/juvenile for 1–3 mm size class individuals is achievable using the closed recirculating system developed at NTU. It is generally accepted, however, that the costs of producing trochus of harvestable size in a land-based aquaculture system would be prohibitive. The requirements for providing natural food would be such that only a relatively few individuals could be housed in a hatchery (Nash 1993), and recent data from NTU indicate that a 5000 L tank would support no more than a dozen full-size adults or at most a thousand large juveniles of 20–30 mm in size. This situation is unlikely to change unless an artificially produced food can be formulated to feed trochus in the hatchery.

It is therefore necessary to release juveniles into the wild at an earlier stage of development. This can be done either in order to replenish stocks at sites with existing populations or to establish stocks on local reefs that appear suitable for juveniles and adults but may not receive a supply of larvae, for example through unfavourable hydrological conditions or lack of suitable substrata for settlement of larvae (Yamaguchi 1990). Over the last 13 years, this technique has been investigated for juvenile trochus of various sizes in Palau (Heslinga et al. 1983; Kikutani 1991), New Caledonia (Hoffschirr et al. 1989; Hoffschirr 1990), Vanuatu (Amos 1991 1995b; Castell 1995), Japan (Kubo 1989 1991) and Eastern Australia (Castell 1995) (see Table 1). In addition, Kubo and co-workers have done extensive research to refine a method for raising large numbers of juveniles in concrete nurseries constructed on reefs in Japan (Kubo et al. 1991, 1992, 1993, 1994, 1995).

Table 1. Previous research into reseeding.

Study	Date of release	Locality	Size of trochus	Total numbers released	Method of release	Monitoring	Survival	Suspected causes of loss of trochus	Positive outcome presented?
Heslinga et al. (1983)	Apr 1983	Palau	2–6 mm (3 months old)	32 000 (into 1 site)	Broadcast by diver in 0.5 m of water	2 months, 5 months	5.3 and 6.2 per man-hour search respectively. (control=1.2 per man-hour search)	Migration of larger individuals	Yes
Kubo (1989, 1991)	Apr–May 1998 Dec 1988–Jan 1989	Iheya and Okinawa, Japan	8–9 mm 17 mm	9000, 8000 respectively (5 sites studied at one site, 3 separate releases)	Some free release, some attached to coral rubble, some into enclosure	15, 40–60 and 120–130 days	13–75, 0–46 and 0–5% respectively. At one site all escaped from enclosure after 3 days; another site was destroyed by storm after 9 days	Predation, storms	No comment
Hoffschirr et al. (1989), Hoffschirr (1990)	Mar, Jun 1989	Lifou, New Caledonia	19 mm	2228 (Mar)+3481 (Jun)=5409 (into 20 sites)	Released into piles of rocks	Initial 2228 sampled after 2, 7 and 12 weeks; all sampled 1 year later	2-week sample disrupted by cyclone, 21–72% survival; 7-week sample incomplete; 8.4% after 12 weeks. 0.3% (=19 trochus) after 1 year	Cyclone	Yes
Kikutani (1991)	Nov 1989–May 1990	Palau	7.1–34 mm	11 244 (number released at each site ranged 250–2400)	Free-released into 'natural juvenile habitat'	2, 5, 10 and 40 days	Recovery rates decreased significantly during the month of monitoring	Predation, migration	No
Amos (1991)	May 1991	Efate, Vanuatu	>20 mm, <20 mm	1000, 400 respectively (into 4 sites)	Protected by rubble and/or cage	4, 7, 12, 27 and 49 days, 13 months	87.5, 51, 44.25 and 44.25%. 28% after 13 months (unpubl.).	Predation (M. Amos, pers. obs.)	Yes
Castell (1995) (pers. comm.)	Nov 1993–Feb 1994	Orpheus Is., Australia	13–44 mm	943	Placed upright on substratum	3, 30, 111 days	41, 26, 12.4%	Predation	No comment
	Oct 1994	Orpheus Is., Australia	4–8 mm	430	Placed upright on substratum	1, 3 days	61, 50%	Predation	No comment
	Dec 1994	Orpheus Is., Australia	4–8 mm	460	Placed upright on substratum	1, 2 days	56, 35%	Predation	No comment
	Feb 1994 Apr 1994 June 1994	Orpheus Is., Australia	Several experiments 10–12. 19–20, 26–27 mm	Total of 672	Tethered to steel rods with 0.5 m lengths of cotton	Different experiments monitored for 2, 4 and 8 days	Varied from 17% (12 mm, 8 days) to 90% (all sizes, 1 day)	No comment	No comment
	May 1994	Efate, Vanuatu	30 mm	600	Placed upright on substratum	2, 40 days	63, 6.7%	Predation	No comment
	May 1994	Efate, Vanuatu	30 mm	60	Tethered to steel rods with 0.5 m lengths of cotton	3 days	81.5%	No comment	No comment
Amos (1995)	Jan 1995	Efate, Vanuatu	Large (12–35 mm), small (8–12 mm)	1400	Protected by rubble and/or plastic mesh	Fortnightly for 200 days	88% (14 days) 32% (200 days)	Predation	Yes

In all the studies there were significant reductions in the numbers of juvenile trochus recaptured in relation to the numbers released (with the possible exception of Heslinga et al. (1983), in which it is not possible to determine this from the information provided). This may be ascribed either to the mortality of juvenile trochus or to their departure from the area sampled. Losses of juveniles were considered to have been caused by such processes as predation (Kubo 1989, 1991; Castell 1995), strong wave action (Hoffschirr et al. 1989; Hoffschirr 1990; Kubo et al. 1991) and the size-specific migration of trochus (Heslinga et al. 1983). Nevertheless, outcomes of reseeding were considered positive in four of the seven studies.

Castell (1995) found that larger juveniles (19–23 mm) survived better than smaller ones (4–12 mm). Other authors have expressed the opinion that survival is likely to be greater for larger trochus (Kubo 1991), and this has also been the experience of researchers studying prawns (Liu 1990) and bream (Tsukamoto et al. 1989). This does not necessarily mean, however, that releasing larger animals will be more cost-effective than releasing smaller ones. Each large individual costs substantially more to produce than a smaller one due to the longer time spent in the hatchery. As discussed above, it is generally considered uneconomical to rear commercial quantities of adult trochus in hatchery tanks because of the requirement for large quantities of food as the animals grow larger (Nash 1993). The appropriate size for release must therefore be a compromise between the probability of an individual's survival in the wild and the cost of its production in the hatchery. Data to evaluate this are still unavailable.

Although they have provided valuable preliminary information, there were some features of the methodologies used in the above studies that limit the scope of conclusions that can be drawn from them (Table 2). For example, only three of the studies (Hoffschirr et al. 1989; Hoffschirr 1990; Amos 1991 1995b) ran long enough for the released trochus to grow to sexual maturity and to approach harvestable size (Table 2). Trochus must be able to be harvested before it is possible to consider reseeding to have been a success.

Heslinga and co-workers (1983) were the only authors to compare abundance of trochus at a reseeded site with that at a control (Table 2). Without comparison of harvests from control and reseeded reefs, the effect of reseeding over and above natural processes cannot be reliably assessed. For example, at the time when hatchery-reared juveniles are being released onto selected reefs, populations on other reefs may be increasing by natural recruitment. Under these circumstances the investment in reseeding would be wasted, because similar increases of harvest may have occurred without intervention. Unfortunately, however, the control and reseeded reefs were not replicated in the study by Heslinga et al. (1983). There is therefore no logical basis for determining whether the subsequent difference in abundance of trochus on the reseeded and control reef was caused by the reseeding or by natural differences between the sites, such as abundance of predators or supply of recruits.

Given the mixed results of these studies and their preliminary nature, there is still no clear consensus on the effectiveness of the process of reseeding. Much of the basic information required to judge its effectiveness is still lacking (Yamaguchi 1990; Nash 1993).

Research Required

In order to test the hypothesis that reseeding reefs will enhance populations of harvestable trochus on those reefs, harvests from experimentally reseeded reefs need to be compared with those from suitable controls that have received no hatchery-reared juveniles. For the test to have logical validity, there must be replicate reseeded and control reefs and they must be monitored after enough time has passed for the trochus released to have grown to minimum legal harvestable size. This size varies among regions, from 65 mm in Western Australia to 90 mm in New Caledonia and Vanuatu, and is thought to take approximately 2–3 years to attain (Nash 1993). A three-year study aimed at testing this hypothesis was commissioned by the Australian Centre for International Agricultural Research (ACIAR) in 1995. The research will involve the release of juvenile trochus of a range of sizes from 1 to 50 mm and will be replicated in Australia, Indonesia and Vanuatu. It should thus give a clear picture of the effectiveness of reseeding in a broad range of biogeographical regions.

The success of reseeding is likely to vary according to the habitat and microhabitat into which individuals are released. As yet, there has been little objective research into the sorts of habitats that may be appropriate for the release of hatchery-reared trochus. Schiel (1993) found that the effectiveness of restocking reefs in New Zealand with juvenile abalone varied considerably over quite small spatial scales.

Table 2. Methodological approaches.

Study	Choice of sites	Sampling method	Tagging method	Unseeded controls	Replication	Duration
Heslinga et al. (1983)	Field surveys of natural populations. Juveniles among dead fragments of *Acropora* in mid- to outer intertidal reef flat	Before and after release by timed searches	None (trochus considered too small at 15 mm)	Yes	No	5 months
Kubo (1989, 1991)	Attempted to survey abundance of trochus in different habitats, but results inconclusive. Trochus released into rocky and coralline habits at some sites.	After release by exhaustive search (not timed)	Coloured cyanoacryllate glue	No	5 sites, two size classes, but no replication for comparison of size classes	140 days (only 5% remaining at this time)
Hoffschirr et al. (1989) Hoffschirr (1990)	Sites placed in calm oxygenated water, <2 m depth, dead coral rubble substratum.	After release by exhaustive search (not timed)	None	No	20 sites (results presented as total % survival)	1 year
Kikutani (1991)	Coral rubble thought to be natural juvenile habitat	After release by exhaustive search (not timed)	Large size (>30 mm) with aluminium rivets, smaller juveniles tagged with blue dye, white marine tex and pencil mark on inner nacre	No	No	8 months
Amos (1991)	Coral rubble chosen by intuition as substratum providing best protection from predators	After release by exhaustive search (not timed)	Polyethylene tag, pencil mark on inner nacre, coloured cyanoacryllate glue	No	No	13 months
Castell (1995) (pers. comm.)	Study involving release of trochus in different habitats and comparison of survival. No consistent differences among high, mid and outer reef flat.	Exhaustive searches. Effectiveness of searches examined in additional study	Numbered polyethylene tags for juveniles >17 mm	No	Yes	Different experiments, varied 2–11 days
Amos(1995)	Coral rubble chosen by intuition as substratum providing best protection from predators.	After release by exhaustive search (not timed)	Polyethylene tag, pencil mark on inner nacre, coloured cyanoacryllate glue	No	No	200 days

Economic analyses indicated that the undertaking would have been profitable in some areas, but not others (Schiel 1993). In their research into trochus, Heslinga et al. (1983) used surveys of natural populations to determine which types of site had the largest numbers of juvenile trochus. Based on this, they chose sites for reseeding research that were on the mid or outer reef flat with a substratum of finger-sized coral rubble. Castell (1995) released animals into different habitats and monitored their subsequent survival as an indicator of habitat suitability. She found that there was considerable variation in survival of trochus over small spatial scales, but that there was no clear effect of height on the shore. Hoffschirr et al. (1989), Hoffschirr (1990), Amos (1991, 1995b) and Kikutani (1991) relied on intuitive insight for the selection of sites. Coral rubble was chosen as an appropriate substratum by these researchers, but the possible suitability of other substrata was not investigated. More studies of the sort done by Heslinga et al. (1983) and Castell (1995) are needed so that our understanding of the factors affecting the suitability of sites for trochus can be refined.

In addition, there has been little consideration of the possibility of temporal variation in the success of reseeding. Factors affecting success of reseeding were thought to include predation (Kubo 1989; Castell 1995) and wave action or cyclonic activity (Hoffschirr et al. 1989; Hoffschirr 1990; Kubo et al. 1991). Both factors are very likely to vary considerably through time in most regions. There may, for example, be some seasons or periods in the tidal cycle in which reseeding is more effective than others. Studies to examine this effect must involve replicate releases of trochus in each of the periods of time under investigation.

Even if reseeding is found by researchers to enhance populations of trochus, Nash (1993) cautions that it will be an effective management tool only if combined with measures to ensure conservative fishing behaviour (e.g. size limits). This would enable a proportion of the hatchery-reared juveniles to reproduce before being harvested and thus allow the reseeded populations to be self-sustaining. For example, if the aim of reseeding is to establish a breeding population rather than a directly harvestable population, it may be appropriate to release relatively large (40–50 mm) juveniles in relatively small numbers and protect them completely from harvest for several years.

Should reseeding of trochus be demonstrated to be effective, it would also be important to carry out some additional research before embarking on a widespread program of stock enhancement with this species. For example, the effect on other species of introducing large numbers of juvenile trochus to reefs is unknown. In addition, stock enhancement can have some deleterious effects on the target population. Hindar et al. (1991) recommended the use of a large and representative pool of broodstock and cautioned against translocation among stocks. These measures aim to reduce the risk of loss of genetic diversity and transfer of pathogens and parasites among stocks. When reseeding reefs that contain no pre-existing population of trochus, however, the importance of these issues would arguably be reduced.

Yamaguchi (1990) suggested that, in its initial phase, the technique may not be attractive to commercial investors because of the slow growth rates of the snails, but that juveniles should perhaps be produced by government-funded hatcheries and sold at cost to co-operative bodies responsible for the management of reef resources. While predicted profits may or may not be large enough to warrant commercial reseeding, the benefits to the community and environment may ultimately justify the adoption of reseeding as a management tool. In any case, it is important thoroughly to investigate the effectiveness of reseeding before proceeding with large-scale programs. If small-scale studies continue to produce encouraging results, preliminary 'commercial scale' releases on selected reefs should also be done within a research framework (see also Walters 1986, Underwood 1995). In this way, the technique can be further refined and data relevant to the outstanding questions discussed above can also be collected.

Conclusions

Although there have been several preliminary studies into the potential of reseeding as a tool for the management of trochus fisheries over the last 13 years, no clear consensus has emerged. The results of the studies have been variable, there have been some methodological problems, and some issues such as temporal variation in success of reseeding have not yet been investigated (see above). The study commissioned by ACIAR should help to resolve some of the issues raised in this paper.

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Potential of Remote Sensing Data for Identifying Trochus Reseeding Sites

W. Ahmad and G. Hill*

Abstract

During different stages of its lifecycle trochus exhibits preferences for different reef habitats. Likewise, within any reef complex there is variation in the availability and distribution of these preferred habitats. Satellite remote sensing is able to identify and map the relevant reef types. Knowledge of the habitat base available within any proposed reseeding area will be an important consideration in reseeding activities. Where local knowledge is unavailable, or the area is remote, satellite remote sensing offers a reliable means of assessing the habitat base for trochus.

THE large marine gastropod *Trochus niloticus* has been used for thousands of years by the island peoples of the South Pacific as a source of food, craft material and trade. In the 1800s it formed the basis of the mother-of-pearl shell trade controlled from Europe. While the extent of harvesting that characterised these times is not found today, the annual world harvest is still over 5 million kg and trochus remains an important cash crop and food source (Bouchet and Bour, 1980). With increasing human population and technological advances that facilitate the gathering of trochus, there is concern over current levels of harvesting and degradation of the reef habitats that support trochus (e.g. Glucksman and Ludholm 1982; Nash 1986; Yen 1985; Catterall and Poiner, 1987; Honma 1988).

However, the tropical coral reefs where trochus shell is found are remote and in many cases uncharted. As well, it is only certain sections of the reefs that are prime trochus habitat. As *T. niloticus* feeds mainly on algae, the best locations are the high-energy sections of the reefs. These generally form narrow zones along the windward edges of the formations. Trochus are found in both intertidal and subtidal reef areas. Most inhabit water depths of between 5 and 6 metres although the larger commercial sizes often occupy deeper water (e.g. Heslinga et al. 1984; Hahn 1989). The task of estimating the abundance of trochus and the distribution of suitable habitat is a difficult one. This is the reason few surveys have been conducted for the commercial fishery areas (Long et al. 1993).

Remote sensing is a suitable base for mapping trochus habitat. However, mapping the habitat requirements of trochus demands high spatial resolution in a sensor system. For this reason satellite remote sensing has not made a contribution to mapping trochus habitat until comparatively recent times. Bour et al. (1986) and Loubersac and Populus (1986), working in New Caledonia, demonstrated that simulated SPOT imagery was capable of mapping with high accuracy the reef areas used by trochus. These results were confirmed after the launch of SPOT (Bour 1988). In the Australian region, Long and colleagues (1993) used Landsat TM imagery and a relatively simple classification technique as a means of rapidly estimating the trochus stocks of the eastern Torres Strait.

Work by the current team made use of the field data and airphoto-derived maps of Long and coworkers to establish the level of detail possible with Landsat TM imagery if more sophisticated processing approaches were tried. This commenced with the successful classification of the reef surrounding Yorke Island in Torres Strait (Hill and Ahmad 1992). In

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attempting to extend this process to surrounding reefs, it became clear that they displayed different patterns of trochus habitat.

One practical problem, from the image analysis point of view, is that the islands and reefs that support trochus are distributed patchily across broad areas. Because of differences in the size, location and geomorphology of the islands or reefs, they exhibit a diverse range of spectral classes for classification. And water depth alone is not necessarily a good indicator of prime habitat. This state of affairs may lead to long and involved image processing sessions before a satisfactory classification of the full range of classes is successfully achieved.

The general aim of this paper is to assess the usefulness of Landsat TM data for mapping trochus shell habitat for the reefs of the Bourke Isles in Torres Strait, which separates Australia and Papua New Guinea. In specific terms, however, emphasis is given to the description of a methodology that streamlines the process of classifying prime habitat for commercial-sized trochus across a group of reefs. It relies on transfer of a classification mask generated from an intensively surveyed reef to similar but unsurveyed areas.

Study Area

The location of the study area is shown in Figure 1. Torres Strait, being roughly 100 km from north to south and 250 km east to west, occupies an area of approximately 8000 km^2 between Papua New Guinea and Australia. It is a shallow zone, up to approximately 50 m in depth, that features a host of reefs, islands and shoals (e.g. Harris 1989). The Bourke Isles are located in the eastern Torres Strait. These islands are coral cays surrounded by elongated platform reefs. The major reef flats face the prevailing south east trade winds, and it is along the windward edge of these that most large trochus are found.

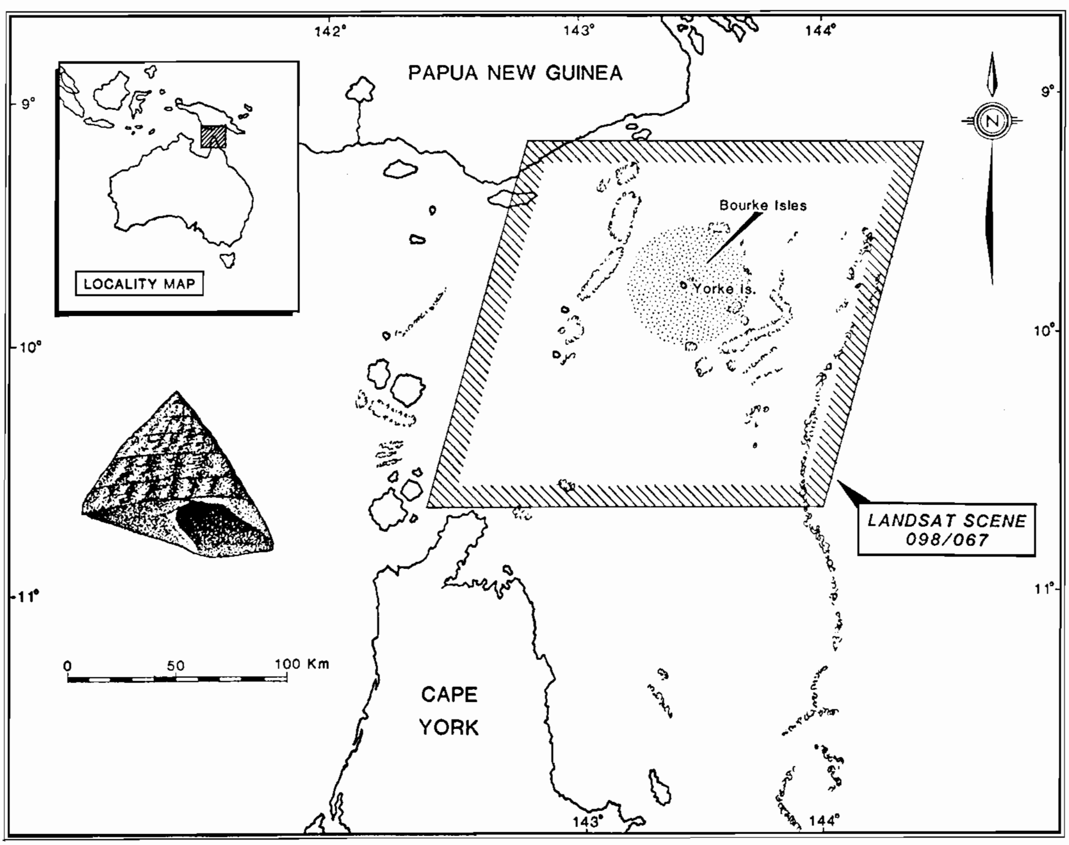

Figure 1. The study area, the Bourke Isles of Torres Strait.

Climate of the area is mild for a tropical region, being dominated by the southeast trade winds that blow from June to November and the northwest monsoon that brings heavy rains between January and May. Maximum annual average temperatures range from 24°C to 30°C with average annual precipitation of 1750 mm (Thursday Island). The dry season provides good conditions for the capture of satellite data with little cloud cover. During the monsoon season, however, there is little likelihood of usable imagery.

Methods

Research was based on a Landsat TM scene (path 98, row 67C) recorded on 31 October 1988. Channel 1 (0.45–0.52 μm), channel 2 (0.52–0.60 μm) and channel 3 (0.63–0.69 μm) were selected because of their water penetration characteristics. As each reef was associated with an island, channel 4 (0.76–0.90 μm) was included to assist with separation of land and water environments.

The project was carried out in two stages. In Stage 1, trochus habitat for the largest island in the group (Yorke Island) was successfully achieved (Hill and Ahmad, 1992). Stage 2 involved the transfer of the classification mask generated for Yorke Island to the remaining three reefs of the study area (Marsden, Kabbikane and Keats islands).Initial classification runs of these reefs using the Yorke Island classification file indicated that the subclasses involved were not directly transferable and adjustments were required. Figure 2 explains the step-wise analysis procedure followed.

Work began with the contrast enhancement of the images for Marsden, Kabbikane and Keats reefs. The data were visually inspected on a high resolution colour monitor and a few homogeneous training areas were selected from the more distinct colour patches visible on the screen. The training areas were added to the previously created classification file for Yorke Island.

To ensure that the spectral variation in the data was sampled and incorporated in the classification procedure, a sequence suggested by Ahmad (1986) was adopted. This procedure relied on the independent sampling and inclusion of themes (spectral classes) based on the use of two-dimensional spectral histograms of the least correlated input channels. Implementation was achieved by defining boundaries that corresponded to the 1–95% values of the individual histograms. By overlaying these boundaries on the two-dimensional cross-plots of the least correlated channels, different boxes were formed and their central values (mean) calculated. These values (additional themes) were then added to the composite classification file.

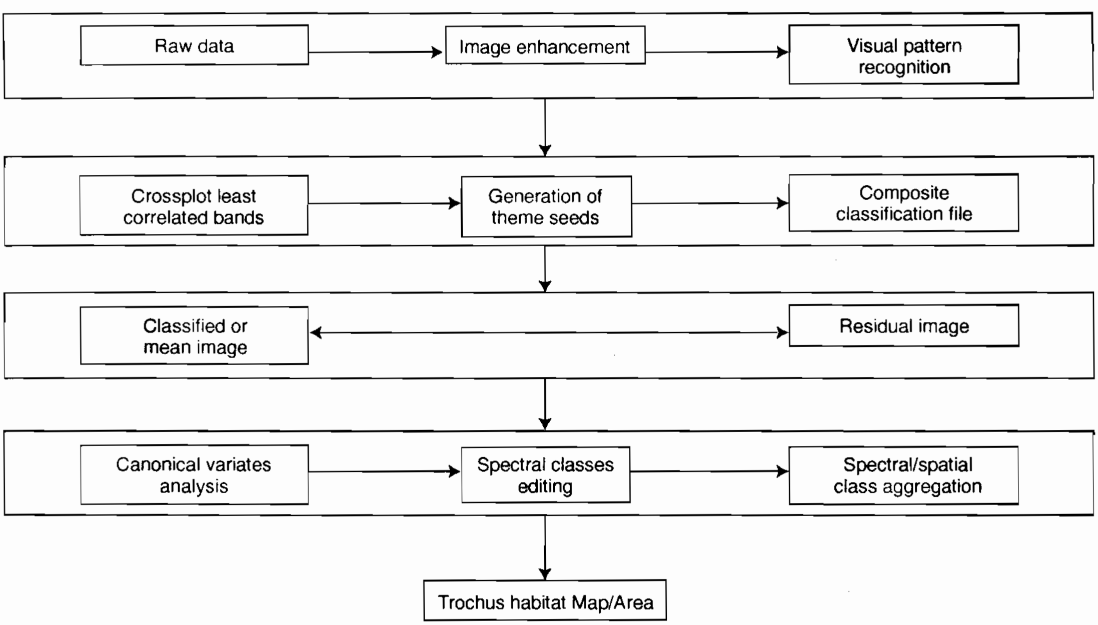

Figure 2. The image processing sequence followed to classify trochus habitat in the Bourke Isles.

As the next step, the composite classification file was used as an input to the minimum distance classifier using the microBRIAN image processing system (MPA 1988, Harrison and Jupp 1990). The three islands were classified separately using the composite classification file noted above. The resultant classified and residual images for the three islands were then evaluated (Jupp and Mayo 1982). In the classified image each pixel's value (per band) is replaced by the mean value for the class to which it belongs. The residual image is the difference between the classified and original data. The more closely the classified image resembles the original data and the more random the appearance of the residual image, the better the classification. This procedure resulted in optimally classified images which did not require any iterative classification.

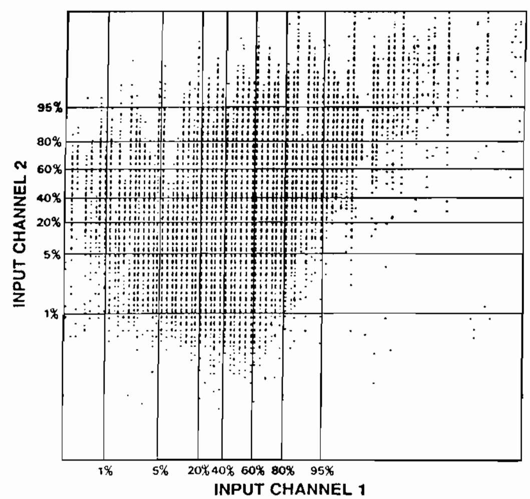

Figure 3. Concept diagram explaining the selection of additional themes for inclusion in classification. For each of the 36 boxes defined for the 1% to 95% range, mean values in each band were extracted and used as additional themes.

The resultant spectral classes were edited and aggregated according to the respective Mahalanobis Distance (Hope 1968) between spectral classes. This was performed by evaluating the Mahalanobis Distance value of the generated spectral classes. The technique calculates the difference between the midpoint values of the original themes and the mean values of the pixels assigned to each of the themes weighted by the within-class inverse covariance matrix. The classes were then labelled using additional analytical techniques provided by the microBRIAN software. This phase involved the aggregation of classes which were spatially contiguous as well as spectrally similar. These techniques included Canonical Variates Analysis (Hope 1968) and Minimum Spanning Tree (Gower and Ross, 1969). A Canonical Variates Plot of the data showing spectral class means of the classified Landsat TM scene together with its associated Minimum Spanning Tree are illustrated in Figure 4.

To finalise the spectral classes which defined trochus habitat, the class amalgamations were fine-tuned by displaying each subclass separately until the optimum stratification for trochus habitat had been achieved. Areas not on the windward side of the reefs were then trimmed manually (Long et al. 1993), the final maps plotted and area of prime trochus habitat per reef calculated. Ground truth maps prepared by the Commonwealth Scientific and Industrial Research Organisation (CSIRO) from fieldwork and interpretation of black-and-white aerial photography were available for each of the reefs under study (Long et al. 1993). These were used to assess the accuracy of the Landsat-based classifications.

Results

Table 1 provides a summary of the areas of trochus habitat estimated for each of the Bourke Isles and how these varied from the groundtruth maps prepared by CSIRO. Figure 5 illustrates the classified images produced.

Table 1. Areas (ha) of prime trochus habitat for the reefs of the Bourke Isles, Torres Strait.

Island	CSIRO map	LANDSAT map
Yorke	168.6	165.8
Keats	20.7	18.2
Kabbikane	17.4	16.9
Marsden	13.1	12.1

As can be seen from Figure 4, the reef edge classes potentially representing trochus habitat form a distinctive cluster in spectral space. In the present study, one group (classes 6, 19 and 20) formed the required zone on the larger Yorke Island, while a separate grouping (classes 2, 3, 4 and 20) represented the smaller islands. Spectral classes 2, 3 and 4 were in the appropriate reef zone on Yorke Island. However, they were located on either the leeward side of the reef or submerged shoals, neither of which are preferred by trochus (Long et al. 1993). It is likely that other

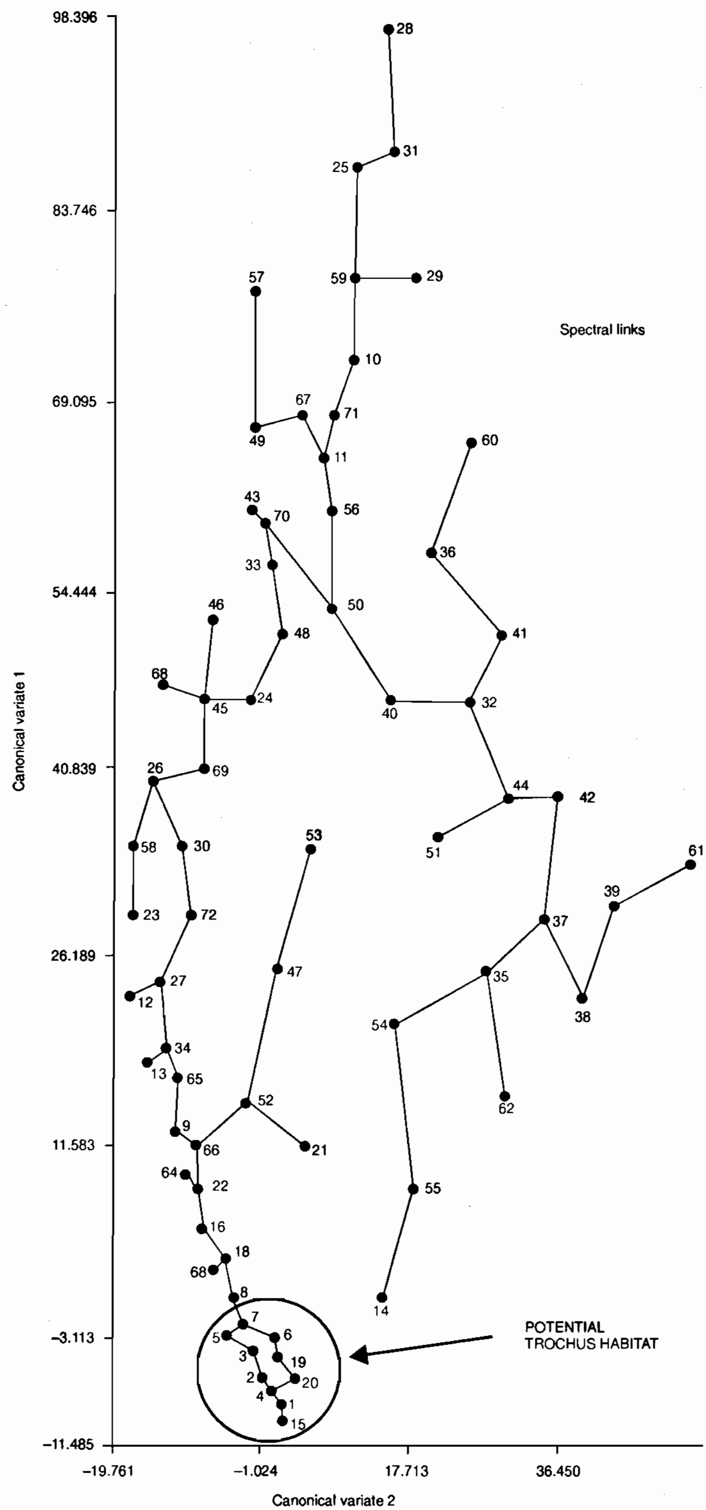

Figure 4. Canonical variates analysis illustrating the spectral links between classification classes for the Bourke Isles reefs.

classes associated with those selected for the Bourke Isles would form optimum trochus habitat in other situations (e.g. classes 1 and 15, 5 and 7). Further research will be required to assess this. It is clear, however, that for the Bourke Isles the key reef zone associated with prime trochus habitat and the subclasses which apply to each island are well-defined by the Landsat TM imagery.

The Landsat TM-based classification produced estimates that were between 1.7% (Yorke Island) and 12.1% (Keats Island) below those produced from the air photos and fieldwork. The results compare favourably with the Landsat-based estimates produced by Long and colleagues (1993) from density slicing of a green on red (TM2/TM3) ratio channel. These varied from a 12% overestimate for Keats island to a 29% underestimate for Marsden Island.

Discussion

The consistent but small underestimates of the Landsat classification, compared to the photo interpretation and field survey methods of CSIRO, are due in part to the generalisation that typifies maps produced by photo-interpreters. In the Landsat-based approach, large coral bommies and isolated patches of coral (in the trochus zone) were classified as a different habitat. On the existing maps these had been included (as minor noise) within the surrounding trochus habitat. Textural problems of this nature are common to all remote sensing classifications that rely on spectral information alone. As pointed out by Long et al. (1993), the narrowness of the key habitat areas (down to 30 m wide) for large trochus must also create classification errors for imagery that features 30-m pixels. They also attribute some of the error in their work to reliance on aerial photography recorded 17 years before the satellite imagery. Problems of this sort aside, the results achieved in the current study demonstrate a quite acceptable level of accuracy.

Trochus prefer sections of reefs where wave action deposits rubble, retards coral growth and scours fine sediments. Because of variation in reef size, position and structure there is variation in where this prime trochus habitat is located from reef to reef. It follows, therefore, that the spectral signatures of these areas are not completely transferable. In particular, it means that water depth is not the only factor of importance in mapping exercises. This would appear to be borne out by the results of Long et al. (1993) for the same area. However, the research completed here demonstrates that the methodology developed allows modification of spectral classes to suit each reef system, providing field data are available. Mapping and monitoring large areas can therefore be performed relatively quickly once the initial classification phase has been completed.

The island nations and communities of the South Pacific where trochus is exploited for economic and traditional uses are currently becoming more actively involved in remote sensing technology (e.g. ESCAP–UNDP 1989, 1991). One difficulty they face, however, is that the standard applications of satellite-based remote sensing are not particularly useful in these environments because of the tiny land areas involved. It is in the assessment of coastal and marine resources that remote sensing offers the greatest scope for useful applications. These resources offer great potential for development and, conversely, overexploitation. To date little progress has been made in mapping reef resources. As pointed out by Quinn et al. (1985:549), 'developing countries do not generally have a satisfactory enough inventory of their shallow water habitats to develop and manage them effectively'.

While research based on Landsat MSS imagery has indicated some success in this field it was not until the advent of the higher resolution systems that detailed assessments became possible (e.g. Loubersac and Populus 1986, Loubersac et al. 1988, Bour 1988, Vercelli et al. 1988). The results of the current work provide a basis for high-resolution mapping an important traditional resource that is under considerable pressure. For many sections of its range there is no database on distribution of suitable habitat or abundance of trochus.

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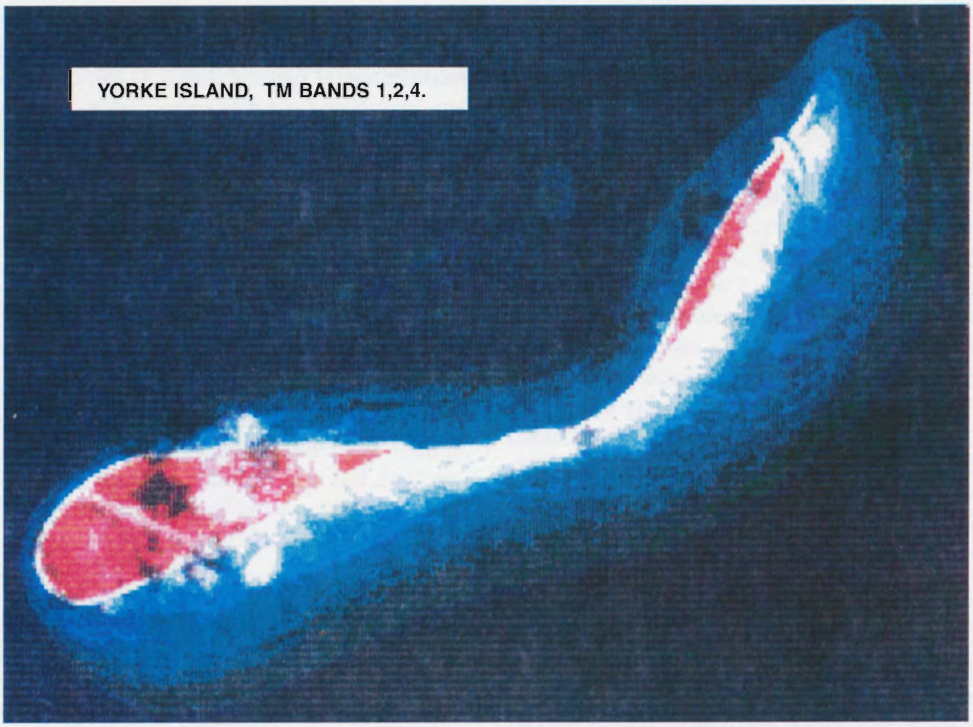

Figure 5. Classification results for trochus habitat

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