



# Technical Potential for Rock Lobster Propagation in Aquaculture Systems

RLEAS Publication No. 3

Edited by Dr Piers Reid Hart and

Dr Robert van Barneveld

August, 2000



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## Proceedings of the 1999 Fisheries Research and Development Corporation Rock Lobster Propagation Workshop

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### **Workshop Organiser**

Dr. Piers Reid Hart

**Tasmanian Aquaculture & Fisheries Institute**

### **Venue**

University of Tasmania,  
Geology Lecture Theatre

&

Room 229,  
Hobart Campus

**January 29 - 31, 1999**

### **Language coordinator**

Melanie Green

### **Conference Services & Venue Hire**

Helen Stewart

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The information contained within this document represents a final report to the Fisheries Research and Development Corporation for Project No. 98/300. The outcomes were used as a basis for a research proposal to the Fisheries Research and Development Corporation which culminated with the establishment of Project No. 99/315 within the Rock Lobster Enhancement and Aquaculture Subprogram.



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## Table of contents

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Non Technical Summary .....	4
Workshop Objectives .....	6
Welcome by Professor Colin Buxton .....	8
Introduction by Dr. Patrick Hone .....	10
Outline of previous and future research on aspects of <i>Jasus edwardsii</i> biology and behaviour pertinent to management of broodstock ( <i>Alistair MacDiarmid</i> ).....	14
Summary of the work done in New Zealand at NIWA Mahanga Bay, on rock lobster ( <i>Jasus edwardsii</i> and <i>J. verreauxi</i> ) propagation ( <i>Lennard J. Tong and Graeme A. Moss</i> ) .....	21
A note on sex ratios and pigmentation of eggs ( <i>Stuart Frusher</i> ) .....	30
Phyllosoma rearing of the Japanese spiny lobster, <i>Panulirus japonicus</i> , using small rearing vessels ( <i>Hirokazu Matsuda</i> ) .....	33
Biological Barriers to Aquaculture of Rock Lobster ( <i>Takeshi Murai and Taku Yoshimura</i> ).....	35
Developmental status of aqua-culture lobster fisheries in Japan ( <i>Taku Yoshimura</i> ) .....	36
Importance of three major factors for successful phyllosoma culture ( <i>Jiro Kittaka</i> ) .....	37
Propagation of southern rock lobster ( <i>Jasus edwardsii</i> ) in Tasmania ( <i>Arthur J. Ritar</i> ).....	42
Effect of photoperiod on survival, growth, feeding and cannibalism in early developmental stages of <i>Jasus edwardsii</i> phyllosoma larvae ( <i>Michel Bermudes</i> ).....	46
Closed cycle breeding of crustacea with special reference to rock lobsters ( <i>Mike Hall</i> ) .....	48
Investigation of the gut system; Application for lobster aquaculture ( <i>Satoshi Mikami</i> ).....	50
Summary of Research – Rock Lobster Digestive Systems/Physiology ( <i>Danielle Johnston</i> ) .....	51
Present and future directions in Western Australian rock lobster propagation ( <i>Bruce Phillips</i> ).....	54
Rock lobster research at the Australian Institute of Marine Science ( <i>John Benzie</i> ) .....	55
The 1999 Japan/Australia rock lobster Workshop in Perth funded by DISR ( <i>John Benzie &amp; Taku Yoshimura</i> ) .....	57
The biological feasibility of rock lobster propagation from egg to puerulus ( <i>Lennard. J. Tong</i> ) .....	59
Rock Lobster Propagation research – an aquaculture perspective ( <i>P Jungalwalla</i> ).....	60



The Cooperative Research Centre (CRC) for Aquaculture ( <i>Dr. Peter Montague</i> ).....	62
Summary and overview of the day's discussion ( <i>Robert van Barneveld</i> ).....	63
Discussion of economic and biological feasibility ( <i>Robert van Barneveld</i> ).....	66
Development of a research plan ( <i>Robert van Barneveld</i> ) .....	70
Appendix 1: Workshop Agenda .....	71
Appendix 2: Report on a study tour to New Zealand and the United States ( <i>Dr. Bradley Crear</i> ) .....	75
Appendix 3: Contact List .....	95



**PRINCIPAL INVESTIGATOR:** Dr P.R. Hart  
University of Tasmania  
Tasmanian Aquaculture & Fisheries Institute  
Marine Research Laboratories  
Nubeena Crescent  
Tarooma TAS 7053  
Telephone: 03 62277242 Fax: 03 62278035

## OBJECTIVES

1. Identify potential international and national, research & industry partners who are interested in collaborative research into culture of rock lobster from eggs
2. Identify barriers to the development of commercial culture of rock lobsters in Australia and determine the R & D required to overcome these barriers
3. Identify what resources (expertise, facilities, funds) are required to conduct the necessary R & D
4. Develop a research plan mapping out R & D projects, the collaborative partners, timelines and resources
5. Develop a collaborative, multi-institutional project proposal for FRDC

## NON TECHNICAL SUMMARY

The Australian rock lobster fishery is an important natural resource making up 25% of Australia's total fishery landings and presently worth around \$450 million per annum. However, most Australian rock lobster fisheries are fully exploited and therefore, future expansion relies on some form of aquaculture.

There is considerable interest worldwide in rock lobster aquaculture. In Australia, the Fisheries Research and Development Corporation (FRDC) has established a National Subprogram for Rock Lobster Enhancement and Aquaculture (RLEAS) in order to develop a cohesive and nationally coordinated approach to the research and development issues.

In the short term it is essential that research is conducted on the viability of puerulus collection, ongrowing and release for enhancement of wild fisheries and the establishment of aquaculture. In the longer term, the viability and sustainability of rock lobster aquaculture depends on closing the life cycle and developing an economically viable method for raising larvae and puerulus through to marketable size.



After a series of national workshops it was decided that closing the life cycle commercially would only be possible with a considerable research thrust involving high risk over a long period of time. The FRDC felt that there was not enough information within Australia to make the necessary decisions regarding research structure and the most cost/effective way to fund it.

Outside Australia the culture of rock lobster to puerulus has been achieved in both Japan and New Zealand. However, survival rates are extremely low. Recently a number of Australian research scientists have expressed interest in becoming involved in this form of research.

The present workshop brought together eminent rock lobster scientists from Japan, New Zealand and Australia with the aim of identifying the areas of most concern in terms of both biology and economics and areas of mutual interest. The format was:

Day 1. Formal presentations from individuals working in relevant research areas.

Day 2. Discussion of the economic and practical feasibility of propagating rock lobsters commercially, followed by an inspection of the research facilities at TAFI.

Day 3. Documentation of research and development priorities, identification of research providers and establishment of timelines and a framework for the preparation of a research submission to the FRDC.

The outcome of the workshop was that, while many questions remain unanswered, sufficient information is currently available to suggest that rock lobster propagation is not only practically feasible, but there are strong indications that a rock lobster hatchery could be economically viable. On this basis, it was agreed that a research and development plan should be pursued and a funding application submitted to the FRDC for an initial 1 year research project with the expectation of a longer and more elaborate project to follow. The second project will be for the development of commercial mass culture techniques.

**KEYWORDS:** Rock lobster, aquaculture, larval rearing, propagation



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## Workshop Objectives

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Most wild stocks of rock lobster in Australia are fully exploited and have no further potential for expansion. However, recent information suggests that production of rock lobsters may be increased through some form of aquaculture. It is already possible to capture wild rock lobster puerulus from artificial collectors and ongrow them in land-based holding systems. However, this creates some controversial issues in relation to sustainability of the natural resource. Consequently, expansion of output by this means is limited. Therefore, artificially closing the life cycle (propagation) is the only option for unlimited sustainable expansion of rock lobster production from Australia.

In 1997/98 the FRDC organised a number of workshops to discuss the issue of rock lobster aquaculture and how the research should proceed. Research projects were formed into a Subprogram which covered all the perceived issues except that of propagation. Propagation research was considered high risk and long term. The investment required was considerable and therefore must be particularly well focussed and planned.

Rock lobster propagation has only been achieved in the laboratories of Japan and New Zealand. Very few puerulus have been produced and there is a paucity of information on the subject available in the literature. If the problem of large scale commercial production of puerulus is to be solved, then it requires a collaborative approach and a pooling of existing resources and information.

In Australia, lobster propagation research is limited to Tasmania where the Tasmanian Government has invested in a small-scale propagation project. However, there are:

- nodes of expertise in Australia which require co-ordination; and
- opportunities for tapping overseas experience.

The fully collaborative national project required to achieve these objectives will be highly complex with a number of components and organisations. These organisations control major resources that must be co-ordinated and brought to bear on the problem of rock lobster propagation.

The FRDC workshops concluded that the best way to achieve this co-ordinated collaboration was to organise a workshop with a number of specific objectives:



- to identify potential international and national, research & industry partners who are interested in collaborative research into rock lobster propagation;
- to identify barriers to the development of commercial propagation and the R & D required to overcome them; and
- to identify the available resources and the requirements for propagation R & D.

The final outcome of the workshop would be a research plan mapping out R & d components, collaborative partners, timelines and resources which would form the collaborative, multi-institutional project required to tackle the problem of rock lobster propagation.



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## Welcome by Professor Colin Buxton

*Director, Tasmanian Aquaculture and Fisheries Institute*

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It gives me great pleasure to welcome all of you here today. Especially to our interstate and international visitors, welcome to Hobart and to this our 1999 FRDC Rock Lobster propagation workshop, hosted by the Tasmanian Aquaculture and Fisheries Institute.

The Rock Lobster Fishery in Australia is an important resource and constitutes some 25% of Australian Fishery landings with a present worth of about \$450M a year. Most of these fisheries are fully exploited; some are slightly over-exploited and recent increases in value have arisen, mostly through the sale of live product and through inventive marketing strategies. I think it is safe to say that there appears to be little scope for significant increased production in the wild fishery, but there is scope to increase production through aquaculture.

We are gathered here today to contemplate Rock Lobster Aquaculture and more specifically the very early life history stages of growth from eggs to puerulus. There is rapidly growing interest in rock lobster aquaculture around the world, recently extended to Australia. In response to this the Fisheries Research and Development Corporation (FRDC) has established a Rock Lobster Enhancement and Aquaculture sub-program to examine this potential and to co-ordinate our efforts.

Currently there are three options for rock lobster aquaculture:

- The first is on-growing of sub-adult animals, an activity that has been in progress in South Australia since 1994 with some success. There are obvious problems associated with method relating to perceived impacts on wild fish stocks.
- The second option is the on-growing of wild caught puerulus in a hatchery situation which has been mooted in several states around Australia. Without over simplifying this issue it is predicated on the notion that the impacts on the wild stock will be smaller or insignificant because natural mortality in these early life history stages is quite high. Notwithstanding this statement there are concerns particularly in the wild fish sector that puerulus harvest could be detrimental to the fishable stock.
- The third option, the subject of our meeting over the next couple of days, is control of the life cycle and the culture of rock lobster from eggs hatched in the laboratory. As you know, spiny rock lobsters have a complicated life cycle, the eggs hatch into a phyllosoma that spends about 24 months at sea undergoing 11 moults before settling on to the reef. While laboratory culture of



phyllosoma has been achieved in Japan and New Zealand, successes are described as only moderate, as only a few individuals survive.

The purpose of this initiative is to workshop our knowledge of this subject and to share research experiences and results. I am firmly of the opinion that such a collaborative effort is the only way to fast track our understanding of this very difficult field of science.

I believe we have assembled an impressive range and depth of expertise under the one roof to discuss rock lobster propagation. I have absolutely no doubt that your deliberations will be informative and stimulating and that this meeting will lead to greater things.

I would like to thank the FRDC for their sponsorship and support of this workshop and the organiser Dr Piers Hart who has made it all happen. With that, it gives me great pleasure to declare this workshop open.



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## Introduction by Dr Patrick Hone

*Programs Manager, Fisheries Research and Development Corporation*

*(Summarised by Robert van Barneveld)*

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The Fisheries Research and Development Corporation have invested in this workshop so that a considered approach to the question of rock lobster propagation can be facilitated. FRDC want to know if rock lobster propagation is a feasible option and if it is something in which FRDC should invest its money. To answer these questions we need to examine the biological feasibility of propagation and the economic feasibility of it. Through the workshop we wish to identify gaps in our knowledge and research topics we need to address.

Assuming the workshop can successfully collate available information on the biological and economic feasibility of rock lobster propagation it is then important to develop a structure for proposed research into the propagation of rock lobster. We need to address ways to include New Zealand expertise and Japanese expertise, and we want to make sure we collaboratively capture the existing Australian expertise so that we can break down the interstate rivalries that sometimes occur. We also want to be inclusive with other funding agencies and other people who have an interest in this research field (eg. CRC for Aquaculture, other Government agencies, other agencies with an interest in this particular area of research).

Rock lobster enhancement and aquaculture is an area that has been attracting interest for some years across the world. To date, however, aquaculture with the spiny lobster on a commercial scale is limited with the exception of live-holding of adult lobsters in sea cages. Recently, the FRDC received a range of research proposals from different groups across Australia to pursue investigations into rock lobster aquaculture. As a national funding body, FRDC identified an opportunity for a coordinated approach to this research effort and formed the Rock Lobster Enhancement and Aquaculture Subprogram which is managed by Dr Robert van Barneveld. The subprogram currently addresses a number of research areas which are encompassed in the following projects (including project 98/300 which represents this workshop):

**98/300: Propagation of rock lobster – development of a collaborative national project with international partners**

*Principal Investigator:* Dr Piers Hart



*(Tasmanian Aquaculture and Fisheries Institute, Marine Research Laboratories, Nubeena Crescent, Taroona, TAS, 7053)*

*Project Objectives:*

1. Identify potential for the development of commercial culture of rock lobster in Australia and determine the research and development potential required to achieve this potential.
2. Identify what resources (expertise, facilities, funds) are required to conduct the necessary research and development.
3. Identify potential international and national research and industry partners who are interested in collaborative research into culture of rock lobster from eggs.
4. Develop a research plan mapping out research and development projects, the collaborative partners, timelines and resources.
5. Submit a proposal to FRDC under the rock lobster enhancement and aquaculture subprogram.

**98/301: Facilitation, administration and promotion of the FRDC Rock Lobster Enhancement and Aquaculture Subprogram.**

*Principal Investigator:* Dr Robert van Barneveld  
*(Barneveld Nutrition Pty Ltd, PO Box 42, Lyndoch, SA, 5351)*

*Project Objectives:*

1. Coordinate the FRDC Rock Lobster Enhancement and Aquaculture Subprogram (applications, workshops, communication).
2. Conduct an annual research workshop to present research outcomes from the subprogram and to define research objectives for subsequent years.
3. Facilitate travel of the subprogram project Principal Investigators, a nominated industry representative and the Subprogram Leader to biannual scientific meetings.
4. Facilitate travel of industry representatives and the Subprogram Leader to biannual Steering and Management Committee meetings.
5. Coordinate the preparation of a subprogram newsletter, media releases and workshop publications.
6. Integrate with other FRDC and externally funded rock lobster research programs (eg FRDC Project 98/300 – Propagation of rock lobster – development of a collaborative national project with international partners and the FRDC Rock Lobster Post-Harvest Subprogram).



**98/302: Towards establishing techniques for large-scale harvesting of pueruli and obtaining a better understanding of mortality rates**

*Principal Investigator:* Dr Bruce Phillips  
(Fisheries WA, WA Marine Research Laboratories, PO Box 20, North Beach, WA, 6020)

*Project Objectives (Revised June, 1999):*

1. To determine appropriate puerulus to legal size survival rates and potential harvesting ratios, that if implemented in the western rock lobster fishery, might result in “biological neutrality” being achieved.
2. To establish techniques for large scale harvesting of pueruli for rock lobsters.
3. To collaborate with scientists conducting puerulus collection research in Tasmania.

**98/303: Feed development for rock lobster aquaculture**

*Principal Investigator:* Dr Kevin Williams  
(CSIRO Division of Marine Research, Marine Laboratory, 233 Middle Street, Cleveland, Qld, 4163)

*Project Objectives:*

1. Review the state of knowledge of crustacean and fish nutrition of relevance to the development of formulated feeds for rock lobsters.
2. Develop a ‘best guess’ formulated diet for juvenile and adult rock lobsters (for tropical, *Panulirus ornatus*, western, *P. cygnus* and southern, *Jasus edwardsii* species) for comparative trialing against existing ‘fresh’ diet.
3. Determine the response of post-pueruli/juveniles and adult rock lobsters to key nutrients critical for the development of cost-effective formulated diets for rock lobsters.
4. Make recommendations on the direction of future nutrition research.

**98/304: Pilot study of disease conditions in all potential rock lobster aquaculture species at different growth stages**

*Principal Investigator:* Assoc Prof Louis Evans  
(Curtin University of Technology, Aquatic Sciences Research Unit, GPO Box U1987, Perth, WA, 6001)



*Project Objectives:*

1. To establish a national network of rock lobster health and disease personnel.
2. To conduct a symposium on health and disease management in lobster aquaculture and long-term holding facilities
3. To collate and document current state of knowledge on rock lobster diseases.
4. To provide a disease diagnosis service for existing FRDC rock lobster projects in Queensland, South Australia and Western Australia.

**98/305: Determination of optimum environmental and system requirements for juvenile and adult rock lobster holding and grow-out**

*Principal Investigator:* Assoc Prof Mike Geddes

*(University of Adelaide, Department of Zoology, GPO Box 498, Adelaide, SA, 5001)*

*Project Objectives:*

1. Assess the interactions between stocking density and feed delivery system on maintaining and improving condition and on the growth performance of adult rock lobsters in existing sea-based holding systems in different seasons.
2. Determine the effects of temperature, salinity and photoperiod on the growth rate and survival of juvenile rock lobsters in existing land-based holding systems.
3. Determine the effects of stocking density and shelter on the growth rate and survival of juvenile rock lobsters in existing land-based holding systems.
4. Evaluate existing system design and management regimes for land-based captive grow out of juvenile rock lobsters and for sea-based holding of adult rock lobsters.

With the expertise assembled from across the world for this workshop, FRDC has every confidence that a meaningful outcome will be achieved and a highly relevant research program will eventuate.



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# Outline of previous and future research on aspects of *Jasus edwardsii* biology and behaviour pertinent to management of broodstock

*Alistair MacDiarmid*

*NIWA, Greta Point, PO Box 14-901, Wellington, NZ*

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*Female mating window:*

*Alistair MacDiarmid*

In *Jasus* species the spermatophore deposited by the male on the female's sternal plate during copulation is short-lived and must be used immediately by the female to fertilise her one annual clutch of eggs. Therefore, a mate must be available when the female's crop of eggs is mature and ready to be extruded. How long is this mating window and what are the consequences for the female if a suitable mate is not found? We addressed these questions in a laboratory experiment carried out at the NIWA Greta Point campus.

Males and small and large mature females were obtained from the fishery in February 1997, well before the female moult, housed apart in tanks and provided with suitable shelter and food. After females moulted in March/April they were randomly assigned to one of 6 treatments, where mates were made available to small and large females prior to, on, or 5, 10, 15 and 20 days after the predicted day of mating (based on moult date and carapace length). We monitored the tanks daily for courting, copulation, egg bearing and mortalities and measured the size of any resulting egg clutches. Under this regime we hoped to determine the length of the female mating window, the period in which females could court, mate and successfully fertilise a full clutch of eggs. Was it a period of days or weeks?

Comparison of results from control females, those which had access to a male for 30 days prior to copulation and thus could mate whenever they were ready, and 0 day delay females, those which had a mate available on the predicted day of mating, showed that there was an experimental artefact. Courting, copulation, egg extrusion and clutch weight were all depressed by 10-30% in 0 day delay females. These results are a reflection of the imprecise tool we had available to predict the actual day of mating for individual females. For example, the actual day of mating of control females varied from 3 days before to 5 days after the predicted day. This experimental artefact needs to be kept in mind when interpreting results from the remainder of the experiment. We found that courting, copulation, egg extrusion and clutch size are all depressed compared to 0 day delay females if males are unavailable either 5 or 10 days after the predicted day of mating. Clutch sizes were depressed by 50%



after a delay of 5 days and by 98% after 10 days. Any further delay results in no reproductive behaviour at all. These results indicate a narrow mating window for female red rock lobsters. Mating must take place within a day or two of when the females crop of eggs are ripe for a normal sized clutch of eggs to be produced.

*Effect of non-mating:*

*Alistair MacDiarmid, Rob Stewart & Megan Oliver*

What is the fate of females which don't have a male available at the required time and never extrude a clutch of eggs? Do they suffer any long term consequences? This is of potential importance to any broodstock management programme. To address this question we kept mated and unmated females from the experiment described above until the 1998 mating season and monitored the tanks daily for moulting mortalities. We found that unmated females attempted to resorb eggs in the ovary, resulting in strong pink staining of the haemolymph and muscle tissue with egg derived pigments. They were also weak and lethargic possibly because the large mass of eggs in the body cavity prevented feeding and normal organ function. In contrast, females which extruded a full batch of eggs had a clear white abdomen with normal coloured haemolymph.

One year after mating, females normally have large ovaries packed with developing oocytes. In contrast, 1 year after their predicted day of mating, the ovaries of surviving non-mated females show evidence of atrophy with only small regions of normal egg development. Although most of the surviving non-mated females mated in the subsequent breeding season, their clutches were very small compared to normally breeding females.

*Effects of male size on female fecundity:*

*Alistair MacDiarmid & Mark Butler*

Sperm limitation, when female fertilisation success is constrained by the supply of sperm, is generally perceived to be an uncommon feature of reproduction in species which directly transfer gametes during copulation. Male size, previous copulations, and the balance of expected reproductive return and future mating opportunity may, however, limit the amount of sperm males transfer to females. We use laboratory experiments where mate size could be manipulated and its consequences on spermatophore size and clutch size determined, to show that in two genera of spiny lobsters (Crustacea: Palinuridae) male reproductive output limits the size of clutches brooded by females. In *Panulirus argus* from the Florida Keys we show that while male size affects spermatophore area, males also vary the amount of ejaculate positively with female size. Furthermore, the area of the spermatophore has a greater influence on the consequent clutch weight than does female size. In *Jasus edwardsii* from New Zealand, female size, male size and mate order all effect clutch weight. In



both species clutches fertilised by small males in the laboratory are significantly smaller than clutches fertilised by large males. These results suggest that to ensure they receive sufficient sperm, females should either mate several times prior to oviposition (not possible in *Jasus*), mate as early as possible in the reproductive season, or choose large, preferably unmated males as partners and thus compete with other females for preferred males. Sperm-limited female fecundity has the potential to limit egg production of captive females if large males are not available.

*Sperm discharge and regeneration in male spiny lobsters:*

*Jenny Mauger & Alistair MacDiarmid*

From the male perspective ejaculation of sperm carries associated costs not only of production, but also of lost mating opportunity, especially in species with short breeding seasons because there may be insufficient time to recharge sperm stores. Male cost of mating is low if only a small proportion of the sperm are discharged per mating or if their recharge rates are rapid. Male costs of mating are high if a large proportion of the sperm are discharged per mating or if their recharge rates are slow. Previous laboratory experiments with *Jasus edwardsii* where males mated with up to 5 females in succession showed there was a corresponding decrease in clutch size hinting that male costs of mating may be high in this species.

We have been working on a laboratory experiment designed to determine whether large male *J. edwardsii* do indeed deposit larger spermatophores than small males and determine the subsequent recharge rates of sperm in the vas deferens after single and multiple matings. These results will ultimately be important in determining the capacity of small and large males to mate successively over the breeding season. We use a unilateral gonopore blocking approach to address these questions.

The male spiny lobster extrudes his sperm through twin gonopores located at the base of the fifth pair of walking legs which allows separate manipulation of the left and right testes and vas deferens. A direct comparison of extruded spermatozoa, from the unblocked side and non-extruded spermatozoa from the blocked side can be examined in the same animal. This approach is not possible in vertebrates such as mammals, birds, fish and reptiles where there is a single opening.

Following a trial of 4 “off-the- shelf” glue products to block one of the gonopores, we opted for Araldite as it was the only glue which remained attached. The Araldite set to form a cap with no noticeable toxicological side effects to the animal.

The male gonopore was blocked prior to exposure to its mate. Tanks were checked daily for females in berry, indicating mating had occurred, and the integrity of glue caps on non-mated males was also



checked. Once the male had mated, he was removed from the mating arena and his testes and vas deferens sampled up to 21 days later. Measures of sperm allocation and recovery are being made by examining the difference in wet weight between the left and right testes and vas deferens, the difference in sperm concentration in vas deferens and the difference in sperm maturation in testes.

We are running four experiments concurrently. In the main experiment small and large males are mated once with small females and then sampled 0, 3, 7, 14 and 21 days later to assess sperm depletion and recovery in the testes and vas deferens. Just prior to the start and at the conclusion of the experiment, 5 control males were sacrificed, their left and right vas deferens surgically removed and weighed to compare with the non-mated gonads of the captive animals. The maturity and stage of development of sperm in each sample will later be determined by histology.

In the second experiment, small and large males were mated once, this time with a large female, then sampled the following day. Data from this experiment will reveal sperm depletion or allocation to large females compared with small females used in the first experiment.

The third part of this experiment investigates serial depletion of sperm in testes and vas deferens over a series of three matings. Small and large males were each mated with three large females and sampled the day of the third copulation.

The fourth concurrent experiment provides background variation of sperm concentration in vas deferens and maturation in testes in the wild. Seasonal samples are being taken monthly for the duration of a calendar year from the capture fishery.

Preliminary results from the main experiment indicate a quicker recovery for smaller males than larger males, whereby smaller males perhaps recover vas deferens wet weight in seven days. Even after 21 days recovery from a single mating, the vas deferens of large males were still 25% lighter than their pre-mating wet weight. Problems encountered with glue caps becoming dislodged in the 1998 mating season mean that the level of replication is not sufficiently high for robust results. For instance glue caps were lost through the mechanical action of the articulating limb against the sternum popping off the cap. The strong muscular contraction of ejaculation was thought responsible for losing other glue caps. In the larger males, the tips of some glue caps were eroded by abrasion against the bottom of their tanks and some ejaculate lost through the “blocked side”. Obviously, in these situations, the data could not be used to compare pre and post mating differences in sperm discharge and recovery and further replication is planned for the 1999 mating season.



*Female mate choice:*

*Alistair MacDiarmid*

Under the circumstances described in the sections above we expect there to be strong selective pressure, especially on large females, to choose the largest mates available. In 1997 we completed an experiment designed to determine if and when, large and small female *Jasus edwardsii* exercise a size based choice of mate. We examined two sizes of female at 4 different times during the moult/mate cycle. The experiment was carried out in two large outdoor tanks at NIWA, Wellington. In each tank were 4 shelters, one in each corner. One shelter was left empty while the other contained a large male, a small male or a female. These lobsters were constrained from moving far by tethering them to the shelter. A single test female was placed in each of the experimental tanks each morning and its position noted 24 hours later. The positions of choice lobsters were rotated around the tank to guard against artefacts caused by “corner” effects. We were looking for association of females with large males greater than we expected by chance alone.

Before we started the experiment we knew there were two possible hypotheses that might explain female choice of large males if it indeed occurred: mate choice and predator avoidance. Because females moult prior to mating they may simply cohabit with large males to avoid predators, especially by small females immediately after moulting while their new exoskeleton is still very soft. In contrast under the mate choice experiment we expect the opposite to occur.

Prior to moulting large females showed no preference in where they sheltered but this changed immediately after moulting when 74% sheltered with large males. This increased slightly to 76% with late post moult females. Once they had mated large females showed no particular choice of shelter. Small females showed no significant preference for large males until just before they mated. After mating small females again showed no preference.

Which of our hypotheses best explains these data. We found little support for the predator avoidance hypothesis. Female choice, especially by small “vulnerable” females, of large males increases with increasing time since moulting, opposite to what might be expected under the predator avoidance hypothesis. The data match our expectations under the mate choice hypothesis.

It is interesting to note the close agreement between our recent lab observations of female choice of large males and previous field observations in which 73% of post-moult/pre-ovigerous females cohabited with a large male. It suggests that much of the field patterns of dispersion of females amongst males during the mating season is driven by female choice of mate exercised well before the overt signs of courtship are displayed. Thus females may spend a substantial period of time prior to



mating searching for a suitable mate. This might be one mechanism which ameliorates the effects of reductions in the abundance of large males in fished populations on female fecundity.

*Male mate choice:*

*Alistair MacDiarmid*

Males might also be expected to choose the largest available female to mate. Larger females may represent a better investment in terms of numbers of eggs available to fertilise against the time invested in courtship. Larger females have 20% larger eggs than small females and consequently their stage 1 phyllosoma larvae are larger, though laboratory experiments indicate that this makes no difference to larval swimming speed or survival. Males may choose to with the largest available females because larger females are either older, and thus proven survivors, or faster growers than smaller females, attributes beneficial to survival and/or growth of the males offspring.

To test these ideas we conducted a set of experiments similar to those described above for female choice. Both pre-moult and post-moult females of four different sizes were tethered alone in concrete block shelters in the 4 corners of a large outdoor pool. A single test male (small 100 mm CL or large >180 mm CL) was placed in each of the experimental tanks each morning and its position noted 24 hours later. The positions of tethered lobsters were rotated around the tank to guard against artefacts caused by “corner” effects. We were looking for association of males with large females greater than we expected by chance alone.

We found no strong selection by males for large females. Large male choice of large females did increase from 25% of pre-moult females to 45% of post-moult/pre-ovigerous females but this was non-significant (n = 20). Selection by small males for large post-moult/ pre-ovigerous females was even lower (30%).

*Conclusions:*

1. Females have a narrow temporal “window” in which to successfully mate.
2. Females which have no opportunity to mate at all either die or suffer long-term damage to their ovaries.
3. Clutches fertilised by small males are significantly smaller than clutches fertilised by large males.
4. Males may take up to 21 days to recharge their vas deferens.
5. Females select large males as preferred mates.
6. Females select potential mates up to 60 days prior to mating.



7. Large females, with more to lose if a large male is not available when she is ready to mate, select potential mates, on average, 14 days prior to small females.
8. Males show no strong size selection of mate.

#### *Future Research Directions*

Over the next 3 years we have funding to undertake research in the following areas:

1. Complete the investigation of male sperm discharge and recovery rates. (Jenny Mauger & Alistair MacDiarmid)
2. Use infra-red video cameras to determine how females choose the largest available male. (Alistair MacDiarmid)
3. Complete investigations into the effects of non-mating on female survival and reproduction. (Alistair MacDiarmid, Rob Stewart, Megan Oliver)
4. Determine the role of olfaction in mate selection and courtship behaviour of spiny lobsters, *Jasus edwardsii* and *J. verreauxi*. (Tania McPherson & Alistair MacDiarmid)
5. Determine the effects of male/male competition on the amount of sperm males transfer to females. (Alistair MacDiarmid & Mark Butler)
6. Determine the mechanism and outcome of female/female competition to mate with the largest available male. (Alistair MacDiarmid & Mark Butler)



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# Summary of the work done in New Zealand at NIWA Mahanga Bay, on rock lobster (*Jasus edwardsii* and *J. verreauxi*) propagation

*Lennard J. Tong and Graeme A. Moss*

*NIWA, Box 19-401, Kilbirnie, Wellington, New Zealand*

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## *Introduction*

In two periods (1990-92; 1994-97) over the past 8 years, staff at the Mahanga Bay research centre, Wellington, New Zealand, have carried out studies on *Jasus edwardsii* with the aim of rearing juveniles from the egg. The research covered three phases: the manipulation of egg development to extend the season for the supply of larvae; determining the food and physical requirements of the early and mid-stage phyllosoma larvae; and to develop a rearing system that was low maintenance and minimised handling and damage to the larvae. We also attempted to rear larvae through to pueruli but only achieved this on one occasion (Booth 1996). In 1997-98 we begun a pilot study to look at rearing the larvae of *J. verreauxi* and from June 1998 began a long term project to rear this species from the egg to juvenile.

Our commercial partners stopped publication of most of results from the first period of work. In the second period we repeated and/or revisited some of the studies as well as carrying out new work and have since published, and submitted or prepared papers for publication. This review will therefore be in the form of abstracts of all the papers that have been written or prepared to date followed by a section where we discuss the implication of our findings to large scale larval rearing. Finally we will discuss directions for further research in New Zealand.

Apart from relatively small commercial inputs most of the work was funded through the New Zealand Public Good Science Fund (PGSF). We would also like to acknowledge the support of our colleagues at Mahanga Bay and Mr Stephen Brooker of Oceanculture New Zealand Ltd.

## *Broodstock management*

**Effect of temperature on embryo and early larval development of *J. edwardsii* and a description of a method to predict larval hatch times.** (Tong and Moss 1997; Tong, Moss, Pickering, and Paewai *in prep.*)

Rock lobsters (*Jasus edwardsii*) were mated in the hatchery. The ovigerous females with known egg extrusion dates were held at 4 temperatures and embryo development monitored at weekly intervals. Times to hatch were approximately 65 days, 95 days, 155 days and 163 days at 18°C, 15°C, 12°C and ambient (8 to 14°C) respectively. For the embryos, the appearance of the median eye, the eyes and the



chromatophores, and an eye index, were used to calculate the biological zero i.e. the temperature at which development ceases which, for *J. edwardsii*, was 7.53°C. A formula was then derived to predict time to hatch at a range of temperatures using the cumulative difference between the rearing temperature and the biological zero.

The eye index (mean of length and width of eye) can be used to calculate the effective accumulative temperature (EAT) using the regression equation ( $\text{Log EAT} = 2.01 + 0.358 \text{ Log Eye Index}$ ). By measuring the eye index of an embryo and calculating the EAT the following equation can then be used to predict days to hatch.

$$\text{Days to hatch} = \frac{\text{EAT at hatch (699)} - \text{EAT at time index measured}}{\text{Rearing temp} - \text{Biological zero (7.53)}}$$

Data was also collected on the time between females moulting and mating, the total number of larvae successfully hatching for a range of female sizes and Stage I phyllosoma larval size in relation to the embryo rearing temperature.

#### *Physical requirements of stages i-viii(ii) phyllosomas*

#### **Effect of temperature on the inter-moult period, feeding rate and survival of Stage I phyllosomas.**

(Tong, Moss, Paewai, and Pickering *in prep.*)

*Jasus edwardsii* naupliosoma larvae were hatched from embryos reared at three temperatures, 18°C, 15°C and ambient (8.0°C to 14°C). The Stage I larvae were then reared at temperatures from 12°C to 24°C and fed brine shrimp raised on the flagellate *Rhodomonas reticulata*.

Larvae reared at 24, 21, 18, 15 and 12°C moulted to Stage II larvae after 7, 8, 11, 17 and 33 days respectively and the variation about these times was small. Survival rates through to Stage II normally ranged from 70% to 100%. The exceptions were larvae from 18°C embryos which had a survival rate of only 40% when reared at 24°C and ambient reared embryos reared at 12°C, where the survival was 10% and 15%. The number of brine shrimp consumed each day declined with reducing temperature.

#### **Effect of temperature on the inter-moult period, feeding rate, and survival of Stage II, IV, VI, and VIII phyllosomas.** (Tong, Moss, and Paewai *in prep.*)

Stages II, IV, VI and VIII *Jasus edwardsii* phyllosomas were reared individually at 3 temperatures and fed either 4, 8, 12, 16, 24 or 36 large (2.7 to 3.3 mm) brine shrimp day per day. At optimum feeding levels and at 24°C, 21°C and 18°C, Stage II larvae moulted at 6, 8 and 11 days respectively, Stage IV larvae at 9, 10 and 12 days, Stage VI larvae at 12, 14 and 17 days and Stage VIII (instar 2) at 18, 21 and 25 days. The consumption rate (brine shrimps eaten per day), increased with an increase in temperature, but overall consumption during the intermoult period was lower. The intermoult period increased as the brine shrimp ration was reduced. Survival was



generally >70% but at 24°C and with more than 16 brine shrimp, survival was <40%. Phyllosomas up to Stage VI can be reared successfully at 21°C with a moult rate approximately 25% faster than at 18°C and Stage VIII larvae can be reared at 24°C.

#### **The effect of brine shrimp numbers on the growth and survival of early stage phyllosomas.**

(Tong et al. 1997)

Stages I to VI *Jasus edwardsii* phyllosomas were fed daily with a fixed number (1, 2, 4, 8, 12 or 16) of 2-3 mm brine shrimps (*Artemia salina*) to determine the optimum requirement for growth and survival. For Stages I and II the threshold below which food became limiting, measured as a significant delay in moulting, was <2 brine shrimps per day. For Stage III the threshold was 4 brine shrimps per day, for Stages IV, V it was 8 brine shrimps per day and for Stage VI, 12 brine shrimps per day. Growth at the moult was reduced when food was limiting. The feeding rate reduced immediately prior to the moult and this was most evident for Stages V and VI. The results are discussed in relation to large scale culture of phyllosomas.

#### **Effect of brine shrimp size on the feeding rate, growth and survival of early stage phyllosomas.**

(Tong and Moss *in prep.*)

Stages I, III and V phyllosoma larvae of the rock lobster *J. edwardsii* were fed daily with 1 mm brine shrimps (*Artemia salina*) at densities of 5, 10, 20 and 40 ml<sup>-1</sup> while controls were fed on 2-3 mm brine shrimps. Stage I larvae moulted at the lowest density in the same time as the controls. Stage III and V larvae fed at a density of 20 ml<sup>-1</sup> or less took significantly longer to moult than the controls. At the highest brine shrimp density inter-moult periods were not significantly different from the controls. There was no difference in the survival rates which ranged between 70 and 100% for all stages. The feeding rate increased significantly as the density of brine shrimps increased. Phyllosoma larvae up to Stage V can be reared successfully on 1 mm brine shrimps and, although greater numbers of brine shrimps are required, the costs of food production are reduced.

#### **Effects of food density on the growth and survival of early-stage phyllosomas.** (Moss et al. *in press*)

Individual Stage I, III and V phyllosomas were also fed daily, fixed numbers (2, 4, and 8 respectively) of brine shrimps in different volume containers (10, 20, 40, 60, 80, 120 and 240 ml) to alter the prey density while maintaining prey numbers. At the levels tested (equivalent to 25-200 brine shrimp L<sup>-1</sup>) prey density had no significant effect on intermoult period or post-moult size but did affect consumption rates.



### **Development of formulated diets for phyllosomas.** (Moss 1997)

Stage II, IV and VI *Jasus edwardsii* phyllosomas and Stage IV *J. verreauxi* phyllosomas were fed a range of formulated diets and their growth and survival compared with larvae fed natural foods (*Artemia* juveniles and mussel gonad). Alginate was used to bind the ingredients into the formulated diets. Survival was highest on the natural foods. None of larvae fed formulated diets made with dried ingredients survived or moulted. However formulated diets made with *Artemia* incorporated into the alginate did promote survival and moulting. Flattened sheets of alginate and *Artemia* (“pizza’s”) are suitable for feeding to phyllosomas cultured in upwelling tanks.

### **Fitness of Stage I phyllosomas.** (Rea et al. *in prep.*)

Fitness of Stage I *J. edwardsii* and *J. verreauxi* phyllosomas was determined by establishing the point of no return (PNR) and point of reserve saturation (PRS). PNR is defined as the maximum period of starvation before first feeding that will allow the larvae to survive and moult to the next stage. PRS is the minimum feeding time to allow sufficient energy reserves to be accumulated for survival and moult to the next stage.

PNR and PRS varied between the species. *J. verreauxi* required food from the first day after hatch while *J. edwardsii* could delay feeding for 4-5 days. Likewise *J. verreauxi* required a longer feeding period to ensure survival through to the moult. Feeding and starvation are reflected in the size, shape and cell volumes of the hepatopancreas.

### **Effects of light intensity on the growth and survival of early-stage phyllosomas.** (Moss et al. *in press*)

Stage I, III and V *Jasus edwardsii* phyllosomas were fed daily with a fixed number (2, 4, 8, 12, or 16) of 2-3 mm long brine shrimps (*Artemia salina*) under four different continuous light intensities (10, 0.1, 0.001 and  $\leq 0.0002 \mu\text{mol s}^{-1} \text{m}^{-2}$ ) to determine the optimum requirement for growth and survival. Phyllosomas of each stage fed higher rations of brine shrimps had a significantly shorter intermoult period and larger post-moult size. Light intensity had a significant effect on the post-moult size of Stage I phyllosomas but had no effect on the intermoult period.

### *Summary and implications for culture*

#### **Broodstock**

From our experience the issue of ensuring a supply of newly hatched larvae over a long period can be achieved by: 1) mating a wide size range of females to ensure different mating times and; 2) raising the berried females at different temperatures so that egg development is accelerated or slowed. Using these techniques we have obtained larvae over a 7 month period.



If longer periods are desirable it would not be too difficult to control the reproductive cycle by manipulating the photoperiod and temperature. In Japan, Kittaka (1997) has reversed the cycle of several southern hemisphere species and successfully obtained larvae over many years.

The temperature during embryogenesis does influence subsequent larval development and growth. Larvae hatched from *J. edwardsii* embryos, reared at 18°C, were significantly smaller than those embryos reared at 15°C. Larval size and energy reserves may affect survival and LD 50's do vary quite considerably among larvae from different females (pers comm. Alsitair MacDiarmid). The larger size of newly hatched larvae from 15°C and ambient embryos was further reflected in the size of Stage II larvae which were larger than those from embryos reared at 18°C.

### *Larval rearing*

#### **Temperature**

An increase in temperature accelerated development and reduced the intermoult period for Stage I-VIII larvae. When considering the long larval life of *J. edwardsii* in the wild and in culture systems a reduction in time could make rearing phyllosoma through to pueruli cost effective. At 24°C and 21°C the rate of development of Stage I larvae was about 36% and 27% faster respectively than at 18°C. The fact that larvae from embryos reared at 15°C also had a high survival at the higher temperatures suggests that the faster development rate could be obtained without high mortality. A temperature of 21°C was also suitable for Stages II to VI and 24°C for Stage VIII larvae indicating that overall, a faster development time can be achieved. Larvae reared at higher temperatures ate significantly more brine shrimp each day but the shorter culture time, may offset the costs to heat water and to produce more brine shrimp to sustain the higher feeding rates.

#### **Feeding**

For Stage I to VI phyllosoma larvae, 2-3 mm brine shrimps reared on microalgae are a suitable food. Brine shrimps are readily produced, are easy to manage in a culture system and their nutritional value can be altered and/or enhanced with natural and artificial diets (Dhont and Lavens 1996). An increase in the number of 2-3 mm brine shrimps supplied to Stages I to VI phyllosoma larvae of *J. edwardsii* led to increased consumption, resulting in a reduction in the intermoult period and significantly increased growth at the moult. For Stage V phyllosomas the prey density had a significant effect on the food capture rate, with numbers captured decreasing as density decreased.

Large brine shrimps can be replaced with smaller 1 mm brine shrimps for early-stage phyllosomas. One millimetre brine shrimp are cheaper to produce and Stages I, III and V larvae can capture sufficient



brine shrimps of this size to moult to the next stage. However higher densities of 1 mm brine shrimps are needed to obtain consumption rates that equate to feeding 2-3 mm brine shrimps, especially for Stages III and V larvae.

However brine shrimp do not appear to be completely suitable for the later-stage phyllosomas and the only success with rearing through to pueruli has been feeding brine shrimps and mussel gonad. For early-stage larvae fed 1 mm brine shrimps there are minimum rations required to be able to capture sufficient food to moult at the optimum rate and there is evidence that there is a maximum number which can be caught and handled in 24 hours. For Stage VIII larvae the consumption of 2-3 mm brine shrimps averages up to 28 per day but the intermoult period between instars also increases, suggesting that these larvae are capturing and handling near to the maximum. It is important that phyllosomas not only consume enough food to moult rapidly but that the food is of suitable nutritive value for the larvae build up the energy reserves required for the next stage.

Artificial diets made from *Artemia* bound in alginate (pizzas) are attractive to phyllosomas. For late-stage larvae, feeding brine shrimps in a pizza allow the consumption of high numbers of brine shrimps while reducing the capture time. The pizzas provided a large food package which, unlike mussel gonad, did not immediately break down and decompose.

Supplying phyllosomas with an optimum ration of brine shrimps, or other prey item, at the appropriate density, in conditions to maintain that density, will be important in managing large scale phyllosoma larval culture systems to maximise both growth and survival and to minimise cost.

### **Light intensity**

The low light intensities had little or no effect on the growth and survival of Stage I, III, or V phyllosomas. The low light intensities or continuous dark commonly used for culturing phyllosomas assist in maintaining an even distribution of brine shrimps and early-stage phyllosomas in the tanks so that no behavioural separation of predator from prey occurs. There is the suggestion of an interaction between optimum prey ration and prey density which means that maintaining sufficient densities and minimising aggregative behaviour of the prey items and larvae in tanks will be important for culturing phyllosomas.

### *Tank technology*

#### **Description of an up-welling tank for phyllosomas. (Illingworth et al. 1997)**

An up-welling tank system suitable for rearing phyllosomas has been developed. Four tanks, one of which remains empty, are interconnected by ports which allow larvae to be transferred without



handling and the vacated tank to be cleaned. The vertical water movement in each tank maintains the larvae in suspension and the overall design and management allows for feeding live brine shrimps (*Artemia*). Mean survival rates over 60% to Stage VIII have been obtained but only one puerulus has been reared.

### **Large scale culture vessel**

A large scale up-welling system has been constructed from two 500 l moulded fibreglass tanks painted black and tested with *J. verreauxi* phyllosomas. The volume of each rearing chamber is 400 l. Larvae reached Stage VI-VII before being terminated. Transfer ports allow the passive movement of larvae from one tank to the other for ease of cleaning. A third tank acted as a reservoir and allowed water to be pumped through the up-welling tanks on a partial re-circulation basis (5-10% per hour). An in-line diatomaceous earth filter and an ultra-violet irradiation unit were used to maintain water quality.

### *The future*

Kittaka (1997) has obtained the highest survival of phyllosoma for *J. verreauxi* and has reared several hundred pueruli. Kittaka considers it to be the hardiest of spiny lobster species that he has worked with. *J. verreauxi* also has a shorter larval life than *J. edwardsii*. We have therefore chosen *J. verreauxi* as our experimental animal to continue to define the feeding and physical requirements of phyllosoma larvae which may then be applicable to other species.

Our short term (2 years) aim is to determine whether we can reduce the inter-moult periods but maintain good growth by changing feeding schedules and/or manipulating food quality. The studies will be carried out on early and mid-stage larvae. If we obtain positive results then we will attempt to carry out similar experiments on individual larvae of Stage VIII and upwards.

At the same time we will determine the effects of temperature on feeding rates, growth and survival. As a sub-tropical species *J. verreauxi* larvae may be able to tolerate higher temperatures (>24°C) which may assist in accelerating growth and development. We will also continue to carry out trials with artificial foods (pizzas with brine shrimp) and enhance the quality of those brine shrimp with booster diets.

We will continue to use our upwelling system but may incorporate algae into the water supply to maintain the nutritional quality of the brine shrimp and act as a possible cleansing agent in the re-circulation system.



Long term we would like to look at the effect of light on the eyes and physiology of phyllosoma, in particular the later stages which appear to be more sensitive. For example does light damage/stress the larvae?

We will also carry out a preliminary study of phyllosoma diseases and how to manage them in a recirculation system.

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## A note on sex ratios and pigmentation of eggs

*Stuart Frusher*

*Tasmanian Aquaculture and Fisheries Institute (TAFI), University of Tasmania (UTAS), Nubeena  
Crescent, Tarooma, Tasmania 7053 AUSTRALIA*

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We recently held some adult lobsters in tanks here for absolutely nothing to do with broodstock management but I think that some of the observations that were made were probably relevant to people who are fishing to hold lobsters for broodstock. I'm afraid that it was very much bucket biology in that we just tossed things in the tank to see what was happening. The reason for doing this was because we had concerns in the wild fishery. In Tasmania we have a uniform size limit, but we have substantial differences in growth rates. In the South of Tasmania females have grown so slowly they rarely make it to the legal size limit and so we essentially have a male only fishery which greatly reduces the number of males. In the remaining population, sex ratio's can be as high as six females to one male and these ratios are quite in favour of the females so we were concerned whether there was a potential for lack of reproduction in this area. The south of the state is also a wild fishery and produces the most amount of eggs for this fishery. The other reason that we were interested and we held animals was that we were concerned about what the fate of those eggs were. Were, for instance, females able to extrude and hold the eggs and therefore at sizes we'd be fooled by these animals thinking that reproduction was fine, because you could go down there and you could see all these animals carrying berry. Were they actually releasing the eggs as though they were mated and then they were just naturally shed, or were they absorbing them and then prove our results very similar to what Alistair MacDiarmid found.

The first thing that we did was set up some tanks and we had 4 females to one male and we replicated that. We also had 8 females to one male and 12 females to one male. We held them in 800 litre tanks. We were going down there weekly, just cutting up some mackerel, tossing them in to these animals, a couple of days later going and picking up what was left. We did that weekly, sometimes fortnightly, so we certainly weren't looking after these animals with tender loving care. What we did find after the first lot of experiments was that, in the tanks with 4, 8 and 12 all the females were berried. So while that solved our problems in the south coast we had no reason for concern. We were intrigued by how many females had been mated by one male. We then, in these 800 litre tanks, had 17 animals - one male, 16 females and in most of those cases there were 14, 15 and 16 females we were a little concerned that the tank size itself may have been restricting what was happening so we actually added 30 females to a single male in some four cubic litre tanks. The final result was that a single male were



able to mate with between 14 and 16 females. In the last experiment we also took the eggs from the female and compared the fecundity relationship which we had for the wild fishery and we could find no significant difference between those females that had been mated in the culture system. What we did note though, was that (there was quite a large number of animals held) every now and then we'd get an animal that had a very low crust size and in most cases this was related to the fact that this animal had damaged or regenerating limbs and we noted in the field that whenever we picked up an animal (we had a code for what we called a partial berry) whenever they had what was obviously not a full berry you always had a close look at this animal and would see that it had either lost a number of limbs or it was regenerating legs.

As it said, it was real bucket biology type of stuff, but what stunned me was not only could you hold these animals over quite a long period in time, not only could they mate and reproduce, but they were held in very poor condition so *jasus edwardsii* is a very robust animal.

The other work we did was to look at the fate of the ova and what we did here was fairly similar but in no way as detailed as Alistair did, and that was essentially holding females with and without males and again we found that what was occurring was that those animals which did not have a male became very red under the tail, the absorption of the carotenoid pigments. So when we noticed this in the field although at a very low number of animals initially, we were concerned that there may have been a disease, but after having done this work we realised that what was happening was that these animals hadn't been mated, there wasn't a male available for them. We then went on to hold these animals for 12 months to see what would happen if we then mated them in 12 months time, but unfortunately due to a system failure we lost which ones had and hadn't been mated. However, we put that aside and answered the questions we would have from a wild fisheries perspective, that a male did go into those ones 12 months later and what actually occurred was that we noticed that we had two groups of lobsters there. We had lobsters which developed eggs which were red (the red colour of the carotenoid), and we had lobsters which developed eggs which were without carotenoids.

Until Alistair's talk this morning, our assumption as to why this occurred was that we believed that this animal had been mated, had extruded its eggs but because it was being fed trash fish with low carotenoids, wasn't able to produce any and therefore didn't produce it in the eggs. This is one which probably reabsorbed the carotenoids because it hadn't been mated and therefore had the carotenoids that we thought was in the eggs. However, this is why Alistair said this doesn't seem to be the case. It appears that maybe those animals which died during that mishap we had, were maybe those ones which weren't mated so I can't really explain why, but really it does highlight why diets and carotenoids in particular are important for the calibration of the eggs. We were hoping to hatch eggs



out to see what would happen but we had another mishap about a month into this and we lost all the lobsters. We did squash some of these pale eggs and they were definitely developing, so carotenoids in diets is normally quite an expensive item but it may be that they don't need it as these animals were developing and continuing hatching. These are our observations. They certainly weren't carried out with an aquaculture reason but I believe that they do have implications for broodstock management.



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# Phyllosoma rearing of the Japanese spiny lobster, *Panulirus japonicus*, using small rearing vessels

Hirokazu Matsuda

Fisheries Research Institute of Mie, 3564-3 Hamjima, Shima, Mie 517-0404, Japan

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## *Introduction*

For the past 15 years, we have been conducting several experiments to optimise rearing conditions (eg. temperature, salinity, photo-period and food conditions) of *Panulirus japonicus* phyllosomas, using small rearing vessels. The first successful attempt to obtain a puerulus in the laboratory was achieved in 1988, and 21 pueruli were produced in the 1997-98 season. During the workshop, methods using small rearing vessels will be explained, and growth and survival of *P. japonicus* phyllosomas under these rearing systems will be shown.

## *Basic culture methods*

One hundred twenty mL glass cups are used for the rearing of the small size phyllosomas (< approx. 13 mm in body length) and 400 ml cups are used for the larger ones. Dihydrostreptomycin is added into rearing water at 10 µg per liter of rearing water to prevent the development of leptothricosis, *Leucothrix* sp. (filamentous bacteria) on the surface of the exoskeleton of phyllosoma. Phyllosomas are transferred daily into clean cups containing fresh seawater. After transferring phyllosoma to the new series of rearing vessels, Artemia and gonad of the mussel, *Mytilus galloprovincialis*, are given. The laboratory photoperiod is controlled and 12 hours of light and 12 hours of darkness are provided.

## *Growth and survival*

*P. japonicus* phyllosomas hatched out in July and August in each year (early summer to late summer) under natural environmental conditions. The newly hatched phyllosomas were approx. 1.5 mm in body length. Phyllosomas reared in the 1997-98 season reached the puerulus stage after moulting 22-29 times (av. 26.2 times, N=5). The body length in the final instar phyllosoma ranged from 28.5 to 33.1 mm (av. 30.3 mm), and the duration of the phyllosoma phase ranged from 248 to 326 days (av. 289 days).

Though survival of small and middle size phyllosoma was very stable, a high mortality of phyllosomas over 18 mm in body length was observed. The mortality was mainly observed at the time of ecdysis. The mortality was probably related to the increase in the moult increment. Moults



from phyllosoma to phyllosoma mainly occurred just before or after turning on the light, and metamorphoses from phyllosoma to puerulus were observed after turning off the light.



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## Biological barriers to aquaculture of rock lobster

*Takeshi Murai and Taku Yoshimura*

*Sekai National Fisheries Research Institute, 49 Kokubumachi, Nagasaki 850-0951, Japan*

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### *Introduction*

Although the artificial propagation of several rock lobster species has been progressing, the technique for mass production has not yet been established in any species and it takes at least half a year to produce a puerulus. Thus, wild post-larvae or juveniles have to be used as the seeds for commercial culture until these problems are solved. First of all, the effects of removing significant quantity of the wild puerulus on the native stock and the conflict with lobster fishermen should be examined to overcome the barriers. This issue is examined using yellow tail, eel and red sea bream being cultured in Japan as the examples. In order to make the aquaculture of rock lobster economically feasible for industry, more studies on the environmental conditions for intensive culture and development of an artificial feed are needed. A certain view for a growout facility is presented from the standpoints of stocking density and disease prevention. I also make some suggestions for future studies to develop an artificial feed especially on protein sources, vitamins, minerals, pigment, attractant, binder and so forth.



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# Developmental status of aqua-culture lobster fisheries in Japan

*Taku Yoshimura*

*Seikai National Fisheries Research Institute, 49 Kokubumachi, Nagasaki 850-0951, Japan*

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## *Introduction*

The Japanese spiny lobster, *Panulirus japonicus*, is one of the most valuable crustaceans and the main target of lobster fisheries in Japan, although six species of *Panulirus* distribute. Because of the great demand for lobsters in the market, over eight times the domestic catch (1300 t) has been imported in recent years from all over the world, mainly from Australia.

The Ministry of Agriculture, Forestry, and Fisheries (MAFF) has established "resources management fisheries" and "aqua-culture fisheries" (sea farming, stock enhancement and the construction of nursery grounds) as two major schemes for advancement of coastal fisheries. A large number of studies and projects have been conducted, mainly in collaboration with prefectural governments, universities, and affiliated organisations of MAFF. For the establishment of "aqua-culture fisheries" for the lobster, intensive efforts to achieve mass-production of artificial seed are continuing by the Japan Sea-Farming Association (JASFA), Fisheries Research Institute of Mie, and Science University of Tokyo. In addition, MAFF and the prefectural governments that have lobster fisheries have examined the technology and performed ecological studies for the construction of artificial nursery grounds. I present the current situation with regard to the study of spiny lobster aqua-culture and future plans in Japan based upon recent research in addition to results of ecological studies on planktonic and early benthic stages by the Seikai National Fisheries Research Institute.



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## Importance of three major factors for successful phyllosoma culture

Jiro Kittaka

*School of Fisheries Sciences, Kittasato University, Okkirai*

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Seed, water and food are three important factors in aquaculture. Because of the difficulty of larval (phyllosoma) culture of spiny lobster, seeds (pueruli) were not available in the hatchery for many years. Recent developments in aquaculture have made it possible to culture phyllosomas from hatching to metamorphosis. The species so far cultured comprise *Jasus lalandii* (1987), *Jasus* hybrid (1987), *J. edwardsii* (1990), *J. verreauxi* (1991), *Palinurus elephas* (1989), and *Palinurus japonicus* (1989). However, progress in phyllosoma culture is slow and culture of several hundreds of pueruli has not yet been achieved in any hatchery. The question remains whether mass culture of phyllosomas is possible or not? Before answering this question we have to identify both environmental and nutritional requirements of phyllosomas.

Complete phyllosoma culture was achieved by introduction of microalga *Nannochloropsis oculata* in the culture water. During the exponential growth phase of the microalgae, water quality is well maintained at non-detectable levels for ammonium-N and  $10^3$ - $10^4$  CFU/ml for the total bacterial count (Colony Forming Units on ZoeBell 2216E agar medium inoculated aerobically at 20°C for 2 days). Under such conditions, phyllosomas fed actively and showed high survival rates. COD (Chemical Oxygen Demand) increased at 0.015-0.06 mg/l/day with *N. oculata* propagation. The increase became much higher at 0.04-0.10 mg/l/day without *N. oculata*. The safety level for COD was temporarily set at 1.2 mg/l, which provided a change of culture water every 2 weeks. Phyllosomas of *J. verreauxi* metamorphosed into pueruli after 17 moults with survival rate of 12.6% for 189-273 (average 234) days (Experiment in 1990-1991).

A comparable survival rate was shown for *J. verreauxi* phyllosomas cultured with a recirculated system. The phyllosomas metamorphosed into pueruli after 17 moults with a survival rate of 5.3% for 189-295 days. *J. edwardsii* also showed good survival at 88.6% during the period from 5<sup>th</sup> instar to 14<sup>th</sup> instar over 103 days (Experiment in 1997-1998).

Culture water for 1,000 *P. japonicus* phyllosomas showed ammonium-N at <25 µg/l, COD at 1.4-1.8 mg/l, and total bacterial count at  $1-2 \times 10^0$  CFU/ml after 80 days of culture without changing culture water from the first instar to the 9<sup>th</sup> instar. Total quantity of food supplied was  $1,400 \times 10^3$  Artemia nauplii, 4,000 pieces of mussel (*Mytilus edulis*) gonad and 1,000 fish (*Pagrus major*) larvae. A 1,000 l circular tank with a conical bottom was used for the culture. The phyllosoma culture tank was



connected to a 500 l filter tank with coral sand as filtering material. The culture water in this recirculation system was characterised by a very low number of bacteria compared to those found in *N. oculata* culture water. This may imply that eutrophic bacteria are not dominant while oligotrophic bacteria may dominate in culture water with a filter system. The latter may be difficult to detect in ZoeBell 2216E culture medium (Experiment in 1998-99).

Complete culture of phyllosomas was achieved by feeding mussels. Nutritional quality of mussels is considered to be satisfactory for phyllosomas. However, better results were attained by feeding fish larvae. *J. verreauxi* phyllosomas were divided into two groups. Each group comprised 44 phyllosomas at 14-16<sup>th</sup> instars. Feeding was either with mussels and hatched larvae of sailfin sandfish (*Arctoscopus japonicus*) in combination, or with mussels only. Thirty-four and 15 phyllosomas respectively, metamorphosed into pueruli after an average 267.8 and 274.3 days respectively. Better survival rate and shorter phyllosoma period for the former group was considered to be due to feeding with fish larvae (Experiment in 1997).

Improved nutritional value of fish larvae was also shown for phyllosomas of *J. edwardsii*. One thousand five hundred 1st instars were cultured in a recirculating system and fed with Artemia nauplii and mussels. At the fifth instar, 127 phyllosomas survived. After the 5th instar, hatched larvae of red sea bream (*Pagrus major*) and goldstriped amberjack (*Seriola lalandi*) were occasionally fed. Tidepool gunnel (*Pholis nebulosa*) were fed for 33 days beginning at 9<sup>th</sup> instar to 12<sup>th</sup> instar. The survival rate was 96% and inter-moult period from 9<sup>th</sup> instar to 12<sup>th</sup> instar was 10 days on average. Fish larvae were not fed after 12<sup>th</sup> instar. Number surviving at instars 13, 14, 15, 16, and 17 were 115, 110, 106, 86 and 66 respectively, with inter-moult period of 23, 33, 30, and 31 days respectively. It is apparent that both survival and growth were poorer without feeding fish larvae (Experiment in 1998-1999).

Lipid analyses for larvae of *A. japonicus* showed high lipid content, particularly HUFA. EPA and DHA content were 38300 and 37000 ng/mg dry weight respectively. Artemia nauplii are a common food used for larval rearing. EPA and DHA content of Artemia nauplii are 4700 and 76 ng/mg dry weight respectively (S. Teshima, Ishikawa and Kittaka unpubl.).

Compared to the difficulty of larval rearing, breeding of rock lobster is well controlled in laboratory. Broodstock of *J. edwardsii* and *J. verreauxi* were shipped from New Zealand to Japan. They have survived for many years and hatch phyllosomas once a year in the same season. Breeding experiments in the early 90's showed spawning in October-April and hatching in December -June for *J. edwardsii*, and moulting in December-January, spawning in April-May, and hatching in June - August for *J.*



*verreauxi*. After 1995, the experimental site was moved to Nemuro, Hokkaido, the most north eastern part of Japan. Broodstock were also moved to the new laboratory. At the beginning, *J. edwardsii* comprised 1-3 years old animals and *J. verreauxi* reached maturity after 5 years. The latter had spawned in 1990.

In Nemuro, *J. edwardsii* spawned in November-December and hatched in February-April, and *J. verreauxi* moulted in January, spawned in April and hatched in July-August. The reproductive cycle shown in moulting, spawning and hatching seems to have become later, and these periods have become shorter. This may be due to the lower water temperature in Nemuro compared to Sanriku. The following facts have been found in the breeding experiments:

1. *J. edwardsii* becomes mature 2-3 years after metamorphosis in the laboratory fed mussels exclusively.
2. Matured females of *J. verreauxi* seem to be induced to moult several days after moulting of males.
3. Juveniles of hatchery reared *J. verreauxi* (metamorphosis in 1991) grew to 100-112 and 111-119 mm in carapace length at 6 years old in 1997, fed mussels exclusively. All individuals have survived in 1999 at 8 years old. It will be interesting to find when they will carry eggs.

#### *Future research*

Feasibility of spiny lobster aquaculture was proposed due to their lower rates of cannibalism in dense communal culture compared to the heavy cannibalism shown by the American lobster (*Homarus americanus*) (Table 2; Kittaka, 1990). Both environmental and nutritional requirements of phyllosoma's have been almost resolved. However, some problems have remained in providing a suitable composition of microflora and effective artificial food. The following experiments will be required to clarify these problems in phyllosoma culture.

1. Investigation of microflora and water quality in the culture water with a filter system in comparison with *N. oculata* culture.
2. Improvement of low survival rate for late-stage *J. edwardsii* phyllosoma's by feeding fish larvae.
3. Ecosystem culture methods for early-stage phyllosoma's.
4. Phyllosoma culture experiments for *Palinurus japonicus* and *Palinurus elephas*.

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## Propagation of southern rock lobster (*Jasus edwardsii*) in Tasmania

Arthur J. Ritar

*Tasmanian Aquaculture and Fisheries Institute (TAFI), University of Tasmania (UTAS), Nubeena  
Crescent, Taroona, Tasmania 7053 AUSTRALIA*

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Preliminary investigations were undertaken in 1997 into the propagation of lobsters followed by more detailed studies in 1998. In addition to the information presented below, M. Bermudes is conducting PhD studies on environmental parameters and digestive physiology on early stages of phyllosoma.

### **1997 investigations**

For ovigerous females caught in August (egg extrusion in May) and held in cool water (9-10°C), hatching of phyllosoma larvae was delayed to late November compared with females held at ambient temperature which hatched in late October. Two experiments were conducted on larval culture.

#### *Experiment 1: density of Artemia in culture vessels*

*Artemia* nauplii enriched with DC Selco were fed daily to phyllosoma at rates of 3, 9 or 27.ml<sup>-1</sup>. Larvae were held in static water with daily water exchange. The highest density of *Artemia* appeared optimal for larval growth and survival. Larvae became heavily fouled with filamentous bacteria (predominantly *Leucothrix mucor*) probably causing suffocation and did not survive beyond stage 3 of development.

#### *Experiment 2: water and light conditions*

In this 2 x 2 factorial experiment, water conditions (clear water filtered to 0.2 µm or water containing the alga *Tetraselmis suecica* at 100,000 cells.ml<sup>-1</sup>) and light conditions (complete darkness or a photoperiod of 16 h light: 8 h dark) were examined. Larvae were fed at 27 *Artemia*.ml<sup>-1</sup>. In the best combination of treatments, larvae survived to stage 6 (moult at Day 75). Inclusion of algae in the water resulted in better survival and growth than in clear water. Survival and growth was also better for larvae held in the photoperiod. Poor water quality (high ammonia, low dissolved oxygen) again appeared to be the overriding factor causing mortalities.

### **1998 investigations**

#### *Broodstock management*

#### *Experiment 1: effect of phototherm regime from before moult to larval hatch*



Male and female adults captured from the east coast or from Maatsuyker Island were held from late March (shortly before moult) either under ambient conditions or a compressed cycle to accelerate the change in water and light conditions. The mass of hatching larvae from each female was recorded daily and larval samples were stored frozen for subsequent analysis of biological parameters and fatty acids. Haemolymph samples were collected before and close to moult and again after hatch for later analysis of reproductive hormones.

Animals in the compressed cycle experienced cool water temperatures earlier and so moult was delayed by two weeks compared with animals in ambient conditions (8 June & 24 May, respectively). Mating was not monitored but egg extrusion occurred about three weeks after the moult. Animals in the compressed cycle experienced warm water conditions earlier and so larval hatch occurred about seven weeks earlier (and over a shorter period) than animals in ambient conditions (11 September to 10 October compared with 24 October to 15 December, respectively). It is therefore clear that the entire reproductive cycle can be controlled by photothermal manipulation. However, the effect on larval quality (size, composition) will be assessed shortly.

#### *Experiment 2: female broodstock held at different temperatures*

Ovigerous females (extruded eggs in May) captured from the east coast in early June were held at ambient temperature, 18°C or 10-11°C until hatch. Other ovigerous females were captured from the same area in early September and maintained at ambient temperature (external controls). The mass of hatching larvae from each female was recorded daily and larval samples were stored frozen for subsequent analysis of biological parameters and fatty acids.

Time to larval hatch occurred earlier for females held at 18°C (7 Aug to 25 Aug) than the external controls (24 Sept to 30 Nov), females at ambient temperature (6 Oct to 6 Nov), or females at 10-11°C (6 Nov to 18 Dec). Embryonic development is therefore accelerated or retarded by holding broodstock in warm or cool water, respectively. It is curious that females caught in September and held at ambient temperature have a much wider window of hatching than those held at ambient from June. The effect of these treatments on larval quality (size, composition) will be assessed shortly.

#### *Larval culture*

To overcome the problems of poor water quality when culturing larvae in the static water system, a flow-through system was adopted in 1998. This entailed sea water passing through successive filtration to 1 µm, then entering a header tank where the temperature was controlled to 18°C before ultraviolet disinfection and then into culture tubs. A series of jets close to the base at the perimeter and near the centre of the tubs allowed circular flow of water. Water exited the tubs through a screen



(to prevent the loss of feed) on a drain fitted to the side of tubs. This allowed water turnover of 3-4 times each hour and so water quality remained consistently high with no obvious problems of microbial infection on the bodies of larvae. Two experiments were conducted on larval culture in 1998.

#### *Experiment 1: feed types*

Newly-hatched larvae were fed either ongrown *Artemia* (1.5 mm) enriched with *T. Isochrysis* at a rate of 4.ml<sup>-1</sup>, chopped flesh (about 700-900 pieces of 0.5-1.0 mm) of mussel (gonad), oyster (gonad) or clam (whole). Very few or no larvae progressed to stage 2 when fed clam or oyster. Larvae progressed to stage 2 most quickly and were largest when fed *Artemia*.

#### *Experiment 2: Artemia density*

In Phase 1 of this experiment, newly-hatched larvae (replicated for three females) were fed daily on ongrown *Artemia* (1.5 mm) enriched with *T. Isochrysis* at a rate of 1.5, 3 or 6.ml<sup>-1</sup> until stage 3 when the experiment was terminated. In Phase 2, the stage 3 larvae from Phase 1 were re-randomised (within female of origin) and each tub was fed 400 pieces of chopped mussel gonad and enriched ongrown *Artemia* (1.5 mm) at a rate of 1.5, 3 or 6.ml<sup>-1</sup>. At the time of writing, the larvae in Phase 2 are at stage 4-5. There were no differences between treatments in the time to moult to subsequent stages or in the size of larvae. However, the larvae from one female were significantly (10-15%) larger than larvae from the other two females at each stage of development. This indicates that the quality of newly-hatched larvae (including their size) is important and suggests that broodstock management is an area worthy of investigation

### **Conclusions and future directions**

#### *Larval culture*

Cultured larvae in 1998 in the flow-through system at 18°C first reached stages 2, 3, 4, 5, 6 and 7 at Days 9, 19, 30, 40, 57 and 73, respectively, but later stages have not yet been examined. This is earlier than larvae at the same stage of development (to stage 6) in the previous year. Larvae were also larger than at the same stage in 1997. These effects are mainly attributable to the better water quality of the flow-through than the static water system but also to the better distribution of larvae and food within the culture system. Nevertheless, the hydrodynamics within the culture system need to be improved to reduce larval mortality and to achieve even better mixing of the entire contents. The system can be scaled down for smaller experimental units or scaled up for mass rearing of large numbers of larvae. Even so, systems employed by researchers in Japan and New Zealand still need to be examined thoroughly.



Future investigations on larval culture should examine feeds (size and density of *Artemia*, enrichments for *Artemia*, shellfish) in more detail including analyses of feeds and larvae after treatment, as well as water quality (ammonia, dissolved oxygen, pH, salinity). An understanding of the digestive physiology, feeding behaviour, photosensory behaviour and respiration will assist in developing suitable culture regimes. It is important to understand the relationship between biological parameters of larval quality and their biochemical markers.

#### *Broodstock management*

It is possible to control the entire reproductive cycle by manipulation of the water temperature and photoperiod. This allows out-of-season production of larvae for hatchery culture experiments. Although the larvae produced out-of-season appear vigorous, analyses of the stored samples needs to be undertaken to determine the comparative physiology of larvae produced at different times of the year and under different phototherm regimes. However, the most important environmental condition is probably nutrition of the broodstock especially in the months approaching mating. Investigations should be undertaken to understand and maximise gamete development over this period. Studies on nutrition will likely yield valuable gains in larval quality to improve the success of culture. A rapid test (such as a stress test incorporating extreme salinity or temperature or a test for swimming ability) should be developed to assess the viability of larvae before they are cultured.



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# Effect of photoperiod on survival, growth, feeding and cannibalism in early developmental stages of *Jasus edwardsii* phyllosoma larvae

*Michel Bermudes*

*TAFI, UTAS, Nubeena Crescent, Taroona 7053 Tas. Australia.*

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Understanding the effect of environmental factors on spiny lobster larvae is essential to define culture conditions and to set experimental baselines for subsequent work on tank design, nutrition and genetics.

Newly hatched *Jasus edwardsii* larvae were reared to the fourth instar under five photoperiods: 0, 6, 12, 18 and 24 hours of light per day. In each treatment, 5 groups of 21 larvae (7 from each of 3 different females) were cultured in shallow plastic bowls with 200ml of sea water and 25ppm of oxytetracycline. Larvae were transferred to clean bowls and water with fresh oxytetracycline every second day. Larvae were fed daily to satiation on newly hatched artemia nauplii enriched for 24 hours with DC Super Selco and *T. Isochrysis galbana* alternately. Illumination was provided by a 20 watt quartz halogen light reflected on a white ceiling above the culture vessels. Slight temperature fluctuations ( $18 \pm 0.5$  °C) were expected between treatments and temperatures were recorded hourly with data loggers. Survival, moult increment, intermoult period (first calculated in degree-day due to above-mentioned temperature differences and transformed back into days at 18°C for interpretation) and feed intake at mid-instar were recorded together with cannibalism of newly moulted larvae which was observed from the second instar onward.

Survival significantly declined from  $97.9 \pm 0.5\%$  (overall mean  $\pm$ SE) at stage 2 to  $83.6 \pm 2.0\%$  at stage 4 ( $P < 0.0001$ ) with no effect of photoperiod at any one stage ( $P = 0.325$ ). Cannibalism (mean % of mortality  $\pm$ SD) accounted for  $13.3 \pm 16.3\%$  to  $74.3 \pm 18.3\%$  of the total mortality observed from stage 2 under 12L and 24L treatments respectively, but was not related to photoperiod ( $P = 0.733$ ). There was an effect of photoperiod on intermoult duration ( $P < 0.01$ ) which was not consistent across stages of development ( $P < 0.001$ ). Indeed, photoperiod did not effect intermoult duration (mean days  $\pm$ SE) in stage 1 larvae ( $P = 0.1471$ ; overall mean =  $12.61 \pm 0.05$ ) which by stage 2 took longer to develop with increasing dark phase ( $P < 0.01$ ; 24L:  $12.84 \pm 0.08$ , 0L:  $13.68 \pm 0.22$ ). A change in response to photoperiod was again observed at stage 3 with larvae under 0L and 24L requiring a longer period to moult to the fourth stage ( $15.01 \pm 0.42$  and  $15.32 \pm 0.29$  respectively) than larvae reared under 6L and 12L ( $14.00 \pm 0.23$  and  $14.14 \pm 0.23$  respectively;  $P < 0.05$ ). The response in body length growth (in % body length increment) was also influenced by photoperiod but not until stage 3 when larvae reared



under 24L grew significantly less than larvae under 6, 12 and 18 hours light treatments ( $P < 0.05$ ). The ability of the larvae to feed was markedly affected by the length of the photoperiod. Feeding activity (mean artemia nauplii/larva/day  $\pm$ SD) of stage 1 phyllosoma increased with longer light phase ( $P < 0.001$ ; 0L: 11.68  $\pm$ 5.06, 24L: 20.80  $\pm$ 2.76). Although not as significant, a similar trend was observed at stage 2 ( $P < 0.05$ ; 0L: 22.33  $\pm$ 6.10, 24L: 28.54  $\pm$ 6.62) and by stage 3 the effect of photoperiod on feeding activity was no longer appreciable ( $P = 0.5403$ ).

Although photoperiod did not affect survival, there was a marked effect on growth. Importantly, the response to increasing photoperiod changed with ontogenetic development. This implies that the adjustment of photoperiod to the requirements of each early stage of development would maximise growth.



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## Closed cycle breeding of crustacea with special reference to rock lobsters

*Mike Hall*

*Australian Institute of Marine Science (AIMS), PMB No. 3, Townsville Mail Centre, Qld 4810*

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Lobsters are divided into two infra-orders, the clawed lobsters of the Astacidea and the spiny lobsters of the Palinura. They represent the most valuable crustacean commodities. On average, lobsters are 65 – 70% more valuable per unit weight than either crabs or prawns. As with other fishery sectors, the yield from wild harvest cannot completely satisfy market demand and there is growing interest in increasing harvests by culturing selected species.

One of the most critical points for successful breeding of crustacea is the spawning of broodstock and larval rearing. Farm prawn culture now accounts for approximately 30% of total world prawn production. In contrast, neither crabs nor lobsters are cultured commercially to any significant extent. The reason for this is due to breakthroughs in larval rearing technologies that were first successful in penaeid prawns. The challenge to aquaculturists for the culturing of lobsters is success in rearing through the extended larval phase. In the Astacidea lobsters have a post-maternal larval phase which lasts 1-2 weeks. In contrast the Australian *Panulirus* and *Jasus* species the larval phase lasts from 4 to 23 months.

The collection of *Palinura puerili* from the wild for captive grow-out would largely remove the lengthy and costly larval rearing component. However, the key to the complete development of any farming sector is the closed life cycle breeding of the selected species. It will require significant technological breakthroughs for the closed life cycle breeding of the *Palinura* to become a practical and economic reality. One potential breakthrough would be truncating the length of the larval phase. The various transformations through the larval stages are under physiological and endocrinological control. All the major endocrine organs, including the X-organ sinus gland complex in the eye stalk, the Y-organ and the mandibular organ, are present in the first postembryonic instar of decapods. The sinus glands of lobsters have a similar hormonal profile to that found in penaeid prawns, including the crustacean hyperglycaemic, molt inhibiting and vitellogenesis inhibiting hormones, or CHH/MIH/VIH family. The Y-organ is the source of ecdysteroids and the mandibular organ of methyl farnesoate. These hormones are likely to modulate metamorphosis. In Astacidea lobsters episodic pulses of ecdysones are associated with larval transformations. Steroids, retinoids and thyroid hormone are known to function as critical signals in invertebrate metamorphosis. Various ecdysone reactive nuclear receptor superfamilies, such as EcR, USP, DHR78, DHR38 and BR-C in *Drosophila*, play



central roles in directing the process of metamorphosis. In addition various biogenic amines also modulate larval metamorphosis in marine invertebrates. Finally a number of chemical clues have been shown to be critical for the transition from the final larval phase to juvenile metamorphosis in marine invertebrates. Since larval transformations in other invertebrates are regulated by hormonal inputs the possibility may exist to dramatically shorten the larval phase of *Palinura* lobsters by means of chemical or molecular intervention. If the larval phase can be significantly shortened then closed life cycle breeding of the *Palinura* lobsters may become a reality. Hormonal manipulation will be required to achieve this goal.



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## Investigation of the gut system: Application for lobster aquaculture

*Satoshi Mikami*

*Australian Fresh Corporation, QDPI Bribie Island Aquaculture Research Centre*

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Diet is an important factor affecting survival of phyllosomas, and one of the key issues in successful lobster aquaculture. Observation of the functional morphology of the digestive system can indicate the suitability of particular food types and contribute to better understanding of the requirements for larval and growout diet. Also, knowledge derived from the study on the digestive system can be used as an indication of whether developed diets are properly utilised by phyllosomas. The functional morphology of the gut system of phyllosomas was examined by light and electron microscopy, and possible flow pathways of food particles were deduced for the structure of the phyllosoma gut system. The morphology of the inner mouthparts of phyllosomas differs from adult lobsters. The paired paragnaths and labrum in phyllosomas are well developed, forming a semi-enclosed chamber. They control the amount of material ingested into the semi-enclosed chamber, where mastication by the mandible occurs. The proventriculus (stomach) is a straight tube having no functional gastric mill or cardio-pyloric valve, but the spines and filter-press (except in newly hatched phyllosomas) are well developed. The structure of both the mouthparts and proventriculus suggests that phyllosomas can only ingest soft-bodied food particles with a high water content. Food particles filtered through the filter-press of the proventriculus, which are mainly in liquid form, enter the midgut gland. From ultrastructural observation of the midgut gland, the final phase of absorption appears to be intracellular, occurring mainly within the R-cells (resorption cells), which are the most abundant cell type in the walls of the midgut gland. Nutrients of food particles are selectively (actively) transported from the lumen of the midgut gland, across the apical cell membrane, to enter the R-cells where they are stored in vacuoles and metabolised intra-cellularly. Food particles rejected by the filter-press enter the lumen of the midgut directly. The lumen of the midgut is relatively short, and food particles are moved toward the hindgut by vermiculation of the midgut, and excluded as lipid rich faeces via the anus.



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## Summary of Research – Rock Lobster Digestive Systems/Physiology

*Danielle Johnston*

*UTAS, PO Box 1214, Launceston, Tas, 7250*

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### *Introduction*

Declining broodstock numbers (as indicated by several seasons of low puerulus settlement) and the ever increasing global demand for rock lobsters has generated considerable interest in their culture, particularly in Western Australia (*P. cygnus*) and Tasmania (*J. edwardsii*). Previous attempts to culture rock lobsters have been hampered by the extended phyllosoma larval phase (up to 1 year) and the fact that pueruli do not feed during their long-distance swim to shore (exhibit “secondary lecithotrophy”). For this reason, the post settlement post-pueruli phase is the most suitable for culture at the present time. This is reflected by efforts to devise cost-effective large-scale methods to capture/harvest pueruli, which is being funded by FRDC (principal investigator Bruce Phillips). However, despite the targeting of post-pueruli and their importance for the success of rock lobster culture, very little is known of their biology. In particular, knowledge of their diet, digestive capabilities and nutritional requirements are essential for the successful culture of rock lobsters. This information is also critical for efforts to close the life cycle, which has been prioritised by industry as one of the key areas for research over the next 5 years by the CRC for Aquaculture.

The following areas of research are proposed based on the current lack of knowledge on larval nutrition and its importance in the success of rock lobster culture. They would ideally suit collaboration with nutritionists and dietary formulation experts.

### *Digestive Enzymes and Alimentary Tract Structure*

The occurrence and concentration of digestive enzymes within the gut is important for determining which dietary components can be utilised for metabolic purposes (Glass and Stark, 1995). The diet and digestive enzyme complement of the adult rock lobster *Jasus lalandaii* has been studied by Barkai *et al.* (1996). They reported that *J. lalandaii* are primarily carnivorous, preferring mussels and a large variety of marine arthropods which was reflected in their high concentrations of proteases and chitinases. They are however opportunistic and can ingest large quantities of sponges which are digested by strong gelatinases. Similar work is needed on pre and post settlement pueruli and juveniles as dietary preferences and digestive abilities form the basis for formulating artificial diets which are necessary for commercial operations. Combined with documentation of changes in mouthpart and gut structure between the larval and juvenile phases (Nishida *et al.*, 1990), research on



digestive enzyme complements in relation to diet will provide a comprehensive understanding of larval nutrition for commercial enterprises. Once established, growth studies with diets specific for puerulus and juveniles would be trialed to ensure rates are optimal. This research will complement studies undertaken by Kevin Williams on an FRDC funded project to develop an artificial diet for juvenile and adult western, southern and ornate rock lobsters. Hence this research would be value adding to already existing FRDC projects and will address critical aspects of larval biology which are necessary to the successful culture of puerulus and juveniles stages and hence closure of the life-cycle.

#### *Digestive Gland Cytology/Energetics*

Studies on structural changes of the digestive glands of *J. edwardsii* during development from puerulus to post-puerulus revealed that lipid inclusions increase in R-cells, whilst fat bodies in the body decrease (Nishida et al., 1995). Fat bodies provide the source of energy for the long distance swim by the puerulus and subsequent developmental changes after settlement (Takahashi et al., 1994), whereas lipid inclusions are most likely used for energy by the post-puerulus. Aside from work done by Lemmens (1994a, b) on the changes in energy reserves and metabolic rate during the transition from *P. cygnus* phyllosoma larva to puerulus to juveniles, very little work has been done of the energetic needs and nutritional dynamics of other rock lobster species. Lemmens (1994a, b) determined the energetic demands and metabolic rate of post-settlement pueruli were low compared with pre-moult pueruli and juveniles and proposed it provides the puerulus with a means of extending the duration of the non-feeding stage and increase the chance for survival beyond metamorphosis to the first feeding stage. Further investigation on the relationship between energetics and nutritional requirements of post-puerulus, juveniles and adult rock lobsters is needed. In particular, the shift in importance from hepatopancreas lipid reserves to external nutrition for energetic requirements during the transition from post-puerulus to juveniles and adults is important for optimising growth in these 3 stages. Furthermore hepatopancreas energy reserves and moisture content of *P. cygnus* and *J. edwardsii* can be indirectly used as a condition factor to roughly determine their overall health during culture. Both these aspects are valuable for ensuring the success of rock lobsters as aquaculture contenders.

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## Present and future directions in Western Australian rock lobster propagation

*Bruce Phillips*

*Curtin University of Technology, GPO Box U1987, Perth, 6001*

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### *Broodstock Management*

No studies are currently in progress. In the 1970's CSIRO showed that it was relatively easy to bring *P. cygnus* into reproductive condition and then to have breeding females produce eggs throughout the year. *P. cygnus* was also raised from pueruli to reproductive maturity in the laboratory.

### *Larval Rearing*

Several attempts at larval rearing have been made by students. The most recent was a PhD student at the university of WA about 5 years ago. He achieved only partial success. However, his principle objective was not the completion of the cycle and rather to study the larvae.

A application for propagation of *P. cygnus* is currently being prepared. It will seek partial funding from the Aquaculture Development Fund in WA, and also FRDC funds through the FRDC Workshop.



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## Rock lobster research at the Australian Institute of Marine Science

*John Benzie*

*AIMS, PMB No. 3, Townsville Mail Centre, Qld 4810*

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The Australian Institute of Marine Science is particularly interested in research on closing the life cycle of lobsters using physiological and molecular approaches to manipulate reproduction and larval development. The Institute is also interested in general aspects of larval rearing. These areas are also those in which the Institute has developed strengths and a proven record of research in penaeid prawns. Research on some aspects of hormonal regulation of physiology of rock lobsters has already been undertaken at AIMS.

### *Research Issues & Directions*

The key bottleneck for lobster aquaculture is the hatchery-nursery phase. Depending on the species this phase lasts 4 to 23 months. Successful propagation will depend on a variety of factors relating to egg quality, larval husbandry and disease. One solution to overcoming the difficulties of rearing delicate larvae will be:

#### **Shortening the larval rearing phase:**

1. this is critical for the economic viability of aquaculture rearing
2. important for the reduction of larval mortality

Various approaches might be used including:

- **Use of model species with short larval phase: e.g. *Panulirus ornatus*, 4-10 month larval phase**
- **Investigation of gene and hormonal controls on moult transformations of the phyllosoma stage**
- **Identify the 'signal' that induces puerulus phase to settle out of the plankton**

A necessary part of this research undertaken in parallel is to:

**Develop rearing technology for mass-rearing of delicate planktonic larvae.**



AIMS is experienced in these research areas relevant to the development of the culturing of rock lobster and keen to apply these skills to rock lobster species. AIMS has excellent larval rearing facilities for tropical lobster, but is keen to work on several species in collaboration with other labs.

*Research experience relevant to the proposed rock lobster work are:*

#### **Reproductive and general physiology of crustacea**

- Optimisation of environmental conditions for successful spawning improvements in broodstock diets.
- The characterisation of egg quality.
- The development of specific spawning induction technologies.
- Research into biogenic amines and free amino acids in rock lobster.

#### **Molecular genetics of crustacean reproduction and development**

- The isolation of reproductive genes from prawns.
- The identification of hormones from individual endocrine tissues.
- Investigations into the molecular biology of larval development.

#### **Selective breeding**

- The development of closed-cycle breeding.
- The production of captive reared broodstock for selective breeding.

#### **Husbandry skills**

- Supply of good quality seawater.
- Mass rearing of crustacean larvae.
- Capability for long-term holding of broodstock.



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## The 1999 Japan/Australia rock lobster workshop in Perth funded by DISR

*John Benzie & Taku Yoshimura*

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This workshop arose out of suggestions made to the 8<sup>th</sup> meeting of the Australia and Japan joint science and technology consultative committee in 1997 and their wish to encourage proposals for meetings on marine biotechnology and mariculture. That initiative led to the development of a workshop on rock lobster aquaculture and was co-ordinated by myself and Mr. Taku Yoshimura from the Sekai National Fisheries Research Institute of Japan.

The objective of that workshop was to bring together scientists from Australia and Japan with expertise in rock lobster biology in order to identify areas in which there was a mutual interest by Japanese and Australian scientists in undertaking joint research. The workshop was attended by 5 scientists from Japan and 6 in Australia. We had Mr. Shima and Mr. Sekine from the Manami Isu branch of the Japan Sea Farming Association. Unfortunately they could not join us here. We also had Mr. Matsuda and Dr. Murai who talked to you this morning, and Mr. Yoshimura. The others you probably know from Australia, myself, Mike Hall, Bruce Phillips, Kevin Williams, Piers Hart and Satoshi Mikame. So all of these people bar two have come across to this meeting in Hobart.

The main focus of the discussions was propagation and larval development and culture. This included general discussions on rearing methods, diet, disease and hormonal control development, but the biology of the larvae in the wild and puerulus collection was also discussed given their importance to developing aquaculture techniques. Key aspects of the biology of larval, post-larval and juvenile biology of rock lobsters such as aspects of diet, disease, the closed life-cycle rearing environments and particularly the length of the larval phase were identified as major impediments to the development of rock lobster aquaculture. Importantly, it was identified that the same problems exist in Japan as Australia and that the solutions to these problems could be achieved more quickly through information sharing and joint research. It wasn't possible to identify specific proposals at the workshop - it is very early days, however it was agreed that greater scientific exchange and joint research would advance the development of rock lobster aquaculture in both countries.

Given that, the workshop will make three recommendations to Governments in each country:

- that greater scientific exchange be developed between Australia and Japan with a view to developing joint research projects in the key areas identified in the report in order to speed development of rock lobster aquaculture and sea farming in both countries. Now this is a very early start to this process, and we will need to undertake a number of meetings and exchanges to develop particular proposals.



- that a second workshop on rock lobster aquaculture be held, preferably in Japan in 2-3 years time in order to advance the development of further joint work.
- that support be provided as a priority for joint research proposals or proposals for scientific exchange between Australia and Japan through established mechanisms eg. biological agreements between Australia and Japan, and also if possible by additional means for rock lobster aquaculture over the next few years and we hope that this will lead to more exchange, more discussion and as trust develops, hopeful particular proposals can develop rock lobster aquaculture in both countries.

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# The biological feasibility of rock lobster propagation from egg to puerulus

*Lennard. J. Tong*

*National Institute of Water and Atmospheric Research, Wellington, N. Z.*

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## *Abstract.*

There are six key areas in which research will be required before larval rearing and pueruli production can be considered to be routine.

- Controlling and maintaining consistently high egg and stage I larval fitness and quality.
- Nutrition and the importance of key components (eg fatty acids) in the diet through all phyllosoma stages.
- The development of a suitable larval artificial diet with appropriate nutritional and physical properties.
- Measuring the effects of light and temperature (eg stress) on the feeding physiology, growth and survival.
- Determining the relationship between feeding behaviour and the rearing system especially stage VIII plus.
- Identifying, monitoring and controlling diseases of phyllosoma.



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## Rock lobster propagation research – an aquaculture perspective

*P Jungalwalla*

*TASSAL Pty., Ltd.*

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Lobsters have long been identified as valuable in seafood markets. As with other wild-fisheries, the gap between supply and demand seems to be increasing, and markets are becoming more sophisticated in the demands they make upon seafood suppliers.

One of the responses has been a growing interest from elements of the wild fishery sector in on-growing some of the catch taken under quota. The primary goal has been to add value to the wild catch, in terms of either size, condition, or timing of delivery, before taking it to market. More recently, interest has widened to include the capture and on-growing of juveniles, down to puerulus stage.

Spurred perhaps by field trials undertaken by industry, research projects on nutrition, growth and health of lobsters have been undertaken by various research agencies. Both wild fishery and aquaculture sectors of industry have endorsed this research because it is likely to increase commercial opportunity.

These developments duplicate the transition of other species from wild-catch to aquaculture (also from hunting to farming). The goals, and indeed the strengths of aquaculture are higher survival and growth rates as compared to the wild, together with the ability to smooth out annual and seasonal fluctuations of product supply. The logical aim, as for any species farmed, is a closed life cycle, giving some control over all stages of production. In most examples of successful aquaculture, control over propagation of the species has been a key requisite for sustained growth into a significant aquaculture industry (eg. oysters, salmonids, prawns)

As proficiency in on-growing wild caught lobsters increases, it is likely to create a viable niche for small to medium enterprises adding value to wild caught resource. These would provide excellent training ground for lobster husbandry, and could become important contributors to regional economies. However, the full realisation of lobster aquaculture as an industry will be constrained if seed stock cannot be reliably and economically generated. Given that the life cycle of the lobster species of commercial interest to us is “barely” closed, propagation research becomes a key investment if the strategic goal is the establishment of a lobster aquaculture industry.

From an aquaculture perspective there is some comfort from information gathered to date showing that lobsters are largely amenable to culture, and that significant improvements in growth and survival



are likely to be achieved by research. What is daunting is that despite significant international effort there is poor control over spawning and larval rearing, and that the larval stage is complex and in some cases extraordinarily long. It is noted that prawn farming was greatly boosted when research provided technological breakthroughs in spawning and larval rearing. Similar breakthroughs in reliability of larval survival or shortening of the larval stages for lobsters would undoubtedly be a watershed in the industry's eagerness to invest in lobster aquaculture.

Whilst established aquaculture industry sectors (eg. salmon, tuna, oysters, & prawns) do fund sector specific research, it is uncommon for industry to directly fund research for new species development. It is considered the role of governments (national and regional) to fund and to carry out at least the preliminary research aimed at developing new industry. Industry generally needs to see at least the potential for aquaculture of new species before investing to any extent.

It is suggested that both the established aquaculture industry as well as new investors would endorse research targeted at the propagation of selected species of lobster, and would monitor the outcomes with interest.

### *Discussion*

To define whether funding should be directed towards propagation or puerulus collection you need to define "who wants what to happen". If the wild fishery sector in general felt that its future lay in diversifying from pure hunting to also include farming, then they would support puerulus on-growing. For the aquaculture sector, the interest and the expertise is in farming animals in the water, and producing and marketing seafood. Don't forget, there is a big difference in attitude. The wild fishery traditionally goes to the market and says "I've got a bucket of dead fish, who's going to buy them from me?" Conversely the aquaculturist's attitude in the market place is "Tell me what you want, and when, and I'll get it there for you." If the two sectors collaborate, then there is merit in putting more effort into puerulus collection and on-growing. If, however, wild fisheries aren't interested except for enhancement, and want to monopolise the wild resource, there is a case for saying that aquaculture should back away from gathering any more puerulus other than for research and put full investment into propagation. It would take longer to set up an industry, but it would effectively bypass the wild resource issue.



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## The Cooperative Research Centre (CRC) for Aquaculture

*Dr. Peter Montague, Executive Director*

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The Aquaculture CRC is essentially a research provider to the aquaculture industry. It is a network linking research institutes and industries together. The aims of the network are to deliver new technology through research by skilled people for the benefit of Australian Aquaculture. It brings an investment of \$2.4 M/year from the Commonwealth Government as well as contributions from industry & research institutions of both money and collaborative services/activities. It also receives grants from FRDC & other sources. In return it has obligations including the delivery of quality research, innovative forms of education, technology transfer into commercial practice, commercialisation of intellectual property for the benefit of the Australian economy. It also has a role to mediate or become involved in international collaborations within aquaculture. The quality of the research to date has received very high commendations and the PhD training has been highly commended.

The CRC research does not presently include any activity in rock lobster. The funding life of the CRC ends in Sept. 2000, but the CRC has filed an application for a further 7 years funding from Canberra. Seeking \$3M from the Commonwealth Government for the first 5 years, tapering off to 0 by 2006. The idea is to look towards building up sources of income from commercialisation, industry support etc. Most of the program focus is on major established industries within Australia. The propagation of rock lobster is considered worthwhile & included in the budget with funding expected of around \$100,000. Tasks & funds are based on the most effective method to get results with a focus on collaboration and activities.



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## Summary and overview

*Robert van Barneveld*

*Leader, Rock Lobster Enhancement and Aquaculture Subprogram*

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This workshop was convened to address a number of fundamental questions:

1. Is it practically, biologically and economically feasible to close the lifecycle of rock lobster and propagate them in an aquaculture system?
2. Do we have the capacity to propagate rock lobster from egg to puerulus? If yes, is it practical and feasible?

As a result of discussions conducted as part of this workshop, a number of important points have been raised, including:

- What proportion (given larval stages are seen as a bottle neck) of production costs can I afford to devote to the larval production phase? While we might be in a position to answer, it is not a simple question. It is likely we require a business plan for further development.
- If you look at the entire production system, we know how long it takes to grow to a particular phyllosoma stage, but do we know how much time can be apportioned to larval development. It may be possible to improve the growth rate of juveniles to a marketable size so that we have more flexibility in the larval stages. This is quite a complex question but we need to devote some time to it.
- Do aquaculture systems need to be developed for every species in the first instance? Is that going to result in a lot of overlap in the research that we're doing? Can we select a target species, work to closing the system of that species and then apply what we know to other species, and deal with specific problems then? At least that approach will provide a base on which to work.
- From a commercial production point of view, if it is feasible to grow a non-native species in a particular area, some benefits may be that you can easily distinguish aquaculture output from wild output and you overcome, say the problem of people collecting small/undersized rock lobsters from the wild. In this instance, however you might face the problem of having a non-native species introduced into the wild.



The research presented to date can be summarised as follows with the following apparent gaps in our knowledge base:

*Pre-broodstock:* There have been suggestions that the history of a lobster can have a big influence on its fecundity and its ability to reproduce. What is the availability of the rock lobsters for use in aquaculture? It would seem that if aquaculture licenses are granted that is not going to be too much of a difficulty to address this issue.

*Broodstock:* While it has been suggested further research is required, compared to the larval stages this section appeared to be better understood. Given that we know condition etc. can be influencing production of broodstock, we might be in a position or closer to a point of utilising this information commercially. Even from generic observations of sex ratios, management conditions etc., this is useful take-home information.

*Larval stages:* This area is complex. While some areas are well-defined (eg. Len Tong gave a good biological limitations overview, with mortality the biggest problem largely due to the design of the experimental systems). Nutrition and environment were discussed separately but should be considered as an interactive system. Nutrition involves intake and if those other systems influence intake the goal posts are moving all the time. Need to work with that as a whole unit. In relation to nutrition, do we progress research with live feeds? Some systems in Australia such as abalone are unable to utilise natural feeds and as a consequence, the level of knowledge surrounding manufactured feeds is well advanced. Development beyond the puerulus phase in terms of how propagated rock lobster can be used in enhancing wild stocks requires further consideration. One of our Japanese colleagues suggested significant implications in relation to enhancement in terms of the potential for the introduction of foreign diseases and the potential impact of puerulus collection on wild stocks. We also have to consider if looking at short term alternatives such as collection of puerulus from the wild are appropriate given the impact on a production systems if the wild puerulus don't settle and you don't have a supply of stock.

The ultimate aim is to deliver sufficient technology to allow a practical and economical outcome in terms of rock lobster aquaculture. However, we don't have to answer every question immediately. We need to answer those questions which would limit our ability to go out and start a commercial rock lobster aquaculture enterprise. It is very clear that the research areas are broad, and the topics will be expensive and time-consuming to address. We are not here to list out things we would like to know. We must define which factors are most critical in allowing someone to practically go and



propagate rock lobsters. Because we have limited funds and we need to complete the research in a reasonable time frame, it is important that we establish a research base and then fine tune it to become more specific in targeting research.



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## Discussion of economic and biological feasibility

*Robert van Barneveld*

*Leader, Rock Lobster Enhancement and Aquaculture Subprogram*

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*Aim:* To deliver sufficient knowledge and technology to allow the practical and cost-effective production or enhancement of rock lobsters in an aquaculture system.

*Focus:* We are trying to make sure Australia has the capacity to propagate rock lobsters. We are realistically trying to identify 2-3 critical knowledge gaps that absolutely prevent us from moving further forward and how we can benchmark our research progress in relation to this (eg. what are our targets, etc?).

*Objectives:* As a group, list the processes required to establish a commercial rock lobster propagation enterprise. Having defined a base level of knowledge, identify the gaps that currently exist in our information.

### *1. Information on biological processes*

#### *Pre-Breeding*

- Ensure broodstock are collected/maintained at adequate size/condition.
- Optimise nutrition, environmental conditions for maintaining condition of broodstock.
- Know the history of individual broodstock (New Zealanders discussed genetic differences, pre-history etc). Identify the best strains, know the source of stock and their pre-history. Optimise and monitor conditions.
- Optimise health.

#### *Breeding*

- Ensure correct ratio of males:females
- Provide optimum conditions for incubation, mating
- Mate at correct time for successful fertilisation
- Maximise gamete production
- Disease control, maximise hygiene



#### *Hatch-out stage*

- Provide a system to catch/select larvae
- Provide adequate conditions for hatching out

#### *Larval phase*

- Maximise survival
- Optimise growth
- Synchronise development (eg moults)
- Minimise length of larval phase & selection to achieve high quality puerulus
- Most efficient culture system

#### *Puerulus Phase*

- Maximise moults
- Appropriate settlement conditions

#### *Nursery/Juvenile Phase*

- Optimise growth
- Maximise survival
- Minimise length of juveniles
- Grade
- Transport from nursery and deliver to market or grow-out or wild
- Optimise conditions & preparation for grow-out
- Wean onto a suitable diet

#### *Others*

- Health (optimise)
- Efficiency of mass production systems (particularly larval phase)
- Optimum nutrition
- Minimise cost/unit production
- Minimise environmental impact
- Select appropriate site

#### *Broodstock selection*

- Selection of broodstock
- Maintenance for broodstock



- Initiate breeding program
- Optimise genetic strategy
- Optimise genetic integrity

Based on the above knowledge base, the following research priorities were proposed:

#### *Pre-Breeding*

Issue irrelevant in the short term. Low priority.

#### *Breeding*

Possibility of deterioration of health of broodstock. Is nutrition responsible for quality of eggs/sperm. Need an index of condition of eggs and it's correlation to larval viability and oocyte development. Do we see differences in larval quality between broodstock ? If so, why ? However, as berried females are available from the wild this is not an issue in the short term.

#### *Hatch out*

Select the best quality larvae (floaters). Limited additional research.

#### *Larval Phase*

Maximise survival. Optimise growth and survival, nutrition, health, and system design. Minimise the length of the larval phase (mechanisms of moulting). Experimental systems are critical to the success of the entire research program based on the accompanying points.

Larval rearing is the major problem in rock lobster propagation. This was identified as the major issue at both the economic and research meetings. The larval phase can be broken down into optimising growth & survival and narrowed down still further to interactions between nutrition, feed intake, system design and health. A second major area was that of shortening the larval cycle. Understanding mechanism of moulting is very important as is the effects of nutritional changes.

## *2. Information on economic feasibility*

Discussions on economic feasibility were based on experiences with other aquaculture enterprises. It was suggested that if juvenile rock lobsters could be produced for between \$AUD0.60 and \$AUD1.20 then potential existed for the establishment of economically viable hatcheries.



There was some disagreement over the approaches used to examine economic feasibility. There was also some concern that details of these discussions could be misinterpreted and used as a commercial guide to the costs of establishing a rock lobster hatchery. For this reason, details of these discussions have not been recorded in these proceedings.



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## Development of a research plan

*Robert van Barneveld*

*Leader, Rock Lobster Enhancement and Aquaculture Subprogram*

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In the development of a research plan as part of this workshop, we have three aims:

1. To develop a strategy for a research submission/s to FRDC
2. To define the components of the research proposal
3. To integrate the development of this proposal with the World Aquaculture meeting and the Subprogram workshop.

The general outcomes from the economics exercise/biological exercise overlapped considerably, in particular the length of the larval cycle, stocking density, feeds, survival, aspects of systems design (recirculation or flow through). Biologically, larval phases and nutrition and system design were indicated as being the key factors requiring investigation, including manipulation of the larval cycle in terms of moults, moult periods and moult hormones.

I think there is probably potential for three main projects:

1. Large collaborative project focusing on larval phase and looking at nutrition, system design, environment.
2. The second thing that was quite obvious was that there appears to be a preliminary project at this stage on looking at ways to influence the moults and reducing the moult cycle.

Finally I would like to see the subprogram utilise this outline to start documenting what we already know.

The workshop was successfully used to develop a research proposal that was subsequently funded by the FRDC (Project No. 99/315) through the Rock Lobster Enhancement and Aquaculture Subprogram.



Appendix I:  
Workshop Agenda



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## Workshop Agenda

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**DAY 1. January 29<sup>th</sup>, 1999.**

### REVIEW OF CURRENT ROCK LOBSTER PROPAGATION RESEARCH

#### GEOLOGY LECTURE THEATRE, University of Tasmania

0900 Welcome and opening address by Director of TAFI, Prof. Colin Buxton

0915 Introduction by project manager of FRDC, Dr. Patrick Hone

#### STATUS AND DIRECTIONS OF ROCK LOBSTER PROPAGATION RESEARCH IN NEW ZEALAND

0930 **Alistair MacDiarmid, NIWA**  
Outline of previous and future research on aspects of *Jasus edwardsii* biology and behaviour pertinent to management of broodstock

10.00 **Lennard Tong & Graeme Moss, NIWA**  
Summary of the work done in New Zealand at NIWA Mahanga Bay, on rock lobster (*Jasus edwardsii* and *J. verreauxi*) propagation

10.30 - 11.00 Morning tea

#### STATUS AND DIRECTIONS OF ROCK LOBSTER PROPAGATION RESEARCH JAPAN

11.00 **Hirokazu Matsuda, Fisheries Research Institute of Mie**  
Phyllosoma rearing of the Japanese spiny lobster, *Panulirus japonicus*, using small rearing vessels

11.30 **Takeshi Murai & Taku Yoshimura, Sekai National Fisheries Research Institute**  
Biological Barriers to Aquaculture of Rock Lobster

12.00 **Jiro Kittaka, Research Institute for Marine Biological Science**  
Importance of three major factors for successful phyllosoma culture

12.30 - 1.30 Lunch

#### STATUS AND DIRECTIONS OF ROCK LOBSTER PROPAGATION RESEARCH IN AUSTRALIA

1.30 **Arthur Ritar, TAFI**  
Propagation of southern rock lobster (*Jasus edwardsii*) in Tasmania

2.00 **Michel Bermudes, TAFI**  
Effect of photoperiod on survival, growth, feeding and cannibalism in early developmental stages of *Jasus edwardsii* phyllosoma larvae.

2.30 **Mike Hall, AIMS**  
Closed cycle breeding of crustacea with special reference to rock lobsters.

3.00 Afternoon tea

3.30 **Satoshi Mikami, Australian Fresh Corporation, QDPI**  
Investigation of the gut system; Application for lobster aquaculture

#### FUTURE DIRECTIONS FOR ROCK LOBSTER RESEARCH

4.00 **John Benzie & Taku Yoshimura**  
Report on Japan/Australia meeting in Perth

4.15 **Lennard Tong, NIWA**  
The biological feasibility of rock lobster propagation and key areas for research

4.30 **Pheroze Jungalwalla, TASSAL Pty., Ltd.**  
How industry (aquaculture) views rock lobster propagation research

4.45 **Robert van Barneveld, FRDC**  
Summary

7.00/7.30 Dinner at Mures Upper Deck Restaurant



**DAY 2. January 30<sup>th</sup>, 1999.**

**IDENTIFICATION OF RESEARCH ISSUES AND OPPORTUNITIES FOR COLLABORATION & RESOURCING**

**ROOM 229, GEOLOGY BUILDING, University of Tasmania**

9.00 **Dr. Patrick Hone, FRDC**

Introduction

9.15 Split into 2 groups to discuss:

a. biological issues, risks and research needs (P. Hart), and

b. Economic feasibility, risks, research structure (P. Hone)

Issues & discussion points will be projected in MS PowerPoint<sup>®</sup> so that a picture emerges as we progress.

10.15 The 2 groups will swap.

11.15 Group discussion of issues raised

1.00 Lunch in the Bistro & farewell to our overseas guests

Afternoon (dependant on what time we finish discussions) **Dr. Piers Hart, TAFI**

Tour of the Marine Research Laboratories, Tasmanian Aquaculture & Fisheries Institute (TAFI)

**DAY 3. January 31<sup>st</sup>, 1999.**

**DEVELOPMENT OF THE RESEARCH PLAN FOR FRDC**

**ROOM 229, GEOLOGY BUILDING, University of Tasmania**

9.00 **Dr. Robert van Barneveld**

Introduction

9.15 **Dr. Peter Montague**

The Aquaculture CRC in relation to rock lobster

9.30 Identify sub-projects

Identify sub-project leaders

Determine milestones for drafting project proposals

5.00 Close



Appendix II:  
Study tours of New Zealand and the  
United States – Brad Crear



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# Report on a study tour to New Zealand and the United States

*Dr. Bradley Crear*

*TAFI*

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*The study tour was part of the FRDC project 98/300 (Propagation of rock lobster - development of a collaborative national project with international partners).*

## **Summary**

The tour was originally planned to attend a workshop on rock lobster culture at the Harbor Branch Oceanographic Institute in Fort Pierce, Florida and to determine the latest developments in the American clawed lobster (*Homarus americanus*) and Caribbean spiny lobster aquaculture industries. However, the workshop was cancelled at the last moment. A tour of lobster aquaculture facilities (research and commercial) in New Zealand was incorporated into the itinerary.

New Zealand has a strong research and commercial involvement in rock lobster (*Jasus edwardsii*) aquaculture. Two commercial companies, relying on the ongrowing of wild caught puerulus, have commenced operation. Their biggest problem at present is the inability to capture sufficient numbers of puerulus. Phyllosoma research in New Zealand has had several exciting results (high survival through early larval stages, development of a culture system). However, techniques to produce commercial numbers of puerulus from larvae have not been developed. Research has now focused on the eastern rock lobster, *J. verreauxi*, as it may have a slighter easier larval cycle.

There is interest in spiny lobster culture in the United States, especially in the area of puerulus ongrowing. However, there appears to be little opportunity in the continental United States, due to low puerulus settlement rates. High settlement rates in some areas of the Caribbean, would suggest that opportunities exist for ongrowing in some countries. However, a lack of infrastructure in most countries means that it is probably not feasible to run commercial facilities at the present time. The most likely avenue for aquaculture of spiny lobsters is through post-harvest impoundment and ongrowing of fishery caught lobsters.

Contrary to reports of renewed interest in clawed lobster aquaculture, no commercial facilities could be found to visit. However, several of the people contacted whilst organising the tour, commented that they were in an advanced stage of setting up projects. In contrast, one of the better known commercial



clawed lobster facilities, Kona Cold Lobsters (Hawaii), had closed down their larval rearing and on-growing projects, and were concentrating on processing wild caught live clawed lobsters (mostly from Canada).

There appears to be an ongoing, and in some cases new, interest in the area of enhancing the wild fishery through the release of hatchery reared juveniles. The two clawed lobster researchers visited were both working in this area. However, there are no clear cut benefits of enhancement projects at the present time.

## **Itinerary**

### **Australia to New Zealand (15 August)**

Visit scientists at the National Institute of Water and Atmosphere Research Ltd (NIWA) in Wellington and Auckland. Also visit commercial rock lobster aquaculturists at Napier and Tauranga.

### **New Zealand to United States (20 August)**

Visit scientists involved in rock lobster research in Indiana (Purdue University) and Florida (Harbor Branch Oceanographic Institute). Visit scientists involved in clawed lobster research in Massachusetts (New England Aquarium) and Rhode Island (University of Rhode Island).

### **United States to Australia (29 August)**

#### **Dr. Len Tong and Mr. Graeme Moss - NIWA, Wellington**

NIWA has a marine research facility based at Mahanga Bay near Wellington. They have been working on the aquaculture of the southern rock lobster, *Jasus edwardsii*, for a number of years. Much of the research has focused on establishing larval and juvenile culture methods. Much of that research is summarised in articles they have written for the Aquaculture Update Newsletter, a quarterly newsletter published by NIWA. Dr. Len Tong is the leading research scientist on lobster aquaculture at NIWA. Mr. Graeme Moss has also been heavily involved in the research.

Besides rock lobster culture the facility was involved in work on seahorses (*Hippocampus abdominalis*), turbot, flounder and abalone at the time of visiting.

## **Broodstock**

NIWA have been using lobsters cultured at the facility from puerulus as broodstock. Some of the lobsters appear to be around 150 g and it was thought that they were around 3 years of age. Lobsters



of around 100 g have berried up in the tanks in the past. **Comment: The small size that lobsters were achieving maturation suggests precocious maturation may be a concern in the culture of *J. edwardsii* where a market size 200-300 g is being targeted.** NIWA have generally kept broodstock at a 6F:1M ratio but feel it didn't need to be too specific. They mentioned that Dr. Alistair MacDiarmid is conducting some work on male size (Foundation for Research Science and Technology: APP 98-NIW-06-6038 Objective 2).

NIWA had *J. edwardsii* phyllosomas hatching during August. That was much earlier than usual (Sept/Oct), and it was suggested that the reason for the early hatching was the warm winter temperatures ( $\approx 2^{\circ}\text{C}$  above normal over the winter period). NIWA have undertaken research on the affect of temperature on the development time of eggs. The general conclusion was that warmer temperatures increased the rate of development. However, phyllosomas hatched at higher temperatures were smaller. NIWA have also calculated the time to hatch based on a lobster eye index (based on the length and width of the eye). A paper on this area of study is due out soon.

### **Larval culture**

The focus of larval culture research has moved from *J. edwardsii* to *J. verreauxi* (the eastern rock lobster). This was partly due to the highly promising results achieved by the Japanese with the larval culture of this species. It appears to have a slightly shorter larval cycle and is regarded as being a tougher species in the larval stage. NIWA expects *J. verreauxi* phyllosomas to hatch out at around Christmas time. They do not have broodstock at the moment because fishermen have not been able to catch any (dirty water due to excessive rain in northern New Zealand). The female generally moults around August and mates in October.

All newly hatched naupliosoma or phyllosoma are placed into a cylindrical perspex tank (40 cm high, 15 diameter). A light is placed above the tank. Stage 1 phyllosomas are strongly photopositive. Only animals which swim strongly towards the light are selected for culture. NIWA believes those ones are the better, stronger animals. They would like to do some work on looking at the condition (eg. lipid levels) of the different batches of phyllosomas, both within one hatch (swim strongly V not swim strongly)and between hatches. They have never seen naupliosoma not develop into phyllosoma, something which has been observed at TAFI.

The larval rearing is as described by Illingworth *et al.* (1997). Basically it consists of 4 X 40 l upwelling tanks. The system is a recirculating system but it has no biofilter. The reason for the recirculating system was to reduce the cost of heating water. NIWA relies on water exchange to



maintain the water quality (approx. 8 water turnovers per day). NIWA believed that part of the problem with the rearing system last season was poor water quality, therefore they are adding a diatomaceous earth filter this year. NIWA have also produced some larger (semi-commercial) rearing tanks. These tanks are about 400 l and have the same basic principles as the smaller rearing tanks.

NIWA keep the phyllosomas in the dark in this rearing system. The phyllosomas appear to be photopositive for the first couple of days and then become reasonably photosensitive. NIWA have measured light levels which the phyllosomas move away from and it is generally very low levels (no actual figure given), but a level that humans would regard as dark. The phyllosomas become more light sensitive at the later development Stages (very sensitive in Stage 6-7). NIWA felt there was plenty of important research to do in this area.

Phyllosomas are kept at 40/l for the first few Stages, but at later Stages (6-7) they tend to damage each other at that density. NIWA reduces the density to 20/l during those Stages. At high densities the phyllosomas tend to cling together and rip pieces off each other. Beyond Stage 3 they tend to like to cling onto something. The pizza shaped food particles (see below) give them the opportunity to cling to something else rather than the each other. The phyllosomas tend to float around with the food particles while feeding on them.

The diet NIWA feed to their larvae mostly consists of ongrown (2-3 mm in length) *Artemia* (reared on algae - *Isochrysis galbana* or *Rhodomonas reticula*). However, they have trialed many other diets in an attempt to develop an easier and better diet. A flat pellet (called a pizza) has shown considerable promise because it tends to stay up in the water column. The pellets are made from *Artemia* bound together with alginates. The only puerulus NIWA was able to produce was grown with *Artemia* up to Stage 8 and mussel gonads after that. An article covering NIWA's work on the development of diets is in the NIWA Aquaculture Update, Issue 19, 1997.

NIWA have also developed some experimental upwellers. These have been described by Tong *et al.* (1997). Basically the system consists of a series of upwellers; each upweller can accommodate one larvae. There are different sized upwellers for the different sized larvae. NIWA have received some criticism of the design because the upwellers are all held within one tank. Therefore, the upwellers may not constitute replications (pseudo-replication). There may be downstream effect of moulting.

NIWA haven't done too much work on the disease problems of larvae. They have noticed that the larvae do get "black gut". They don't know what it is but it kills the larvae in a short period. A disease



researcher has been appointed to the staff to investigate some of the problems (with fish as well as rock lobster).

### **Future studies on larval rearing**

The specific areas NIWA would like to study with regards to larval rearing include: light levels, nutrition and disease. However, in the near future they are moving onto *J. verreauxi* (for the reasons mentioned above). Hopefully, sometime down the track, they will be able to try to overcome problems with the larval rearing of *J. edwardsii* using techniques developed through work on *J. verreauxi*.

### **Juvenile rearing**

NIWA has been conducting research in this area for a number of years. NIWA Aquaculture Update (Vol 18, pp. 5-6) give a good run down on what they have done and what they would like to do. They have a couple of articles due out soon on aspects of juvenile on-growing: (i) Growth of lobsters fed opened and unopened mussels (James, 1998). (ii) Consumption/conversion ratios (L. Tong in New Zealand Seafood).

NIWA were getting mussel wet weight (mat only) to lobster wet weight food conversion ratios of between 12:1 and 17:1. A commercial farmer had been getting ratios of 22:1.

NIWA have done a fair bit of work on juvenile hides (NIWA Aquaculture Update, Vol 21 pp. 8) and also density. The density trial were conducted for the first 6 months post puerulus. An article is due out on this soon but basically they found that lobsters kept at 50/m<sup>2</sup> had better growth than lobsters at 100/m<sup>2</sup> or 200/m<sup>2</sup>. Survival did not differ between the treatments and was about 90%.

NIWA have done some preliminary work on artificial diets with little success, so they are very interested in the nutrition work occurring in Australia. A commercial company is doing some work on artificial diets at the moment (no details were available).

Dr. Tong and Mr. Moss both indicated their willingness to attend the planned propagation workshop and were keen to foster collaborative projects.

### **DR. JOHN BOOTH - NIWA, Wellington**

Dr. Booth has been conducting research on rock lobster larval recruitment (including puerulus settlement) processes for many years. He has recently become involved in aiding with the design of puerulus collectors for the commercial rock lobster aquaculture enterprises in New Zealand.



### **Commercial puerulus capture**

Due to confidentiality agreements with the commercial operators Dr. Booth could not elaborate on the design of puerulus collectors being developed. However, he did say that much of the collection is taking place under Gisborne wharf (the site of large catches by NIWA over the years) using modified crevice collectors.

### **Puerulus settlement**

Dr. Booth suggested that in Tasmania we should try wharf areas as a source of puerulus. Should be able to contact commercial divers who work around the wharves and ask them if they have seen many juvenile lobsters under the wharves. Dr. Booth is unsure why there has been huge settlement at Gisborne wharf, as the wharf does not appear to be in the right area, as far as water flow is concerned, to act as a trap. He thought that settlement numbers would be in the hundreds of thousands at Gisborne wharf, and that most of those lobsters were dying.

To try to further understand why puerulus settle where they do NIWA is undertaking a study on settlement cues (Foundation for Research, Science and Technology Application: 98-NIW-06-6038, Objective 4 - Rock lobster larval recruitment dynamics). Cues they are investigating include: sound (frequency and amplitude mix) and a range of biotic (algal film, dominant invertebrates, presence of con-specifics) and physical features (hole shape and orientation).

Some preliminary work completed on the presence of con-specifics suggests that puerulus are asocial ie. the presence of other lobsters within collectors does not attract or repel the settling lobsters. He also found that lobsters prefer to go into hides (the holes of house bricks) rather than remaining in an algal substrate (hiding spaces present but not holes). NIWA is now comparing conditioned (algal growth) bricks against unconditioned bricks.

Dr. Booth thought that puerulus numbers dropped off with depth of the water and that they tended to be found in shallower areas (<20 metres depth). He was interested to hear that we had collected large numbers from scallop collectors that were in depths > 25m (to 40 m). He thought that even though the lobsters were attached to the outside of the scallop collectors that they mostly wouldn't drop off as the collectors were being hauled to the surface.



## Other issues

NIWA had done some work on ontogenetic changes in social behaviour. A confidential report on the research has been written, but the paper will be available in the near future.

Dr. Booth felt that there was around 90% mortality of pueruli/juveniles in the first year from settlement. He had no data to back that up.

## Hawkes Bay Aquaculture, Napier, New Zealand

Hawkes Bay Aquaculture is the first commercial rock lobster aquaculture company in New Zealand. The facility is situated at Napier on the east coast of the North Island. The company is capturing puerulus and on-growing them to a marketable size of 200-300 g. HBA caught their first puerulus during winter last year (1997).

## Quota conversion

Hawkes Bay Aquaculture (HBA) has a license to obtain 60000 puerulus per year. This means that 1.5 tonnes of rock lobster fishery quota must be relinquished to allow the appropriate number of puerulus to be obtained (conversion rate is 1 tonne of rock lobster: 40000 individual puerulus or 30 kg of puerulus). **Comment: based on the average weight of puerulus in Tasmania ( $\approx 0.5$  g) it would be much better to convert in the form of weight rather than number. This would mean that 1 tonne of quota would constitute 60000 puerulus.** The manager, Mr. Davies, was very happy to show me around although he didn't want to go into a lot of detail with many of their procedures.

## Puerulus collection

HBA's main problem was obtaining enough puerulus (not on-growing them). At present it is costing HBA approximately NZ\$100000 to collect the 60000 puerulus (\$1.66 per puerulus). The NZ\$100000 includes quota leasing costs of NZ\$30-35000, vehicle (car and boat), collector manufacture and labour. Even with that outlay of funds HBA is not sure it can collect the required numbers each year. Because of the uncertainty of supply Mr. Davies can see the need for a hatchery, and hopes that in the future a group of farmers has a hatchery which produces the puerulus.

HBA have spent a lot of time, with the help of NIWA (Dr. John Booth) in refining the puerulus collection methods. It was originally thought that the deployment of 100 collectors would be sufficient to capture the required numbers of puerulus. However, HBA now has 500-600 collectors out and it was thought that would be enough. HBA has found that there are real pockets of puerulus settlement ('hot spots'). In the 'hot spots' up to 70 puerulus can be collected every few days in 3-4



adjacent collectors. However, in other collectors close by (2 metres) very few or no puerulus will settle. When HBA find a 'hot spot' then it saturates the spot with collectors. Unfortunately the 'hot spots' can move, which means that HBA need to be continually searching for them.

HBA has a few main collection spots: Gisborne Wharf, Napier Wharf and an area out from Napier in about 8-10 metres of water. They are concentrating their efforts in those areas at the present time but are continually searching for new sites (based on what they are learning and from work by NIWA). They haven't tried deep collection sites yet.

### **Puerulus collectors**

The design of puerulus collectors is an ongoing process. HBA has tried a lot of different substrates. The basic conclusion was that when puerulus are settling in numbers, they will settle on just about anything, even unconditioned collectors. However, when there were few puerulus about then they tended to settle on the crevice-type collectors.

HBA has developed an H-shaped metal frame which they can place on the sea floor in shallow areas (< 1m deep at low tide). The crevice collectors are screwed onto the frame. These can be waded out to and the collectors screwed off individually to be checked.

HBA have developed a frame to attach to the side of a dinghy so that they can service their crevice collectors under the wharves (the collectors hang from the wharf). This frame allows the collectors to be serviced easily and without significant amounts of man handling. The frame is used to flip the collector into the dinghy.

HBA is attempting to build collectors based on the crevice design, but that are cheaper and easier to build. They would like to move to a long line setup. They believe that if the collectors are brought on board slowly enough the puerulus will stay attached (similar to Dr. John Booths' theory).

To transport the puerulus back to the growout site HBA has developed mesh containers. Each mesh container is about 20 cm long and 7 cm in diameter. The mesh gives the pueruli something to attach to during transport. They place lots of fifty pueruli into each container and then place 10-12 containers in an esky (with water) for transport. When back at the growout site the lobsters are released into the tanks underwater. Therefore, after the initial handling period at capture and counting the lobsters are not handled again. HBA has found they get mortalities if the puerulus are handled too much. They also believe that the best way of handling the pueruli is by the antennae. HBA have found that they



get poorer survival of puerulus during winter than during summer. No reason could be suggested, except that the water temperature may have been too cold for them.

### **Juvenile growout**

The water intake is situated about 9 metres under sand therefore they do not require any further pre-filtration of the incoming water. The ambient temperature ranges from 10°C to 22°C. The mean temperature over the year is 14-16°C. The growout system is run as a recirculating system during winter which maintains the temperature a couple of degrees above ambient. They run a protein skimmer on the system. No heating is employed. HBA has had no problem with survival of lobsters at 22°C.

HBA is using shallow black plastic tanks. The tanks are about 2 x 1 x 0.2 m but only contain about 7 cm of water. They were designed in conjunction with NIWA (Mahanga Bay). HBA had a mould made. Water flows in one end and out the other. No aeration was being used and they have put abalone shells in the tanks as hides (approx. 6/tank). Stands have been made and the tanks are stacked 3 high. Mr. Davies sees that as being a problem as you can't observe the lobsters well or easily clean out the tanks. This is especially a problem for the younger juveniles (<30 mm CL or 10-15 g). HBA is making a new system that is only one tank in height for the younger juveniles.

The lobsters I saw were mostly less than 30 g. They were stocked at 100 per tank (50/m<sup>2</sup>) but Mr. Davies believes that at the larger sizes they will need to reduce to 25 per tank. HBA is currently doing an experiment with NIWA looking at stocking densities of 50, 100 and 200 per m<sup>2</sup>.

HBA is using mussels (Greenlips - *Perna canaliculus*) as feed for the lobsters. They were using wild collected ones but they were a nuisance (hard to clean up) and some that were collected were toxic (caused lobster mortalities). The toxic mussels were collected from around Napier wharf and had probably accumulated large amounts of toxins, such as from antifouling paint. HBA buys the mussels now from a commercial mussel farmer (NZ\$1 per kg). Feed rate is corrected depending on the amount eaten the day before. During summer (water temperature of 22°C) the tanks need to be cleaned out daily (mussels go off) but during winter (water temperature 10-12°C) the mussels can be left for a couple of days.

After collection, the puerulus/post-puerulus are placed into mesh crates within tanks. The mesh crates give the puerulus something to attach to and also allows the tanks to be easily cleaned. The pueruli are fed mussels from first feeding. Mr. Davies believes that the first 3 moults are the most critical for



survival. HBA is working at maximising survival through that period. Survival is higher (although could be better) from after 3 moults (approx. 2 g) to 10-15 g, and is very high after that.

The average size of the lobsters caught last year (approx. 14 months old) is 30 g. HBA is hoping to achieve market size in 3 years.

**Comment: I was told that HBA had a quite high mortality rate during the first year of operation ( $\approx$  38%). Problems included poor food, poor water quality, cold temperature and freshwater runoff causing salinity problems. Moulting death syndrome was evident in some of the mortalities.**

#### **Aqua - Bay of Plenty, Tauranga, New Zealand**

Aqua - Bay of Plenty (ABP) has been in operation for approximately one year as well. They caught their first puerulus during winter, 1997. They have 6000-7000 lobsters at the moment. ABP is situated at Tauranga which is on the north coast of the North Island, about 2 hours from Auckland. The manager is Mr. Chris Zane.

ABP wasn't as forthcoming in discussing their project as HBA. However, their major problem area appeared to be exactly the same as HBA. That is, the collection of sufficient numbers of puerulus. They have also found that there are 'hot spots' of settlement. ABP are looking in similar places to HBA (under wharves). ABP is also targeting long beaches where there are little or no natural settlement areas. They intend to put collectors at the end of the beaches to catch puerulus that drift along the beach looking for a settlement site. Mr. Zane believed that is why the wharf at Gisborne is so successful. Mr. Zane also believed that the puerulus feed and it would be possible to attract them using the right food source.

ABP use a recirculating system for growout. Mr. Zane would not elaborate on the design. It is being run at 16°C and the lobsters are being fed greenlip mussels (bought from producer).

#### **DR. SIMON HOOKER, DR. ANDREW JEFFS - NIWA, Auckland**

Dr. Simon Hooker is the Business Development Manager in Aquaculture and Marine Ecology for NIWA. He has been involved in rock lobster research (mainly juvenile on-growing) for a number of years. Dr. Andrew Jeffs is involved in research into the condition of pueruli as they return to the coast and possible navigational cues used by pueruli.



Dr. Hooker noted that although there had been only two licenses issued for rock lobster culture in New Zealand, there were many companies waiting in the wings to take up a license. He expected some of those companies to take up licenses in the near future. Dr. Hooker has significant interaction with the two commercial enterprises already in operation.

In some of Dr. Hooker's research larger lobsters (>100 g) have shown a reasonably slow growth rate. He was unsure of the reason for that but thought stocking density may have been part of the problem. Maximising the growth rates of those larger lobsters was of much interest to him. Dr. Hooker has a masters student (Scott Kington - see below) conducting some research in the area of alternative growout diets. Mr. Kington suggested the slow growth rate may be due to the relatively greater carapace thickness of larger lobsters. Therefore, more feed is used to lay down carapace rather than being incorporated into muscle growth.

Dr. Jeffs is involved in work with Dr. John Booth on navigational cues (Foundation for Research, Science and Technology Application: 98-NIW-06-6038, Objective 4 - Rock lobster larval recruitment dynamics). Large numbers of puerulus have been known to settle in the area of the water exit from the New Plymouth power station and it has been suggested that the noise emanating from the power station is acting as a navigational cue. In this project he taped the noise (white noise) being emitted from power station. and broadcasted it at Gisborne. Preliminary results suggested that the sounds being broadcast were not attracting pueruli.

As part of the same project Dr. Jeffs is also determining how the condition of pueruli varies both temporarily (summer Vs winter) and spatially (distance from the coast - up to 300 km). Initial results suggest that the pueruli may have a different method of lipid synthesis to that recorded in other crustaceans. He is writing up the results at the present time.

Both Dr. Hooker and Dr. Jeffs stressed the need for extensive collaboration between the two countries. They highlighted the significant amount of research conducted on the southern rock lobster in New Zealand that Australian researchers could tap into. They also realised there were significant opportunities to piggyback on some of the Australian rock lobster aquaculture research that was being conducted in areas not yet looked at in New Zealand (the production of artificial diets being one such area).

#### **SCOTT KINGTON - Auckland University, Auckland**

Mr. Kington is a Masters student at Auckland University. He is undertaking research on ongrowing juveniles and has Dr. Simon Hooker as one of his supervisors. Some of his work in being conducted



in conjunction with a commercial fishing company which is interested in being involved in rock lobster aquaculture.

Mr. Kington has two growout sites north of Auckland (Leigh Marine Laboratories and a private companies site). At Leigh the water temperature ranges from 14 to 24.5°C whilst at the private site it ranges from 14 to 22°C. He has found that the lobsters appear to suffer from moult death syndrome (stuck in shell) when the water temperature is up around 24.5°C. Slower growth is also evident. He has not seen the same problems at 22°C. The growth rate of lobsters in his studies has been excellent. They have grown from 5 to 50 g over 200 days at an average water temperature of 19°C. Mussels have provided the best growth rates, compared to other natural diets such as pipis and clams. He is also looking at a diet which includes a mixture of fish frames and mussels in an attempt to reduce the cost of production. So far the lobsters have not been very attracted to the fish frames.

Mr. Kington is also doing some research on the affect of shelter on growth. Using larger juveniles (50 g +) and providing animals with half round terracota pots or not. No significant differences in growth or survival have been found as yet.

In Mr. Kington's experiments an overall mortality rate of around 10% has occurred. There was no specific reason for the mortalities (apart from those occurring due to warmer water temperatures).

#### **PROF. PAUL BROWN - Purdue University, Indiana**

Prof. Paul Brown has been conducting research on nutrition of the Caribbean spiny lobster, *Panulirus argus* (Brown *et al.*, 1995). His main interest has been investigating the use of alternative dietary protein sources (soybean meal). He showed an interest in continuing research in the area of spiny lobster nutrition, especially with the use of alternative dietary protein sources, but there is not much funding in the area. Prof. Brown is trialing some similar diets with *H. americanus*, with some success (both wild caught adults and hatchery reared juveniles). The work is being done in conjunction with The Lobster Institute in Maine.

#### **DR. LEROY CRESWELL - Harbor Branch Oceanographic Institute, Fort Pierce, Florida**

Dr. Leroy Creswell has been involved in spiny lobster (*Panulirus argus*) research at Harbor Branch Oceanographic Institute (HBOI) for a number of years, although little is being undertaken at the present time. It has been hard to obtain research funding in the area because of the limited fishery for *P. argus* in the United States. His main interest areas with lobsters are the holding and ongrowing of



fishery caught adults and the aquaculture of wild caught pueruli. Dr. Creswell sees adult on-growing as being the most feasible in the near future.

### **Adult holding and on-growing**

The general idea of this is to grow lobsters through one moult, thereby achieving a 30-40% weight increase (for a 400 g lobster). The moult generally occurs within 60 days of capture (on average). Raceways/seacages are being used to hold the lobsters. Only a small percentage of lobsters are kept for on-growing at the present time. Part of the reason for that is that in many communities the lobsters are caught by wire, therefore they are dead when they are landed. The general idea is to focus on the European market to obtain a better price. The beach price of *P. argus* is around AUS\$9-11.00/kg.

Dr. Creswell has developed a test for moult stage based on the colour of the underside of the tail. This can be used to predict moulting to within a couple of days. It can be used by processors to select lobsters for on-growing. If the lobster is judged to be just post-moult then it would probably not be kept for on-growing as the time period to the next moult would be too long.

One of the problems associated with adult on-growing is the lack of a suitable diet for the lobsters. Dr. Creswell does not see it as a major problem if the lobsters are only held through one moult. Even so, he believes the industry needs access to an artificial diet as there are limited possibilities for using products from other sources (eg. mussels, fish or conch trimmings) as a diet. He has a masters student doing some research on preparing a water stable feed and feed attractants. This research is just starting but so far there have been positive results with the use of betaine as a feed attractant. Dr. Creswell suggested that the lack of a suitable artificial diet is one of the major reasons it is hard to get people interested in investing in the industry.

### **Puerulus aquaculture**

There are three major impediments to the development of puerulus aquaculture; (i) lack of supply of puerulus, (ii) lack of a suitable diet, and (iii) lack of infrastructure in suitable culture areas. At the present time it appears that there is no company working in this area. Dr. Creswell thought that there was a Jamaican company doing some work but he couldn't report on its success or whether it was still in operation. There has been a significant amount of research conducted on puerulus aquaculture (Bill Lellis in the late '80s and early '90s). *P. argus* can be grown to a suitable size from puerulus in 18 months.



As in Australia and New Zealand there is a spatial and temporal variability in the ability to capture *P. argus* puerulus using artificial collectors. Not only is the lack of supply of puerulus a problem area, there is also potential legislative problems with collecting puerulus that would need to be examined. This area has not been tested to any great extent as yet. It has been suggested that any aquaculturists would need to return a certain percentage of females to the wild. There is the possibility that puerulus will be able to be collected in the area of the Texas oilfields. Significant numbers of puerulus have been known to settle on the oil platforms. The animals are carried there by the Gulf stream. However, during winter it is too cold for *P. argus* to survive. Because there is no lobster fishery in the area, collection of puerulus here has the added advantage of not conflicting with other users of the resource.

A lack of infrastructure decreases the feasibility of culturing puerulus in many Caribbean countries. In many cases the cost of power would make the cost of production excessive. For that reason the Jamaican company that was investigating puerulus on-growing was looking at culturing them in cages in a lagoon area. Therefore, Dr. Creswell suggested that puerulus culture may be attempted in some USA mainland states using recirculating systems (maintaining a water temperature above 27°C).

### **Phyllosoma culture**

There has been some research conducted in the 1980's the phyllosoma of *P. argus* which suggested that the larval stage may be up to 12 months long. Dr. Creswell recently conducted a trial to re-examine the issue as he thought the time period could be shortened. Using a Kreisel tank system, and a diet of on-grown *Artemia*, the phyllosoma reached Stage 5 after 5 months (survival was low). He concluded that the larval stage could be up to 12 months in duration. Mostly due to those results there is no interest at HBOI to conduct any more phyllosoma culture research.

### **NEW ENGLAND AQUARIUM, Boston, Massachusetts**

Staff at the New England Aquarium have been raising the American clawed lobster, *Homarus americanus*, to display in the aquarium and for some research on enhancement they were conducting. They also sell the larvae to universities to be used in experimental work. They have a limited ability to rear larvae due to space constrictions. The enhancement work is using small artificial reefs (casitas) as habitat to improve survival of wild juveniles and will use the same structures as substrates for reseeded the hatchery reared juveniles. They are also trying other substrates such as bricks and pieces of PVC tubing. The aquarium uses standard clawed lobster larval rearing and juvenile culture methods.



### **Larval rearing**

Kreisel tanks (approx. 40 l) are used as the larval culture vessel and the system is kept at 17-18°C as part of a recirculating system. They have a protein skimmer and a UV filter in the system. The larvae are fed enriched *Artemia* for the first two stages and then ongrown frozen *Artemia* for the following 2 larval stages.

### **Juvenile culture**

The juveniles are raised in individual compartments and are progressively placed into larger compartments as they grow (to ensure growth is not restricted by culture area). Feeding and cleaning of the individual lobsters are very time consuming practices. They are fed frozen ongrown *Artemia*. The lobsters I saw on display were grown at ambient temperature and were about 30 g after 3 years.

### **Broodstock**

To enhance the time period over which the larvae are available for display and research, some broodstock are maintained at a low temperature ( $\approx 10^{\circ}\text{C}$ ) to retard the rate of development of the eggs. Just prior to hatch the broodstock are brought up to normal hatching temperature ( $\approx 20^{\circ}\text{C}$ ), to ensure a swift and complete hatching.

### **DR. KATHY CASTRO, University of Rhode Island**

Dr. Kathy Castro is also working with clawed lobster enhancement. Dr. Castro gave me a copy of the proceedings of a workshop on lobster enhancement (Gendron, 1998), which is a great synopsis of worldwide research conducted in the area. Dr. Castro has just finished a larval rearing run and had only that week released a large number of juveniles to the wild.

### **The project**

The initial catalyst for this project was an oil spill (minor) which was suspected to have killed lobsters in the area. Therefore, the government decided to investigate the possibility of enhancing the damaged fishery population through the release of hatchery reared juveniles.

### **Artificial reef**

The initial results of this trial were reported in Cobb *et al.*, 1998. Six artificial reefs have been established, each 20 m X 10 m and each reef was divided into two 10X10 m segments. Each segment had a different rock size (10-20 cm stones and 20-40 cm stones) so that different size hides were



created. The idea was that small lobsters may settle in the smaller hides produced by the small stones. As the lobsters grow they may move to the larger hides produced by the larger stones. The stones were layered 1 to 2 high.

Dr. Castro found a beneficial affect of the reefs to the lobster population in the first year. There was a significant increase in the lobster population of the bay, including numbers in the artificial reef areas that were similar to that of nearby control reefs. The artificial reefs had significant biofouling within 6 months. Tagged juveniles were released during August 1998. One of the aims is to see if the hatchery reared juveniles survive. Therefore, they were doing a survey a week after release. Another aim is to see if there is a significant increase in the population of seeded reefs compared to unseeded reefs. Thus, only 3 of the artificial reefs will be seeded with hatchery reared juveniles.

Visual census, trapping and airlift operated suction pump have been used to do surveys of the population.

### **Larval rearing**

Dr. Castro uses standard clawed lobster larval rearing and juvenile culture methods. The lobsters are tagged at Stage VI or VII (approx. 5mm carapace length or 0.2 g) using coded microwire tags. Each batch released is tagged with the same code (batch code not individual code). There have been no problems with the tagging (ie. tag induced mortality) but there has been some problems with picking up the tag using the sensor. The animals need to be dissected to be sure of the tag, however because the tag is small it can be difficult to find.

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## **CONTACTS**

### **Wellington, New Zealand**

Dr. Len Tong (NIWA)  
E-mail: l.tong@niwa.cri.nz

Dr. John Booth (NIWA)  
Ph: 0011 64 4 386 0300  
Fax: 0015 64 4 386 0574  
E-mail: j.booth@niwa.cri.nz

### **Napier**

Phil Davies (Hawkes Bay Aquaculture)  
Ph: 0011 64 6 834 0998  
Fax: 0015 64 6 834 0992

### **Tauranga**

Chris Zane (Aqua - Bay of Plenty)  
Ph: 0011 64 7 577 6692  
Fax: 0011 64 577 6693

### **Auckland**

Dr. Simon Hooker (NIWA)  
Ph: 0011 64 9 375 2054  
Fax: 0015 64 9 375 2051  
Mobile: 0011 64 25 997801  
E-mail: simon.hooker@niwa.cri.nz

Scott Kington (University of Auckland)  
Ph: 0011 64 9 4222 6111  
Fax: 0015 64 9 4226113  
E-mail: s.kington@auckland.ac.nz

### **Purdue University**

Dr. Paul Brown)  
Ph: 0011 1 765 494 4968  
Fax: 0015 1 765 496 2422  
E-mail: pb@fnr.purdue.edu

### **Harbor Branch Oceanographic Institute**

Dr. Leroy Creswell  
Ph: 0011 1 880 333 4264 Ext: 405  
Fax: 0015 1 561 466-1506  
Email: creswell@HBOI.edu

### **University of Rhode Island**

Dr. Kathy Castro  
E-mail: kcastro@uriacc.uri.edu



## Appendix III: Contact List



## Contact list

PERSON	INSTITUTION	CONTACT DETAILS
<b>AUSTRALIA</b>		
Dr. Trevor Anderson	JCU, Department of Aquaculture, James Cook University, Townsville Qld. 4811	Ph. 07 4781 4585 Fax: 07 4781 5586 Email: Trevor.Anderson@jcu.edu.au
Dr. Robert van Barneveld	Barneveld Nutrition, PO Box 42, Lyndoch, SA, 5351	Ph. 08 85246 477 Fax. 08 85246 577 Email: robvanb@dove.net.au
Dr. John Benzie	Australian Institute of Marine Science, Cape Ferguson, Townsville, 4810	Ph. 074 753 4211 Fax. 074 772 5852 Email: j.benzie@aims.gov.au
Mr. Michel Bermudes	TAFI, Marine Research Labs, Nubeena Crescent, Crayfish Point, Tarooma, 7053 Tas.	Ph. 03 62277291 Fax. 03 62277298 Email: Michel.Bermudes@dpiwe.tas.gov.au
Prof. Colin Buxton	Director, TAFI, Marine Research Labs, Nubeena Crescent, Crayfish Point, Tarooma, 7053 Tas.	Ph. 03 62277247 Fax. 03 62278035 Email: Colin.Buxton@dpiwe.tas.gov.au
Dr. Bradley Crear	TAFI, Marine Research Labs, Nubeena Crescent, Crayfish Point, Tarooma, 7053 Tas.	Ph. 03 62277295 Fax. 03 62277298 Email: Bradley.Crear@dpiwe.tas.gov.au
Mr. Graeme Dunstan	CSIRO Division of Marine Research, GPO Box 1538, Hobart, 7001, Tas	Ph. 03 6232 5274 Fax. 03 6232 5123 Graeme.Dunstan@marine.csiro.au
Mr. Andrew Ferguson	SARLGA, PO Box 800, Victor Harbor, SA, 5211	Ph. 08 85525521 Fax. 08 85522660
Mr. Stewart Frusher	TAFI, Marine Research Labs, Nubeena Crescent, Crayfish Point, Tarooma, 7053 Tas.	Ph. 03 62277271 Fax. 03 62278035 Email: Stewart.Frusher@dpiwe.tas.gov.au
Mr. Caleb Gardner	TAFI, Marine Research Labs, Nubeena Crescent, Crayfish Point, Tarooma, 7053 Tas.	Ph. 03 62277233 Fax. 03 62278035 Email: Caleb.Gardner@dpiwe.tas.gov.au
Dr. Geoff Gooley	Mgr, Aquaculture Program, MAFRI, Private Bag 20, Alexandra, VIC 3714	ph. 03 5774 2208 fax 03 5774 2659 Email: G.Gooley@nre.vic.gov.au
Dr. Piers Hart	TAFI, Marine Research Labs, Nubeena Crescent, Crayfish Point, Tarooma, 7053 Tas.	Ph. 03 62277242 Fax. 03 62277298 Email: Piers.Hart@dpiwe.tas.gov.au
Dr. Mike Hall	Australian Institute of Marine	Ph. 074 753 4211



	Science, Cape Ferguson, Townsville, 4810	Fax. 074 772 5852 Email: m.hall@aims.gov.au
Dr. Patrick Hone	FRDC, Deakin House, Unit 1, 50 Geils Crt., Deakin, ACT, 2600	06 285 4485/ 0419 628400 Fax. 06 285 4421 Email: fishrdc@actonline.com.au
Dr. Danielle Johnston	UTAS, PO Box 1214, Launceston, Tas, 7250	Email: djohnsto@aims.gov.au
Dr. Clive Jones	Queensland Department of Primary Industries, Walkamin, Q 4872	Ph. 61 (07) 4093 3733 Fax. 61 (07) 4093 3903 Email: jonescm@dpi.qld.gov.au
Mr. Pheroze Jungalwalla	TASSAL Pty., Ltd., GPO Box 1645, Hobart, 7001, Tas.	Ph. 03 6224 0355 Fax. 03 6224 0373 Email: jungalwalla@tassal.com.au
Mr. Matt Kenway	Australian Institute of Marine Science, Cape Ferguson, Townsville, 4810	Ph. 074 753 4211 Fax. 074 772 5852 Email: m.kenway@aims.gov.au
Dr. Roy Melville-Smith	Principal Research Scientist W. A. Marine Research Labs. P.O. Box 20, North Beach, 6020, WA	Ph: 08 92468406 Fax 08 94473062 Email: rmsmith@fish.wa.gov.au
Dr. Satoshi Mikami	Australian Fresh Corporation Pty QDPI, PO Box 2066, Woorim, Bribie Island, 4507, Q.	Ph. 07 3400 2000 Fax: 07 3408 3535 MikamiS@prose.dpi.qld.gov.au
Dr. Peter Montague	Aquaculture CRC, University of Technology, Sydney, P.O. Box 123, Broadway, NSW, 2007	Ph. 02 95141385 Fax. 02 95141491 Email: p.montague@uts.edu.au
Dr. Bruce Phillips	Curtin University of Technology, GPO Box U1987, Perth, 6001, WA	Ph. 08 9266 9266 Fax. 08 9266 2255 Email: rphillip01@cc.curtin.edu.au
Dr. Arthur Ritar	TAFI, Marine Research Labs, Nubeena Crescent, Crayfish Point, Taroona, 7053 Tas.	Ph. 03 62277294 Fax. 03 62277298 Email: Arthur.Ritar@dpiwe.tas.gov.au
Mr. Neil Stump	TRLFA, PO. Box 138, St. Helens, 7216, Tas.	Ph. 03 6376 1041 Fax. 03 6376 1041
Dr. Kevin Williams	CSIRO Cleveland, PO Box 120, Cleveland, 4163, Q.	Ph. 07 3826 7284 Fax. 07 3826 7222 Kevin.Williams@marine.csiro.au
	<b>NEW ZEALAND</b>	
Dr. Alistair MacDiarmid	National Institute of Water and Atmospheric Research, PO Box 14-901, Wellington, New Zealand	Ph: +64 4 386 0300 Fax: +64 4 386 2153 E-mail: a.macdiarmid@niwa.cri.nz
Mr. Graeme Moss	National Institute of Water and Atmospheric Research, PO Box 14-901, Wellington, New Zealand	Ph. +64 4 388 8596 Fax. +64 4 388 9932 Email: g.moss@niwa.cri.nz
Dr. Len Tong	National Institute of Water and	Ph. +64 4 388 8596



	Atmospheric Research, PO Box 14-901, Wellington, New Zealand	Fax. +64 4 388 9932 Email: l.tong@niwa.cri.nz
	<b>JAPAN</b>	
Prof. Jiro Kittaka	School of Fisheries Sciences, Kittasato University, Okkirai, Sanriku-cho, Iwate 022-01, Japan	Ph: +81 1532 8 2152 Fax: + 81 1532 8 2282 suiken96@marimo.or.jp
Mr. Hirokazu Matsuda	Principal Research Scientist Fisheries Research Institute of Mie 3564-3 Hamjima, Shima, Mie 517-0404, Japan	Ph: + 81 5995 3 0016 Fax: + 81 5995 3 2225 E-mail : fishmie@lilac.ocn.ne.jp
Dr. Takeshi Murai	Principal Research Scientist Seikai National Fisheries Research Institute, 49, Kokubu- machi, Nagasaki-shi, Nagasaki 850 Japan	Ph : +81-95-833-2694 Fax : +81-95-821-4494 E-mail : tmurai@snf.affrc.go.jp
Mr. Taku Yoshimura	Chief Scientist, Seikai National Fisheries Research Institute, 49, Kokubu-machi, Nagasaki-shi, Nagasaki 850 Japan	Ph: +81-95-833-2697 Fax : +81-95-821-4494 E-mail : taku@snf.affrc.go.jp

