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FRDC PROJECT 2002/045
January 2007

Fish for the future
Department of Fisheries
Government of Western Australia

Australian Government
Fisheries Research and Development Corporation

ROCK LOBSTER ENHANCEMENT & AQUACULTURE

CURTIN
University of Technology
Western Australia

2002/045 ASSESSING THE POSSIBILITIES FOR THE NATURAL SETTLEMENT OF WESTERN ROCK LOBSTER

Principal Investigator

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OBJECTIVES:

1. To investigate in the laboratory, the number, size and positioning of holes suitable for post-pueruli shelters in an artificial reef environment.

2. To estimate the number of shelters needed to make a measurable impact in a study area, and a preliminary estimate of what would be needed to provide an impact in a regional commercial catch and effort (CAES) reporting area.

3. To design, in conjunction with coastal engineers, suitable pueruli/post-pueruli enhancement structures that could be built in the future to test as a device to enhance local rock lobster populations.

4. To undertake a cost benefit analysis for the various options for enhancing western rock lobster.

NON TECHNICAL SUMMARY:

OUTCOMES ACHIEVED TO DATE

Through a series of laboratory experiments we have determined the hole occupancy patterns of the post-pueruli of the western rock lobster, including preferred distances between holes and the position of holes occupied on the underside, vertical side or upper side of a block. We have also determined how these patterns are affected by the presence of larger post-pueruli lobsters or the presence of fish predators.

Working with engineers, we have designed a system using the data provided from the laboratory results, which can be used to test the enhancement of the pueruli and post-pueruli survival during their first year in the benthic environment. Two different sized devices were designed. The smaller device is approximately 0.25 m high and 0.4 m square, weighing 85 kg and containing 50 holes. Approximately 200 of these devices would be required for a 10,000 hole field test (0.3 hectare).

The larger device is 0.4 m high and 1 m square, weighing 1,000 kg and containing 240 holes. Approximately 42 devices would be required for a 10,000 hole field test. The two types of devices need to be field-tested to confirm which is the most suitable with regard to stability, ease of operation, and effectiveness in terms of the distribution and size of holes.

A cost benefit analysis was impossible to achieve; fixed costs such as the price of concrete for manufacturing the blocks are known, but too many other inputs (such as the manufacture costs of the blocks) to such an analysis are unknown at this time.

A research plan for a field test of the enhancement devices has been developed and will be discussed with industry.

Previous studies have shown that western rock lobster pueruli and post-pueruli shelter in small holes and crevices and that suitable shelter in the inshore environment is limited. It is considered that by providing suitable shelter, the survival of this life-stage could be substantially enhanced, leading to increased production in the wild capture fishery. This project was developed to design suitable shelters, which can be used to test this proposal.

A series of laboratory experiments were conducted to investigate the number, size and positioning of holes that might be suitable for post-pueruli shelters in the marine environment. The study found that:

- The number of post-pueruli in an experiment changed the hole occupation distribution patterns;
- Post-pueruli preferred the under face of the block and preferred to occupy holes in the vertical faces rather than the top of the block;
- Post-pueruli preferred the outer holes on a block face, rather than the central holes;
- Post-pueruli preferred holes further away, rather than closer to, similar sized post-pueruli, but would occupy holes close together rather than no hole at all;
- Smaller post-pueruli preferred the outer holes of a block face rather than those nearer to larger post-pueruli in the centre of the face; and
- The presence of predators did not affect hole occupancy distribution patterns.

From these results, we estimated the number and dimensions of settlement habitats needed for a scientific field test to demonstrate that it is possible to increase survival of the post-pueruli. An area of 0.3 ha was estimated as a suitable sized area for such a demonstration. Based on settlement data that has been collected in previous studies in Western Australia, it is estimated that up to 30,000-pueruli settle per hectare in very good seasons of settlement. This means that suitable settlement habitats should offer at least 30,000 holes per hectare if each hole were to accommodate a puerulus. However, settlement normally occurs over an extended period and therefore not all of the holes need to be of the smallest size (12 mm in diameter and 50 mm deep). This study has assumed that the allocation of holes per hectare of coastline needs to be 15, 000 of 12 mm diameter holes, 10,000 of 20 mm and 5,000 of 30 mm. The holes would be 50, 80 and 120 mm deep, respectively. The distribution of the 12 mm holes will be at least 30 mm from its nearest neighbouring hole, while the larger diameter holes will be 60 and 120 mm from their nearest neighbouring holes.

The project team, in collaboration with Marine Engineers in the Centre for Marine Science and Technology at Curtin University of Technology, has proposed two types of device suitable for field-testing as puerulus survival enhancement devices. Both devices are concrete blocks with holes in the side only.

The smaller of the two block designs would be approximately 0.25 m high and 0.4 m square, weighing 85 kg and containing 50 holes. 200 of such devices would be required for a 10,000 hole (0.3 hectare) trial. The larger device would be approximately 0.4 m high and 1 m square, weighing 1,000 kg and containing 240 holes. 42 of such devices would be required for a 0.3 hectare trial. Both devices are robust, capable of tolerating wave action in depths of less than 10 m, and will be capable of surviving deployment for periods of at least three years before recovery.

A cost-benefit analysis was impossible to achieve; fixed costs such as the price of concrete for manufacturing the blocks are known, but too many other inputs (such as the manufacture costs of the blocks) to such an analysis are unknown.

A research plan for a field test of the enhancement devices has been developed and will be discussed with industry.

KEYWORDS: Pueruli, post-pueruli, enhancement, *Panulirus cygnus*, western rock lobster, enhancement designs

ACKNOWLEDGEMENTS

Dr Kim Klaka, a Senior Research Fellow and Mr Peter Henley, Research Officer, in the Centre for Marine Science and Technology, at Curtin University of Technology for advice and assistance with the design of artificial reefs. Mr Ivan Lightbody of the Department of Fisheries for workshop assistance. Mr Sam Norton for making models of the concrete settlement block structures; Mrs Sandy Clarke, for assistance with computer modifications to some photographs, as well as formatting and printing the final report.

1.0 BACKGROUND

The idea of enhancing rock (spiny) lobster fisheries is not new. Plans for translocation of juveniles, reseeding areas with aquacultured pueruli and post-pueruli, or increasing survival of naturally settling pueruli or post-pueruli by introducing artificial structures, have all been proposed, especially in Japan (see review by Nonaka et al., 2000). Field experiments with *Panulirus argus* have clearly demonstrated that with appropriately designed artificial structures, recruitment to local populations can be increased (Butler and Herrnkind, 1997).

In Western Australia, there has been considerable discussion over the years about the possibility of enhancing the Western Rock Lobster Fishery. These discussions have taken place at the level of the Rock Lobster Industry Advisory Council (RLIAC), the Western Australian Fishing Industry Council (WAFIC), the Rock Lobster Enhancement and Aquaculture Reference Group (RLEAARG), and many times at the level of individual fishers at the annual coastal tours. These discussions have often been focussed on the translocation of young juveniles, but many alternative options have been presented. Unfortunately, although considerable discussion has taken place and research in this area has been identified as a priority, little progress has been made towards achieving this goal.

Several significant studies of the habitat requirements of western rock lobster post-pueruli were undertaken under FRDC funding in the late 1980s (see Fitzpatrick et al., 1989; Jernakoff, 1990 and Jernakoff et al., 1994). These studies had multiple objectives amongst which was the determination of natural and artificial habitat requirements of post-pueruli and an investigation into the suitability of various artificial habitats as a means of enhancing post-puerulus survival. These studies found that post-pueruli sheltered in small holes and crevices of hard surfaces at depths of up to 30 m, while no post-pueruli were found in seagrass beds or sand habitat. It was found that at a carapace length of 16-20 mm, post-pueruli lobsters moved into a reef den/ledge habitat.

More recently, research has been undertaken to investigate how large numbers of pueruli could be harvested for a potential future rock lobster aquaculture industry and to investigate, if harvesting did occur, how biological neutrality could be achieved, so as to avoid negatively affecting egg production in the wild (FRDC Project 1998/302). Whilst it has always been assumed that there is very high mortality of post-pueruli, the modelling work undertaken in Project 1998/302 quantified the extent of that mortality as being somewhere between 80 to 98% within the first year and indicated that as few as 0.9% survive to recruit to the fishery at approximately 4.5 years of age (Phillips et al., 2003). It is believed that one of the reasons for this high mortality is that the non-gregarious pueruli and early post-pueruli stages require suitably sized shelters to protect them from the many inshore predators.

It was suggested (Project 1998/302) that suitable shelter in the inshore environment is limited and that only a very small proportion of the animals that settle are able to secure such shelter. By providing suitable shelters for pueruli and post-pueruli during their first year after settlement, it is highly likely that survival could be substantially enhanced, thereby benefiting the wild fishery through increased production.

This project was developed to design suitable shelters, which can be used to test this proposal. The field tests validating the substrate design and enhancement options were not included within the current project, but are seen as being undertaken as a new project.

1.1 NEED

The Rock Lobster Enhancement and Aquaculture Reference Group to the Department of Fisheries, has recommended the funding for small-scale experiments in rock lobster habitat enhancement as an industry priority.

The Rock Lobster Industry Advisory Committee (RLIAC) has endorsed a timetable and plan for possible research funding for western rock lobster enhancement and aquaculture. The idea behind this plan is to develop the technology, so that if it is thought possible and desirable to undertake these developments, the information will be available for Western Australia to commence these activities. This is one of the projects identified and supported in the plan.

Rock lobster enhancement is one of the priority areas of the FRDC Rock Lobster Enhancement and Aquaculture Subprogram (RLEAS). There is a need to use and build on the available information that exists on the habitat requirements of western rock lobster pueruli and post pueruli so that it can be applied in a way that might lead to enhancing production in the wild fishery. A pre-proposal outlining the proposal assessing the possibilities for enhancing the natural settlement of western rock lobster was endorsed by the RLEAS.

1.2 OBJECTIVES

The objectives of the current project were:

1 To investigate in the laboratory, the number, size and positioning of holes suitable for post-pueruli shelters in an artificial reef environment.

2 To estimate the number of shelters needed to make a measurable impact in a study area, and a preliminary estimate of what would be needed to provide an impact in a regional commercial catch and effort (CAES) reporting area.

3 To design, in conjunction with coastal engineers, suitable pueruli/post-pueruli enhancement structures that could be built in the future to test as a device to enhance local rock lobster.

4 To undertake a cost benefit analysis for the various options for enhancing western rock lobster.

2.0 METHODS and RESULTS

2.1 OBJECTIVE 1 **To investigate in the laboratory, the number, size and positioning of holes suitable for juvenile shelters in an artificial reef environment.**

2.1.1 INTRODUCTION

The puerulus stage is the last nectonic stage, which settles out of the plankton at the end of the oceanic cycle. It is a non-feeding stage; a few days after settlement it moults to the first benthic stage. Post-pueruli and juvenile are terms that are used synonymously, though generally a post-puerulus would be considered to be a very young juvenile, whereas a post-puerulus of a year or older would be considered to be a juvenile. There is no physical characteristic that defines a post-puerulus from a juvenile. The juvenile stage, up to 30 mm carapace length, is referred to as a post-pueruli. Post-pueruli of this size are not gregarious and tend to seek shelter in small holes in hard surfaces.

A series of experiments were conducted to explore the preferences of post-pueruli to occupy holes in limestone blocks. This is necessary because studies of the European spiny lobster, *Palinurus elephas,* in the Mediterranean Sea (Diaz et al., 2004) have shown that in general, the types of artificial reefs usually set out for fish do not provide suitable microhabitat or shelter for post-pueruli lobsters, until they are over one year old juveniles.

Determining the appropriate type of shelter to be deployed is also a requisite step in situations where shelter enhancement may be desirable. Although no published studies have specifically compared results among a suite of potential artificial shelters, the natural shelter preference for a particular species and comparisons among studies employing a single type of shelter will frame an appropriate starting point. Clearly, shelters scaled to the body size of developing lobsters and ones that accommodate the ontogenetic social shift toward aggregation at larger juvenile sizes, will successfully concentrate lobsters and improve their survival (Eggleston et al., 1990, 1992, 1997; Mintz et al., 1994; Arce et al., 1997; Sosa-Cordero et al., 1998). Small, scattered artificial shelters that mimic the types of shelters used by the most vulnerable, small lobsters are most likely to alleviate demographic bottlenecks to recruitment (Butler and Herrnkind, 1997, 2000; Arce et al., 1997; Herrnkind et al., 1997, 1999).

The studies by Fitzpatrick et al. (1989), Jernakoff (1990) and Jernakoff et al. (1994) showed that on limestone reefs, post-pueruli inhabited small holes in the reef surface, mainly on the reef face/top but also in ledges and caves. However, attempts to determine which of the surfaces were the preferred habitats of the post-pueruli were unsuccessful. It was reported that in some months there were significant differences between the densities of pueruli and post-pueruli (of less than 25 mm) on the cave, ledge and reef face habitats, but in other months there were not. On the occasions when they were not equal, the densities in the caves and ledges were higher than on the face. Post-pueruli showed a preference for holes of 12 mm diameter, 50 mm depth, and for shelters covered by seagrass and/or algae (Fitzpatrick et al., 1989).

A series of laboratory studies were initiated to investigate the number, size and positioning of holes suitable for post-pueruli in an artificial reef environment.

In the first year, tests were conducted in the laboratory to determine whether post-pueruli show a preference for:

(i) holes spaced at particular intervals apart (i.e. are there nearest-neighbour effects in the way that post-pueruli position themselves in available holes);

(ii) holes on the upper, lower or side surfaces of artificial reef;

(iii) holes close to those occupied by larger post-pueruli, or to those of similar sized post-pueruli; and

(iv) whether the results of (i), (ii) and (iii) were affected by the introduction of a predator or predators.

2.1.2 METHODS

2.1.2.1 Source of experimental animals

Panulirus cygnus pueruli used in the experiments were collected from sandwich type collectors (Phillips et. al., 2001a) placed in the inshore area (depths <5m) at Seven Mile Beach, near Dongara (Figure 1).

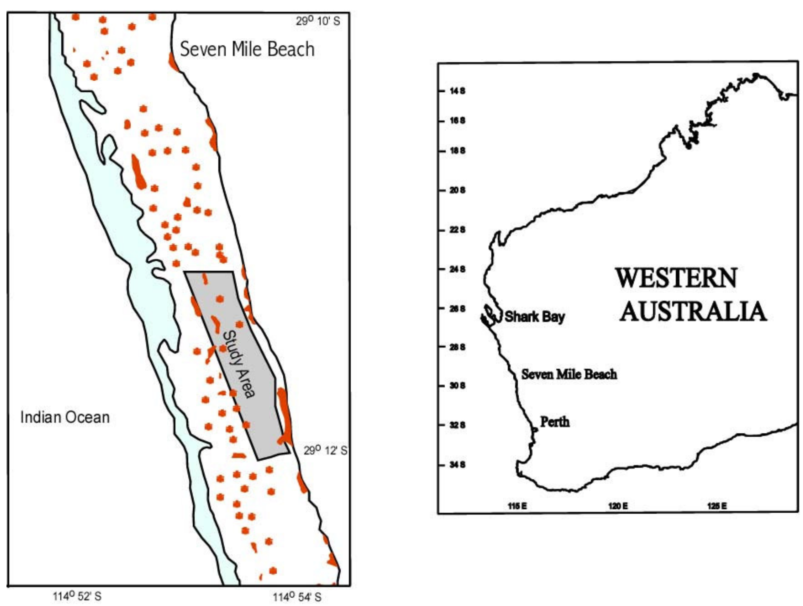


Figure 1. Study area at Seven Mile Beach (near Dongara), Western Australia, where pueruli of *Panulirus cygnus* were collected. Small patches of shallow inshore reef are indicated by hatched dotted areas.

When removed from the collectors, the animals were placed in an insulated container with ambient seawater, an aeration stone and tassels to provide a surface to which the animals could cling during transport back to the laboratory. If the collection and transportation to the research laboratory took more than one day, the animals were fed with mussels.

2.1.2.2 Laboratory set up

On arrival at the research laboratory, the pueruli were held in a 450 L experimental tank with a good water supply (1L/min) but without shelter. Pueruli and post-pueruli remained in this tank and were fed on mussels until they were required to be used in the experiments. Dead and sluggish animals were removed from the holding tank as soon as they were identified.

Limestone blocks, cut from naturally occurring limestone from the Neerabup area near Perth with dimensions of 24 x 24 x 24 cm, were used for the experiments. Holes 12 mm in diameter and 50 mm deep (deep enough for pueruli and post-pueruli to take refuge) were drilled into all sides of the blocks. This configuration limited the maximum number of holes in each side to 25 (5 rows x 5 columns) (Figure 2).

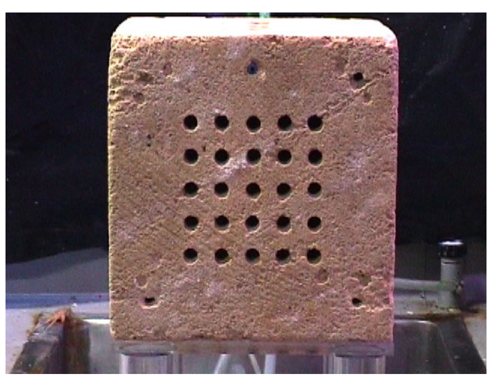


Figure 2. The side of a typical limestone block (24 x 24 x 24 cm) used in the experiments. Each face had 25 holes (5 x 5 formation) drilled to a depth of 50 mm.

Three (replicate), fibreglass tanks (450 L) were used in the experiments. They were supplied with ambient temperature seawater, pumped by a flow through system. Blocks were positioned in their tanks on clear 'Perspex' stands so that the block formation could be placed equidistant between the base, top and sides of the tank. Each tank was covered with 70% shade cloth and kept in a room blocked from all external lighting. Tanks were situated underneath white fluorescent lighting for 12 hours a day (06:00 to 18:00) (Figure 3).

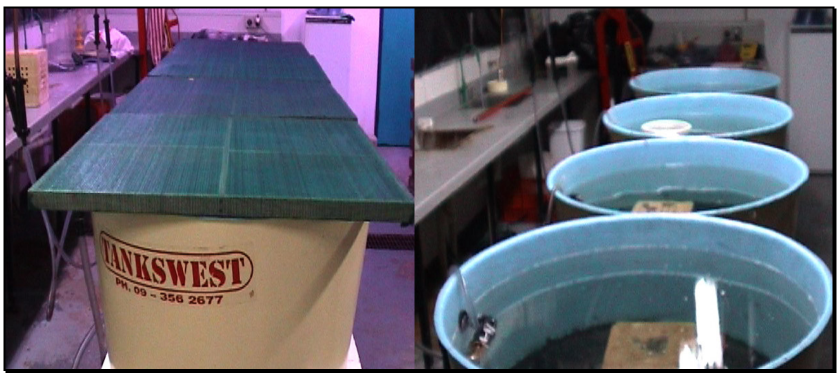


Figure 3. Tanks used in the experiment were covered with 70% shade cloth (left) that was removed when it was necessary to record the location of the pueruli and post-pueruli of *Panulirus cygnus*.

Blocks were positioned as evenly as possible, from the side of the tanks, with the top face of each block 110 mm from the surface and the bottom face 120 mm from the bottom of the tank.

Tank covers were removed each morning and the positions of post-pueruli on the block or on the bottom of the tank were recorded. This included a record of which particular holes they occupied and a count of the number of stragglers (pueruli or post-pueruli that had not occupied a hole). Counting the number of pueruli on the top and vertical faces was easy because of visibility of their antennae; however, it was necessary to use a mirror to check for post-pueruli occupying holes on the bottom side of the block. Mortalities were also recorded where possible, but in most cases the carcases were cannibalised by other animals and therefore could not be recorded.

At various stages in the experiment it was necessary to rotate the blocks in the tank. This was done to investigate whether post-pueruli chose a particular face of the block, regardless of its orientation, or whether they chose particular holes on the face. Rotating of the block was performed after the recording of post-pueruli positions each morning and the post-pueruli were not disturbed.

Over the course of the experiments, hourly water temperatures of the tanks were recorded using a "Tidbit" data logger. All tanks were supplied by the same water source and had the same temperature. Only one tank had a temperature logger. Temperatures recorded for the study period have been summarised as mean, maximum and minimum temperatures each month.

The level of light striking each face of the blocks in each of the three tanks was measured using a "Lutron" Light Meter LX-103.

Pueruli and post-pueruli *P. cygnus* are nocturnal and it was therefore expected that they would move around the tanks at night exploring and foraging. This justifies why the counting of animals in their shelters was conducted in the mornings, as at this time it should reflect the settlement behaviour of pueruli and post-pueruli at the conclusion of the previous nights activities. Post-pueruli used in this experiment were not uniquely identifiable.

Experiments were conducted over a minimum ten-day period. Each of the three experiments conducted used a different block formation.

The three formations used at various times in the three tanks were:

A. A single block with each side having 25 holes (5 rows by 5 columns) (Figure 4A);

B. Two blocks conjoined horizontally. Each of these blocks only had holes in the top and bottom face. One block had a hole formation as depicted in Figure 4A and the other, a hole arrangement with only 9 holes (3 rows x 3 columns) (Figure 4B); and

C. A block with only its vertical sides having holes. The hole arrangement was 25 holes (5 row x 5 column formation) but had two holes drilled with a larger diameter so as to hold larger post-pueruli (Figure 4C).

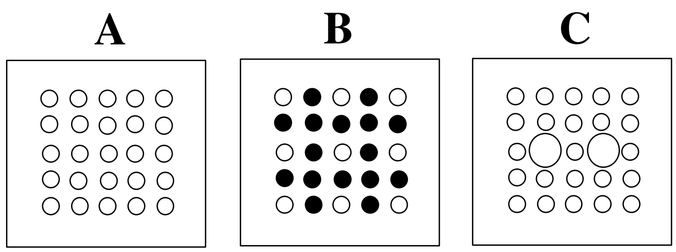


Figure 4. The three different hole arrangements drilled into the faces of limestone blocks used in the experiments. The 'solid' holes indicate that these holes had been filled to prevent occupation.

2.1.2.3 Design of the experiments

Each of the three experiments was designed for a different purpose:

- **Experiment 1** – to test on which face of a limestone block post-pueruli preferred to occupy holes;
- **Experiment 2** – to test if post-pueruli preferred to occupy holes close together or far apart; and
- **Experiment 3** – to test if smaller post-pueruli preferred to occupy holes close to larger post-pueruli, or far apart from them.

At a later stage in each of the three experiments (1C, 2B and 3B) two predators were added to each tank to test whether hole occupancy behaviour of the post-pueruli changed in the presence of a predator(s). The predators added to the tanks were blowfish (banded toadfish) *Torquigener pleurogramm* ranging in size from 105 to 115 mm (head to tail length).

All experiments were replicated in triplicate. The number of pueruli in the tanks at the beginning of the various experiments were not always the same and as the experiment progressed the number of post-pueruli in different tanks was frequently different because of deaths and ‘stragglers’. When these differences occurred, ‘tank’ was included as a factor in performing the analysis of variances (ANOVA) to test whether or not the number of post pueruli in the tank had a significant effect on the post-pueruli hole occupancy behaviour.

A summary of the experiments is presented in Table 1.

Table 1 Summary description of experiments to study the characteristics of pueruli and post-pueruli occupation patterns in artificial habitat structure.

Experiment	Date	P-puerulus added	Date puerulus collected from field	Blocks	Turned	Faces Open	Blow Fish	Large Holes	Post puerulus	Holes open	Food placement
1A	10/9/02 – 23/9/02	Tank 1 (10) Tank 2 (20) Tank 3 (40)	7/09/02	1	N	All	-	-	-	-	Top of tank
1B	24/9/02 – 4/10/02	Tank 1, 2 & 3 (c\o)	7/09/02	1	Y	All	-	-	-	-	Top of tank
1C	2/12/02 – 13/12/02	Tank 1, 2 & 3 (20)	1/12/02	1	Y	All	2	-	-	-	Bottom of tank
2A	28/10/02 – 15/11/02	Tank 1, 2 & 3 (20)	9/10/02	2	Y	Top & bottom only	-	-	-	9 in each face of one block, all in other block	Bottom of tank
2B	18/11/02 – 29/11/02	Tank 1 & 3 (17) Tank 2 (15)	12/11/02	2	N	Top & bottom only	2	-	-	9 in each face of one block, all in other block	Bottom of tank
3A	22/1/03 – 31/1/03	Tank 1, 2 & 3 (40)	7/01/03	1	N	Vertical only	-	8	8	-	Top of tank
3B	3/2/03 – 15/2/03	Tank 1, 2 & 3 (c/o)	7/01/03	1	N	Vertical only	2	8	8	-	Top of tank

Note ‘ – ‘ denotes NA

2.1.3 RESULTS

In this report only those questions relevant to the design of the enhancement apparatus have been addressed. Other questions that arise from the results of the experiments, such as hole occupancy behaviour during the night leading to the eventual selection of a particular hole by the post-pueruli, may be addressed at a later time.

2.1.3.1 Environmental conditions

The monthly mean water temperatures in the tanks showed a steady increase from 16 to 23°C from the winter to the summer (Figure 5).

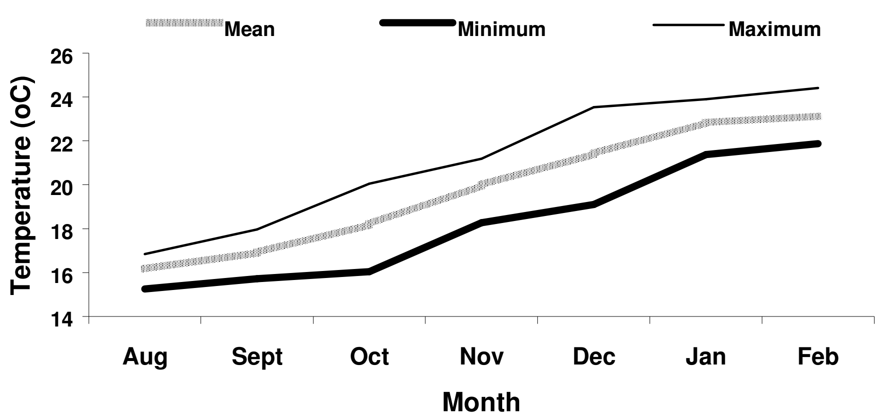


Figure 5. Mean, minimum and maximum water temperatures for the tanks over the course of the experiment.

A summary of the incidence of light (corrected for Fluorescent light) for each face of the block in each tank is shown in Table 2. All sides had similar light intensities, but light intensity on the top and bottom differed substantially due to shading and exposure to the light source that was centred above the tanks.

Table 2 Light intensity measured in lux (corrected for fluorescent light) for each face of the blocks in the three tanks.

Face \ Tank	1	2	3
North	16.8	25.2	6
West	19.2	16.8	22.8
South	19.2	10.8	22.8
East	25.2	20.4	16.8
TOP	43.2	39.6	34.8
Bottom	2.4	1.8	1.2

2.1.3.2 Experiments

2.1.3.2.1 Validation of the data

Due to the complex nature of recording the positioning of the pueruli and post-pueruli in the holes of the dimensional blocks, considerable time was spent validating the data. Plots of stragglers over time were made for each tank and any significant differences in hole occupancy from one day to another were examined in more detail by re-examining original datasheets. The accuracy of the data was improved by calculating the total number of post-pueruli recorded for each tank over time (total in holes + stragglers) and comparing those totals against the numbers recorded as being present at the start of the experiment. Any discrepancy was again balanced against the raw data sheet and if required, an appropriate correction was applied.

2.1.3.2.2 Analysis of the experimental data

The experiments were designed to determine various aspects of post-puerulus hole occupancy behaviour. These aspects were whether post-pueruli preferred a particular face of a block, particular holes on those faces, preferred to settle close together or further apart, and whether the presence of a predator(s) affected their hole occupancy behaviour.

When pueruli were first placed into the tanks it was seen that there was a period of time before they would move into holes of the block (‘initial’ period). It was also noticed in the experiments that after a period of time, the number of post-pueruli residing in holes began to decrease (‘final’ period). This was due to mortality and possibly them losing their instinctive sheltering behaviour over time in their artificial environment.

In analysing the settlement behaviour of post-puerulus it is important to use a period that largely demonstrates the post-puerulus preference to reside in particular holes on particular faces. The term ‘stable’ has therefore been used to describe that part of the data set that was deemed appropriate because of their shelter-seeking behaviour, for use in the analyses. ‘Initial’ and ‘final’ periods were omitted from all analyses.

Time (t) has been included in the following analyses to account for the change in behaviour of post-pueruli in occupying holes in the experimental blocks, as they grew in size. Time refers to the number of days since the pueruli were caught on the collectors.

For each of the performed experiments, three analyses were required to determine various aspects of the post-puerulus hole occupancy behaviour.

Performing the ANOVA and checking the significance of the time factor determined whether or not a repeated measures analysis was required for any of the analyses. If time was not significant, then a repeated measures analysis was not required and the simple ANOVA was sufficient.

2.1.3.2.3 Analysis 1

Did pueruli or post-pueruli prefer a particular side of the block and did they prefer particular holes on those faces?

A logistic model was used to determine whether post-pueruli had a preference for a particular face or hole on a face. The probability that each hole of each face of each block is settled over time was modelled using the following covariates and factors:

- Y_{ijktf} is a binary variable indicating whether hole (i,j) of face f in tank k contained a post-pueruli at time t;
- N_{kt} is the number of post-pueruli in tank k at time t, that are residing in holes (i.e. excluding stragglers);
- t is the number of days since the pueruli were collected from the field;
- F is an effect due to the face type. Face types include vertical, top and bottom;
- P_{kt} is a binary variable indicating whether the tank k had predators (1) or not (0) at time t; and
- $F*Hole_{ijfkt}$ is an effect due to hole (i,j) on the face (vertical, top or bottom).

To solve the GLM for the associated effects, we have used the quasi method. This method simultaneously estimates the dispersion parameter, which is necessary when that parameter is believed to be different to 1. One by one, the most non-significant factor is removed from the model and a reduced ANOVA performed until only significant factors remain.

It should be noted that the face effect was included in the model for 'part A' only, since the other two parts of the experiment only had vertical faces open. It should also be noted that each of the vertical faces of a block was assumed to perform similarly and hence, are represented by one face when presenting related effects of these faces.

2.1.3.2.4 Analysis 2

Do post-pueruli prefer to settle close together or further apart?

The distance that post-pueruli reside from each other (or of smaller post-pueruli from large post-pueruli) was examined in terms of the number of holes between those occupied by smaller post-pueruli and larger post-pueruli. That is, it is said that hole (i',j') is z holes from hole (i,j) if, ($|i-i'| = z$ or $|j-j'| = z$) and ($|i-i'|, |j-j'| \leq z$). This measuring definition is further clarified with Figure 6.

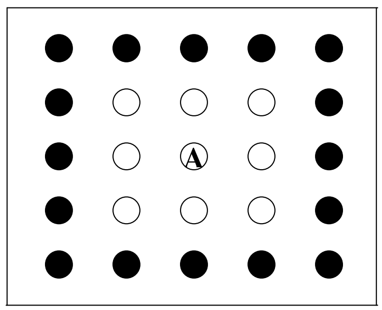


Figure 6. In reference to hole A, the hollow holes are said to be a distance of z = 1 whilst those filled holes are said to be at a distance of z = 2. The above diagram represents one side of a 3 dimensional block.

A logistic model was used to determine whether post-pueruli showed a preference for occupying a hole a particular distance from others. The proportion of holes at a distance *z* from (i,j) that contain post-pueruli (Y_{ijkft}) was modelled using the following covariates and factors:

- Y_{ijzktf} is the proportion of holes at a distance of *z* holes from hole (i,j) in block (tank) *k* at time *t*, to be occupied by post-pueruli;
- N_{kt} is the number of post-pueruli in tank *k* at time *t*, that were residing in holes (i.e. excluding stragglers);
- *t* is the number of days since the post-pueruli were collected from the field;
- *z* is the distance from hole (i,j) (only distances of 1 and 2 holes were considered);

- E_{ij} is the number of holes that (*i,j*) had less than the maximum that a hole can have at a distance *z* (i.e. only holes on the same face were considered and hence, holes closer to the edge of the face had less surrounding holes than those holes closer to the middle of the face);
- P_{kt} is a binary variable indicating whether the tank k had predators (1) or not (0) at time *t*;
- $Hole_{ijfkt}$ is an effect due to hole (*i,j*);
- *F* is an effect resulting from whether the position of the face of the block was vertical, top and bottom; and
- $F*Hole_{ijfkt}$ is an effect due to hole (*i,j*) on the face (vertical, top or bottom).

In the above description only $z \in \{1,2\}$ were considered since every hole in a block face has at least some other holes within these distances; at z>2, this is not true for every hole and could lead to false conclusions from the analyses that followed. It should also be noted that only holes that contained animals were used as observations for this analysis.

As stated previously, the model was fully reduced by removing each non-significant factor one at a time until only significant factors remained and a quasi method was used to solve these GLMs.

Treatment contrasts have been presented for each hole of each face position based on the analyses of each experiment. In this context, treatment contrasts represent the difference in effect for each hole compared to one specific hole, occupied by a post-puerulus at any point in time. These contrasts were then used to calculate the probability of each particular hole being occupied, at any point in time of the experiment, by using an inverse logit equation (SPLUS 2000). The higher the contrast, the higher the probability of a post-puerulus occupying a particular hole at any point in time during the experiment.

Comparisons between the occupancy of holes by post-pueruli were made between faces and within faces using either paired or two-sampled one-tailed t-tests.

2.1.3.2.5 Analysis 3

Does the presence of a predator(s) affect the hole occupancy behaviour of post- puerulus?

This was dealt with by including a binary factor into each of the analyses presented in section 2.1.3.2.3 (i.e. which side and holes do post-pueruli prefer) to account for the effect of adding a predator; 0 indicating that no predator was present and 1, that predators were present.

2.1.3.3 Experiment 1

A single block with all faces having the maximum number of holes (Figure 4A). This experiment allows for the testing of the various aspects of post-puerulus hole occupancy with all face types being available (top, vertical and under).

2.1.3.3.1 Analysis 1

Referring firstly to that part of the experiment that had no predators (Figure 7), it can be seen that the post-puerulus hole occupation was ‘stable’ from Day 7 to Day 20. Prior to Day 7, many of the animals are likely to have been pueruli. By Day 7 all may be assumed to have moulted to post-pueruli.

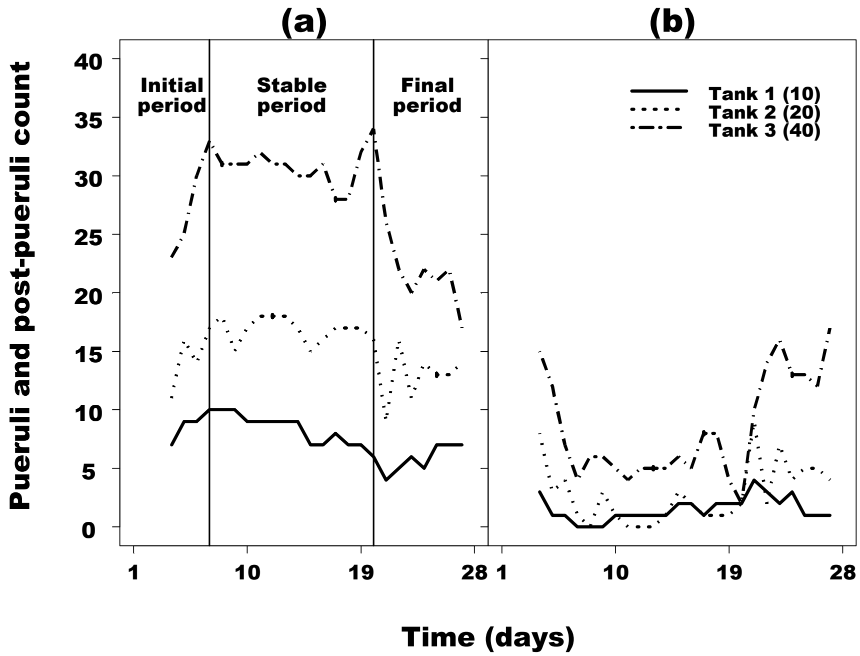


Figure 7. Plot of the number of pueruli and post-pueruli of *Panulirus cygnus* occupying holes in the block in each tank (a) or straggled (b) over time, for 'Experiment 1' that included no predators (part 1A & 1B). Time refers to the number of days since the pueruli were collected from the field. The numbers of pueruli/post-pueruli in each tank at the start of the experiment are presented in brackets.

Restricting data to the 'stable' period (days 7 to 20), the ANOVA for testing the significance of various factors on whether or not a post-puerulus settles in a particular hole is presented in Table 3. Due to the unequal number of puerulus in each tank at the beginning of the experiment, tank has also been included as a factor.

Table 3 ANOVA estimating the probability of whether a particular hole was occupied on either the vertical, bottom or top faces in Tanks 1 – 3. The data set is restricted to that period in which there were no predators and unequal numbers of post-puerulus in the tanks.

	Df	Deviance	Resid. Df	Resid. Dev	Pr(Chi)
Holes	1	228.07	6298	4525.07	< 0.01
Time	13	0.58	6285	4524.49	0.99
Face	2	272.57	6283	4251.91	< 0.01
Tank	2	4.64	6281	4247.28	0.10
Particular holes	24	141.89	6257	4105.38	< 0.01
Face:Tank	4	32.28	6253	4073.10	< 0.01
Face:Particular holes	48	96.17	6205	3976.93	< 0.01
Tank:Particular holes	48	69.03	6157	3907.90	0.02
Tank:Face:Particular holes	96	151.76	6061	3756.14	< 0.01

The non-significant factors in the model in Table 3 were removed and the fully reduced model is presented in Table 4.

Table 4 ANOVA estimating the probability of whether a particular a hole was occupied on either the vertical, bottom, or top faces in Tanks 1 – 3. The data set is restricted to that period in which there were no predators in the tanks and an unequal number of post-pueruli in the tanks. A fully reduced model is presented.

	Df	Deviance	Resid. Df	Resid. Dev	Pr(Chi)
Holes	1	228.07	6298	4525.07	< 0.01
Face	2	272.57	6296	4252.50	< 0.01
Tank	2	4.61	6294	4247.88	0.10
Particular holes	24	141.89	6270	4106.00	< 0.01
Face:Tank	4	32.26	6266	4073.74	< 0.01
Face:Particular holes	48	96.17	6218	3977.56	< 0.01
Tank:Particular holes	48	69.00	6170	3908.56	0.03
Tank:Face:Particular holes	96	151.69	6074	3756.87	< 0.01

The estimated effect of each face of the block in each tank is presented in Table 5. The estimated effect for each hole of each face for the optimal model associated with Table 5 is presented in Figure 8.

Table 5 Contrasts for the face blocks in tanks containing 10, 20 and 40 post-pueruli.

Post-Pueruli per tank	**Face**	**Value**	**s.e.**
10	Top	0	NA
	Under	1.29	0.33
	Vertical	0.16	0.31
20	Top	-2.01	0.69
	Under	2.00	0.39
	Vertical	0.37	0.39
40	Top	-0.46	0.74
	Under	1.39	0.72
	Vertical	0.45	0.72

10 post-pueruli/tank					20 post-pueruli/tank					40 post-pueruli/tank				
Top					**Top**					**Top**				
0 (NA)	-15.4 (797.2)	-15.4 (797.2)	-15.4 (797.2)	-15.4 (797.2)	-1.3 (1.2)	-16 (797.3)	-16 (797.3)	-16 (797.3)	-1.3 (1.2)	-0.6 (1.1)	-2.3 (1.3)	-16.9 (797)	-2.3 (1.3)	-1 (1.1)
-15.4 (797.2)	-0.8 (1.2)	-15.4 (797.2)	-15.4 (797.2)	-15.4 (797.2)	-16 (797.3)	-16 (797.3)	-16 (797.3)	-16 (797.3)	-16 (797.3)	-16.9 (797)	-16.9 (797)	-2.3 (1.3)	-2.3 (1.3)	-2.3 (1.3)
-15.4 (797.2)	-15.4 (797.2)	-15.4 (797.2)	-0.8 (1.2)	-15.4 (797.2)	-16 (797.3)	-16 (797.3)	-1.3 (1.2)	-16 (797.3)	-16 (797.3)	-16.9 (797)	-2.3 (1.3)	-1 (1.1)	-1.5 (1.2)	-1.5 (1.2)
-15.4 (797.2)	-15.4 (797.2)	-15.4 (797.2)	-15.4 (797.2)	-15.4 (797.2)	-16 (797.3)	-16 (797.3)	-16 (797.3)	-16 (797.3)	-16 (797.3)	-16.9 (797)	-2.3 (1.3)	-16.9 (797)	-16.9 (797)	-1.5 (1.2)
-15.4 (797.2)	-15.4 (797.2)	-15.4 (797.2)	-15.4 (797.2)	2.4 (0.9)	-16 (797.3)	-16 (797.3)	-16 (797.3)	-16 (797.3)	-16 (797.3)	-1.5 (1.2)	-16.9 (797)	-1.5 (1.2)	-1.5 (1.2)	-1.5 (1.2)
Under					**Under**					**Under**				
-0.8 (1.2)	-0.8 (1.2)	-15.4 (797.2)	0.9 (0.9)	0.9 (0.9)	0.9 (0.9)	0.6 (0.9)	0.6 (0.9)	0.3 (0.9)	0.6 (0.9)	0.6 (1)	-1 (1.1)	0.3 (1)	0.3 (1)	-0.3 (1.1)
-15.4 (797.2)	-0.8 (1.2)	0.9 (0.9)	0 (1)	-0.8 (1.2)	0.9 (0.9)	-0.6 (1)	0.6 (0.9)	-0.1 (0.9)	1.5 (0.9)	-1 (1.1)	0.6 (1)	-1.5 (1.2)	0.3 (1)	0.6 (1)
-15.4 (797.2)	-0.8 (1.2)	0 (1)	-15.4 (797.2)	-0.8 (1.2)	-0.6 (1)	0.6 (0.9)	-0.6 (1)	0.3 (0.9)	-0.1 (0.9)	0.3 (1)	-1 (1.1)	-0.3 (1.1)	0 (1)	-1 (1.1)
-0.8 (1.2)	1.8 (0.8)	0.9 (0.9)	-15.4 (797.2)	0 (1)	0.3 (0.9)	-1.3 (1.2)	-0.1 (0.9)	0.3 (0.9)	1.2 (0.9)	-0.6 (1.1)	-0.3 (1.1)	-1 (1.1)	-0.3 (1.1)	-1 (1.1)
-0.8 (1.2)	-0.8 (1.2)	-0.8 (1.2)	0 (1)	0 (1)	1.8 (0.9)	0.9 (0.9)	-0.6 (1)	0.9 (0.9)	0.9 (0.9)	0.3 (1)	0 (1)	-0.3 (1.1)	-0.6 (1.1)	0.9 (1.1)
Vertical					**Vertical**					**Vertical**				
0.3 (0.8)	-15.4 (398.6)	-2.2 (1.1)	-2.2 (1.1)	-0.8 (0.8)	-0.1 (0.8)	-2.1 (1)	-0.9 (0.8)	-0.9 (0.8)	-0.2 (0.8)	-0.8 (1)	-1.6 (1)	-0.8 (1)	-1.1 (1)	-0.4 (1)
-2.2 (1.1)	-15.4 (398.6)	-15.4 (398.6)	-2.2 (1.1)	0 (0.8)	-2.1 (1)	-2.8 (1.2)	-2.1 (1)	-1.6 (0.9)	-1.6 (0.9)	-0.8 (1)	-2.3 (1)	-1.8 (1)	-3 (1.1)	-1.5 (1)
0.1 (0.8)	-15.4 (398.6)	-1.1 (0.9)	-15.4 (398.6)	-15.4 (398.6)	-0.6 (0.8)	-16 (398.7)	-1.1 (0.8)	-1.6 (0.9)	-1.1 (0.8)	-0.4 (1)	-1.8 (1)	-1.1 (1)	-1.6 (1)	-0.1 (1)
-2.2 (1.1)	-2.2 (1.1)	-15.4 (398.6)	-15.4 (398.6)	-1.5 (0.9)	-2.1 (1)	-0.9 (0.8)	-16 (398.7)	-2.1 (1)	-0.6 (0.8)	-1.6 (1)	-2.6 (1.1)	-1.1 (1)	-1.2 (1)	-1.4 (1)
0 (0.8)	-2.2 (1.1)	-2.2 (1.1)	-0.2 (0.8)	-1.5 (0.9)	-0.7 (0.8)	-0.7 (0.8)	-0.7 (0.8)	-0.9 (0.8)	-0.9 (0.8)	-0.4 (1)	-0.7 (1)	-1.2 (1)	-1.1 (1)	-0.6 (1)

Figure 8. Probability coefficients for post-pueruli of *Panulirus cygnus* occupying a specific hole on a particular face of blocks in tanks with 10, 20 and 40 post-pueruli/tank using the optimal model presented in Table 5.

There was a tendency for post-pueruli to occupy holes in the outermost holes of each face rather than the central holes (Figure 9). The results of a one-tailed two-sample t-test showed that outer hole contrasts were greater than that of inner holes (the greater the contrast the higher the probability) for each face of blocks in tanks with 10, 20 and 40 animals per tank (Table 6).

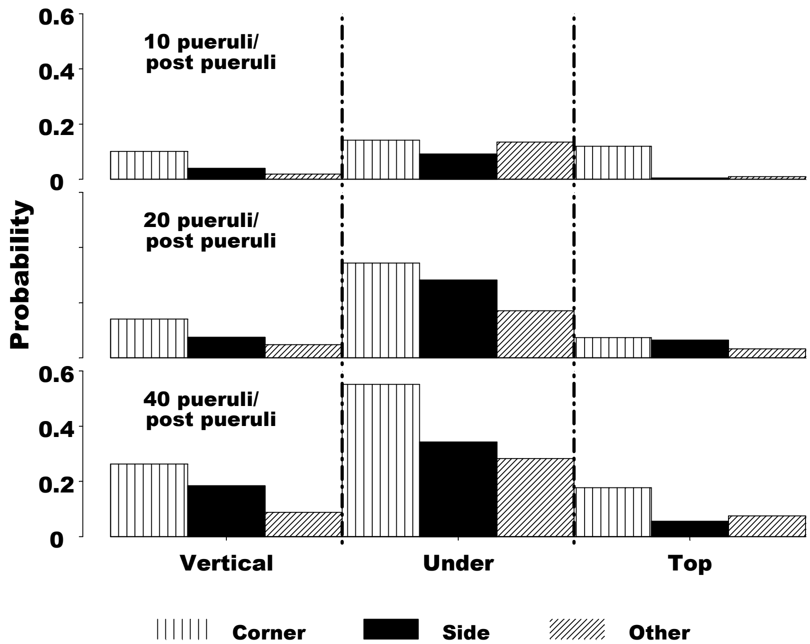


Figure 9. Probability of corner, side, or central holes being occupied by post-puerulus of *Panulirus cygnus* at any stage of experiments 1 A & B, for each face of the blocks in tanks with 10, 20 and 40 animals.

Table 6 P-values for a one-tailed, two-sample t-test comparing the probability contrasts between inner and outer holes of post-pueruli occupying holes in each face of blocks in tanks with densities of 10, 20 and 40 per tank.

Pueruli per tank	Face	p
10	Top	0.68
	Under	0.50
	Vertical	< 0.01
20	Top	0.46
	Under	< 0.01
	Vertical	0.02
40	Top	0.43
	Under	0.20
	Vertical	< 0.01

2.1.3.3.2 **Analysis 2**

Referring to that part of ‘Experiment 1’ that included two predators (part C) (Figure 10), it can be seen that the number of post-pueruli that settled in each block in each tank was ‘stable’ at day 3 onwards. It is considered that most of the animals would have been pueruli during the first two days of the experiment.

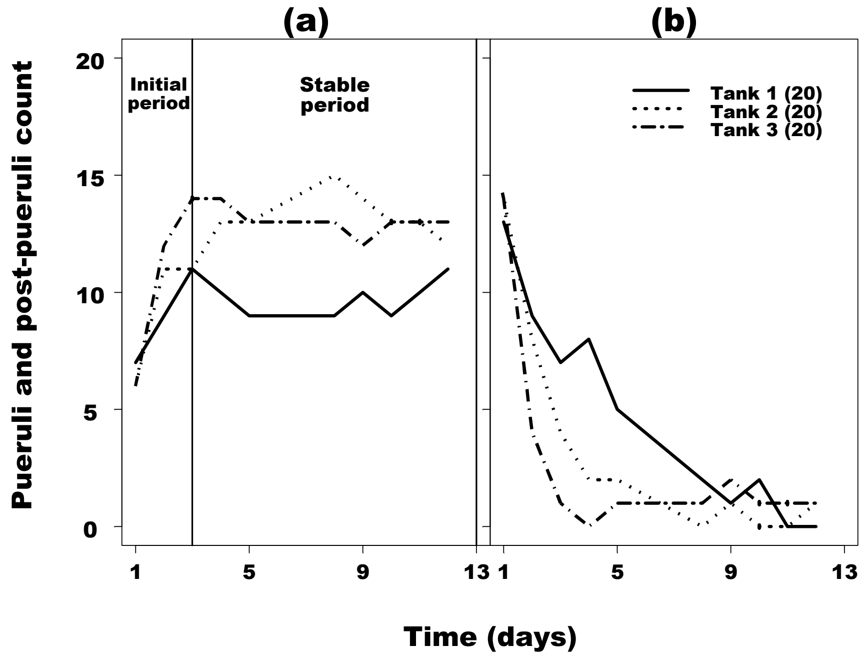


Figure 10. Number of pueruli or post-pueruli of *Panulirus cygnus* that occupied holes in their respective blocks in each tank (a) or straggled (b) over time, for that part of 'Experiment 1' that included two predators (part C). Time refers to the number of days since the pueruli were collected in the field. The numbers of pueruli/post-pueruli in each tank at the start of the experiment are presented in brackets.

From section 2.1.3.2.5.2 it was seen that the number of puerulus in the tanks at the beginning of the experiment, had a marginally significant (p=0.10) effect on the post-puerulus settlement behaviour. Therefore, to study the effect of adding predators to each tank attention was restricted to only those tanks that had similar numbers of post-pueruli. Hence, we include all the data associated with only Tank 3 (i.e. 40 post-pueruli) so that settlement preferences could be thought of in terms of what settlement distribution post-pueruli would take on. Secondly, two separate analyses, one each for when there were and were not any predator(s), were performed to test if time was a significant factor on settlement distance. Time was seen to not be significant for when a predator(s) did and did not exist (p = 0.99 for both). Hence, time has been removed from further consideration.

The full analysis for testing the preferred distance of pueruli to settle from other similarly sized animals, with and without predators is presented in Table 7.

Table 7 ANOVA for the proportion of holes at a distance z, for each hole that contained a post-puerulus. Only Tank 3 data has been used in this analysis.

	Df	Deviance	Resid. Df	Resid. Dev	Pr(Chi)
Post-Puerulus Count In Holes	1	0.25	1112	124.74	0.62
Time	1	2.44	1111	122.30	0.12
Distance from hole	2	5.37	1109	116.92	0.07
Number of holes at distance z less than maximum?	1	0.16	1108	116.76	0.69
Predator	23	14.17	1085	102.59	0.92
Particular hole	2	29.22	1083	73.37	< 0.01
Face	22	1.14	1061	72.23	1.00
Predator:Particular hole	2	0.20	1059	72.03	0.90
Predator:Face	41	16.19	1018	55.84	0.99
Face:Particular hole	21	0.85	997	54.99	0.99
Predator:Face:Particular hole	1	0.25	1112	124.74	0.62

The most non-significant factors in Table 7 have been sequentially removed from the analysis and the ANOVA rerun until no non-significant factors remained. A fully reduced model is presented in Table 8.

Table 8 Fully reduced ANOVA for the proportion of holes at a distance z, for each hole that contains a post-puerulus.

Factor	df	Deviance	Resid df	Resid dev.	p
Face	2	31.31	1111	93.69	< 0.01

There was no particular difference in the distance that post-pueruli settled from each other (Table 7, p = 0.12). The only factor that was statistically significant was the choice of face settled (Table 8, p < 0.01). By studying the contrasts for each face, it is seen that post-pueruli prefer holes on the under face (contrast = 14.91, s.e. = 121.75) rather than those on the vertical (contrast =13.41, s.e. = 121.75) or top face (contrast = 0, s.e. = NA).

2.1.3.3.3 Analysis 3

From section 2.1.3.2.5.2 it was seen that the number of pueruli in the tanks on the beginning of the experiment, had a marginally significant (p=0.10) effect on the post-puerulus settlement behaviour. Therefore, to study the effect of adding predators to each tank we restricted our attention to only those tanks that had similar numbers of post-pueruli. Hence, we include all the data associated with only Tank 3. Secondly, two separate analyses, one each for when there were and were not any predator(s), were performed to test if time was a significant factor on settlement distance. Time was seen to not be significant for when a predator(s) did and did not exist (p = 0.99 for both). Hence, time has been removed from further consideration. The ANOVA for the restricted data set is presented in Table 9.

Table 9 ANOVA estimating the probability of whether a particular hole was occupied on either the vertical, bottom or top faces.

Factor	Df	Deviance	Resid. Df	Resid. Dev	Pr(Chi)
Post-Puerulus Count In Holes	1	89.93	3298	2851.37	< 0.01
Face	2	157.70	3296	2693.67	< 0.01
Predator	1	0.19	3295	2693.48	0.66
Particular hole	24	91.93	3271	2601.54	< 0.01
Face:Predator	2	9.46	3269	2592.09	0.01
Face:Particular hole	48	66.81	3221	2525.28	0.04
Predator:Particular hole	24	23.29	3197	2501.99	0.50
Predator:Face:Particular hole	48	41.79	3149	2460.20	0.72

The most non-significant factors were sequentially removed from the analysis and the ANOVA rerun until no non-significant factors remained. The results are presented in Table 10.

Table 10 A fully reduced ANOVA estimating the probability of whether a particular hole was occupied on either the vertical, bottom or top faces.

Factor	df	deviance	Resid df	Resid dev.	p
In Holes	1	89.93	3298	2851.37	< 0.01
Face	2	157.70	3296	2693.67	< 0.01
Particular hole	24	91.94	3272	2601.73	< 0.01
Face:Particular hole	48	66.99	3224	2534.75	0.04

2.1.3.4 Experiment 2

Two conjoined blocks with only the top and under faces with holes; one block with the maximum number of holes (Figure 4A) and the other with fewer but further distantly spaced holes (Figure 4B). As well as the other aspects of post-puerulus settlement, this experiment allows for further testing of the distance preference of post-pueruli to settle closer or further to other similarly sized animals.

2.1.3.4.1 Analysis 1 & 3

In both parts of this experiment for when there was no predator(s) (part A) and that part that had a predator(s) (part B), the number of pueruli starting in each tank were similar and in the range of 15 – 20 pueruli. It was therefore decided to combine analyses 1 and 2.

Using data when no predator was present, in Experiment 2A (Figure 11), it can be seen that the number of post-pueruli that occupied holes in each block in each tank was 'stable' over the period day 22 through to day 35.

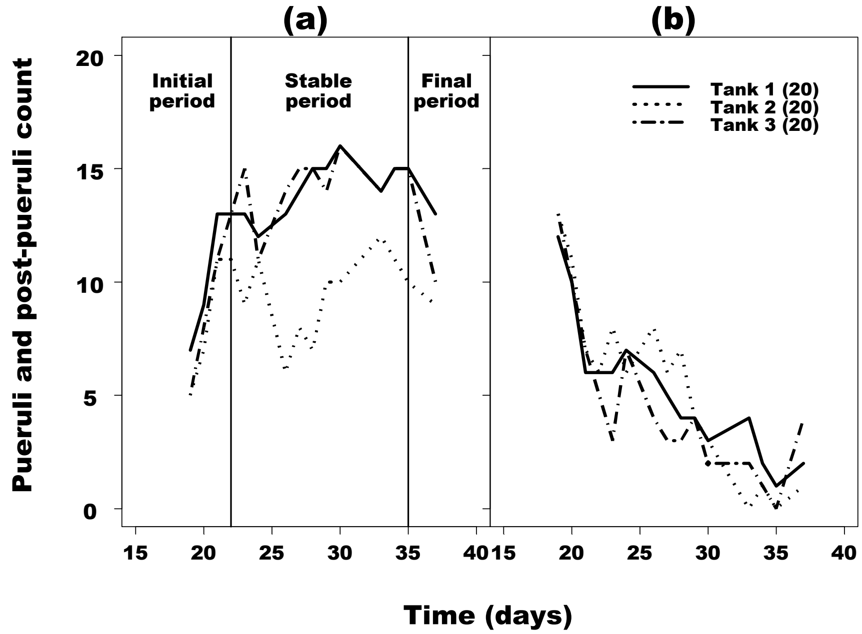


Figure 11. Number of pueruli and post-pueruli of *Panulirus cygnus* that occupied holes in the block in (a) each tank, or (b) straggled, over time. The data relates to that part of Experiment 2 part B during which there were no predators. Time refers to the number of days from when the pueruli were collected in the field. The numbers of pueruli/post-pueruli in each tank at the start of the experiment are presented in brackets.

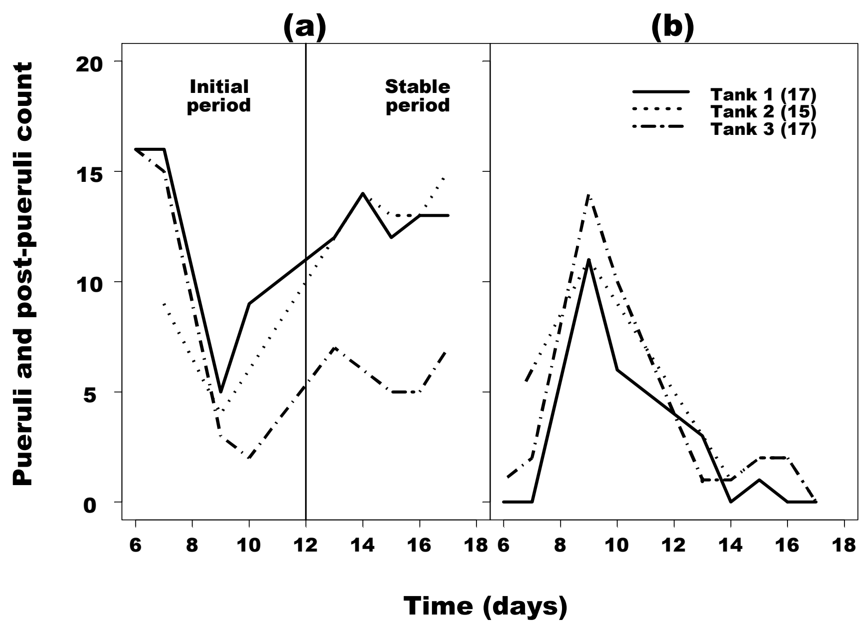


Figure 12. Number of pueruli and post-pueruli of *Panulirus cygnus* that have occupied holes in the block in (a) each tank or (b) straggled, over time. The data relates to that part of Experiment 2 part B during which two predators were present. Time refers to the number of days from when the pueruli were collected in the field. The numbers of pueruli/post-pueruli in each tank at the start of the experiment are presented in brackets.

In that part of the experiment during which there were two predators (Figure 12), the number of post-pueruli that occupied holes in each block in each tank was 'stable' from day 12 onwards.

In combining both parts of Experiment 2 together, it is seen that the effects of time and adding a predator(s) are confounding (the time periods for when there were and were no predators are non-overlapping). Performing two separate analyses, one each for when there were and were not any predator(s), it was seen that time was not a significant factor ($p = 0.99$ for both). Assuming that the effect of time is not significantly different for when there were and were no predators, an ANOVA testing the probability of a hole being occupied using data that is considered stable for when there is and is not a predator, is presented in Table 11.

Table 11 ANOVA testing the probability of a particular hole on the top, vertical or bottom faces being occupied in Tanks 1 – 3.

Factor	Df	Deviance	Resid. Df	Resid. Dev	Pr(Chi)
Post-Puerulus Count in Holes	1	49.93	3262.00	2998.26	< 0.01
Face:Particular hole	49	296.16	3213.00	2702.10	< 0.01
Predator	1	0.01	3212.00	2702.09	0.93
Block	1	30.47	3211.00	2671.63	< 0.01
Predator: Face:Particular hole	49	70.40	3162.00	2601.22	0.02
Block: Face:Particular hole	17	23.16	3145.00	2578.06	0.14
Predator: Block	1	0.09	3144.00	2577.97	0.77
Predator:Block: Face:Particular hole	17	21.49	3127.00	2556.48	0.21

The most non-significant factors in Table 11 have been sequentially removed from the analysis and the ANOVA rerun until no non-significant factors remained. A fully reduced model is presented in Table 12. Note that since the main effect and all but one of the interactions involving predator are non-significant, it was decided to drop predator from the model altogether under the assumption that it is unlikely in this experiment for the main effect of predator to not be significant but one of its interactions is.

Table 12 Fully reduced ANOVA testing the probability of whether a hole was occupied, on the vertical, bottom and top faces for Tanks 1 – 3.

Factor	Df	Deviance	Resid. Df	Resid. Dev	Pr(Chi)
Post-Puerulus Count in Holes	1	49.93	3262.00	2998.26	< 0.01
Face:Particular hole	49	296.16	3213.00	2702.10	< 0.01
Block	1	30.47	3212.00	2671.63	< 0.01

The estimated parameters for each hole of each face for the optimal model associated with Table 12 are presented in Figure 13.

Top **Block 1**

0 (NA)	-1.2 (0.6)	-1.5 (0.7)	-1.5 (0.7)	-1.5 (0.7)
-1 (0.6)	-13 (105.7)	-1.5 (0.7)	-2.7 (1.1)	-1.5 (0.7)
-1 (0.6)	-2.7 (1.1)	-1.9 (0.8)	-1.2 (0.6)	-1.5 (0.7)
-1.2 (0.6)	-1 (0.6)	-1.9 (0.8)	-1.5 (0.7)	-1.9 (0.8)
0.1 (0.5)	-1.2 (0.6)	-0.6 (0.5)	-1.5 (0.7)	-0.3 (0.5)

Block 2

0.5 (0.5)		-0.4 (0.5)		-1.5 (0.7)
0 (0.5)		-1 (0.6)		-1 (0.6)
0.4 (0.5)		-1.5 (0.7)		-0.4 (0.5)

0.6 (0.5)	-0.6 (0.5)	-0.6 (0.5)	-0.1 (0.5)	0.2 (0.5)
-1 (0.6)	-0.1 (0.5)	-0.7 (0.6)	-0.7 (0.6)	-0.4 (0.5)
0 (0.5)	-1 (0.6)	-0.7 (0.6)	-0.6 (0.5)	0.2 (0.5)
-0.4 (0.5)	-0.4 (0.5)	-0.3 (0.5)	-1.9 (0.8)	-1 (0.6)
0.2 (0.5)	-0.3 (0.5)	-0.3 (0.5)	0.4 (0.5)	0.4 (0.5)

0.8 (0.5)		1.2 (0.5)		1.2 (0.5)
0.4 (0.5)		0.8 (0.5)		1.1 (0.5)
0.9 (0.5)		1 (0.5)		0.5 (0.5)

Figure 13. Modelling the output of the probability coefficient of a *Panulirus cygnus* post-puerulus occupying a particular hole on a particular face. The coefficients have been taken from the optimal model presented in Table 12.

A one tailed (outer hole contrasts are greater than the innermost holes), two-sample t-test was used to test whether the apparent tendency for post-pueruli to settle in the outermost holes of each face (Figure 13) was significant. The results of that analysis are presented in Table 13.

Table 13 P-values for a one tailed two-sample t-test comparing the occupation of inner and outer holes for each face of the two blocks, as well as comparing the bottom face effects to the top face.

Comparison	Face	Block	p
Outer > Inner	Top	-	< 0.01
	Under	-	< 0.01
Under > Top	-	25 holes / face	< 0.01
	-	9 holes / face	< 0.01

2.1.3.4.2 Analysis 2

In this experiment it is not necessary to perform the analysis (analysis 2) previously outlined, due to the existence of two blocks in each tank that offer different settlement options to post-pueruli (one block with many holes allows post-pueruli to occupy holes close, whilst the other block with only a few holes only allows post-pueruli to occupy holes further apart). Instead, to determine whether post-pueruli prefer to settle close together or far apart, a one tailed (contrasts of farther spaced holes being greater than those same positioned holes of the closer spaced hole face), paired t-test was performed for the holes of both block types, for each face type (see Table 14).

Table 14 P-values for a one tailed (contrasts of farther spaced holes being greater than those same positioned holes of the closer spaced hole face) paired t-test, for each face of the two block types.

Face	p
Top	0.07
Under	< 0.01

In terms of block, independent of face, post-pueruli preferred the block with the wider spaced holes (contrast = 0.66, s.e. =0.12) to that with the closer spaced holes (contrast = 0, s.e. = NA).

2.1.3.5 Experiment 3 (vertical faces open, 8 large post-pueruli)

A single block with only vertical faces with holes. These faces allow for the settlement of two larger animals (Figure 4C). Only this experiment allows for the testing of the preference of smaller post-pueruli settling close or far from other larger animals. Particular hole preferences and the effects of adding a predator are also analysed for this experiment, as they were with Experiments 1 and 2.

2.1.3.5.1 Analysis 1 & 3

As with Experiment 2, it was also decided to combine analyses 1 and 3.

Holes on the top and bottom of the block were sealed leaving only those on the vertical faces available for settlement. In the first half of this experiment, no predators were in the tank; for the second half, two predators were added. The number of post-pueruli that occupied each block is presented (Figure 14).

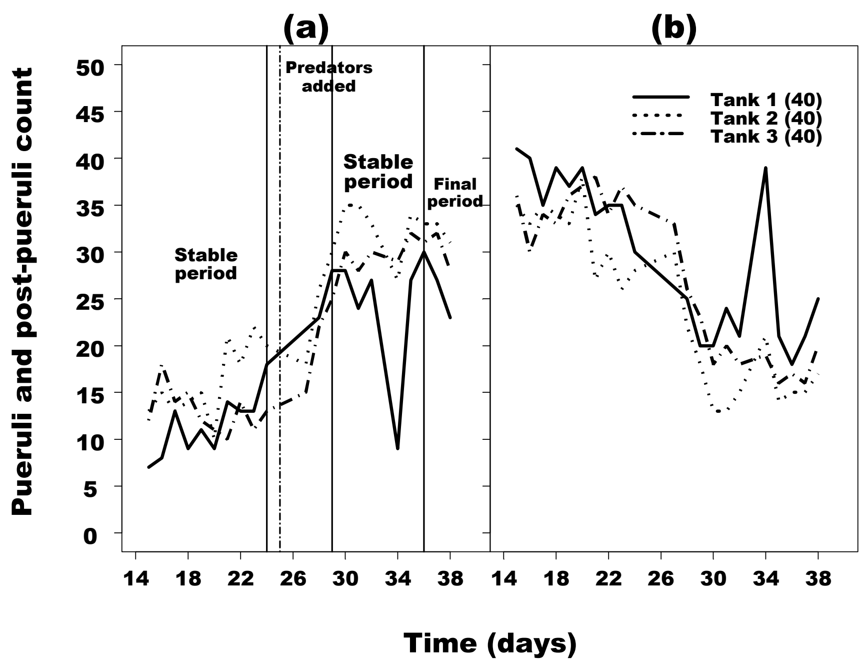


Figure 14. Number of post-pueruli of *Panulirus cygnus* that occupied (a) holes in the block and (b) those that straggled, in each tank, over time. That part of the experiment that did and did not have predators is identified. Time refers to the number of days from when the pueruli were collected in the field. The numbers of pueruli in each tank at the start of the experiment are presented in brackets.

When there was no predator(s), the whole period was considered ‘stable; when there were predator(s) only days 29 through to 36 were considered ‘stable’.

In combining both parts of Experiment 3 together, it is seen that the effects of time and adding a predator(s) are again confounding. Performing two separate analyses, one each for when there were and were not any predator(s), it was seen that time was not a significant factor ($p = 0.92$ and $p = 0.99$, respectively). Assuming that the effect of time is not significantly different for when there were and were no predators, an ANOVA testing the probability of a hole being occupied using data that is considered stable for when there is, and is not, a predator, is presented in Table 15.

Table 15 ANOVA testing for the probability that a hole was occupied or not for each particular hole of the vertical faces.

Factor	Df	Deviance	Resid. Df	Resid. Dev	Pr(Chi)
Post-Puerulus Count In Holes	1	270.12	5098.00	3838.67	< 0.01
Particular hole	24	421.23	5074.00	3417.43	< 0.01
Predator	1	0.10	5073.00	3417.33	0.75
Predator:Particular hole	24	25.56	5049.00	3391.76	0.38

The most non-significant factors in Table 15 have been sequentially removed from the analysis and the ANOVA rerun until no non-significant factors remained. A fully reduced model is presented in Table 16.

Table 16. Fully reduced ANOVA estimating the probability of whether a particular hole was occupied on the vertical faces.

Factor	df	deviance	Resid df	Resid dev.	p
Post-Puerulus Count In Holes	1	270.12	5098.00	3838.67	< 0.01
Particular hole	24	421.23	5074.00	3417.43	< 0.01

The estimated parameters for each hole of each face for the optimal model associated with Table 16 are presented in Figure 15.

Vertical

0 (NA)	-0.8 (0.3)	-1.2 (0.3)	-0.7 (0.2)	0 (0.2)
-0.7 (0.2)	-2.4 (0.4)	-2.7 (0.4)	-3.2 (0.5)	-0.8 (0.3)
-1.2 (0.3)	-1.8 (0.3)	-14.4 (80.5)	-2.3 (0.4)	-1 (0.3)
-0.8 (0.3)	-2.1 (0.3)	-2 (0.3)	-2.4 (0.4)	-0.8 (0.3)
-0.2 (0.2)	-0.8 (0.3)	-1.1 (0.3)	-0.3 (0.2)	0.3 (0.2)

Figure 15. Modelling output of the probability coefficients of a post-puerulus of *Panulirus cygnus* occupying a particular hole. The coefficients have been taken from the optimal model presented in Table 16.

A one-tailed (outer hole contrasts are greater than the innermost holes), two-sample t-test was performed to test whether the apparent tendency for post-pueruli to settle in the outermost holes of each face rather than those to the inside was significant. The test showed that the outer holes were more preferable than the inner ($p < 0.01$).

2.1.3.5.2 Analysis 2

An analysis was performed to test if there was a preference of post-pueruli to occupy holes close to or far from other larger post-puerulus. Firstly, two separate analyses, one each for when there were and were not any predator(s), were performed to test if time was a significant factor. Time was seen to not be significant for when a predator(s) did and did not exist ($p = 0.99$ for both). Assuming that the effect of time is not significantly different between the periods that there were and were no predators, an ANOVA testing the probability of a hole being occupied using data that is considered stable for when there is and is not a predator, is presented in Table 11. The ANOVA for testing the distance preference for post-pueruli to settle from other larger post-puerulus is presented in Table 17.

Table 17 ANOVA testing the distance that smaller post-pueruli prefer to settle from other larger post-pueruli by using the proportion of holes at a distance z that contained puerulus, for each hole that was occupied.

	Df	Deviance	Resid. Df	Resid. Dev	Pr(Chi)
Post-Puerulus Count In Holes	1	0.00	560.00	38.93	0.95
Distance from hole	1	2.20	559.00	36.73	0.14
Number of holes at distance z less than maximum?	2	0.11	557.00	36.63	0.95
Particular hole	16	11.87	541.00	24.76	0.75
Predator	1	0.00	540.00	24.75	0.97
Predator: Particular hole	3	0.36	537.00	24.39	0.95

From Table 17 it can be see that post-pueruli show no tendency to settle close or further from other larger post-pueruli (p = 0.14).

2.1.4 SIGNIFICANCE OF LABORATORY RESULTS

From the results it can be concluded that:

- Different densities of post-pueruli in the experimental tanks changed the occupational distribution of the animals. In general, when densities were low, the bottom face was most preferred, but as densities increased the vertical faces become increasingly occupied. Post-pueruli showed preference for settling on the bottom face of a block. Their second preference was for occupying holes on the vertical faces rather than the top. Holes in the upper face were the least preferred;

- Post-pueruli preferred to occupy the outer holes of block faces rather than holes close to the centre of the face;

- Post-pueruli preferred to occupy holes further away from, rather than close to similar sized post-pueruli, but will settle close together rather than not occupy a hole at all;

- Smaller post-pueruli preferred to occupy the outer holes of a block face rather than holes near to large post-pueruli near to the centre of the block face; and

- The presence of predators did not result in different hole occupancy distribution patterns of post-pueruli from when predators were removed.

All analyses showed that the choice of holes selected for occupancy by post-pueruli was not random ($p < 0.01$).

From Experiment 1 it was seen that increasing the number of post-pueruli in the tank changed the hole occupancy distribution of these animals in the holes of the block. When the number of animals in the tank was at its lowest (10 post-pueruli), it was seen that the bottom face was much more saturated than the other faces. When this number was increased to 20 animals, the holes of the vertical faces then began to become more filled. At the top level (40 post-pueruli in a tank), the holes in the top face began to be occupied by more post-pueruli. Experiment 1 also showed that there was a statistical difference in the hole occupancy probabilities between the different faces ($p < 0.01$).

Mapping the ‘particular hole’ contrast of the probability of post-pueruli occupying holes in each face type, it was consistently found that the higher contrast for each face (indicating a higher probability) was associated with the outermost holes, as opposed to holes closer to the centre of the face of the block.

Adding predators to the tanks did not make any difference to the patterns of hole occupancy of post-pueruli. In Experiment 1, the effect of the predators on settlement distribution ($p = 0.67$) and settlement distance preference ($p = 0.69$) were not significant. This was also true for the settlement pattern in Experiment 2 ($p = 0.80$).

Due to the significantly different number of post-pueruli occupying the holes of the block before a predator was added (approximately 15 for each tank) to that when predators were added (approximately 30 per tank), it was not seen appropriate to combine the two parts of the data sets together so as to test for the effect of adding predators. It was noticed that the number of stragglers on the floor of the tanks was reduced once predators were added, which explains the significant increase ($p < 0.01$) of post-pueruli occupying the holes in the blocks.

It was not possible to quantify any effect that may exist for the positioning of the food, since there are no data sets that varied the positioning of the food but at the same time, kept all the other factors constant e.g. the faces open changed with the food position or predators added. However, examining the results of Experiments 1A & B (food on bottom of tank and all faces open) with those of Experiment 2A (food on top and only top and bottom faces open) (Table 1), suggests that the food position had no effect on the faces or holes occupied by the post-pueruli, because the optimal face for both these experiments was the bottom face.

Another factor in these experiments was whether or not animals chose a face of a particular block due to its position (under, top or vertical) or because of some ‘special attribute’ of the block face. To test this, Experiment A (no rotating of block) and Experiment B (block rotated) were analysed for whether or not rotating the block changed the face preference (under, top or vertical) of post-pueruli. Rotating the block did not change the face preference of post-pueruli in these experiments ($p = 0.77$) (Table 18). and this was assumed to be true for all other experiments.

Table 18 ANOVA testing whether or not the rotation of blocks affected the significance of which face post-pueruli preferred to occupy. Parts A and B of Experiment 1 have been used for this analysis.

Factor	Df	Deviance	Resid. Df	Resid. Dev	Pr(Chi)
Post-Puerulus Count in Holes	1	228.07	6298	4525.07	< 0.01
Block Rotated	1	0.07	6297	4525.00	0.80
Face	2	272.57	6295	4252.43	< 0.01
Tank	2	4.64	6293	4247.79	0.10
Particular hole	24	141.89	6269	4105.90	< 0.01
Block Rotated:Face	2	0.53	6267	4105.37	0.77
Face:Tank	4	32.21	6263	4073.16	< 0.01
Face:Particular hole	48	96.23	6215	3976.93	< 0.01
Tank:Particular hole	48	69.01	6167	3907.92	0.03
Face:Tank:Particular hole	96	151.80	6071	3756.12	< 0.01

Whether or not smaller post-pueruli preferred to occupy holes close to similar sized post-pueruli was investigated by different experiments. Experiment 2 concluded that there was a preference for post-pueruli to settle further apart rather than close together. However, it was concluded in Experiment 1 that there was no statistically significant preference. These two experiments differed in that Experiment 2 used two blocks, one with 25 holes per face and the other with only 9, whilst Experiment 1 used only one block with 25 holes per face. The reason for this difference in result is not obvious, since although Experiment 1 used twice as many animals as Experiment 2, Experiment 1 had twice as many holes in the one block than did Experiment 2 with the two blocks. The single block in Experiment 1 had all faces open but the two blocks in Experiment 2 had only the top and bottom faces open.

Experiment 3A showed that post-pueruli prefer to occupy holes further apart rather than close to larger post-pueruli. Part B of this experiment indicated no such preference. This contradiction may be best explained by the fact that part B had about twice the number of settling post-pueruli than part A and hence, small post-pueruli may prefer to settle away from other larger animals but in a high density situation would prefer to settle close to large post-pueruli rather than no hole at all.

2.1.5 RELEVANCE OF THE LABORATORY STUDIES TO FIELD ECOLOGY

There are inevitable problems in the interpretation of the results from laboratory studies. However, it is necessary to conduct such studies to identify the influence of specific factors so that these can be considered when designing equipment and field experiments.

It may also be useful to examine some conclusions from studies of similar stages of the Red Rock Lobster (Australia's southern rock lobster), *Jasus edwardsii* in New Zealand, (Booth, 2001 and Booth and Ayers, 2005). In their studies, they found that juvenile lobsters chose to occupy holes (shelters with sides) over open horizontal gaps. Furthermore, that small lobsters preferred to occupy holes with two openings rather than one, but larger lobsters >30 mm CL preferred one opening over two. It was shown that for post-pueruli sized lobsters, small holes closely fitting the body size of the lobster provide a high degree of protection from both predators and environmental forces. All this indicates that holes are likely to be more effective than crevices in the construction of artificial reefs for this species.

2.1.5.1 Selection of location of holes in the blocks

Although the area of first preference of the post-pueruli was the holes on the underside of the blocks, there is no real equivalent location in the field in the shallow coastal areas occupied by the post-pueruli. There are no loose rocks or stone in these areas. The shallow limestone reefs are the remains of areas from which the softer areas have been dissolved or eroded, and what remains is contiguous with the substrate. However, some of these limestone reefs may be "hollow" or have extensive crevices or caves within them, and these would presumably be the areas most sought after by the post-pueruli. Most of these areas are inaccessible to divers, but where they are accessible, they have been found to contain post-pueruli (Jernakoff, 1990).

2.1.5.2 Selection of daytime shelters made just before "dawn"

Of interest during this study of *P. cygnus*, was the nighttime activity of the post-pueruli as they left their shelters to seek food and when they sought shelter around dawn.

These activities are not easily described, but we have attempted to film the activities using a video camera and infrared lighting (these results have not been included in this report). It is sufficient to say that there is intense activity with regard to hole selection, control of the food, and the selection of the hole that they select at the end of the night and occupy during the day. There was definitely shelter selection!

Because the light dark cycle in the aquarium room did not allow for variable light levels, there were no dusk or dawn cycles. It is therefore possible that some of the post-pueruli chose to select holes very rapidly after the lights came on, and would have chosen different holes if there had been a dawn light cycle.

2.1.5.3 Holes or crevices

We did not offer a choice of holes or crevices in our tests. However, field results from Jernakoff (1990) suggest that either is selected in the field, but that pueruli and early post-pueruli show a clear preference for holes over crevices.

Ideally refuges should be conditioned for a period of time prior to use. Due to time constraints, the blocks used in these studies were only submerged for a few days in seawater prior to the experiments. However, we were trying to determine the location of the holes selected by the post-pueruli as places to reside during the day. We believe that while conditioning would have made the holes in the blocks more acceptable to the post-pueruli, further conditioning would not have influenced the selection of particular holes. It is possible that the apparently large number of stragglers could have been a response to the non-conditioned state of the blocks.

2.1.5.4 Rough surfaces compared to smooth

The sides of the block and the holes in the block were not especially smooth or rough. However, they were consistent and therefore this was unlikely to be a factor.

2.1.5.5 *Hard-walled shelters*

The blocks were definitely hard walled!

2.1.5.6 *Applicability of the laboratory results*

It is impossible to tell how well the laboratory conditions replicated conditions that the post-pueruli experience in the field. The depth of water would be different. There is no tide or wave action in the laboratory and it is impossible to say how well the light conditions simulated those in the field.

Nevertheless, the results do provide useful information, which assists with the design of the field apparatus for enhancing the survival of the pueruli and post-pueruli.

In designing the field apparatus, it is necessary to develop equipment that will permit the monitoring of the numbers of pueruli and post-pueruli occupying the device. For this reason, it may not be possible to produce a device with holes on the underside, as these may prove impossible to check by divers. However, a more permanent device could incorporate caverns or tubes with holes in them, which could produce a similar effect.

2.2 OBJECTIVE 2 **To estimate the number of shelters needed to make a measurable impact in a study area, and a preliminary estimate of what would be needed to provide an impact in a regional commercial catch and effort (CAES) reporting area.**

To conduct a scientific field experiment to determine if the enhancement devices are effective it will be necessary to set out a series of shelters in an area of about 1 ha.

Based on settlement data at Seven Mile Beach, up to 30,000 pueruli settle/ha year in very good seasons of settlement (Phillips et al., 2003). This means that the device should offer up to 30,000 holes if each hole were to accommodate a post-puerulus. However, peak settlement normally occurs over a number of months, mainly from August to January. Therefore not all of the holes need to be of the smallest size (12 mm diameter and 50 mm depth).

Because of the extended period of settlement, combined with natural mortality, the allocation of holes per ha along the coastline should possibly be around 15,000 of 12 mm diameter hole, 10,000 of 20 mm and 5,000 of 30 mm.

We would suggest that any future trial of these habitat structures would need to be constrained to a total of approximately 10,000 holes (0.3 ha), due to constraints surrounding the tagging and monitoring of settling pueruli.

A preliminary estimate of what would be needed to provide an impact in a regional commercial catch and effort reporting area was not undertaken as we concluded that field-testing of the artificial reef designs needs to be carried out before a proper estimate of their impact in a regional area can be undertaken.

Additional information and discussion of this topic is made under 2.3 Objective 3.

2.3 OBJECTIVE 3 **Design, in conjunction with coastal engineers, suitable pueruli/juveniles enhancement structures that could be built in the future to test as a device to enhance local rock lobster populations**

2.3.1 DESIGN OF SETTLEMENT HABITATS

These should preferably be inexpensive, handled with existing fishing boats and equipment, designed in a way that will enable them to be removed from the ocean at the end of any testing.

Objective 3 was to design, in conjunction with coastal engineers, suitable pueruli/post-pueruli enhancement structures that could be built in the future to test as a device to enhance local rock lobster populations. During this reporting period we have been working closely with Dr Kim Klaka, a Senior Research Fellow and Mr Peter Henley, Research Officer, in the Centre for Marine Science and Technology, at Curtin University of Technology, on the design of enhancement structures.

The following design criteria for a proposed structure were decided on:

- 30,000 holes per ha with a range of hole sizes required.
- The allocation of holes per ha of coastline being 15,000 holes of 12 mm in diameter, 10,000 of 20 mm in diameter and 5,000 of 30 mm in diameter. The holes should be 50, 80 and 120 mm deep respectively.
- Experimental work in the laboratory (Objective 1) has shown that the underside of surfaces is the most sought after side for colonization by the pueruli, followed by the sides, with the uppermost horizontal surface seldom occupied. Corner and edge holes showed the highest occupancy rate.
- The distribution of the 12 mm holes should be at least 30 mm from its nearest neighbouring hole. The larger diameter holes

should be 60 and 120 mm from their nearest neighbouring holes.

- The device must be capable of being deployed from a vessel of opportunity – in all likelihood a lobster boat – and must be recoverable at the end of the experiment. It must be capable of surviving deployment for periods of up to 3 years, after which it must be recoverable with no long-term impact on the environment.
- A possible site for the experiment is Jurien Bay, in Western Australia, although this still has to be confirmed. This is an off-beach site, partially protected from swell by off-lying reefs with water depths of up to 15 m.
- The design must remain on the seabed, with minimal drift (<10 m) over the deployment period and it must possess a low likelihood of capsize. It must not be seriously affected by sand banking or erosion.
- The trial is to be constrained to 10,000 holes (0.3 hectare) in the first instance, due to the practical tagging and monitoring constraints. The tagging will allow us to monitor the length of time that lobsters remain on the artificial reef, and most importantly if they survive and enter the commercial fishery.

2.3.2 THE PROPOSED ENHANCEMENT DEVICE

An initial guess for the device would have a maximum mass for deployment of 200 kg, based on handling constraints. A 200 kg device would provide of the order of 100 holes. Therefore 100 devices for a 10,000 hole trial would be required. At an optimistic deployment time of 15 minutes per device, this amounts to 25 hours, plus mobilisation time. The location for deployment is subject to wind and wave climate that would rarely provide a weather window of more than 3 consecutive days. It is concluded that using 200 kg devices with diver deployment could result in long and expensive delays waiting for suitable weather to start or complete the deployment. The approach taken has been to avoid intricate designs that require careful positioning and orientation, thus obviating the need for divers during deployment. Two alternatives then present themselves:

2.3.2.1 Option 1: use several small devices

Small devices can be deployed quickly and in rough seas compared with larger devices. Such an approach would widen the weather window, shorten the required window length and permit a wider range of vessels to be considered for the deployment. A small device has a large number of holes per kilogram and a high proportion of corner and edge holes (which had higher occupancy rates in laboratory experiments). However, a small device may be more susceptible to wave-induced drift and sand accumulation.

2.3.2.2 Option 2: use fewer, larger devices

If larger devices were used, the deployment vessel may require enhanced winching capability on the stern deck. Vessels with such capability are not so readily available in WA and are likely to be more expensive to charter than smaller general-purpose vessels. The number of holes per kilogram is low, as is the proportion of corner and edge holes. On the other hand, a larger device is less susceptible to wave-induced drift and sand accumulation.

It is proposed that a prototype of each of the two options be designed and built in order to move to the feasibility stage.

2.3.3 PROPOSED DESIGN CONCEPT

The design concept for both options is the same. It is a concrete block with holes in the sides only, not top or bottom. In this preliminary design, holes have not been made in the under surface. The initial designs are principally to examine in the future, the influence of the high-energy environment in Western Australia in the winter on the stability of the block design. Whilst laboratory experiments showed bottom facing holes to have the highest occupancy rate, a configuration with an open bottom face would be more expensive, prone to capsize and susceptible to sand accumulation. In the event of capsize, the advantage of the bottom-facing holes would be lost and the risk of damage increased due to the lower stability of the design. A design with only side-facing holes has a lower probable occupancy rate (based solely on laboratory experiment results), but has a much higher probability of retaining its correct orientation and of the holes remaining clear. In the absence of any clear indication of a bottom-faced design yielding overall probability of higher occupancy, the simpler side-holes design concept is adopted.

The device is approximately square in plan form, with length nominally twice the height. This configuration would tend to fall naturally on its base after free-fall and would be unlikely to capsize due to wave action, thus ensuring the hole faces would be functional during the trial. Lifting rings would be moulded in for deployment and recovery. Manufacturing costs would be low, allowing for extra devices to be built and deployed. This would provide a redundancy factor for any devices that break on impact (highly unlikely) or land capsized (also unlikely). The engineering equations governing the relationship between the device size, mass and number of holes have been developed and modelled numerically using "Matlab" software, to optimise the device during the next stages of design. Indicative size estimates for both options are given below, subject to optimisation in the subsequent detail design stage.

2.3.3.1 Option 1

The smaller device would be approximately 0.25 m high and 0.4 m square, weigh 85 kg and contain 50 holes (Figure 17 a and b). Approximately 200 of such devices would be required for a 10,000 hole trial (0.3 hectare). The relatively low mass permits them to be deployed by two people dropping them over the side of a workboat at a rate of approximately one every minute, with no specialised equipment required. Deployment time would be 3.5 hours, plus mobilisation time.

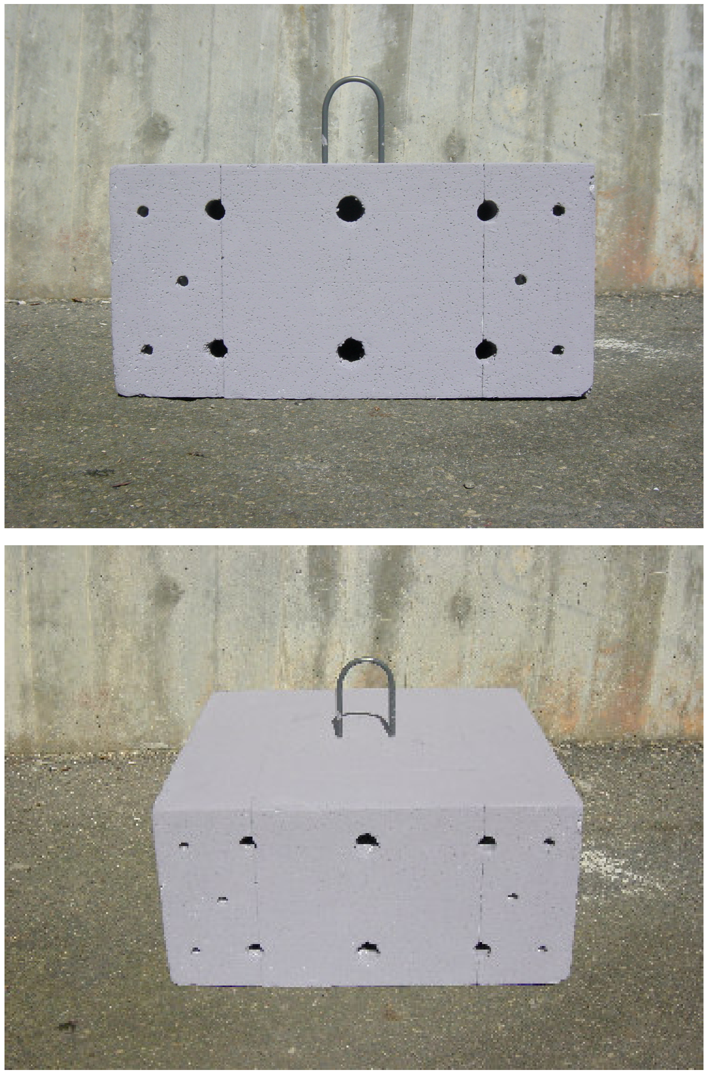


Figure 17 a, b. The smaller enhancement device as seen from different angles. Dimensions are 0.25 m high and 0.4 m square, weigh 85 kg and contain 50 holes.

2.3.3.2 Option 2:

The larger device would be nominally 0.4 m high and 1 m square, weigh 1,000 kg and contain 240 holes (Figure 18 a and b). Required coverage would be obtained with 42 devices. Deployment time at 10 minutes per device would be 7 hours, plus mobilisation time, with specialist lifting gear required.

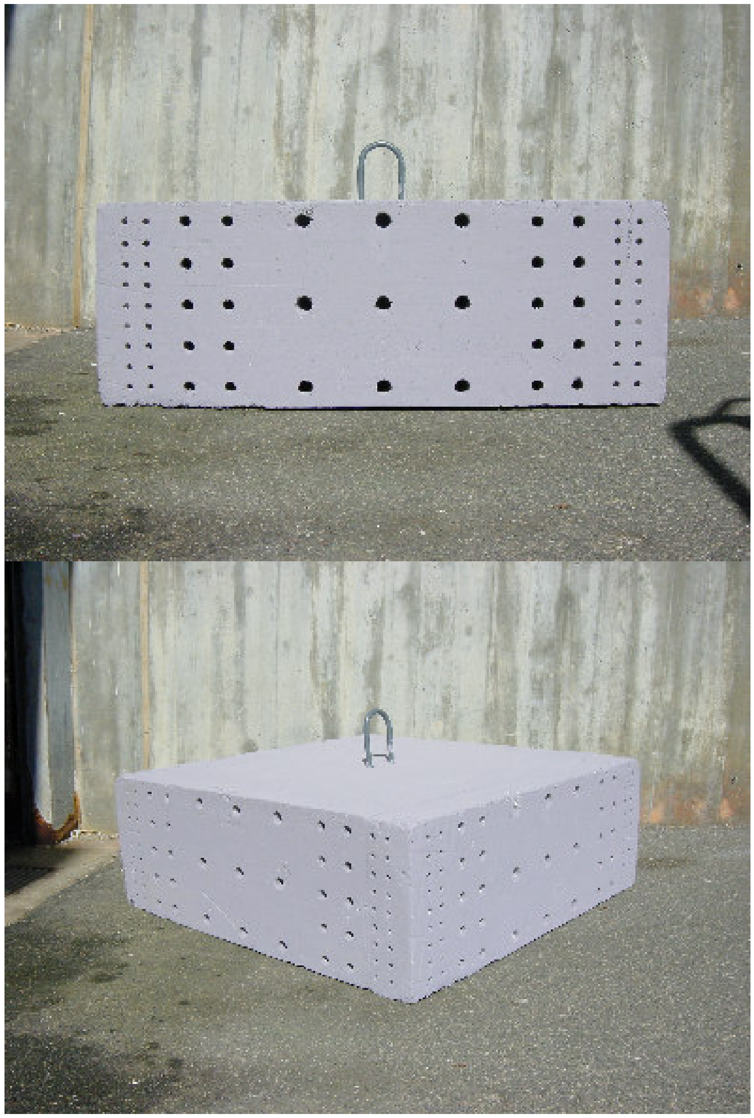


Figure 18 a, b. The larger enhancement device, with dimensions 0.4 m high and 1 m square and weighing 1,000 kg as seen from different angles.

2.3.3.3 Distribution of the holes

An additional factor to be considered is the distribution of the holes in the block. Several configurations are possible, see Figure 19 a-f. The benefit of having sides with a single sized hole instead of a mixture of holes such as Figure 18 a, b and 19 a, b is that it may reduce construction costs. Secondly, because of wasted space when mixing three different sized holes on one face of a block, having only holes of one size on a block face would increase the total number of potential settlement holes over the whole block.

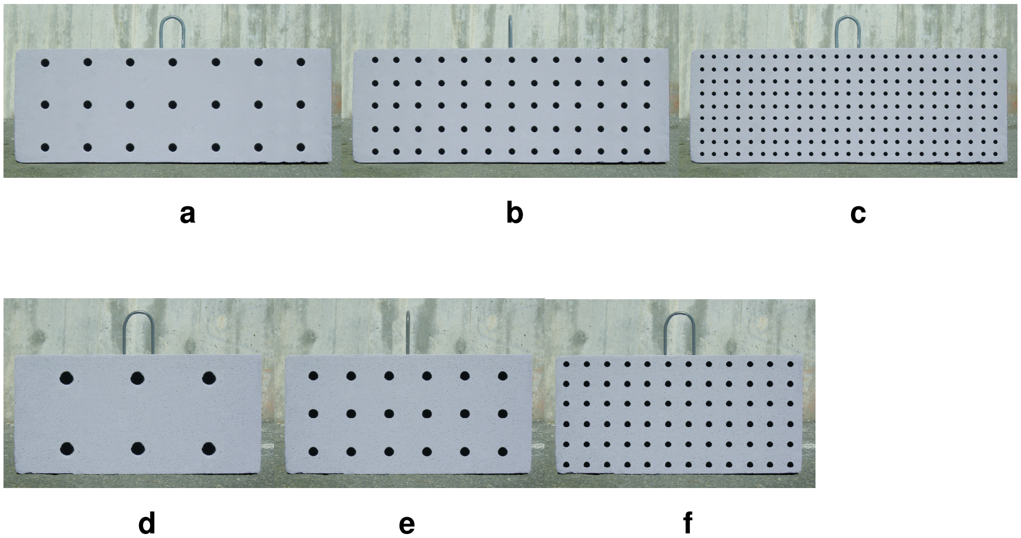


Figure 19 a-f. Large (a-c) and small (d-f) blocks, showing arrangements of small, medium and large size holes on a single face.

At this stage there is no way of knowing which arrangement of holes might be the most effective in achieving maximum levels of occupation by pueruli and/or retention of post-pueruli over the first post larval year.

The best most effective distribution of the holes would need to be examined during a field test of the blocks.

2.3.4 RUBBER-CLAD CONCRETE

It was proposed to build models of the two concrete devices. However, the all-concrete option proved difficult as it was hard to find a manufacturer confident of being able to build the product with the correct number and size of holes.

An alternative was suggested. This model had a concrete core, for weight, but was clad with a thick rubber coating made from reconstituted car tyres (Figure 20 a, b). Before serious consideration of this option, it was considered prudent to investigate the chemical components of the cladding. Particular concern revolved around the very pungent odour of the rubber. This analysis was undertaken by Dr Steve Fisher, a fisheries chemist attached to the

Research Division at the Department of Fisheries. He provided the following reports:

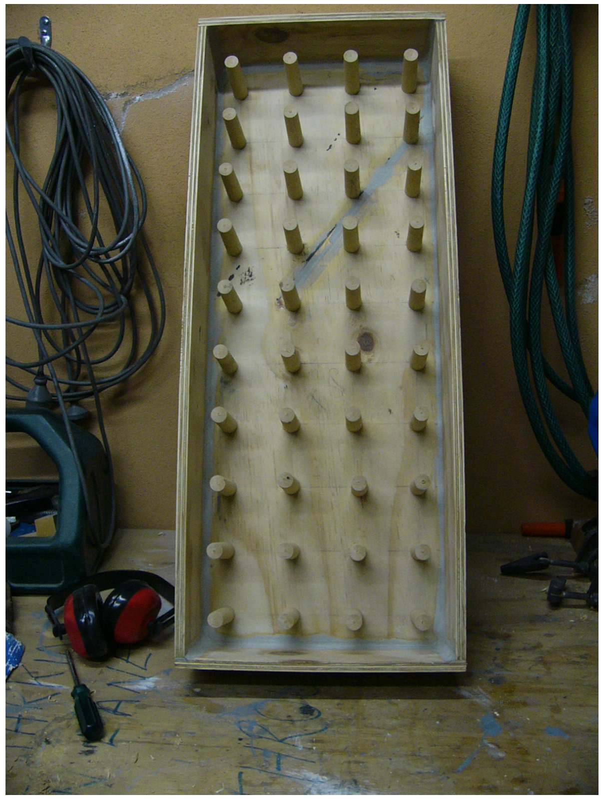
'*The product is shredded rubber from car tyres from which the steel belts have been removed. A single pack (aromatic) polyurethane binder is added to hold the rubber together.*

An analysis was made of the volatile compounds emitted by the product.

The major components of the headspace gas are hydrocarbons in the carbon number range C8 to C11. This is a fairly complex mixture, so I can't identify all of the components unequivocally, but volatile aromatic hydrocarbons such as the xylenes and other alkylbenzenes are clearly present. These are common constituents in petroleum products, especially gasoline (petrol): you might be familiar with the acronym BTEX, which refers to Benzene, Toluene, Ethylbenzene and Xylenes.

Rubber is a very efficient adsorbent for organic chemicals including volatile petroleum hydrocarbons such as BTEX, especially when these are vaporised. Therefore, the xylenes and alkylbenzenes identified in the headspace may have been adsorbed from the atmosphere in which the rubber material has been stored. These compounds might be absent from a similar sample of the rubber material stored in a "clean" environment.'

In the light of this advice it was resolved to recommend the full concrete versions of the enhancement devices rather than the rubber coated option.



(a)

(b)

Figure 20. (a) Method for creating the holes into rubber cladding, and (b) sample of the rubber cladding material with the largest of the holes The University of Western Australia has performed leachability tests on the product and found that very little leached out using water. Of the metals in the leachate, zinc was the most abundant, albeit at very low concentrations.

2.4 Objective 4 To undertake a cost benefit analysis for the various options for enhancing western rock lobster

2.4.1 THE OPTIONS FOR ENHANCEMENT

The options for enhancement of the stocks and production of western rock lobsters are:

- Pueruli or post-pueruli raised by aquaculture from breeding stock (propagated) could be released into selected field sites where suitable food and shelter are available. It is not yet scientifically possible to propagate western rock lobsters and even if it was, it is likely that the majority of such animals would be held under aquaculture conditions and raised to a marketable size. Studies of *Jasus edwardsii* in Tasmania (Mills et al., 2004) have indicated that it is practical to release very small juveniles into the wild with minimal mortality, at least initially after release.

- Holding of wild caught undersized juveniles under aquaculture conditions and raising them to legal size. This has always been an option. However, it runs contrary to the present operational model for the fishery, which is based on allowing these animals to be returned to the ocean and a proportion of these to survive to become part of the breeding stock.

- Translocation of wild caught pueruli, post-pueruli or juveniles of any size up to just below legal size, to other field locations where shelter and food are available. This has been proposed many times but there has been very little research on transferral techniques and their effect on post-transfer survival and growth in the western rock lobster. Jernakoff (1990) carried out preliminary field studies on the effects of transfer and found that post-pueruli of *P. cygnus* often refused to enter shelters after transfer, although their response was affected by the release technique. Given the potentially high rates of post-transfer predation (Howard, 1988; Jernakoff, 1990; Butler et al., 1997) further study of transferral techniques that maximize post-settlement survival would be needed.

- Setting of devices to increase the survival of naturally settling pueruli and the subsequent post-pueruli. This is the basis of enquiry of the current study.

2.4.2 COSTS OF THE VARIOUS OPTIONS

The costs of the various options described above are almost impossible to estimate at this time. For example, the first option requires information on the cost of the production of the pueruli/post-pueruli, transport to the proposed sites of release, release operations, monitoring of the results of releases, etc. All are unknown.

It is not even possible to produce a reliable costing for the type of possible enhancement being investigated in the current study. The possible use of a design using a concrete block with specified hole numbers, depths and hole arrangement is proposed. However, this device has not been built, nor tested in the field.

Models were constructed of the two design types and quotes from industry were sought for their construction. The quotes varied widely, partly because it was not possible to specify the number of units to be constructed. In addition, none of the companies approached had any experience of construction anything like the device that we have in mind. Concern was expressed as to whether it would be constructed with sufficient precision to suit our purposes. It was felt that they thought we were after a “polished” article when we were rather hoping for a rough finished block.

All of this means that we do not have a firm price on construction of the blocks. In reality the only fixed value is on the known price of concrete, but with some inventiveness it may be possible to obtain this almost for nothing using “left overs” from larger jobs, which are usually dumped as rubbish. It is also possible that after preliminary testing of the blocks, a full concrete design may be found to be unnecessary.

Therefore without true costs of the enhancement devices, there are no real criteria on which to base a solid analysis of the problem, and all of the other unknown costs of transport to the site, installation, monitoring, etc., become irrelevant.

One possible alternative is the use of natural limestone rocks or blocks. However, our enquiries revealed that this would be likely to be even more expensive than concrete.

3.0 DISCUSSION

Because these have been laboratory experiments there has to be a number of reservations associated with the results. These may have included for example, the small size of the experimental tanks and the inability to replicate the physical and chemical complexity of the benthic environment. Nevertheless, recommendations for the design for artificial reefs for enhancing pueruli and post-pueruli can be made from this work.

One technique by which natural populations of western rock lobsters could potentially be enhanced is by the deployment of artificial structures to shelter vulnerable pueruli and post-pueruli. The success of such an approach depends on the natural ecological limits to recruitment of the species and the economic practicality of obtaining and releasing sufficient numbers of artificial shelters on the seafloor, against the value of the increased yield from the fishery. This could of course be a locality-by-locality increase in yield rather than an entire coastal effect.

In Western Australia it is estimated that between 80 and 98% of the small lobsters die within the first year and as few as 0.9% survive to recruit to the fishery at approximately 4.5 years of age (Phillips et al., 2001b). It is believed that one of the reasons for this high mortality is that the post-pueruli stages require suitably sized shelters to protect them from the many inshore predators, and these shelters are in short supply. Density-dependent relationships between puerulus and catch indicate mortality is highest during good years of settlement (Caputi et al., 1995). These relationships vary between the different zones, depending on the abundance of puerulus and availability of shelter.

Other rock (spiny) lobsters are in a similar situation. The high natural mortality of post-pueruli and early benthic juveniles (e.g., < 50 mm CL) compared to larger lobsters and their dependency on crevice shelters appears to limit the recruitment of *Panulirus argus* in the Caribbean, and *Panulirus marginatus* in Hawaii in a density-dependent manner, presumably again through predation (Butler and Herrnkind, 2000).

Field experiments with tagged *P. argus* indicated that providing appropriately scaled artificial shelters designed to mimic small, widely distributed natural shelters (i.e., sponges and small coral heads) at the critical period when juveniles vacate algae to dwell in crevices increases their survival by protecting them from predation, thus increasing local recruitment (Butler and Herrnkind, 1997). However, the placement of artificial structures at sites with ample natural shelter (Mintz et al., 1994; Arce et al., 1997; M. Butler and W. Herrnkind, unpub. data), or at sites where post-larval supply is minimal (Riclet, 1998), has little or only a minimal effect on local abundances of juveniles.

Western Australia is particularly interesting, because the levels of puerulus settlement along the coast are normally significantly higher than needed to maintain the level of recruitment to the fishery, and the breeding stock. This is despite the high levels of natural mortality that occur in the pueruli and post-pueruli period.

The question is: could these high levels of lobster mortality be reduced, for a consequent gain in the production (catch). The answer is probably yes, at least in some areas along the coast. In support of this are the studies by Briones-Fourzan and Lozano-Alvarez (2001). They have clearly demonstrated that artificial habitats (casitas) used by fishers increase the abundance and biomass and catch of lobsters (*Panulirus argus*) in habitat-limited environments in Mexico.

Lobsters need both shelter and food. Some habitats along the coast of Western Australia, such as near Cliff Head, appear to have extensive seagrass beds, which would contain the type of food used by rock lobsters. Diving operations suggest (Phillips, *pers. comm.*) that suitable shelter in large quantities is not available at this site.

The new project proposed will be to demonstrate the enhancement devices ability to increase survival of the western rock lobsters over their first year as a post-puerulus. A suitable site to test the enhancement devices needs to be selected and this selection needs to be made with industry consultation. Field research is needed to fully evaluate both the efficacy of artificial population enhancement techniques and their impact on the surrounding ecosystem before the enhancement approach is advocated. Jurien Bay has been suggested as a possible candidate because of other scientific studies proposed for the marine environment of this area. An assessment of the impact of the enhancement devices might be more easily accomplished at this site.

Cost of the devices will be a prime consideration in the long-term development of enhancement operations. However, the first consideration should be to determine if successful enhancement is possible.

4.0 BENEFITS

To be able to provide the commercial and recreational sectors and the research community with (1) an estimate of the amount of artificial substrate that would be required to make a scientifically measurable impact to catches in the wild fishery and (2) to assess the likely scope, form and cost benefit of artificial reef construction, and any potential ESD issue that might need to be addressed.

5.0 FURTHER DEVELOPMENTS

This current project has produced designs of enhancement devices that may be suitable for deployment. However, field tests are needed to validate the substrate design and enhancement options were not included within the current project, but are seen as being undertaken as a possible extension to this project or as a new project. Such tests would require a minimum of three years duration and are likely to be expensive because of the OHS requirements of diving studies and the need for ecological assessments because of environmental concerns by government. It has been decided to delay application of the new project to FRDC until the 2005 round of applications. This will permit completion of the 2002/045 project and its dissemination and full discussion with industry. The results will be presented to the RLIAC, the Western Rock Lobster Council, and other fishing Associations in Western Australia at the earliest opportunity.

A preliminary research proposal has been developed and will be submitted to the Western Australian FRAB for consideration of a new project commencing in 2006, to test these devices in the field to demonstrate their ability to increase survival of post-pueruli. The devices, which it is proposed to test, have been specifically designed for the scientific evaluation of whether enhancement of the post-pueruli in the first year after settlement can be achieved.

Some preliminary trials would need to be conducted to test the stability of the blocks, measure their rates of siltation of the holes and "conditioning" on the outer surface, and our ability to monitor occupation by pueruli and post-pueruli.

This would be followed by a scientific examination of an attempt to enhance recruitment in an area of 0.3 of a hectare in a coastal reef area. This is the size of the area we have assessed as the maximum that we could successfully enhance and monitor for the test. A control site would also be required.

This could be achieved with 200 small or 42 of the larger blocks.

Project objectives

- To field test two enhancement devices for stability, ability to withstand wave energy, and ability to attract occupation by pueruli and post pueruli of the western rock lobster.
- To assess the best design in terms of the distribution of the holes in the block.
- To assess the ability of the preferred enhancement device, to increase survival of post-pueruli western rock lobsters during their first year after settlement.
- To examine the impact of the preferred enhancement device on the ecology of the area in which it is located.

Once this project has accomplished these aims, the information will then exist to provide managers and stakeholders with the opportunity to assess the pros and cons of modifying the environment to achieve increased production in the western rock lobster fishery.

This new study will build on these results by conducting field tests to validate enhancement design and options for their positioning within the fishery. The exact number of enhancement devices to be installed has yet to be determined, but the tests will be confined to a 0.3 ha site in an area that is thought to be habitat limited for small settled animals. At this stage Jurien Bay is the preferred site, but this has yet to be discussed with other scientific organisations working on projects in this area. In addition, two sandwich collectors will be deployed at the preferred site as an independent monitor of puerulus settlement. At the conclusion of the experiment, all devices will be removed from the ocean.

The tests will include diving surveys to determine densities of post-pueruli lobsters settling on the collector devices and permit micro tagging of the post-pueruli to allow estimates of survival between settlement and the end of the experiment. This will require the study to run over a minimum of two years and will include ecological assessments in the areas where the enhancement devices are located. It is planned to involve other scientific organisations in the ecological impacts aspects of this study, to enhance the value of the overall results of the tests.

6.0 CONCLUSIONS

This project has identified possible enhancement devices to increase survival of the pueruli and post-pueruli stages of *P. cygnus* in Western Australia. Field-testing is required before it will be possible to establish whether these devices are effective.

Enhancement of wild stocks has been viewed as a positive fisheries management tool for over 100 years (Molony et al., 2003), but has not been used for lobsters in Australia up to this time. However, reseeding to compensate for puerulus exploitation is proposed in Tasmania (Mills et al., 2004) and as pointed out by Booth and Cox (2003) in their examination of marine fisheries enhancement in New Zealand, rock lobster is one of the species likely to be of interest for enhancement over the next 10 years.

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8.0 APPENDICES

Appendix I

INTELLECTUAL PROPERTY:

Technology transfer category: Published and disseminated

Objectives

1. To communicate the results of a study aimed at enhancing the natural settlement of western rock lobster

Target/Audiences

- Managers of the fishery: i.e. commercial and recreational fisheries program staff, RLIAC, the Western Rock Lobster Council and the Recreational Fisheries Advisory Committee (RFAC)
- Commercial and recreational western rock lobster fishers
- Rock Lobster Enhancement and Aquaculture Subprogram (RLEAS)
- The national and international research community

Appendix II

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