Report to the National Seafood Centre

Improving Packaging Technology, Survival and Market Options for Kuruma Prawns

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CENTRE FOR FOOD TECHNOLOGY



Bribie Island Aquaculture Research Centre

FISHERIES RESEARCH & DEVELOPMENT CORPORATION



Project 92/125.32

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Summary

The viability of Australian Kuruma prawn production, and live export, is directly linked to in-transit survival and the market price achieved for the product. Fluctuations in the market price over which the Australian Kuruma industry has no control, include exchange rates and status of the Japanese economy. A sound knowledge of the elements influencing in-transit survival, product quality, and the establishment of a strong market image are, controllable factors that may positively influence market price stability. Thus, an opportunity exists for the industry to control and guarantee quality, and ensure that a market reputation is established and maintained.

The purpose in seeking to improve packaging technology, survival and market options for Kuruma prawns is to assist this industry sector in establishing and maintaining a strong and reliable market position.

The upper lethal temperatures for Kuruma prawns have been confirmed through extensive practical trials. Results suggest that synchronisation of moulting deteriorates as temperatures exceed 28°C with severe effects (pond mortality) occurring above 32°C.

This project has examined the effect on "in pack" survival of growing prawns under high ambient pond temperature conditions and examined several alternate processing protocols. It was found that a distinct decline in prawn survival rate (post 36 hours dry storage) occurred as ambient pond/growout temperatures increased. Modified preparation and packaging operations that were trialed gave no real increase in survival rates for prawns held at high ambient pond temperatures. These results question the capacity to achieve adequate survival under high ambient pond temperatures.

A refined live prawn packaging module was modified and rigorously tested. The new module showed superior insulation and temperature control properties when compared to current designs and is recommended for the packaging and transport of live Kuruma prawns to overseas markets

The feasibility of growout during the cooler months in more tropical climates and relocating prawns for further growout or "climatic agistment in more southerly locations, has been addressed. The results from these preliminary trials suggests that trials would be worth considering on a commercial basis.

Overall, this research has provided information and additional tools for management to use in stabilising production related issues that impact on market quality.

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Background

Live Kuruma prawns are a valuable export commodity. Since the successful development of methods for the preparation and packaging of live prawns, exports of this product have expanded annually, with an anticipated export production of up to 300 tonnes in 1996 with sales value in excess of A\$20M.

One of the reasons for the success of the Australian product on the Japanese market has been the effectiveness of the packaging system and the high survival rate. This packaging system involves an outer cardboard carton with an inner lining made up of 7 separate sheets of polystyrene.

Australia now has an established position as a high quality supplier in this market. However some aspects of Kuruma prawn growth and marketing required attention as a matter of some importance in order to maintain and enhance our reputation in the light of increasing production.

In 1995 production of Kuruma prawns in Ayr, northern Queensland, resulted in a loss of market confidence and poor prices following high mortalities of this product in the Tokyo markets. This highlighted the need to investigate the suitability of current methods for packing Kuruma prawns in warm climates. There was also the need to examine the effect of high ambient pond conditions on survival and to determine whether modified preparation and packaging methods would minimise the effects of these warm ambient conditions. The authors of the current proposal therefore submitted a funding application to FRDC (1995-96) to address these issues. This earlier application was rejected by QFIRAC because the industry did not consider the anticipated problems significant at that time. However, Kuruma prawn farmers now recognise these issues as critical.

There is considered to be a general correlation between the warm ambient conditions (temperatures in the mid to high 20's) and a reduction in the survival rate in-transit. Whether this is related to higher packing and transport temperatures or to an intrinsic condition of the prawns at these temperatures is uncertain. Thus, there is a need to determine which is the case and formulate a strategy that may permit Kurumas to be successfully grown in warmer climatic conditions, such as in northern Queensland. In the event that the condition of the prawns in the warmer climate is such that high survival rates are unachievable, and hence cannot be improved through developments in the preparation and packaging operations, we need to determine whether it is possible and feasible to transport these prawns in bulk to sites in cooler climatic areas for "conditioning". This concept certainly has precedents in other agricultural industries.

In considering each issue, the identity of the Australian product and the need for highly functional packaging is central. While a more advanced form of packaging was proposed by IFIQ more than three (3) years ago the industry has not been sufficiently advanced to justify the production of the packaging. It has also been suggested that smaller net weight (ie. 6 or 8kg instead of 10kg) packages will offer

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Bribie Island Aquaculture Research Centre access to a broader range of buyers and hence strong prospects for higher prices (Kitada, Austrade Osaka).

At a meeting of Kuruma prawn farmers (7th March 1996) a consensus was reached on the need for Australian producers to be technically advanced and identified on a country basis as a supplier of quality produce. The meeting also agreed for the first time to use a common size of inner package, making the production of an advanced outer package more likely to be viable.

A new generation packaging module was designed by the Centre for Food Technology (CFT) in co-operation with "Rmax" (Huntsman Chemical Company Pty Ltd), and differed from existing packaging in that it is compact, has superior insulation and is of similar cost. It also contains a number of technical innovations allowing greater control of temperature as well as the capacity to vary the temperature control system and the net package contents.

With minimal modification the module should also be suitable for the transport of other live crustacea such as redclaw, lobsters, and spanner crabs as well as chilled and frozen seafood.

Need

In order to maintain and further develop Australia's market position for Kuruma prawns, the Australian product needs to be identifiable as superior and reliable. A technically advanced, universally adopted package which helps guarantee in-transit temperature stability will have a major influence in insuring a stable market position for Australian producers. This package needs to perform under a range of environmental and climatic conditions in order to protect product quality. An instruction manual covering its use and a temperature monitoring system is needed to assure that in-transit conditions are optimum.

Survival rate and condition are key elements in determining the market price of Kuruma prawns. More than 5% mortality will cause a price decline. Prawns grown and packed at higher ambient temperatures are much more likely to die in-transit. There is an urgent need to determine the survival prospects for Kuruma prawns grown at high temperatures and their suitability for live transport.

Methods of preparation and packaging of prawns that are harvested under high ambient temperature conditions need to be developed and trialed. Attempts to package Kuruma prawns in warmer conditions have, to date, resulted in poor performance in the market place and jeopardise the future of this industry.

It is also essential to determine the possibility of transporting immature and mature prawns from one farm to another. This will allow the future potential of conditioning prawns for live transport, by climatic agistment, to be evaluated.



Objectives

The objectives of the project were:-

- To evaluate the temperature stability of a technically advanced live prawn (seafood) package design (prototype and finished item), under a range of storage, transport and climatic conditions.
- Ascertain/confirm the upper lethal temperature limit for Kuruma prawns.
- Determine if modifications to the preparation and packaging operations are likely to improve the survival rate of packaged prawns from high ambient temperature growout conditions.
- Establish the potential feasibility of transporting live Kuruma prawns from one location to another to optimise growout conditions and hence quality through climatic agistment.

All objectives were achieved. In some cases current progress has surpassed the original project objective. The first objective "To evaluate the temperature stability of a technically advanced live prawn (seafood) package design....." has led to an aesthetically and technically advanced package that implies market advantage as it can be separated for the individual marketing of 3kg units in self contained insulated packs. The module is also being evaluated by other industry sectors (redclaw and rock lobster).

Methods

Package development testing:- An extensive testing program focused on monitoring temperature changes at up to 6 positions in and around the new package. Prawns for the study were simulated by using pre-cooled gel packs equivalent to the total nett product weight. A detailed study of the performance of gel coolants ranging from 250gm to 750gm as well as coolant configuration within the package were also conducted. In the original proposal, testing was to extend to a range of constant external temperatures as well as a range of simulated in-transit conditions. However, the study was modified to test shipments from local commercial prawn farms to the Tokyo markets using data logging equipment to monitor actual in-transit temperatures.

Upper lethal temperature studies:- To determine the upper lethal growout/pond temperatures, Kuruma prawns were maintained in laboratory scale tanks and subjected to experimental temperatures of 28°C to 36°C (in two (2) degree increments). The survival, moulting rate and feed consumption rate were then measured over a period of four weeks. The temperature which gave an LD50 (mortality of 50% of the population) within this period is to be considered the upper thermal tolerance limit.





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High temperature processing and packing:- By setting the temperature limit for harvesting just below the temperature after which survival drastically declined (32°C), it was possible to determine whether Kuruma prawns would have a high level of survival during transport and/or if processing and packing modifications could improve their survival. To achieve this, prawns were held at temperatures of 23°C, 27°C and 31°C for an acclimatisation period (four days) before being subjected to a range of packaging conditions including contemporary methods as well as significantly slower and faster preparation procedures. Prawns were then held for a period of 36 hours to simulate transport before being unpacked to record the survival rate for each method used. The post transport survival of prawns was also studied by subjecting the active/surviving prawns from each treatment to an intank holding period of seven (7) days.

Transport and climatic agistment:- To establish the feasibility of transporting large numbers of prawns from one location to another, prawns were stored at both high and low densities in oxygenated seawater at temperatures of 25°C and 16°C for extended periods. In the original proposal, 48 hours was to be used to simulate transport time. However, for this study, the simulated transport time was reduced to 12 hours as this better represents the time required for transportation to and from the most northern prawn farms. Water quality (pH and ammonia) was monitored during transport, while survival rates were recorded after the 12 hour period. The effects of water conditioners/ammonia scrubbers and anaesthetics on prawn survival post transport were also studied by the addition of these to the transport water.

Detailed Results

The detailed results of the project are presented, and discussed in the general circulation part of this report (Part 2).

Benefits

The implied benefits of this research are highlighted in the summary and individual discussions of results but may quickly be summarised as follows;

- Technically advanced packaging which also offers market advantage has been tested and introduced to the industry through this research. This will assist producers in enhancing and maintaining quality and market position.
- The question of whether kuruma prawns can be grown and packaged in tropical conditions has been largely answered through this project. The upper lethal temperature for prawns in ponds is now confirmed and the effect of high ambient conditions on the survival rate of packed prawns is also presented in this report.
- While there are portions of the year when it is unsuitable or less than optimal for the growing of kuruma prawns in one location, the growing season may be



extended if prawns can be relocated to a more suitable environment when conditions deteriorate. This study has shown that under controlled conditions there are reasonable prospects of relocating these prawns to improve prawn condition or potentially extend or modify the growing season.

Intellectual Property

There are no issues of intellectual property addressed through this project.

Staff

Bruce Goodrick, B Sc - Food Tech, Seafood Team Leader, (Project Leader). David Hewitt, PhD, Fisheries Biologist. Stuart Frost, B Sc - Food Tech (Hons), Seafood Technologist. Stephen Grauf, ADAB, Seafood Technician (Funded by the project).

Final Cost

The final cost of the project was \$33,750 consisting of \$13,750 in salaries (Grauf and Goodrick), \$3,600 in travel, and \$16,400 for operating costs. Operating costs consisted of purchase of prawns and feed, packaging materials, freight, and casual wages.

Distribution

NSC 10 copies (1 copy unbound, plus disc copy)

Acknowledgments

We would like to acknowledge the assistance of Rmax (Huntsman Chemical Company Pty Ltd) for the design and manufacture of the new package design; Moreton Bay Prawn Farm and Tomei for commercial export trials; Moreton Bay Prawn Farm, Gold Coast Marine Hatcheries and Luxe Enterprises for supplying prawns, technical and support staff at the Bribie Island Aquaculture Research Centre and the Centre for Food Technology.

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SECTION 2

COLLECTIVE RESULTS (ACHIEVED OBJECTIVES).

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Improving Packaging Technology, Survival and Market Options for Kuruma Prawns.

Project Objective 1: To evaluate the temperature stability of a technically advanced live prawn (seafood) package design (prototype and finished item), under a range of storage, transport and climatic conditions.

Bruce Goodrick and Stephen Grauf.

Introduction

Since the early 1990's, Australian aquacultured Kuruma prawns have been packed in 1kg inner boxes which are then placed in a cris-cross fashion inside a larger outer box for export to Japan (Goodrick et al., 1992&1993). The outer boxes are normally composed of cardboard with polystyrene inserts for the walls, bottom and lid. To maintain constant temperature and survival during transit, a thin polystyrene insert is placed on top of the inner boxes and a gel type coolant placed upon this (Goodrick et al., 1996). Historically, this packing arrangement, although not optimum, is capable of maintaining $15^{\circ}C+/-5^{\circ}C$ for periods of up to 22 hours.

In 1996, as an interim measure, the live prawn industry moved towards improved packaging, designed by the Centre for Food Technology (CFT). This box was of a smaller profile and nett content, with the inner boxes placed in a parallel fashion to give a larger mid-section air space. The larger air space allowed greater air flow from the coolant to give more even temperature distribution between the inner boxes. This "interim" box design still incorporated a cardboard/fibreboard outer shell with polystyrene inserts located inside for insulation purposes The design is capable of maintaining 10-15°C for periods up to 30 hours. While this interim box is an improvement on the original 10kg package (which showed internal temperature variations of up to 3°C), scope remained to further improve the temperature stability.

It has been suggested that smaller nett weight (ie. 6 or 8kg instead of 10kg) packages will offer greater access to a broader range of buyers and hence strong prospects for higher prices (Kitada, Austrade Osaka). Kuruma prawn farmers reached a consensus that there is a need for Australian producers to be technically advanced and identified on a country basis as a supplier of quality produce. It was also agreed, for the first time, to use a common size of inner package, making the production of an advanced outer package more likely to be viable. In answer to these issues a new generation packaging module has been designed by the Centre for Food Technology (CFT) in co-operation with Rmax (Huntsman Chemical Company Pty Ltd). This module differs from existing packaging in that it is compact, has superior insulation and is of comparable or lower cost. The package itself also contains a number of technical innovations allowing greater control of temperature as well as the capacity to vary the temperature control system and the nett package contents. With minimal modification the module should also be suitable for the transport of other live crustacea such as redclaw, lobsters, and



spanner crabs as well as chilled and frozen seafood. This study outlines the current prototype package and the thermal capabilities of this design.

Materials and methods

Prototype package design.

Polystyrene sheets (R-max) of varying thickness were cut and glued into a basic box configuration to accommodate 6kg (3*1kg*2) of packaged product. Primary format and dimensions of the prototype were as follows:- a box base section (564mm x 370mm x 280mm), a mid section (564mm x 370mm x 75mm) for the coolant and a lid section (564mm x 370mm x 20mm). An integral feature of the design is the incorporation of a score line ("snap-off") that allows the box to be split in the middle thus forming two (2) completely independent 3kg boxes (Diagram 1).

Diagram 1: Dimensions of prototype live prawn box.



Test equipment and temperature recording.

As the cost of using live Kuruma prawns for performance testing was prohibitive, it was decided to simulate prawns by using gel packs equivalent to the wet weight of prawns. To accomplish this, two 450gm water based gel packs (Chillpack Pty Ltd) were packed with hoop pine sawdust into 1kg cardboard inner boxes, as per normal live prawn packing procedures. The inners containing gel packs were then pre-cooled in a 14°C cold room for several days prior to testing. T-type thermocouples were connected to an

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external datalogger (Datataker DT-500) programmed to record temperature every 15 minutes for a period of 36 hours. Thermocouples were placed in a set position within the box. Positions were held constant throughout testing of all modifications. Thermocouple positions were as follows:- on the surface of the simulated prawns (gel pack), inside the bottom, middle and top inner boxes; the air space between the top inner and the interior of the box wall; and beneath the coolant in the middle section of the box (Diagram 2). A thermocouple was also attached to the exterior surface of the prototype lid to measure ambient air temperature during each trial.

Data logged from each trial was down loaded from the datalogger and transferred to a spreadsheet program (Microsoft - Excel 5.0). Standard format graphs were produced from the sets of data to allow evaluation of each modification.

Diagram 2: Exploded view of prototype box and thermocouple positioning for performance testing



- 1:- On top of simulated prawn pack in bottom inner.
- 2:- On top of simulated prawn pack in middle inner.
- 3:- On top of simulated prawn pack in top inner.
- 4:- Air space between top inner and interior box wall.
- 5:- Under coolant in middle (coolant) section.
- 6:- Centre of exterior box lid (ambient air temperature).

Preliminary trials.

Preliminary trials of the prototype package were undertaken to determine the performance of various sizes of gel coolants. Gel coolants weighing 250g, 500g and 750g, both with and without 3mm Bubble-Wrap[™] plastic insulation were trialed. Coolants were frozen to -30°C in an industrial freezer (24hrs), then tempered (by leaving

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coolants in ambient temperature air) to -5°C before placing them into the prototype box. The effectiveness of each coolant was determined via thermocouples and datalogging equipment as previously described. Data from these trials indicated that the 500g insulated coolant would give best results with the new system.

Prototype performance testing

To establish the effectiveness of modifications made to the prototype design; replicated trials were performed. An exhaustive range of design modifications were trialed, however for simplicity only the major changes have been included in Table 1. For each trial, frozen and pre-tempered 500g insulated coolants were placed into the modified box, while thermocouples and data logging equipment were again used to record temperature profiles over a 36 hour period. Data collected from trials of each modification to the prototype was evaluated against the interim box and the unmodified prototype box.

Table 1: Major modifications tested

1) Different thickness in base material under the coolant in the middle section and effect of raising the coolant off the base.

2) Air gaps of varying depths under the bottom inner box and above the top inner box.

3) Size and frequency of the holes around the edge of the base in the middle section.

4) Shape of middle section base. For example, a fluted base versus a flat base versus a base with grooves in a chocolate block configuration etc.

5) Removal of the dividing wall in the middle section and varying the thickness of this wall

6) Varying the thickness of the dividing wall in the base section while maintaining the overall box dimensions thereby increasing the air flow down the sides of the inner boxes.

Final prototype testing (commercial export tests).

To test the performance of the final prototype, five shipments of 60 kg (live prawn weight) were dispatched to the Japanese markets from two domestic prawn farms. Prawns were harvested, cooled and packed into inner boxes using standard commercial practices. Inner boxes were then placed into the final prototype box with the insulated gel coolants. Small data loggers (Cox Tracers and Hastings Stowaway XTI's) were used to monitor internal box temperature as well as outer ambient air temperature. Data loggers were removed and returned via mail on unpacking and sale in Japan.



<u>Results</u>





Gel coolant trials.

Figure 1 depicts the temperature data for both insulated and uninsulated forms of gel coolants. At an ambient air temperature of 20°C, uninsulated 250g, 500g and 750g gel coolants maintained their cooling capacity for 12hrs, 18hrs and 20 hours respectively. The use of plastic insulation (3mm Bubble-WrapTM) with 500g packs extended coolant life for an extra 6hrs, from 18hrs to 24hrs. As live prawn consignments take approximately 30 hours to reach the market floor in Japan, at which time the prawn temperature should be around 15°C, it was decided to use the 500g insulated. coolant as the standard coolant for the prototype testing of the box.

Prototype testing

The testing of the interim box was carried out using the methods described on pages 2 and 3. These results are shown in Figure 2. As can be seen from this figure, the inner pack temperatures vary up to 1.5°C. While this result is an improvement on the original 10kg package (which showed internal temperature variations of up to 3°C), scope remained to further improve the temperature stability.



Figure 2: Temperature record of interim box.



The first prototype design (Figure 3) produced inner pack temperature variations of up to 5°C. This box had no air circulation systems or moulded polystyrene insulation in the design. Results from trials on this box indicated that modifications were needed. Temperature records of some of these modifications are shown in Figures 4 and 5.

After more than 2000 hours of testing and modifications (Table 2), a prototype box was produced that contained inner pack temperature variations of approximately 1°C. This box also allowed greater opportunity to effectively market the product in that it has the capacity to be divided into 2 self contained modules at any stage of the marketing chain.

From the final design R-max produced a moulded prototype box. Complexities in the design resulted in some mechanical teething problems in the manufacturing process related to the extraction of the box from the die. This problem was overcome by modifying the angles of the interior walls and reinforcing the corners in the coolant section.

Final prototype testing (commercial export tests).

The modified boxes were tested and gave comparable results to the final prototype box. A commercial run of boxes was produced and these were taken to two domestic prawn farms where they were packed and shipped along with shipments of the same prawns in the standard interim boxes.







Figure 3: Temperature record of the original prototype box.

Figure 4: Temperature record of the prototype with no holes against middle wall of coolant section.



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Figure 6: Temperature record of the final prototype box after all modifications.



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Bribie Island Aquaculture Research Centre The commercially produced boxes, which were packed and shipped to Japan, contained temperature loggers and a typical temperature record of a shipment is shown in Figure 7. It can be seen from this graph that it is possible to determine the external conditions (temperature) of the shipment and the resulting effects on internal temperature which shows the effectiveness of the new module in maintaining stable temperatures.





Discussion

Objective 1 of this project sought to evaluate the temperature stability of the new package. In doing so it became necessary to make a number of modifications to the package and evaluate the effects of the modifications on the performance of the package. The prototype testing proved to be a far more extensive process than originally anticipated. With the number of modifications, greater than 2000 hours of testing was required to achieve a product with a high level of temperature stability.

The package now encompasses a high level of temperature stability with the use of gel pack coolants of assorted sizes, used to achieve different performances in terms of time in transit and seasonal temperatures. The final emphasis in modifying the box was to achieve adequate air flow around the internal packages, across the base, at the sides and across the top of the internal packs. The main aim was to prevent temperature stratification between the upper and lower boxes. The top boxes tend to be cooled due to their close proximity to the coolant, while the middle and bottom boxes are subject to



heat entering from the outside. Therefore it is essential to ensure adequate internal airflow and heat transfer to the coolants in the upper compartment.

Having achieved a prototype design that worked and a temperature difference of less than 1°C between the top and bottom packs, we then proceeded to produce the manufactured product, which also proved to be a challenge.

In the polystyrene moulding process, heating and cooling of the mould are critical to the strength, durability and appearance of the expanded material. Because of the intricate nature of the design, the heating and cooling of the die proved to be an engineering challenge. As a result, a number of mechanical modifications were made to the die to ensure adequate bead bonding and facilitate the removal of the moulded box from the die.

The final commercial sample was evaluated with shipments to Japan in June 1997. Feedback from the Japanese indicated that the module was outstanding and very suitable in terms of its thermal properties and market advantages, as the module could be split into smaller 3 kg units. The capacity to adjust the coolants to compensate for changes in the environmental temperatures in Japan and Australia was acknowledged, especially as Japan moved further into summer. In the past, this has been a problem as increased mortality can occur in upper or lower areas of the box through excess cooling (top) or excessive heat entry (bottom) during the Japanese summer.

The commercial shipments were received well by the Japanese. Apart from the above mentioned features, the colour, shape, label and general aesthetics of the package were also considered to be very good. It is expected that the new package will be in use by Kuruma farmers in 1998 after existing packaging stocks have been depleted.

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Improving Packaging Technology, Survival and Market Options for Kuruma Prawns

OBJECTIVE 2: Ascertain/confirm upper lethal temperature limit for Kuruma prawns.

David Hewitt.

Introduction

The metabolic rate of poikilothermic (cold blooded) animals such as prawns increases with increasing temperature. The metabolic rate of *Penaeus esculentus* and *P. monodon* has been found to increase linearly between 20 and 35°C (Subramanian and Krishnamurthy, 1986; Dall, 1986). This rise in metabolic rate with temperature is accompanied by a demand for additional energy. In the animal itself, these additional energy requirements are expressed as increases in food uptake and oxygen demand for the oxidation of nutrients in the food. As the temperature reaches the upper thermal limit for the animal, mortalities occur which are attributable to the failure of enzyme systems, denaturation of proteins and/or asphyxiation. However, even before the thermal limit is reached other factors can act to reduce the overall condition of the prawn and therefore its ability to tolerate further stress such as harvesting, processing and transport.

In 1995, Kuruma prawns (*Penaeus japonicus*) that were produced in north Queensland under high growout temperatures and transported to the Tokyo markets, attracted low prices due to high mortalities during shipment. Events of this nature have highlighted the need to investigate the effect of warm ambient pond conditions on survival. Taiwanese farmers believe that survival is affected when Kuruma prawns are grown under conditions where pond temperatures may rise above 32°C. However, even before survival is affected, the condition of the prawn in terms of metabolism may be impacted enough to ensure that survival after processing, packing and transport is dramatically reduced.

The aim of the present study was to determine whether Kuruma prawn survival was affected by temperatures equivalent to those experienced in ponds located in north Queensland. By setting the temperature limit for harvesting just below the temperature at which survival drastically declined it was possible to determine whether these prawns would have a high level of survival during transport and if processing and packing modifications could improve survival.



Materials and methods

Live Kuruma prawns (wet body weight $15.6 \pm 0.2g$) were obtained from a commercial prawn farm, maintained in a twelve tonne tank equipped with a sand bottom and a sub sand filter and fed a commercial Kuruma prawn feed (CP J005 *japonicus* grower). After a two week period to recover from the stress of transport, prawns were transferred in groups of eight to 250L black polyethylene tanks equipped with sand bottoms and sub sand filters. Prawns were maintained for a one week period at ambient temperatures (approx. 20°C), then slowly bought up to the required temperature by raising water temperature at a rate of approximately one degree per day.

Experimental temperatures ranged from 28°C to 36°C in two degree increments with two tanks (replicates) per temperature. To achieve the required experimental temperatures, heated water (36°C) from a header tank was proportionately mixed in a sump (located between the experimental tanks) with 28°C water from another header tank. Air lifts cycled water from the sump to experimental tanks and then back to the sump, with overflow draining to waste. A 300W immersion heater was installed in the sump to control small fluctuations in water temperature.

To aid feeding and monitoring, the photoperiod was reversed when prawns were first stocked into the experimental tanks. The dark (night time) period began at 05:00hrs and ended at 17:00hrs. Prawns were fed at 08:00hrs, 12:00hrs and 16:00hrs with 40%, 40% and 20% respectively of the total daily ration. Feed rate was adjusted so that a slight excess of feed remained before the next feeding time. This residual feed was then removed before fresh feed was applied. Feed consumption was measured on two occasions for a five day period. The feed applied was weighed at each feeding period and leftover feed was siphoned from tanks prior to the next feeding time and dried at 105°C. A correction factor was applied for weight loss of feed caused by leaching. To determine the dry matter weight loss of the feed, pre-weighed feed pellets were gently agitated in distilled water at various temperatures for up to twelve hours, then dried and weighed. This generated a curve of dry matter loss over time that covered the four hour period the food remained uneaten.

Mortalities during the acclimatisation period were removed from the tank and replaced with prawns from the sump tank that had the same temperature history. When each tank had reached its experimental temperature (approx. one week) the experiment was started and observations were made for a four week period. All tanks were examined twice daily (night time period in experimental room) for moults and dead prawns, the presence of both was recorded and mortalities were replaced (as above) to negate any possible density effects. Differences in food consumption were determined using a one way analysis of variance.



<u>Results</u>

Analysis of experimental data indicated that there were no differences in the survival of Kuruma prawns at 28°C, 30°C and 32°C but there was 100% mortality of prawns at 34°C and 36°C before the end of the experiment (Figure 1). Accurate prawn growth rates could not be measured during the trial because of the replacement techniques that were used when mortalities were encountered (See: Materials and Methods). The moulting rates for Kuruma prawns were observed to be highest at 32°C, 28°C and 30°C, lower at 34°C and very low at 36°C (Figure 2). The shape of the moulting curves also differed with temperature. At 30°C and 32°C where survival was not significantly different, there was a constant rate of moulting. In contrast, at 28°C there was a degree of moult synchronisation with 69% of prawns moulting within a 9 day period, followed by a 8 day period where only one individual moulted, followed again by a 9 day period where 69% of prawns moulted.



Figure 1. Effect of water temperature on the survival of Kuruma prawns.

Growth rate in prawns is a combination of moulting rate and moult increment and although moulting rate was higher in the three lowest experimental temperatures without a knowledge of moult increment, growth rate cannot be estimated.





Figure 2. Effect of water temperature on the rate of moulting of Kuruma prawns.

Food consumption increased significantly (p<0.01) from 28°C to 30°C, was not significantly different between 30°C, 32°C and 34°C (p>0.05) but was significantly lower at 36°C (Figure 3).

Figure 3. Effect of water temperature on the food consumption of Kuruma prawns (mean \pm 1 SE).



Discussion

Upper lethal water temperatures for Penaeid prawns generally vary between species. Taiwanese prawn farmers suggest that 32°C is a critical bottom water temperature in Kuruma prawn culture as mortalities begin at this point. O'Brien (1996) found that juvenile *P. esculentus* showed satisfactory survival at temperatures up to 30°C when salinity was between 15 and 45 parts per thousand (ppt) but the upper temperature could be raised to 35°C if stress could be avoided by keeping salinity between 25 and 45ppt. The tropical prawn *P. monodon* suffered no mortality between 23°C and 34°C but temperatures below 23°C caused mortalities (Deering 1991, unpublished data).

In this study, using a laboratory scale system, mortality did not increase significantly until temperatures of greater than 32°C were reached. However, this work was performed in the absence of the water quality stress factors which are part of the normal growout pond environment. Such factors as diurnal fluctuations in dissolved oxygen, pH and temperature may well reduce the temperature that can be tolerated by the prawns.

As previously indicated it was not possible to measure growth of the prawns in this study. Although, when taken together, the moulting and the food consumption data suggests that prawns were still growing well at temperatures up to 32°C; but at 34°C a high level of food consumption did not result in a high growth rate as moulting was low. This may be due to a reduction in the digestibility of the feed at higher temperatures but is more likely a result of a dramatically increased metabolic load. The two degree increase to 36°C resulted in a significant drop in food consumption, moulting and ultimately, survival.

The upper lethal water temperature tolerated by Australian Kuruma prawns needs to be known so that pond management and design strategies can be put in place to prevent such high bottom temperatures eg. deeper ponds, greater water exchange rates. However, the temperature which is tolerated in the pond, and that from which prawns can be harvested and still survive, may not be the same. It is a common experience that Kuruma prawns harvested from ponds in southern Queensland under high temperature conditions (pond temperatures over 27°C) exhibit higher mortalities during live transport than prawns taken from 23°C ponds.

The moulting of Kuruma prawns in ponds has frequently been observed to be a synchronised event, with the majority of prawns moulting over a three day period. This synchronisation was observed at the 28 °C temperature in this study, but not at the higher temperatures. There appears to be a breakdown of the process at elevated temperatures but the reason for this is unknown. When prawns moult in synchrony harvests can be organised/timed to target inter-moult prawns. Research has shown that stage B (early post moult) prawns are most likely to suffer mortality during transport (Hewitt, unpublished data). If moulting is not synchronised, there will always be a constant proportion of prawns in the post-moult stage, putting a greater onus on manual sorting, itself not 100% effective, in order to minimise transport mortality. Thus, the breakdown of moult synchronisation as a result of increasing pond temperatures may have had a major bearing on the high level of mortality seen during this work.



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Improving Packaging Technology, Survival and Market Options for Kuruma Prawns.

OBJECTIVE 3: Determine if modifications to the preparation and packaging operations are likely to improve the survival rate of packaged prawns from high ambient temperature growout conditions.

Stuart Frost, David Hewitt, Bruce Goodrick and Stephen Grauf.

Introduction

Survival, appearance and quality are critical factors affecting the market price for live Kuruma prawn (*Penaeus japonicus*, Bate) export. Premium prices for this product are achieved when the survival rate of prawns arriving at the market is reliably in excess of 95% (Goodrick et al., 1993). Ensuring high prawn survival rates at the market is paramount to the viability, profitability and future expansion of this small industry.

When exported, Kuruma prawns are normally cooled in seawater to a point at which they are unable to right themselves (10-14°C) and then packed with chilled dry sawdust into cardboard cartons (Shigeno 1979). Once anaesthetised by the cold, handling them during packing has no effect upon their respiration rate (Paterson, 1993a). When packed in this manner and providing that temperatures of 12-15°C are maintained, the prawns can survive for up to 30 hours out of water (Goodrick et al., 1993). However, Kuruma prawns may need to be cooled to lower temperatures in winter than in summer. The ability of low temperatures to immobilise Kuruma prawns (and hence maintain their survival during live export) is related to the prawns habit of burying into (and remaining in) the substrate during periods of extreme cold (Paterson 1993b). This raises the question of what happens when the prawns are grown in tropical regions.

There is considered to be a general correlation between warm ambient conditions such as temperatures in the mid to high 20's and a reduction in the survival rate in-transit. This has highlighted the need to address the suitability of current Kuruma prawn packing methods when applied in warm climates. This study examines the effect of warm ambient pond conditions on survival as well as the potential to modify preparation and packing methods to minimise the adverse effects of warm ambient conditions.



Materials and methods

Raw Materials - Prawns

P. japonicus used for experimentation were obtained from two sources:- Gold Coast Marine Hatcheries Pty Ltd, Woongoolba and Luxe Enterprises Pty Ltd, Ningi; on the southern coast of Queensland. Prawns of either sex, ranging in weight from 6.8 to 18.2 grams (average 11.1 +/- 2.61g) and moult stage C-D1 (Smith and Dall, 1985) were used. Prawns were harvested and transported in seawater, using an insulated 800L live transport tank (aerated via bottled oxygen), to the Bribie Island Aquaculture Research Centre laboratories. Travel time between farms and the research laboratory ranged from 25 to 80 minutes. Prawns to be used for experimentation were then kept in laboratory scale seawater tanks (1000L) with sand bases (5 cm deep). The stocking density per tank was 40-45 prawns per square metre. All tanks utilised a flow through water system, and each tank contained an sub-sand filter system. Water salinity in each tank ranged from 28 ppt to 32 ppt, while water temperature was controlled to within 0.1°C via temperature process controllers.

Acclimatisation

A series of seawater tanks each containing prawns (n=90) were brought up to temperatures of 23°, 27° and 31°C, by increasing water temperature at a rate of 1°C per 24 hour period. Prawns were then allowed to acclimatise (in tank) for four days at each respective temperature prior to harvesting. During temperature increment and acclimatisation, the prawns were fed daily with commercial Kuruma feed (CP J005 *japonicus* grower). The feed pellets were broadcast by hand into the tanks each evening and uneaten feed was removed the following morning.

Harvesting

As per industry practises, prawns were harvested after dusk. Prawns were removed from the tanks via a 12mm mesh net that had been placed between the sand base and the filter. Each of the four corners of the mesh net were removed from the sand and pulled up flush against the sides of the tank. Prawns were then crowded by moving the ends of the net towards the centre of the tank. The net was then pulled up out of the tank with the prawns in situ. This method removed all prawns both those actively swimming or still buried. The prawns were then transferred into floating harvest trays. When required small batches of prawns (n=40-45) were transferred from the harvest trays into 35L mesh based plastic tubs and carried a short distance to the cooling baths.

Cooling regime

Both standard commercial methods (controls) and modified methods (treatments) were used to inactivate the prawns for packing. The 35L mesh based tubs were placed in one of a series of temperature controlled, aerated 120L seawater baths for a period of 15 minutes. Each bath in the series contained seawater (28ppt salinity) at differing temperatures to allow a temperature drop of 4-7°C at each stage. The cooling regimes (controls and treatments) used for each batch of acclimatised prawns are presented in Table 1. A treatment utilising a food grade aquatic sedative (AQUI-S[™]) was also included for this study.



| Description | Acclimitisation | climitisation Seawater Bath Temperatures | | | | Final Storage |
|--------------------------|-----------------|--|------|------|------|---------------|
| | Temperature | No.1 | No.2 | No.3 | No.4 | Temperature |
| | (°C) | (°C) | (°C) | (°C) | (°C) | (°C) |
| Control 1ª | Ambient | 18 | - | - | 14 | 14 |
| Control 2 | 23 | 18 | - | - | 14 | 14 |
| Control 3 | 27 | 22 | 18 | - | 14 | 14 |
| Control 4 | 31 | 26 | 22 | - | 16 | 17 |
| Treatment 1 | 31 | 26 | 22 | - | 16 | 14 |
| Treatment 2 | 31 | 26 | 22 | 18 | 14 | 14 |
| Treatment 3 ^b | 31 | 24 | 18 | - | 16 | 14 |

Table 1: Cooling regimes.

^a : Control taken from ambient pond temperatures of 20°C as per commercial harvest procedures, no acclimatisation period provided.

^b : AQUI-S[™] added to first two baths at a rate of 30ppm (30mg/L).

Packing, storage and survival

After cooling in the final water bath, inactive prawns were then sorted to remove soft (post moult) and/or weak animals. The sorted prawns were then packed (n=30) in dry, chilled (4°C) hoop pine sawdust into 1kg standard cardboard 'inner' boxes. Duplicate boxes were prepared for each treatment. The packed boxes were then stored in temperature controlled rooms for a period of 36 hours. After 36 hours the survival rate of the prawns in each box was assessed. Prawns that showed strong reflexive movements (kick, tail splay and head arch) when unpacked were counted as alive. Moribund prawns, that is those that showed only feeble movements of the legs and gill bailer were counted with the dead prawns, as past experience showed that these would not recover.

Holding tank survival

Surviving prawns from each treatment (post packing) were placed into 120L sand bottomed aquarium tanks (water temperature 25+/-0.5°C; 28ppt salinity); each tank holding the surviving animals from one duplicate of each treatment. The tanks utilised a flow through system and a sand substrate. Prawn survival was monitored daily over a seven day period. The animals were fed every evening with a commercial Kuruma feed and water quality and mortalities were monitored and recorded each morning.



<u>Results</u>



Figure 1: Effect of holding temperature and modified cooling and packaging regimes on *P. japonicus* survival post 36 hours dry storage.

20°C = Control 1; 23°C = Control 2; 27°C = Control 3; 31°C^a = Control 4; 31°C^b= Treatment 1 (modified cooling regime); 31°C^c= Treatment 2 (modified cooling regime); 31°C^d= Treatment 3 (modified cooling regime; aquatic sedative AQUI-S[™] added).

The data indicates a significant decline in post-packing survival as pond temperatures increase from ambient (20°C) to temperatures of 31°C. Control treatment prawns packed from water held at 20°C, 23°C, 27°C and 31°C returned survival rates of 83%, 79%, 63% and 61% respectively on unpacking after 36 hours dry storage (Figure 1).

Modified cooling and packing operations showed no real increases in survival rates for prawns from high ambient water temperatures (Figure 1). Prawns held at 31°C and gradually cooled to final storage temperatures of 14°C or 17°C resulted in low survival rates ranging between 43% to 61%. The addition of AQUI-STM to the cooling water (Treatment 3) returned the highest survival rate of 61%; the same survival rate achieved for the 31°C control. Modifying the packaging regime to include a final storage temperature of 14°C (Treatment 1) gave the lowest survival rate of 43% after 36 hours dry storage. The prawns, when cooled more gradually by the addition of an extra cooling step in conjunction with a final storage temperature of 14°C (Treatment 2) returned a survival rate of 53%.





Figure 2: Survival of *P. japonicus* held in seawater tanks after 36 hours dry transport.

A high number of mortalities occurred within the first 48 hours for prawns held in seawater tanks following 36 hours storage (Figure 2). Prawns from high ambient temperatures (31°C) and subjected to modified cooling and packing regimes showed the most significant mortalities. Prawn survival for Treatments 1, 2 and 3 was reduced to less than 10% after 48 hours. The same degree of mortality was not seen in control treatment prawns.

Table 2: Stage of moult of *P. japonicus* mortalities post 36 hours dry storage.

| Treatment | Pond Temperature (°C) | Mortalities (n) | % Post moult (B-C) | % Pre- moult (D0 - D4) |
|-----------|--------------------------|--------------------|--------------------------|------------------------------|
| Control 1 | Ambient (20°C) | 25 | 90 | 10 |
| Control 2 | 23 | 11 | 35 | 65 |
| Control 3 | 27 | 25 | 90 | 10 |
| Control 4 | 31 | 15 | 85 | 15 |

A study of the mortalities from Controls 1, 3 and 4 revealed that a large proportion of prawns (>85%) had just undergone a moult event ie. B-C (early post moult). Mortalities from the second control group (Control 2) were classified as D0 - D4 (pre-moult), this indicating that the prawns were about to undergo a moult event.



Discussion

The normal method of preparing Kuruma prawns for packing and transport involves cooling them in a series of water baths to allow a 6°C temperature drop at each stage and a final storage temperature of approximately 14-15°C (Shigeno, 1979). This method generally returns survival rates in excess of 95% (Goodrick et al., 1993). The poor survival rates obtained for the modified cooling treatments suggest that a final storage temperature of 14°C may in fact be detrimental when packing from high ambient pond temperatures, as seen by the results obtained for Treatments 1, 2, and 3. Gradually cooling the prawns from 31°C to 14°C, by the addition of extra water baths (Treatment 2) resulted in only a slight increase in survival. However, this treatment also had a final storage temperature (14°C) 3°C lower than the high ambient control sample. Samet et al., (1996) found that long cooling periods (in excess of 5 hours) prolonged in-transit survival. Pre-cooling times such as these are not viable for Australian exports due to the short turn around required from point of harvest to air However the findings from the present study and those of freiahtina. Samet et al., (1996) do suggest that extra cooling stages have some effect in increasing in-transit survival rates.

The use of aquatic sedatives, such as AQUIS[™], when added to cooling water shows some promise for increasing prawn survival when packing and transporting from high ambient temperatures. Concentrations of 30ppm increased survival slightly (Treatment 3) despite storage at 14°C which was detrimental to other treatments. Results also indicate that the sedatives may provide a buffer for greater initial temperature drops during prawn cooling.

Control samples of prawns returned lower survival rates than anticipated. Observation of the mortalities from each control treatment revealed that a large percentage of prawns had just undergone or were about to undergo a moult event (Table 2). During the final stages of a moult event, a prawns' metabolism increases and its energy reserves become depleted leaving the prawn significantly weakened (Dall, 1986). When these factors are added to the stress incurred through elevated pond temperatures, harvesting, handling and packing, the animals chances of survival are dramatically reduced. This may suggest that the normal practise of grading out soft shelled animals prior to packing does not eliminate pre-moult prawns which are metabolically stressed and are likely to incur high mortalities. Thus, the development of more discerning methods that establish the condition of the prawn prior to packing and transport should assist greatly in increasing survival rates.

The practice of holding post-transport Kuruma prawns alive in aquarium tanks for the Japanese restaurant trade is not an uncommon one. However, the survival data collected during this study would seem to suggest that this practice would be expensive due to the mortalities that occur. For all treatments, high degrees of mortalities occurred within the first 48 hours after unpacking and introduction into the tanks. This would seem to suggest that prawns, significantly weakened through the stress of packing and transport, have little chance of survival even when held under optimum conditions. As previously indicated high ambient pond temperatures and stage of



moult also have serious implications with regard to transport and post-transport survival. To obtain better transport and post-transport survival rates a more detailed prawn history is required. Aspects such as growout pond temperatures, preparation and packing treatments as well as physiological parameters, for instance, stage of moult and general overall condition of the prawn, should be of great benefit in predicting future survival.

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CENTRE FOR FOOD TECHNOLOGY



Bribie Island Aquaculture Research Centre

Improving Packaging Technology, Survival and Market Options for Kuruma Prawns.

Objective 4:- Establish the potential feasibility of transporting live Kuruma prawns from one location to another to optimise growout conditions and optimise quality through climatic agistment.

Stuart Frost, David Hewitt, Bruce Goodrick and Stephen Grauf.

Introduction

In Australia live Kuruma prawns (*Penaeus japonicus*) are normally shipped long distances via air freight, a method of transit that has been well established now for a number of years. The prawns are transported live using modified traditional Japanese techniques, where they are first cooled in water at 10°C - 14°C to reduce activity and then packed with dry sawdust into cardboard cartons (Shigeno, 1979; Goodrick et al., 1993). This system is applicable only to small quantities of premium priced animals. Thus, the bulk transportation of large numbers of these animals by other means has not been fully investigated, with little published information available.

For short distance and overland transportation of live prawns, water is the most common medium used in either closed or open systems (Richards-Rajadurai, 1989). Bulk wet transportation of live penaeids is limited by the animal's metabolism and transport environment in which they are shipped. The critical factors affecting survival during transport may be listed as: oxygen supply, accumulation of toxic metabolites (CO₂, ammonia etc), pH and temperature of water, stress and injury, stocking density, and time period held in transit (Robertson et al., 1987; Richards-Rajadurai, 1989). These factors, if controlled have the ability to increase survival, allow increased packing densities, and reduce transport costs.

Penaeid metabolic waste production can be controlled via a number of physical methods and by the use of suitable chemicals that remove these waste products. The lowering of transit water temperature reduces activity and metabolic rate, thus oxygen consumption is reduced as is the accumulation of carbon dioxide and ammonia. Aquatic sedatives when used correctly can also reduce activity and metabolism thereby reducing the amount of carbon dioxide and ammonia excreted. Chemical buffers may be added to transit water to maintain constant pH levels, while water conditioners may be added to remove accumulated ammonia and nitrites.

There is considered to be a general correlation between warm ambient growout conditions such as temperatures in the mid to high 20's and a reduction in the survival rate in transit. Whether this is related to higher packing and transport temperatures or to an intrinsic condition of the prawns at these temperatures is uncertain. Thus, there



is a need to determine and formulate a strategy if Kuruma prawns are to be grown in warmer climatic conditions, such as in northern Queensland. It is also necessary to determine whether it is possible and feasible to transport these prawns in bulk to alternate sites in cooler climatic areas for "conditioning". Australian Kuruma prawn farmers have expressed an interest in transporting large numbers of prawns from one area to another to take advantage of optimum growout and climatic conditions. The present study was conducted to establish the feasibility of transporting large quantities of Kuruma prawns in a closed system for periods up to 12 hours.

Materials and methods

Preliminary Trial

To study the effect of prawn activity during transport, live Kuruma prawns (*Penaeus japonicus*) were collected from a local commercial prawn farm (Moreton Bay Prawn Farm) and maintained at a density of 40-45 prawns/m² in a series of seawater tanks equipped with sand bases and sub-sand filter systems. Ambient tank water temperature was brought up to 27-28°C at a rate of 1°C/day to represent the high ambient growout conditions achieved in north Queensland. Small batches of prawns (n=15-30; 16.3 +/- 0.5gm wet body weight) were then harvested, weighed and placed into 4L round bottom plastic vessels with enough seawater (19°C) to provide a transport density of 500g of prawns / 1L water (1:2 ratio). Bottled oxygen was supplied to each vessel; and vessels were subjected to 30 hours simulated transport under conditions of constant light or constant darkness. Water quality (ammonia concentration) and prawn survival in each vessel was monitored and recorded at 6 hourly intervals.

Transport Trials

Pond reared *P. japonicus* (21.9+/- 5.6 g wet weight) were collected from the same commercial prawn farm as above. Prawns were removed from the growout ponds (water temperature 26-28°C) via baited traps, placed into wooden harvest trays, and transported to the laboratory in a 800L live transport tank. During transport, tank water was aerated via a small 12v DC current aquarium pump. Bottled oxygen was also supplied using air lines and a diffuser system. Upon arrival, trays containing the prawns were transferred to a small laboratory scale seawater tank (25°C) for a brief period (45 minutes) before each of the following three (3) treatments was applied.

• 1: Effect of low temperature, low density

Duplicate groups of prawns (each n=15) were removed from the harvest trays, weighed and placed into a pair of 4L round bottom plastic vessels with enough seawater chilled to 14°C to provide a transport density of 100g of prawns / 1L water (1:10 ratio). Bottled oxygen was supplied to each vessel and the internal water temperature maintained by placing each vessel in a 14°C water bath. Vessels containing prawns were then subjected to 12 hours simulated transport. Water quality was monitored at 2 hourly intervals while prawn survival in each vessel was recorded after 12 hours.



• 2: Effect of low temperature, density, and aquatic sedative

Duplicate groups of prawns (each n=15-30) were removed from the harvest trays, weighed and placed into seawater (25°C) dosed with the aquatic sedative AQUI-STM (Institute of Crop and Food Research Inc., New Zealand) at a rate of 30mg/L. A 25 minute contact time was allowed for anaesthesia to occur. Prawns were then removed and placed into a pair of 4L round bottom plastic vessels with enough seawater chilled to 14°C to provide transport densities of 100g of prawns / 1L water (1:10 ratio) and 250g of prawns / 1L water (1:4 ratio). This water was also dosed with AQUI-STM at a rate of 20mg/L. Oxygenation, temperature maintenance, and water quality monitoring were followed as previously described.

• 3: Effect of low temperature, high density, aquatic sedative and water conditioner Duplicate groups of prawns (n=30) were removed from the harvest trays, and treated with the aquatic sedative AQUI-S[™] as described above. Prawns were then removed and placed in 4L round bottom plastic vessels with enough seawater chilled to 14°C to provide a transport density of 250g of prawns / 1L water (1:4 ratio). The water conditioning product Prime[™] (SeaChem Laboratories Inc., Stone Mountain, GA) was added to the chilled transport water at the maximum recommended dosage of 5 drops per 4.5L. Oxygenation, temperature maintenance, and water quality monitoring were followed as previously described.

<u>Results</u>

Table 1: Survival of live Kuruma prawns in simulated wet transport at 19°C and stocking density of 500g/L (1:2) under conditions of constant light and constant darkness.

| Time | Constant light | Constant darkness | | |
|---------|----------------|-------------------|--|--|
| (hours) | % Alive | % Alive | | |
| 0 | 100 | 100 | | |
| 6 | 95 | 81 | | |
| 12 | 81 | 71 | | |
| 18 | 76 | 71 | | |
| 24 | 71 | 62 | | |
| 30 | 38 | 19 | | |

Table 2: Changes in transport water total ammonia concentration during transport of live Kuruma prawns under light and dark conditions.

| | Constant light Constant darkne | |
|------|--------------------------------|---------------|
| Time | Total ammonia | Total ammonia |
| | (ppm) | (ppm) |
| 0 | 0 | 0 |
| 6 | 10 | 10 |
| 12 | 20 | 15 |
| 18 | 38 | 19 |
| 24 | 53 | 29 |
| 30 | 91 | 83 |



Preliminary data collected showed that Kuruma prawns held under conditions of constant light during simulated transport returned survival rates of 95%, 81% and 38% over 6, 12 and 30 hours respectively (Table 1). Survival rates for prawns transported under conditions of complete darkness were significantly poorer; with 81%, 71% and 19% for the same time periods. A study of the mortalities from this treatment revealed that cannibalism had occurred. Cannibalism was also observed in light transported prawns, although to a much lesser extent. Decreases in transport water pH was observed to be more rapid for prawns transported in the dark. The opposite was observed for total ammonia concentrations. The transport water for prawns stored under light conditions showed very rapid increases in ammonia concentration, with 10ppm, 20ppm and 91ppm recorded after 6, 12 and 30 hours (Table 2).













Figures 1 and 2 show data collected from the transport trials. Although not shown, the survival rate for all treatments, including the control, was 100% after 12 hours simulated transport. Water quality parameters did, however, vary between treatments. Total ammonia levels were highest (16ppm ammonia) in the low temperature, high density (1:4) treatment to which the water conditioner Prime[™] was added, lower in the control sample held at 1:10 stocking density at 25°C (12ppm ammonia), and lowest in samples held at stocking densities of 1:10 at 14-16°C. Ammonia levels in the water conditioner treated sample appeared to stay below those of the control for the first 10 hours of transport before rising sharply. Water pH for the control sample decreased rapidly for the first four hours, but after this period, rose to a similar level of other treatments. Data collected also showed that there was little significant difference between treatments in regard to transport water pH.

Discussion

Preliminary data suggests that acceptable survival rates are achievable when Kuruma prawns are transported 'wet' under conditions of constant light for periods up to 12 hours. During daylight hours the natural protective instinct of the Kuruma prawn is to bury into the substrate and remain quiet. When in this rested state the animals' metabolic and respiration rates are significantly reduced (Egusa, 1960). However, if the prawn is unable to bury itself, stress occurs. Under stressful conditions such as this, excessive handling and escape from predation, the resultant metabolic and





respiration processes lead to greater excretion of waste products (eg CO2 and ammonia).

High ammonia levels were seen in transit water for both treatments over the 30 hour transport period. However, the more rapid accumulation of ammonia seen for prawns transported under constant light may indicate that the animals were stressed due to being unable to bury. Observations made throughout the study seem to support this as prawns were seen to be actively trying to bury in the base of the transport vessel.

Kuruma prawns are nocturnal by nature and normally spend this period either feeding or actively searching for food. In the present study, cannibalism was observed in animals transported under conditions of complete darkness; however the same degree of cannibalism was not seen in animals transported under constant light. This observation would suggest that prawn activity and feeding behaviour was increased by the 30 hour period of constant darkness during transport. Additionally, the absence of other food sources and the close quarters involved in bulk transport would have allowed weak or injured prawns to be predated upon.

The primary method of minimising the inevitable decline in water quality during transport is to reduce the metabolic processes of the animal. Both the chilling of transport water and use of anaesthetics are effective means of reducing prawn metabolism and thus, the production of toxic metabolites such as CO_2 and ammonia (Richards-Rajadurai, 1989). Trial data shows that chilling transit water to 14-16°C, and maintaining this temperature for the duration of transport, substantially reduced the accumulation of ammonia. A similar finding was also obtained, when the aquatic sedative AQUI-STM was added to the transport water.

Control of ammonia in shipping water is essential because the toxicity of ammonia increases with increasing pH values. Past use of ammonia oxidising compounds and bacteria added to transport water to remove or lower total ammonia levels have proved successful (Meade, 1985). In the present study, the water conditioner PRIME[™] was effective in maintaining low ammonia levels for a period of time. Ammonia levels were observed to be well below those of the control, (even with twice the density of animals per litre of transport water) for the first 10 hours of transport, before rising sharply. This would seem to suggest that the effectiveness of the conditioner stopped at the 10 hour period. However, it may be feasible to increase the required dose rate of the conditioner to allow for greater effectiveness over long transport periods or when high densities of prawns are to be shipped.

Normally, respiration results in carbon dioxide production. In a closed system CO_2 can build up to create levels toxic to the animal itself; and this build up causes the water to become acidic, resulting in a drop in pH. In the present study there were little or no differences in pH data collected for all treatments. Data showed that carbon dioxide (CO_2) build up and the resultant pH decline was reduced by bubbling bottled oxygen continuously through the transport water. The effect of high oxygen concentrations in the shipping water had no adverse effects on the prawns. This observation is



supported by Robertson et al (1987) who found that supersaturated oxygen levels had no detrimental effect on in-transit survival of *P. setiferus*.

Transport trials involving 12 hour shipments of pond reared *P. japonicus* resulted in 100% survival for all treatments studied. High transport densities of 250gm/L (1:4) had little effect on prawn survival though significant deteriorations in water quality were observed. Results suggest that even without control of the water temperature or quality, prawns could be transported for periods up to 12 hours. However, if extended periods of transport such as interstate travel are to be achieved; temperature reduction, the use of anaesthetics and water conditioners should be considered.

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Best Practice Manual for use of the Advanced Packaging Module for Kuruma Prawns

CENTRE FOR FOOD TECHNOLOGY



Bribie Island Aquaculture Research Centre

FISHERIES RESEARCH & DEVELOPMENT CORPORATION



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Best Practice Manual for use of the Advanced Packaging Module for Kuruma Prawns

Background

Because of Australia's isolation from the Japanese market, the packaging system for Kuruma prawns needs to resolve several issues. The system must provide a stable temperature and humid environment and allow the prawns to be packed in a suitable support medium. The system needs to be insulated from the external environment and contain coolants to compensate for heat entering the module.

Kuruma prawn farmers reached a consensus that there is a need for Australian producers to be technically advanced and identified on a country basis as a supplier of quality produce. It was agreed that a common inner package size would facilitate the production and use of improved outer packaging. It was also suggested that smaller net weight (ie. 6 or 8kg instead of 10kg) packages would offer greater access to a broader range of buyers and hence strong prospects for higher prices (Kitada, Austrade Osaka). In response to these issues a new generation of packaging module has been designed by the Centre for Food Technology (CFT) in co-operation with Rmax (Huntsman Chemical Company Australia Pty Ltd). This system differs from existing packaging in that it is compact, has superior insulation and provides greater temperature stability while still being comparable in cost. The package itself contains a number of technical innovations allowing greater control of temperature as well as the capacity to vary the coolant system. This manual will give instructions on how to best use this box and system.

Preparation

General Conditions

The processing establishment will need to have Australian Quarantine and Inspection Service (AQIS) registration as a live export facility and good hygiene practices should be maintained.

The condition of the prawns to be packed is of vital importance. Current research by the Centre for Food Technology (CFT) and Bribie Island Aquaculture Research Centre (BIARC) will help to establish a *Condition Index* (CI) to assist in predicting the survival of prawns during transport. In general, operators should ensure prawns are in a clean healthy state, with a firm shell before initiating the harvesting operation.

Note: Care should be taken to minimise stress when handling the prawns during harvesting, grading and packing.

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Labelling

Packing labels should be affixed to the outer box prior to the nights packing and contain all the information required by AQIS. Labels should include a correct product description and identification of the exporter. Label design should also take account of the Japanese culture and make appropriate use of colours.

Coolants

Freezing coolants

Gel coolants are recommended as they have a longer duration and less chance of leakages. Coolants should be frozen flat and the most efficient and space saving way to achieve this is to place the coolants in stackable mesh trays. Coolants may also be frozen in the coolant section of the package which can be stacked in the freezer ready for use. The bottom coolant section must however be placed on a mesh base above the floor to allow ventilation of the stack while the coolants are freezina.

Specific Heat Effects

Coolants are frozen at -30°C and prawns are packed between 10-14°C, therefore a temperature difference of up to 45°C can occur. If coolants are placed immediately into the box from the freezer, the specific heat absorbed by the warming coolants can rapidly reduce the prawns body temperature and be detrimental to the survival of the prawns. Coolants should therefore be tempered to approximately -5°C before the coolant section of the box is placed on the base. This can be achieved by placing the coolants on a bench and measuring the temperature between two coolant packs until -5°C is reached. Generally, it is adequate to separate the coolants at ambient temperatures for 15 to 30 mins or leave the coolants in the insulated coolant section at ambient temperatures for 1 to 2 hours prior to use.

Choice of Coolants

When selecting a suitable coolant to use, several factors must be taken into account:-

- Ambient temperatures both in Australia and Japan
- Surface area of the coolant
- Nett weight of the coolant.

The weight of the coolant will largely govern the duration of the coolant, while surface area will determine its cooling power. Insulation such as bubblewrap will reduce the cooling power and extend coolant life.

Table 1 shows the thermal performance of some insulated and uninsulated water based coolants.



Table 1: Thermal performance of various coolants.

| Weight (g) | Dimensions (mm) | Surface Area (mm²) | Insulation | Coolant Life (Hours) |
|---------------|--------------------|-----------------------|----------------|-------------------------|
| 250 | 125*125*20 | 31250 | No | 12 |
| 500 | 180*125*30 | 45000 | No | 18 |
| 500 | 180*125*30 | 45000 | 3mm bubblewrap | 24 |
| 750 | 230*125*30 | 57500 | No | 20 |

Packing

Prawn Packing

After prawns have been cooled in seawater to a point at which they are unable to right themselves (10-14°C), they are packed with chilled dry sawdust into cardboard cartons. These inner packs are then held at 12-14°C prior to master carton packing. The master module is loaded with 6 packs in a horizontal configuration as shown in Figure 1.

When packed in this manner and providing that internal temperatures of 12-15°C are maintained, the prawns can survive for up to 30 hours out of water (subject to prawn condition, season, harvesting and processing).

Figure 1: Inner packs are placed into master module



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After the prawns are packed, the coolant section can then be put in position and taped into place. The specified pre conditioned coolant is placed into this section and the lid fitted and taped (Figure 2). Care should be taken with coolant conditioning to avoid mortalities due to overcooling as described in the specific heat effects section.



Figure 2: Lid and coolant section are slotted onto the base and taped into place.

Master Module Storage

As the master box contains a coolant, it is best stored at 15-20°C until transfer to the airport. Care should be taken not to over-chill the box as this may bring the prawn temperatures down to lethal levels. It is therefore recommended that the storage temperature of these master modules be monitored.

Aircraft Conditions

It is worthwhile checking with your agent to find out whether the shipment will be transported in a heated hold. It is recommended that the shipment be transported in a non heated hold. Even though the boxes are stacked together, creating an insulating effect, this may still have an influence on the coolants and box temperature in the outer layers.

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By sending temperature loggers it is possible to monitor the in-transit temperatures and accordingly adjust the coolant and holding conditions (where possible) to ensure the prawns remain at optimum temperatures throughout the journey.

Temperature monitoring of Shipments

Previous work has demonstrated the importance of monitoring prawn shipments. By receiving detailed temperature data, product can be traced from the time of packing until the point of sale. This information should then be cross checked with information from Japanese agents to determine what is actually happening to the product.

By using temperature loggers, the life of the coolant can be determined and the temperature of your product checked on unpacking. Flight delays, stopovers and aircraft hold temperatures can be monitored and coolant size can be adjusted for seasonal temperatures in Japan.

At the time of preparation of this manual, temperature logging of commercial prawn shipments is being monitored through a DPI New Initiatives funded project and further information or technical assistance can be obtained from Steve Grauf on 07 34068619.



