FISHERIES RESEARCH CONTRACT REPORT NO. 11, 2005

Yabby hybrid growout experiment

FRDC Project No. 97/319.02

Aquaculture Development Fund of Western Australia, Project No. 41

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Department of Fisheries

AQUACULTURE DEVELOPMENT COUNCIL

Australian Government
Fisheries Research and Development Corporation

Fish for the future

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Yabby hybrid growout experiment. FRDC Project No. 97/319.02

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OBJECTIVES

1. Confirm the feasibility of large scale production of all male hybrid progeny
2. Record the growth rate of hybrids
3. Compare the growth rate of hybrids with WA yabbies
4. Develop a *Thelohania* free broodstock population
5. Develop a commercial options paper for transferring the technology to industry, including an economic costing of various production scenarios for producing male progeny.

Non-technical Summary

OUTCOMES ACHIEVED TO DATE

This project will increase returns to yabby farmers through the production of larger and consequently more valuable hybrid *Cherax rotundus* x *Cherax albidus* crayfish.

It has been shown in FRDC project 94/075 that hybridising two species of freshwater crayfish, female *Cherax rotundus* x male *Cherax albidus* produces only male progeny. This hybrid offers considerable potential for controlling reproduction by yabbies in aquaculture ponds. In FRDC project 97/319.02 the production of only male progeny from this hybrid has been confirmed in multiple crosses. In addition, the male *C. albidus* x *C. rotundus* hybrids did not produce progeny when mated with *C. albidus* or *C. destructor* females.

In FRDC 94/075 the all male hybrids were shown to grow faster than mixed sex groups of *C. albidus* in aquaria. In FRDC project 97/319.02 this comparison was repeated in model ponds where growth can be assessed more realistically. The hybrids grew almost twice as fast as *C. albidus* yabbies. The average growth rate of hybrid *C. albidus* x *C. rotundus* yabbies was greater (0.11 ± 0.004 g/day) than that of *C. albidus* mixed sex yabbies (0.06 ± 0.002 g/day). The growth rates of *C. albidus* x *C. rotundus* hybrids and the *C. albidus* yabbies were similar until they reached sexual maturity, at around 14 g, at which stage the growth rate of *C. albidus* decreased. At the conclusion of the experiment the *C. albidus* x *C. rotundus* hybrids (50.7 ± 2.2 g) were almost twice as large as the *C. albidus* yabbies (27.2 ± 1.7 g). The greater growth and increased numbers of yabbies in the larger and therefore more valuable size categories resulted in the final harvest value of *C. albidus* x *C. rotundus* hybrids being 4.8 times greater than that of the *C. albidus* yabbies.

Sensory evaluation showed that the taste of the hybrids is comparable with currently farmed *C. albidus* yabbies.

While the survival of hybrids (63%) in a simulated transport trial was lower than *C. albidus* yabbies (90%), it was greater than that reported previously for transport survival of *C. albidus* (Roe 1994), and due to the low numbers of animals should be repeated when more hybrids are available.

An economic evaluation of three potential methods for producing hybrids 1) Stocking breeding ponds, 2) Cage reproduction and 3) Hatchery production, shows that a small farm producing $5000 of *C. albidus* yabbies/year could expect to increase returns to $23,000/year. This has contributed to the subsequent process for obtaining translocation approval to move *C. rotundus* out of quarantine in WA for commercial aquaculture of hybrids. Unfortunately, unless there was agreement to establish a government operated hatchery, it is unlikely that this approval will be granted.

While stocks of both *C. rotundus* and *C. albidus* are available in NSW, elsewhere in Australia, commercialisation of this hybrid is limited by availability of *C. rotundus*. Space and environmental limitations have restricted the quantity of *C. rotundus* that could be produced in this study. To overcome this challenge a new project with local funding was undertaken by the Department of Fisheries in WA to produce potential broodstock for farmers in larger ponds. Production was limited by management factures and the high incidence of *Thelohania* in the available farmed population of *C. rotundus* interstate.

GENERAL CONCLUSIONS AND RECOMMENDATIONS

The hybrid produced from mating female *Cherax rotundus* x male *Cherax albidus* offers considerable potential for increasing the profitability of yabby farming by providing increased growth and controlling reproduction in aquaculture ponds.

This simple and readily available technology should be applied by farmers to increase profits from yabby farming. It is recommended that commercial trials of this hybrid be established in NSW where these two species already occur. In SA and Victoria relevant state authorities should consider permitting yabby farmers to import *C. rotundus* from NSW, if necessary. In WA a *Thelohania* free broodstock population was being developed to provide animals for industry, but as indicated above survival in experimental wheatbelt ponds in Western Australia was poor.

Please note that at the request of the Aquaculture Development Council (ADC) of Western Australia, the final reports to ADC and FRDC, which co-invested in this research, have been updated in this Research Contract Report, particularly in relation to translocation and commercialisation issues.

Key words : *Cherax*, yabbies, hybridisation, hybrids

GENERAL INTRODUCTION

Background

The FRDC yabby project 94/075 has shown that the major factor limiting growth of yabbies in WA farm dams is density. Yabbies attain high densities in farm dams due to uncontrolled breeding. The combination of high densities with uncontrolled breeding results in a high proportion of yabbies from WA farm dams being below the current minimum market size of 30 g (Figure 1.1).

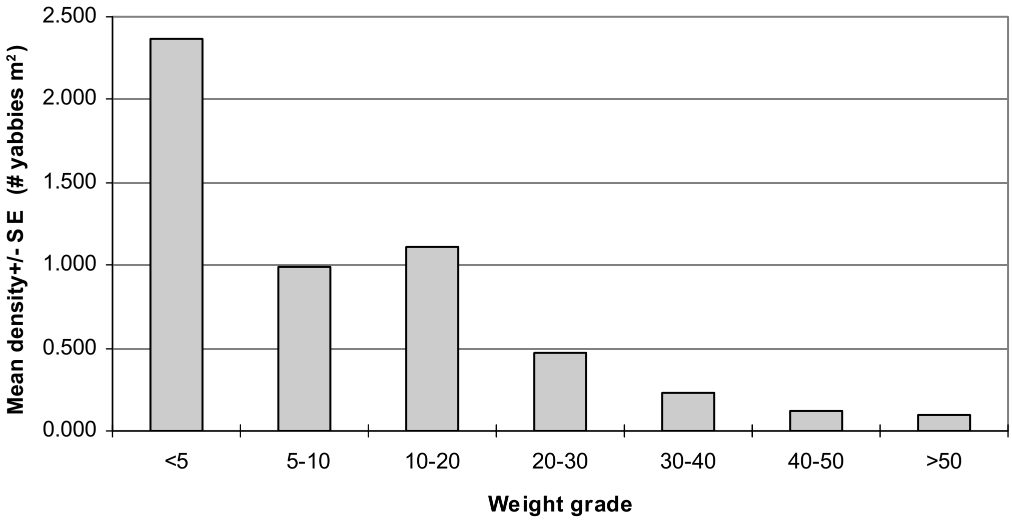

Figure 1.1 Size distribution of yabbies from 21 farm dams.

It has also been shown in the FRDC 94/075 yabby project that male yabbies grow faster than female yabbies and, if the sexes are separated, males grow even faster (Lawrence *et al.* 1998, Lawrence *et al.* 2000).

One approach to solve the problem of uncontrolled breeding would be to stock yabbies which could not reproduce (i.e. only one sex or sterile). A number of farmers recognise the value of monosex production but find manual sexing of yabbies labour intensive and prone to errors.

Department of Fisheries WA scientists working on the genetics component of the FRDC 94/075 yabby project discovered a yabby hybrid that produce only male progeny (Lawrence *et al*. 1998, Lawrence and Morrissy 2000). This hybrid therefore has considerable commercial potential for aquaculture.

The discovery of the yabby hybrid has been published (Lawrence *et al*. 1998, Lawrence and Maguire 1998, Lawrence and Morrissy 2000, Lawrence *et al*. 2000). It has also been widely disseminated via both industry and scientific seminars and media reports (Yabby Roadshow 1998, Genetics in the Aquaculture Industry 1998 - “Genetic improvement of marron and yabbies” Walker and Field (1998 - 99), The 1999 Annual Meeting of the World Aquaculture Society, Sydney, Australia - “Hybrid production and growth of monosex yabbies”, ABC Quantum 1999 - Hybridisation of yabbies to produce male progeny).

Need

Prior to industry commercialising this technology, research is required to:

i) Confirm the feasibility of large scale production of all male hybrid progeny
ii) Record the growth rate of the all male hybrid in conditions similar to commercial ponds
iii) Compare the growth of the hybrid with existing WA yabbies
iv) Undertake disease testing to ensure that a *Thelohania* free broodstock population is available for farmers.

Objectives

1. Confirm the feasibility of large scale production of all male hybrid progeny
2. Record the growth rate of hybrids
3. Compare the growth rate of hybrids with WA yabbies
4. Develop a *Thelohania* free broodstock population
5. Develop a commercial options paper for transferring the technology to industry, including an economic costing of various production scenarios for producing male progeny (this is subject to the project showing that the all male progeny have commercial potential).

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HYBRID CHERAX ALBIDUS **x** CHERAX ROTUNDUS **YABBIES GROW FASTER THAN** C. ALBIDUS **YABBIES**

Abstract

A major factor limiting the commercial farming of yabbies (Cherax albidus) *is uncontrolled reproduction in ponds prior to harvest. It has been shown that hybridising two species of freshwater crayfish, female* Cherax rotundus *x male* Cherax albidus *produces only male progeny. This hybrid offers considerable potential for controlling reproduction by yabbies in aquaculture ponds. However, prior to commercialisation of this hybrid it is necessary to compare its growth and reproductive characteristics, under pond conditions, with that of* C. albidus *which is currently farmed in Western Australia,.*

The hybrids proved to be all males and were compared to a mixed sex C. albidus *population as the latter provides a commercial standard. Mean initial sizes ranged from 0.03 - 0.06 g.*

The average growth rate of hybrid C. albidus *x* C. rotundus *yabbies was greater (0.11 ± 0.004 g/day) than that of* C. albidus *yabbies (0.06 ± 0.002 g/day). The growth rates of* C. albidus *x* C. rotundus *hybrids and the* C. albidus *yabbies were similar until they reached sexual maturity, at around 14 g, at which stage the growth rate of* C. albidus *decreased. At the conclusion of the experiment the* C. albidus *x* C. rotundus *hybrids (50.7 ± 2.2 g) were almost twice as large as the* C. albidus *yabbies (27.2 ± 1.7 g).*

The greater growth and increased numbers of yabbies in the larger and therefore more valuable size categories resulted in the value of C. albidus *x* C. rotundus *hybrids being 4.8 times greater than that of the* C. albidus *yabbies at harvest after 424 days.*

The male only C. albidus *x* C. rotundus *hybrids did not produce progeny when mated with* C. albidus *or* C. destructor *females.*

Introduction

Previous research has shown that hybridising two species of freshwater crayfish, female *Cherax rotundus* x male *Cherax albidus,* produces only male progeny (Lawrence *et al*. 1998, Lawrence and Maguire 1998, Lawrence and Morrissy 2000, Lawrence *et al*. 2000). A similar result was achieved with *C. rotundus* x *C. destructor* (Lawrence *et al.* 1998, Austin and Meewan 1999). The farming of these male only hybrids has been proposed as a solution to uncontrolled yabby reproduction in aquaculture (Lawrence *et al*. 2000). This is important to farmers because the inability to prevent uncontrolled reproduction in ponds or farm dams is a major limitation on yabby farming in Australia. This is because yabbies reproduce prior to reaching market size. This uncontrolled reproduction causes density to increase and gives large numbers of stunted individuals as growth of yabbies, and other species of freshwater crayfish, is reduced at high densities (Mills and McCloud 1983, Morrissy 1992, Morrissy *et al.* 1995, Pinto and Rouse 1996, Jones and Ruscoe 1996, Brown *et al.* 1995, McClain 1995a, McClain 1995b, McClain 1995c). Diversion of energy to reproduction also affects growth rate in freshwater crayfish (Jones and Ruscoe 1996, Lawrence and Morrissy 2000, Lawrence *et al*. 2000).

Farming of male only hybrids offers the opportunity of controlling reproduction and improving growth as has been achieved for some other aquaculture species. Hybridisation has been used previously in aquaculture to control precocious maturity, pond breeding, overpopulation and consequently density-induced stunting in tilapia, a fish species that is now farmed worldwide (Lovshin, 1982).With tilapia, a major step in stock improvement to overcome the over-breeding problem was to investigate hybrids of closely related species, resulting in single-sex progeny (Lovshin, 1982).

In addition, it has been shown that male yabbies grow faster than females and both males and females grow faster in monosex culture (Lawrence *et al*. 2000). Therefore it is possible that the hybrid *C. albidus* x *C. rotundus* yabbies will grow faster than the "pure" *C. albidus* yabby strain which is currently farmed throughout Australia.

Furthermore, the aim of hybridising two species is often to produce a faster growing animal as a result of heterosis or hybrid vigour (Lutz 1997). In aquaculture a number of species that have been hybridised have resulted in a faster growing animal as a result of heterosis (Tave *et al.* 1990, Thien and Trong 1995, Bakos and Gorda 1995, Rahman *et al.* 1995). Therefore the yabbies produced by hybridising female *Cherax rotundus* x male *Cherax albidus* may grow faster than the *Cherax albidus* currently farmed in Western Australia. This trend was evident in aquarium trials (Lawrence *et al*. 1998) but in this study, model ponds were used as they reflect commercial conditions more closely and allow intraspecific competition to be assessed without the high rates of cannibalism evident in aquaria.

Materials and methods

Populations of *C. albidus* were collected from the Avondale Research Station in Western Australia (32° 7'S 116° 55'E) and *C. rotundus* from Karuah in New South Wales (32° 40'S 151° 50'E). The yabbies were transported alive to the Aquaculture Reproduction and Genetics Laboratory, University of Western Australia, in Perth, Western Australia.

Sexually mature individuals were randomly stocked into breeding aquaria and fed to satiation. Crosses were established to produce progeny from *C. albidus* x *C. albidus* and hybrids from *C. rotundus* female x *C. rotundus* male yabbies. To permit breeding the building was heated to maintain a mean temperature of 21°C and photoperiod of 14 light : 10 dark.

Growth rates of *C. albidus* and *C. albidus* x *C. rotundus* hybrid yabbies

Eight ponds, each 16 m^2 in water surface area and 1 m deep, were constructed. Each pond had a plastic liner covered by a 7 cm layer of clay. The clay provided turbidity typical of commercial yabby dams. Each pond contained refuges and aeration. The ponds were individually netted to prevent bird predation and movement of stock. The ponds were randomly allocated either hybrid or mixed sex *C. albidus* yabbies. Each treatment was replicated four times. Juveniles from six crosses of each of the two mating combinations, *C. albidus* x *C. rotundus* or *C. albidus* x *C. albidus,* were pooled for each treatment. Each pond was stocked with 64 randomly selected juveniles (4 animals /m^2). The mean stocking weight (± se) of hybrid juveniles was 0.06 ± 0.013 g and the mean weight of *C. albidus* juveniles was 0.03 ± 0.003 g.

Yabbies in the ponds were fed crayfish pellets each day at the rate of 8 g/pond/day. This feed rate equates to 2.5g/m^2/week which is a conservative feed rate that has been shown to be suitable for ponds without water exchange or aeration (Lawrence *et al*. 1998). Ponds were sampled each month by placing two baited traps into each pond for 2 hours. Yabbies collected from traps were individually weighed and sexed. The economic value of production was calculated at the conclusion of the experiment by dividing yabbies into standard industry size grades and applying standard size grade values to yabbies (Lawrence 1998). Prior to analysis of percent survival by t-test, data was arc sin square root transformed to simulate normality. An F-test was used to ensure that variances were uniform.

The experiment was stocked on the 16th February 1999 and concluded after 424 days on the 19th April 2000 when all yabbies were collected by first trapping then draining ponds.

Fertility of hybrids

Hybrids produced from mating *C. rotundus* female x *C. rotundus* male yabbies and *C. albidus* animals were reared to sexual maturity in aquaria at the Aquaculture Reproduction and Genetics Laboratory. The *C. albidus* x *C. rotundus* hybrid (males) and *C. albidus* (females) were stocked into ten aquaria breeding tanks (1M:1F). The *C. albidus* x *C. rotundus* hybrid (males) and *C. destructor* (females) were stocked into five aquaria as breeding tanks (1M:1F). Controls of *C. albidus* x *C. albidus* and *C. rotundus* x *C. rotundus* were also stocked in aquaria. Yabbies were fed daily with crayfish pellets.

This experiment commenced on the 3rd March 1999 and concluded after 409 days on the 14th April 2000.

Results

Growth rate of *C. albidus* vs *C. albidus* x *C. rotundus* hybrids

The average growth rate of hybrid *C. albidus* x *C. rotundus* yabbies was greater (0.11 ± 0.004 g/day) than that of *C. albidus* yabbies (0.06 ± 0.002 g/day) (P = 0.002). The growth rates of *C. albidus* x *C. rotundus* hybrids and the *C. albidus* yabbies were similar until the latter reached sexual maturity, at around 14 g, at which stage the growth of *C. albidus* decreased (Figure 2.1).

There was a significant difference in the final mean weight of hybrid *C. albidus* x *C. rotundus* yabbies (50.7 ± 2.2 g) and *C. albidus* yabbies (27.2 ± 1.7 g). (P = 0.002). At the conclusion of the experiment the hybrid *C. albidus* x *C. rotundus* were almost twice as large as the *C. albidus* yabbies (Figure 2.1).

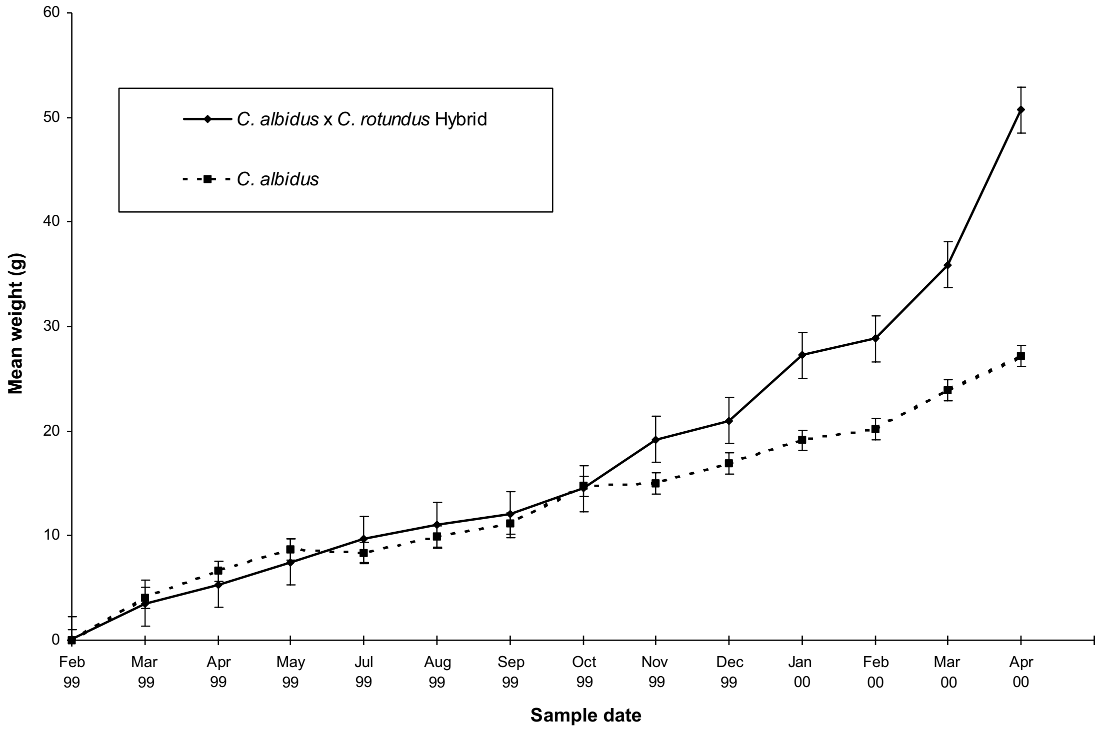

Figure 2.1 Growth of *C. albidus* vs *C. albidus* x *C. rotundus* hybrids over 424 days months (juveniles excluded). (Bars indicate se n=4)

Size distribution of *C. albidus* x *C. rotundus* hybrids and the *C. albidus* yabbies

There were more animals within the larger size classes in the *C. albidus* x *C. rotundus* hybrid ponds than in the *C. albidus* ponds (Figure 2.2). The greater growth and increased numbers of yabbies in the larger and therefore more valuable size classes resulted in the value of *C. albidus* x *C. rotundus* hybrids ($A0.48/$m^2$) being over 4.8 times greater than that of the *C. albidus* yabbies ($A0.10/$m^2$). This equates to an economic value of $A4823 /ha for hybrids (final biomass 675 t/ha) compared to $A1038 /ha for *C. albidus* yabbies (final biomass 188 t/ha).

Reproduction occurred in the *C. albidus* ponds resulting in a bimodal distribution representing juveniles and adults (Figure 2.2). The mean weight of juveniles in the *C. albidus* ponds was 5.4 ± 0.2 g. No reproduction occurred in the *C. albidus* x *C. rotundus* hybrid ponds (Figure 2.2).

Dividing the size distribution of *C. albidus* yabbies according to sex shows that in the larger size categories there were more *C. albidus* x *C. rotundus* hybrid males than *C. albidus* males (Figure 2.3).

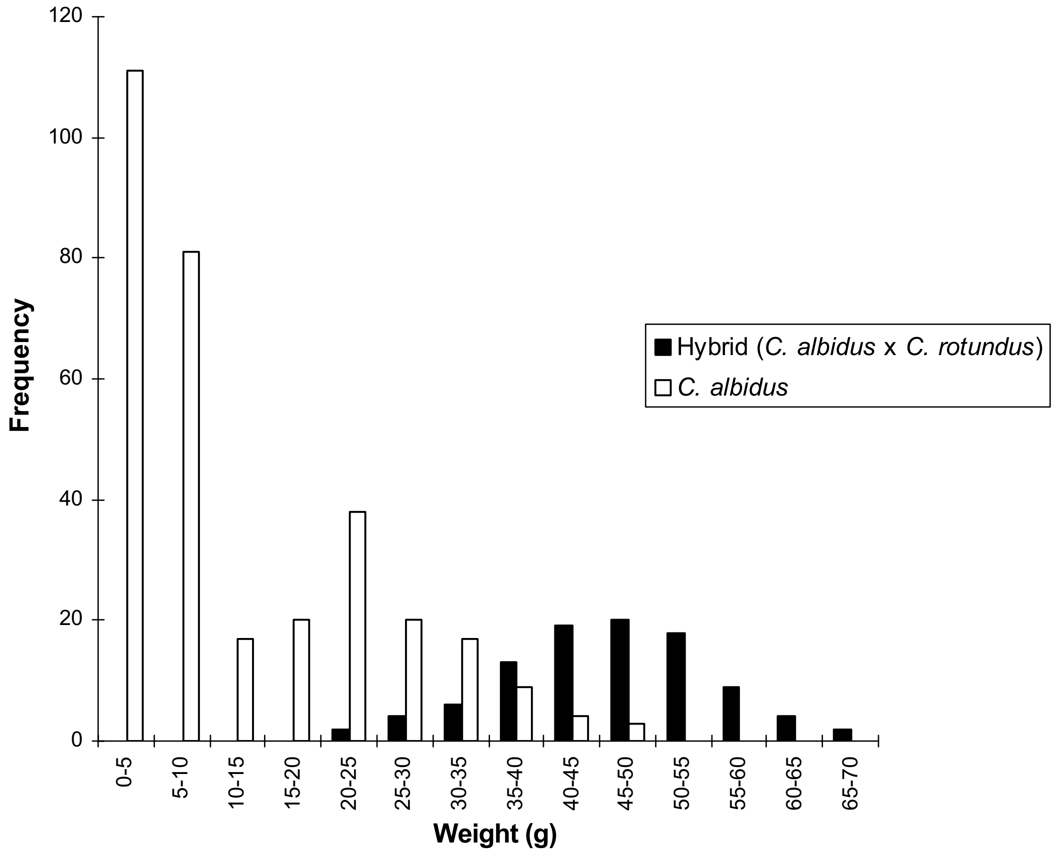

Figure 2.2 Size distribution of *C. albidus* x *C. rotundus* hybrids and the *C. albidus* yabbies (juveniles arising from reproduction in *C. albidus* ponds included).

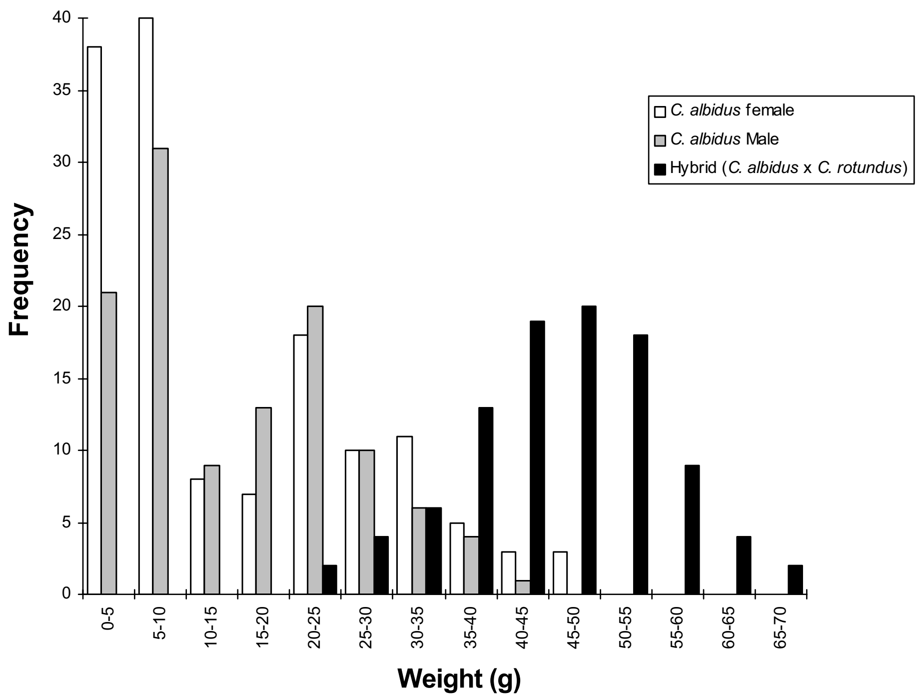

Figure 2.3 Size distribution of *C. albidus* x *C. rotundus* hybrids and the *C. albidus* yabbies according to sex (juveniles arising from reproduction in *C. albidus* ponds included).

Survival

There was no significant difference in the average (± se) survival of *C. albidus* x *C. rotundus* hybrids (38 ± 4.4 %) and *C. albidus* (43 ± 5.3 %) (P=0.467).

Fertility of hybrids

In 17 matings *C. albidus* x *C. rotundus* hybrids either did not mate with *C. albidus* females or the eggs were aborted (Table 2.1). In 6 matings *C. albidus* x *C. rotundus* hybrids either did not mate with *C. destructor* females or mated with *C. destructor* females but eggs were aborted (Table 2.1). Controls of *C. albidus* x *C. albidus* and *C. rotundus* x *C. rotundus* mated and produced off spring during the same period.

Table 2.1 Reproductive success between *C. albidus* x *C. rotundus* hybrids and either *C. albidus* or *C. destructor*.

Mating combination	Not berried (n)	Berried then aborted (n)	Berried then produced offspring (n)
C. albidus x *C. albidus* x *C. rotundus* hybrids	10	7	0
C. destructor x *C. albidus* x *C. rotundus* hybrids	4	2	0
C. albidus x *C. albidus*	0	3	9
C. rotundus x *C. rotundus*	0	4	8

Discussion

This experiment has shown that the male only hybrids produced by mating *C. albidus* x *C. rotundus* grow almost twice as fast as the *C. albidus* yabbies currently farmed in Western Australia. This results in a greater proportion of the final harvest of hybrid yabbies being in the larger and therefore more valuable size grades. These results indicate considerable potential for farming hybrid yabbies as the faster growing and consequently larger hybrids produced a crop which was 4.8 times more valuable than the slower growing and therefore smaller *C. albidus* yabbies which are currently farmed in Western Australia. This equates to an economic value of $A4823 /ha for hybrids (final biomass 675 t/ha) compared to $A1038 /ha for *C. albidus* yabbies (final biomass 188 t/ha). It is likely that the difference would have been greater if the trial could have been extended, under favourable water temperatures, as the growth advantage in hybrid ponds probably arose from both lack of diversion of energy to reproduction and lack of intraspecific competition from multiple cohorts. The latter effect may have been more pronounced as juvenile biomass increased in *C. albidus* ponds. This is related to the fixed feed ration used regardless of biomass. This feeding strategy reflects that currently used in commercial yabby ponds and dams.

In Western Australia, hand sexing yabbies to stock monosex ponds is a popular method of obtaining improved growth rates. Male *C. albidus* yabbies stocked into monosex ponds grow 53% faster than mixed sex populations (Lawrence *et al*. 2000). The results obtained in this experiment show that the male-only yabbies produced by hybridising *C. albidus* x *C. rotundus* grow 83% faster than mixed sex *C. albidus* yabbies.

While uncontrolled backcrossing of hybrids has limited their commercial application in other aquaculture species such as tilapia, this does not appear to be a problem with *C. albidus* x *C. rotundus* hybrid yabbies. In this experiment the male hybrid yabbies either did not mate with *C. albidus* or *C. destructor* females, or where they did mate the embryos were not viable and all eggs were aborted. This will greatly simplify the commercial application of hybrid *C. albidus* x *C. rotundus* yabbies on existing farms.

The growth rates recorded in model research ponds for *C. albidus* in this experiment are within the range reported at similar densities from earthen ponds (Lawrence, unpublished data). Therefore the model research ponds used in this experiment are suitable for freshwater crayfish experiments and results can be extrapolated to commercial farming conditions.

Other attributes beyond growth that need to be considered are taste, survival during transport, disease tolerance and burrowing behaviour in earthen ponds.

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Chapter 3

N. Cole

INCUBATION, EGG FERTILISATION AND EMBRYO DEVELOPMENT IN HYBRID YABBIES

Abstract

Artificial egg incubation and histology were proposed as two techniques which may provide data to explain the occurrence of male only progeny from hybridising female Cherax rotundus *and male* Cherax albidus *yabbies. Artificial incubation of eggs provided some preliminary results, however this trial was not successful in determining the occurrence of male only progeny. No data were collected from histological slides, as time constraints prevented the submission of eggs for cytological examination. The results of this experiment are therefore inconclusive. Further study into both techniques is recommended, as this experiment proved it is possible to artificially incubate* Cherax *eggs. It is recommended that this experiment be repeated as a university student project with further trials involving the variables which may have affected this experiment, namely temperature, egg stocking rates, malachite green concentrations, flow rates and broodstock diet.*

Introduction

Progeny resulting from the female *C. rotundus* x male *C. albidus* yabby cross are all males, however the mechanism by which this occurs is not known (Lawrence *et al.* 1998). It is possible that all embryos are male or, alternatively, female embryos are not genetically viable and therefore do not survive. Close observation of eggs from this cross would enable the monitoring of survival and hatch rates to provide an indication of the number of male hybrids produced and the number of eggs that are not viable. These hypotheses could be tested by counting the number of juveniles that hatch from a known number of hybrid cross eggs. If most of the eggs hatch and produce male juveniles, it may be assumed that all eggs produced by this cross are male. If only half of the eggs hatch, it is likely that the female embryos do not survive.

It is, however, difficult to accurately count eggs on pleopods when they are attached to the underside of the female's abdomen. This is because the female retreats during berry, and manual handling during egg counting may cause egg damage, or the female may abort her eggs (Matthews and Reynolds 1995, Perez *et al*. 1999).

Careful removal and placement of known numbers of *C. rotundus* x *C. albidus* eggs into artificial incubation chambers (cells) would allow for survival to be monitored closely. Artificial incubation would therefore permit the number of viable hybrid males, resulting from a known berry of eggs, to be determined.

An alternative option is to compare the numbers of fertilised and unfertilised (or poorly developed) hybrid eggs from histological sections. This approach would allow for validation of the incubation trial by providing comparative data and estimation of egg mortalities during the incubation process.

Materials and methods

Preliminary development for Experiment 1 (building and trialing incubators) was conducted from December 1998 to October 1999 before Experiment 1 (October 1999 to March 2000).

These trials were both conducted at the Aquaculture Reproduction and Genetics Laboratory, University of Western Australia, in Perth, Western Australia.

Experiment 2 was to be conducted by an external cytology/histology service.

Experiment 1 – Egg Incubation

Design of Preliminary Experiment

C. albidus eggs were placed into the artificial incubation unit as described below. The viable hatch rate of three replicate berries (i.e. eggs from three females) was recorded and the suitability of the unit for experimental incubation evaluated.

Design of the Main Experiment

Once the methodology was developed from the preliminary experiment, a minimum of three *C. rotundus* x *C. albidus* replicate berries were artificially incubated using the same methodology.

The artificial incubation unit was a recirculating system, placed horizontally on the floor of a 500 mm wide x 800 mm long x 310 mm high aquarium tank (120 L). The unit consisted of a rectangle of 15 mm PVC piping (750 mm x 150 mm) connected via 15 mm PVC pipe with a flow rate tap to a power-head filter (1000 L/hour). The PVC rectangular structure had 20 (7 mm) holes drilled into its upper side (10 on each long side), with 20 100 ml, 40 mm diameter syringes (barrels only) pushed into each hole by the tips (top of each tip is 6 mm in diameter), sitting the barrels upright. The base of each barrel (cell) was 12.5 cm^2, and contained a circle of 1 mm^2 mesh (40 mm in diameter), held in place by a flat rubber 'O' ring. The top of each cell was covered by a circular plastic grate, covered with 1 mm^2 mesh to allow water flow through the top of the cells without the eggs escaping.

The aquarium tank was filled to approximately 20% of its capacity (about 24 L) with water, or until the syringe barrels are half submerged. The power-head filter then pumped water into the PVC pipe rectangle, where it flowed up and out of the top of the syringe barrels. The water joined the reservoir in the main tank until it is pumped through the unit again. The flow rate was reduced via the tap to approximately one quarter of the filter's capacity (ie. to 250 L/hour).

Four identical incubation units were used (a total of 80 incubation cells), each housed in a 120 L aquarium. Five additional tanks held the parental (*C. rotundus* x *C. albidus*) crosses at a ratio 3F:4M, and females were checked daily for eggs. Fifteen *C. rotundus* females and 20 *C. albidus* males were required to stock the parental tanks. Any females carrying attached eggs were taken from the tank and the eggs removed.

The eggs were stripped immediately after they had attached to the pleopods by using blunt tweezers, stripping from the base to end of the pleopod, as described by Mason (1977) and Carral *et al*. (1992), approximately four days after the female showed behavioural signs of berry (the tail is tightly curled up). It was assumed the eggs had completed the external fertilisation process by this stage, and the female had not yet aborted any inviable eggs. Eggs were placed into the unit at random, and only eggs which were not damaged by the stripping process were stocked (Rhodes 1981).

In the first two or three days of berry, the eggs are surrounded by glair (mucus) and are not yet attached to the pleopods. It is believed that the external egg fertilisation is completed while the tail is tightly curled. Previous incubation experiments with freshwater crayfish eggs have found the survival rate of eggs is increased with the time eggs were left on the pleopods (Mason 1977, Carral *et al*. 1992).

The eggs were placed into a 10 ppm solution of malachite green for 10 minutes to reduce the occurrence of *Saprolegnia* spp., a fungal infection (Henrion 1996, Rhodes 1981). The reservoir water was also dosed with 100 ml of a 10 ppm malachite green solution once a week. The eggs were then stocked into the cells of the incubation units at a stocking rate of 2.2/cm^2 (6 eggs per cell), as specified in previous experiments (Carral *et al*. 1992). Each unit had the capacity to hold 120 eggs. Dead eggs were removed daily and recorded.

A timer set at 14L:10D controlled light duration. The parental crosses were fed a mix of earthworms, pellets and frozen daphnia daily. The laboratory was heated to approximately 26°C via two wall-mounted air temperature control units. The water temperature of the parental cross tanks stayed relatively constant at 22-25°C. The water in the incubation units was heated to a constant 24°C by a submersible 100 watt heater. The incubation time is known to decrease with increased temperature, however, the chance of fungal infection may also increase.

Due to evaporation, the parental cross tanks were topped up every two to three days. A 20% (24 L) water change was carried out every 4 weeks. The incubation tanks were also topped up regularly to the required level and received a 20% (approximately 5 L) water change and simultaneous siphoning every 4 weeks.

Experiment 2 – Histology

Three replicates of a berry of eggs (three x *C. albidus* controls and three x *C. rotundus* x *C. albidus*) were to be removed from the pleopods as in Experiment 1

(ie. approximately 4 days after copulation). The eggs were to be immediately transferred to 70% ethanol for preservation for approximately 48 hours before being placed in Bouins, an egg fixing agent, and sent to a histology lab for sectioning and slide presentation. From the slides, the numbers of fertilised eggs (viable) verses non-fertilised (or inviable) eggs could be recorded.

Results

Experiment 1 – Egg Incubation

Unfortunately, no viable hybrid juveniles were recorded from the two hybrid egg incubation trials. The treatment of a 10 ppm solution of malachite green did not prevent the occurrence of *Saprolegnia* spp. infestation. The hybrid eggs were observed to swell and go a pale beige/orange colour (the typical colour of dead eggs), before becoming infected. The eggs were removed from the unit once they were infected.

C. albidus eggs initially suffered from fungal infections, however further trials produced juveniles that hatched from non-infected eggs and were counted. The results from experiment 1 are displayed in Table 3.1.

Table 3.1 Artificial incubation hatch rates of eggs.

Cross	Stocking Date	Conclusion Date	Duration (days)	No. eggs hatched	No. eggs stocked	No. eggs remaining	% Survival
C. albidus x *C. albidus*	5 Oct 1999	21 Oct 1999	16	67	100	22 (11 'missing')	67 %
C. albidus x *C. albidus*	23 Feb 2000	20 Mar 2000	26	94	120	26	78 %
C. rotundus x *C. albidus*	24 Nov 1999	11 Jan 2000	48	0	226	226	0 %
C. rotundus x *C. albidus*	11 Jan 2000	17 Jan 2000	6	0	73	73	0 %

Experiment 2 – Histology

Eggs were not submitted for histology as 3 replicates of eggs from the control and test crosses were not collected due to time constraints.

Discussion

Results from this experiment are inconclusive. It was determined that the artificial incubation of *Cherax* eggs is possible, however there are many variables which could have affected the collection of useful data. Further trials to determine the optimum environment for conducting the incubation experiment are recommended.

The hybrid parental cross (particularly *C. rotundus*) may require higher temperatures to breed more often. The average water temperature was 21°C over the year, which may not have been warm enough for the females to breed as often as this experiment required. Further trials involving variation in temperature, egg stocking rates, brood stock diet and malachite green concentrations are recommended as a series of experiments to reduce the variation that occurred in the original experiment. It is considered that the original designs for these experiments could still be useful in determining the mechanism for the production of all male progeny.

From an industry perspective the results of this experiment are academic, as from multiple spawnings, no female has ever been recorded resulting from the hybrid cross

(*C. rotundus* x *C. albidus*). It is therefore recommended that future research into the mechanism contributing to production of all male progeny be investigated as a university student project. This will permit Department of Fisheries WA researchers and industry to focus upon commercialisation of the hybrid.

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F. Johnston and C. Lawrence

POST-HARVEST EVALUATION OF ALL MALE HYBRID YABBIES (CHERAX ALBIDUS x C. ROTUNDUS)

Abstract

Two genetic strains of yabbies were harvested from 8 model ponds, and were used in simulated live transport trials and sensory evaluation. In the simulated live transport trials, there was negligible difference in the survival rate of two genetic strains after six days, and the average survival rate was 90%. After eleven days the survival of the hybrid yabbies was significantly lower than for C. albidus. *However, in most cases, six days is sufficient time for the animals to reach their overseas markets.*

The sensory evaluation could not detect a significant difference between the hybrid and C. albidus *yabbies. Mean values, for each of eight sensory attributes evaluated, were usually similar for the hybrids and* C. albidus.

Introduction

Yabbies are an important freshwater aquaculture species in Western Australia (Cole 2000). An aquaculture market for these animals has developed and significant numbers are being sold, especially on export markets. These animals are particularly suitable for export, because they require very little processing and may be shipped live to maintain product quality. The post-harvesting process, which can be stressful to yabbies, begins once they are taken from their aquatic environment and includes handling and processing, factors that may alter the animal's sensory characteristics such as flavour and appearance (Jones 1990). Commercially, yabbies are harvested with baited traps (see Lawrence 1998) from private water storage dams, allowed to clear sediment from the gill cavity by being held in dam water and then transported out of water, under cover, to a processing facility. Yabbies are held out of water in polystyrene boxes, typically for up to 6 days. Gut contents are purged prior to sale as a live product.

The stocking of monosex yabbies, and hence density control, has resulted in higher yields, larger animals, and up to a 70% increase in income (Lawrence *et al*. 2000). In an attempt to produce a more profitable crop, an all male hybrid was developed from a cross between *C. albidus* and *C. rotundus* (Lawrence *et al*. 1998). The growth rate of this strain was examined over a period of 14 months in model ponds, and compared very favourably with mixed sex *C. albidus* (Chapter 2).

The purpose of the following experiments was to determine if the hybrid males differ in sensory appeal compared with mixed sex *C. albidus*. In addition, the hardiness of these hybrids was examined in terms of simulated live transport conditions to establish if this strain is likely to arrive to an overseas market in saleable condition.

Materials and methods

Harvesting and purging of yabbies

The two genetic strains of yabbies, used in both the sensory evaluation and the simulated live transport trials, were harvested from 8 model ponds (Chapter 2). The yabbies were removed using two methods, trapping with baited traps, and collection after draining. Trapping was used to decrease the number of animals burrowing into the mud, and therefore, decrease bacterial contamination of their gills that is associated with this activity. Draining of all ponds took 3 days, and during this period trapping was continued in those ponds not being drained.

Once caught, animals were weighed and held unfed in fibreglass purging tanks (160 cm x 60 cm x 38 cm) for 18 - 72 hours before they were removed for the following experiments.

Simulated live transport trials

Yabbies used in the simulated live transport trials had been in the purging tanks for 1 to 3 days. They were chilled using ice in the purging tanks to 15°C, removed and weighed prior to being packed into 10 kg capacity polystyrene boxes (545 mm x 400 mm x 145 mm). Two boxes of each genetic strain were used, with 20 animals packed into each box. The largest *C. albidus* animals were selected for this experiment (mean weight 29 ± 2.17 g), as they were smaller on average than the hybrid animals (mean weight 44 ± 0.35 g). Consequently in each polystyrene box the mean biomass of WA (583 ± 43 g) and hybrid yabbies (879 ± 7 g) was different (P = 0.02). The insulated container was separated in half and animals placed in only one half of the box to increase yabby density and mimic more closely the conditions experienced in industry. The animals were packed under a moist foam liner to maintain humidity and prevent damage, and an ice pack was used to lower the temperatures further. The ice pack was wrapped in newspaper to prevent direct contact with the animals, and then sealed in a plastic bag. The resulting volume occupied by the animals was approximately 200 mm x 220 mm x 80 mm. A Tinytag Plus data logger was introduced at this time to monitor the temperature within each box. The boxes were sealed and a 3 mm hole was punched in the box to allow some air flow. These conditions therefore mimicked those of yabbies packed for export. The four resulting boxes were then transported to The University of Western Australia and stored in a coolroom at 11°C. The boxes were left sealed with tape for 6 days and then checked. Dead animals were removed and weighed. The boxes were resealed and checked on days 7, 8, and 11, resealing the box after each mortality check.

Prior to analysis of percent survival by t-test, data was arc sin square root transformed to simulate normality. An F-test was used to ensure that variances were uniform.

Sensory evaluation

Fifty yabbies of each genetic strain, *C. albidus* and the hybrid, were used in the sensory trials. These animals were removed from the experimental ponds and placed in purging tanks for at least 18 hours prior to preparation. The live animals were removed from the tanks and transported in insulated boxes to the Western Australia Marine Research Laboratories where they were prepared. The heads were removed, and the tails were rinsed prior to being steamed for 5 minutes. The cooked tails were then delivered to the sensory laboratory the day before the sensory evaluation. These samples were peeled and then refrigerated before being presented to the respondents.

To assess differences in quality between the two genetic strains of yabbies, a sensory evaluation test was carried out by The Food Centre of Western Australia (Inc.). A triangle test-sampling pattern was used with a questionnaire consisting of continuous rating scales. Thirty respondents were required to complete the questionnaire, and the results were analysed by the statistical package SPSS. The questionnaire assessed appearance, texture, flavour, toughness, dryness, feel, and overall acceptability.

Results

Simulated live transport trials

After six days there was negligible difference in the survival rate of the two groups (90% ± 5, n = 2 for hybrids and 100 ± 0, n = 2 for *C. albidus*) (P = 0.07).

After eleven days the survival of the hybrid yabbies (mean ± se = 63% ± 7.5, n = 2) was significantly lower than the *C. albidus* animals (mean ± se = 90% ± 5, n = 2) (P = 0.003). Analysis of the relative size of animals, within each group, that survived or perished revealed that size was not a factor affecting their survivability (P = 0.97) during the transport trials.

Sensory evaluation

The sensory evaluation could not detect a significant difference between the hybrid and *C. albidus* yabbies (P > 0.05) (Table 4.1). The "null hypothesis" states that there is no significant difference between the samples. When a significant difference is found the null hypothesis is automatically rejected and the alternative hypothesis is accepted. The level of significance is set at 0.05 (i.e. in the table, "*P*" must be greater than 0.05, for the null hypothesis to be accepted). For this test, the nearer "*P*" is to 1.0, the more likely it is that all samples fall in the same "statistical" population. Detailed results from the sensory evaluation are presented in Appendix 1.

Table 4.1 Sensory evaluation of *C. albidus* x *C. rotundus* all-male hybrids and *C. albidus* yabbies (n=45).

Sensory attribute	Rating scale (0-40)	Hybrid *(mean ±SD)*	*C. albidus* *(mean ±SD)*	(P)
General appearance	Very unpleasant(0) to very pleasant(40)	31.4 ± 9.8	33.0 ± 8.1	0.86
Flavour pleasantness	Very unpleasant(0) to very pleasant(40)	30.2 ± 9.0	31.4 ± 9.3	0.90
Eating texture	Very unpleasant(0) to very pleasant(40)	31.7 ± 9.0	32.5 ± 8.9	0.93
Toughness	Very tough(0) to very tender(40)	32.1 ± 10.1	32.3 ± 9.7	0.98
Dryness	Very dry(0) to very moist(40)	35.0 ± 7.7	34.6 ± 9.0	0.96
Mouthfeel	Very unpleasant(0) to very pleasant(40)	33.1 ± 9.6	33.9 ± 8.2	0.93
Aftertaste	Very unpleasant(0) to very pleasant(40)	31.3 ± 9.6	34.2 ± 7.9	0.75
Overall acceptability	Totally unacceptable(0) to totally acceptable(40)	31.6 ± 10.1	33.8 ± 8.4	0.82

Discussion

Simulated live transport trials

The hybrids had an average survival of 90% after six days, which is far greater than the maximum mean survival rate observed by Roe (1994) during her transport experiments with commercial animals. She found the maximum mean survival of *C. albidus* to be 70% after a period of only 90 hours. Therefore, it may be inferred that the losses incurred by the hybrids are at acceptable levels for industry. In most cases, six days is sufficient time for the animals to reach their overseas markets. However, it must be considered that these animals were not packed at commercial densities.

The differences in survival between the *C. albidus* strain and the hybrids could be due to a number of reasons. Results from data loggers in the transport containers indicate that the mean temperature values were similar (range 9.8 - 12.8°C), as were the minimum (range 7.3 - 8°C) and maximum temperatures (22.7 - 23°C) indicating that all animals experienced similar environments once packaged. The sex of the *C. albidus* animals used in this trial was not considered. There is a possibility that females have better survival, however, due to the limited number of animals available for this pilot study, the largest individuals, regardless of sex, were included in this trial.

Due to the larger size, hybrids experienced a higher biomass density that may have resulted in the higher mortality rate. This was considered prior to packaging, however, an increase in the number of animals in the *C. albidus* treatment may have increased mortality rates of these animals due to risk of competition and aggressive interaction. In addition, although attempts were made to package these yabbies at industry densities, which is around 8 kg in a 10 kg box for overseas transport, neither treatment approached this level.

Due to the potential influence of these uncontrolled factors on these preliminary results, and the size of the sample used, it is suggested further trials be carried out. This should include only male *C. albidus* animals that are of comparable size to the hybrids. This would increase the power of the results, and give a clearer understanding of the hardiness of the hybrid animals. Given that the hybrids survived well for 6 days storage, this trial does not demonstrate that they are unsuited to normal commercial storage and transport regimes. However, the difference observed between the two groups after a very severe test (11 days), suggests that further research is warranted when sufficient comparable stocks are available to compare similar size yabbies at normal commercial "loading" i.e. kg of yabbies per 10 kg box. It is difficult to avoid all potentially confounding factors as *C. albidus*, of the same size as the hybrids, will either be older or intensively size selected or grown on a different regime e.g. much lower stocking density.

Sensory evaluation

The results of the sensory evaluation are based on the information supplied in the report Tam (2000) (Appendix 1). The results indicated that there was no significant difference in any of the attributes tested. Although, due to the low numbers of animals available for this study, these results should be considered preliminary findings, it appears that the hybrid strain of yabbies is a potential substitute for the present commercially available *C. albidus* strain. It was suggested that sex and animal size may have been a contribution to these findings, however, in other similar studies there appeared to be no difference in flesh composition of different sexes of animals, or of varying animal sizes within a given grade (Jones 1990).

The results from this preliminary investigation into the quality of the all male hybrid (*C. albidus* x *C. rotundus*), suggest that it is a suitable substitute for the presently available parent strain *C. albidus* in terms of their sensory appeal. Furthermore, although the hybrids exhibited a lower rate of survival in simulated transport trials, these levels may be comparable to those found in commercially available yabbies. Additional studies should be conducted on this hybrid's ability to tolerate the stresses of transportation, where the variables previously discussed are managed as discussed above.

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COMMERCIALISATION OPTIONS, ECONOMIC COSTINGS AND TRANSLOCATION RISK ASSESSMENT OF HYBRID PRODUCTION SCENARIOS

Introduction

Yabbies are farmed widely throughout central and eastern Australia. Since being introduced into Western Australia from Victoria in 1932 (Morrissy and Cassells 1992), the yabby, *Cherax albidus*, has spread and formed the basis of a growing aquaculture industry over an area of 750,000 km^2 ranging from Northampton to Esperance.

Yabby production for 1997/1998 was 231 tonnes, up from 107 tonnes in 1996/1997. More recent production estimates (July-December 1999) indicate rapid expansion of production although this is influenced annually by rainfall (Cole 2000). The majority of production comes from around 4,000 extensively managed (and most pre-existing) farm dams (Lawrence *et al*. 1998). Yabbies have been tried in purpose built ponds, however the capital investment required to set up dedicated ponds does not make this type of yabby aquaculture commercially attractive to most farmers at least in WA.

Lawrence *et al*. (1998) suggest that with increased efficiency and greater exploitation of available farm dams, the yabby industry has the potential to reach 5,000 tonnes per annum in Western Australia alone, worth $50 million in exports. While such production may be possible, there are problems that must be overcome before this potential can be realised.

One of these problems is the production of large numbers of animals below market size (<30 g). As with many freshwater crayfish, larger animals receive a higher market price per kilogram (Lawrence *et al*. 1998), so it would be more profitable for farmers to maximise the number of larger sized animals being produced.

While many believed that undersized or stunted crayfish were the result of underfeeding, McClain and Romaire (1995) proposed that population density is the single most limiting factor affecting crayfish growth. Lawrence *et al*. (1998) have also reported that stocking density was of a greater significance to yabby production than feeding lupins. As yabbies are produced in farm dams that cannot be drained, there is little control over reproduction and consequently, population densities in the dams. This results in reduced growth rates, survival and thus value of production even though the biomass of yabbies may be greater in a dam where reproduction is occurring.

One method of preventing reproduction in farm dams and thus controlling density and increasing production would be to stock dams with single sex individuals. Lawrence *et al.* (1998) and Lawrence *et al.* (2000) reported that both males and females in mono-sex cultures grew faster than males or females in mixed sex cultures. In terms of the production value, stocking with male only yabbies resulted in an increase of 70% of the gross value of animals harvested compared to mixed-sex cultures (Lawrence *et al*. 1998, Lawrence *et al.* 2000). While industry has adopted the idea of mono-sex culture, hand-sexing of yabbies is very labour intensive and not 100 % accurate when stocking small juveniles.

One area of research recently undertaken by Department of Fisheries WA was to investigate the hybridisation of a number of yabby species collected from geographically isolated regions throughout Australia. (Please note: These animals were introduced into Western Australia under a strict quarantine protocol and on the condition that the hybridisation trials took place at an approved quarantine facility and there has been no unauthorised release of any animals from this quarantine facility other than for transfer of *C. rotundus* to a new quarantine facility at Avondale).

This hybridisation study identified a mating combination that results in only male offspring. Juveniles produced from repeated spawnings of female *Cherax rotundus* crossed with male *Cherax albidus* has resulted in only male progeny (Lawrence and Morrissy 2000). The reciprocal cross for this combination (*C. albidus* female x *C. rotundus* male) produced a 1:1 sex ratio (Lawrence *et al*. 1998, Lawrence and Morrissy 2000).

In aquaria, the hybrid grew 30 - 172% faster than the Western Australian yabbies (Lawrence *et al*. 1998). Due to strict quarantine protocols, the performance of the all male hybrid in ponds has only recently been evaluated in research pools with favourable results (see Chapter 2). Should the hybrid demonstrate similar growth in farm dams, industry may have the answer to solving over population and consequent stunting problems that reduce production in farm dams, while removing the need for labour intensive hand sexing to have mono-sex cultures.

By holding populations of *C. rotundus* and *C. albidus* in separate ponds, farmers operating a hatchery to produce hybrids would be able to produce an ongoing supply of broodstock from which female *C. rotundus* and male *C. albidus* can be taken and crossed to produce an all male offspring for ongrowing to market size.

In Australia *Cherax albidus* yabbies are available in SA, WA, NSW and Victoria. However, *Cherax rotundus* is only found in a small region of NSW (see Figure 5.1). In NSW, stocks of *C. rotundus* and *C. albidus* are already available to farmers. Therefore translocation approval would be required for *C. rotundus* to be introduced onto commercial properties in SA, Victoria and WA.

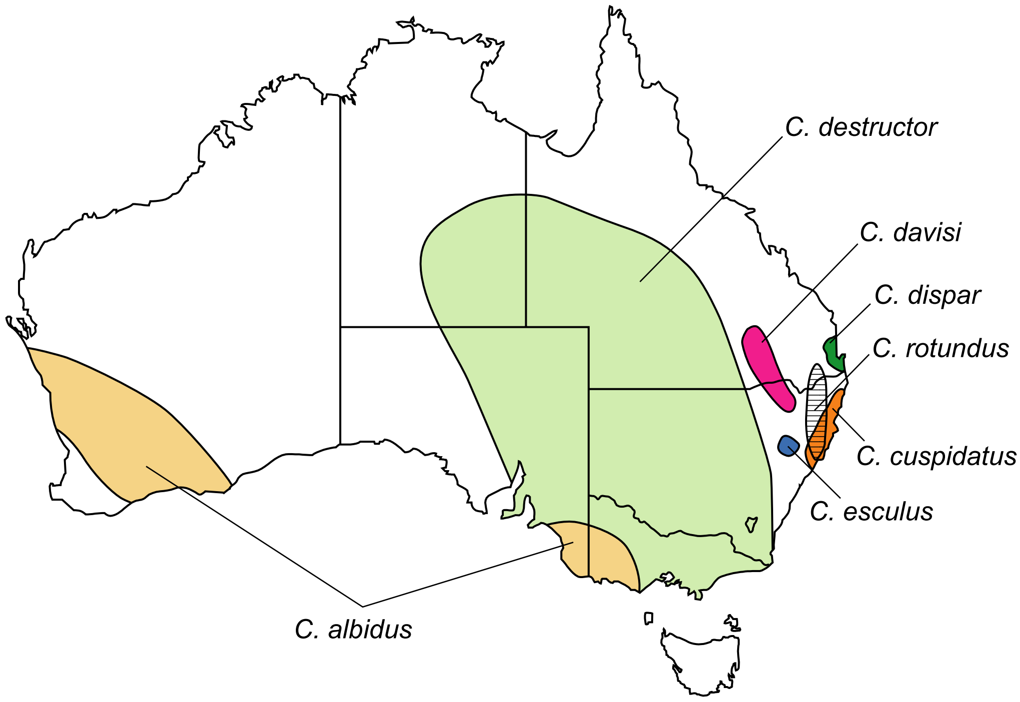

Figure 5.1 Distribution of yabbies according to Reik (1969).

The following outlines the proposed commercialisation strategy of *Cherax rotundus* and the all male hybrid animals.

Commercialisation strategy

1) Obtaining broodstock

Stocks of both *C. rotundus* and *C. albidus* already exist on yabby farms in NSW. In SA and Victoria *C. albidus* is wide spread, however *C. rotundus* is not thought to exist. Although there has been a report of *C. rotundus* in Buxton and Taggerty in Victoria, (in 1963 and 1983 respectively) (Sokol 1988), it is thought to be a misidentification of the Barmah swamp yabby which has similar morphological features (Lawrence *et al.* 1998). Farmers in SA and Victoria would of course have to obtain permission from relevant state agencies prior to introducing *C. rotundus*. In Western Australia farmers should abide by existing Department of Fisheries WA policies that discourage the movement of animals with *Thelohania* within this state. In WA there are two major concerns, namely introduction or spreading of diseases and the risk of *C. rotundus* or viable hybrids entering natural waterways. *C. albidus*, which is not native to WA, is now present within some recreational fisheries for marron, *C. tenuimanus.*

To facilitate the commercialisation of the hybrids, *Cherax rotundus* have been collected in NSW and sent to Western Australia to the quarantine facility, under an approved research permit.

There are two strategies proposed, depending on whether the introduced stock is *Thelohania* negative or positive.

i) *Thelohania Negative*

If, after disease testing, the animals are shown to be free of *Thelohania*, Department of Fisheries WA will transfer *C. rotundus* individuals to a quarantined section of the Avondale research station, where the agency has twenty-five 100 m^2 ponds. (This only went ahead with approval from the Senior Fish Pathologist and after appropriate quarantine protocols were in place. For example, to prevent escape, the ponds required a low solid fence around the edge and were already covered with bird netting to prevent predation).

ii) *Thelohania Positive*

If the animals were *Thelohania* positive, they would be destroyed.

Once at the Avondale research station, reproduction and behaviour of *C. rotundus* (e.g. burrowing behaviour) is being studied which would provide valuable information on this strain. Following this, it is anticipated that commercial hatcheries will apply for permission to culture the progeny of the translocated animals. It is anticipated that prior to the moving of animals from Avondale, disease testing would need to be carried out.

2) Who could apply to use the translocated animals?

Any farmer with a commercial aquaculture license to culture yabbies would be able to apply to culture *C. albidus x C. rotundus* hybrids. Each application for a hatchery to hold *C. rotundus* will be assessed on a case by case basis by relevant State authorities but the location of a commercial farm may have an impact on the success or otherwise of an application. (In WA, approval to farm the hybrid yabbies is likely to be provided in existing yabby farming areas provided they are not shown to cross with other yabbies - see Chapter 2, or have extreme burrowing tendencies. However, farming of *C. rotundus* broodstock is only likely to be approved in a restricted number of licensed locations to permit appropriate compliance checks, on required protocols for preventing escape of *C. rotundus*. Again disease status and burrowing tendencies of *C. rotundus* would have to be assessed. Responses to these translocation issues may differ in other states where yabbies, in general, form native populations in waterways.)

By only considering applications from commercial yabby growers, yabbies will not be spread beyond the area which commercial yabby farms are already located, and thus there should be no greater risk to the native freshwater crayfish from yabbies than there currently is.

Hatcheries would maintain separate populations of *C. rotundus* and *C. albidus*; they could then cross female *C. rotundus* with male *C. albidus* to produce the all male hybrid for growout by farmers.

3) Production of hybrids and economic costing

Hybrids can be produced by mating female *C. rotundus* x male *C. albidus* yabbies. This can be accomplished using one of three methods;

1) Stocking breeding ponds
2) Cage reproduction in dams
3) Hatchery aquaria production

i) *Stocking breeding ponds*

Construction of three breeding ponds would be required for this method. Each pond should be fenced and enclosed by bird netting. One broodstock pond should be stocked with *C. rotundus* and one with *C. albidus* to establish two separate broodstock populations. The third pond would be used to produce male only progeny by placing 100 x female *C. rotundus* from the *C. rotundus* pond and 50 x male *C. albidus* from the *C. albidus* pond into the breeding pond. After spawning the breeding pond can be trapped or drained to remove male only progeny for stocking into growout ponds or dams.

Economic costing:

This technique requires the construction of 2 x 100 m^2 broodstock ponds ($1,000) and 1 x 100 m^2 breeding pond ($500).

Production

Assuming 200 juveniles/female/year annual production of hybrid juveniles from this system would be around 20 000 juveniles per year using a conservative estimate of only one successful spawning per female per year.

ii) *Cage reproduction in dams*

This relatively simple option entails placing cages containing *C. rotundus* and *C. albidus* broodstock into existing farm dams. The animals would mate in the cage and juveniles produced could then escape through the cage mesh into the dam.

Economic costing:

Assuming farmers use three existing dams this technique only requires the construction of breeding cages ($50/cage) in addition to the broodstock ponds. Cages would simplify removal and reuse of broodstock and reduce cannibalism of juveniles by broodstock. The use of breeding ponds would be optional but would allow control of stocking density in farming ponds/dams.

Production

Placing 10 cages each containing 10 females and 5 males into a 2,000 m^3 dam should produce 20,000 juveniles per year assuming 200 juveniles/female/year.

ii) *Hatchery aquaria production*

Hatchery production provides greater control over reproduction, however space limitations generally result in lower production levels than those achieved in ponds or dams. Tanks would be required for keeping broodstock populations and for breeding/hatching juveniles.

Economic costing

Hatcheries vary considerably in complexity and design. However, for a small aquarium facility based upon 30 tanks (20 broodstock holding and 10 breeding), establishment costs in an existing building are around $8,000. Operating costs for hatcheries can be high, particularly where water is heated for year round reproduction. However, these costs vary according to system design and location.

Production

Ten 80 L aquaria can hold 10 pairs of broodstock for producing hybrids. Assuming the facility is heated to permit year round reproduction each breeding tank could accommodate 5 matings per year, assuming 200 juveniles/female, production would be 10 000 juveniles. However, in our experience, survival of juveniles in hatchery tanks is poor with 10% survival being common. Therefore, total annual production from this system would be around 1,000 hybrids/year.

4) Return on investment

Economic evaluation of growing hybrids has shown a 4.8 times increase in gross return, with no additional operating costs aside from producing hybrid juveniles (Chapter 2). Therefore, for a small yabby farm currently producing $5,000 of *C. albidus* yabbies per year, hybrid production would increase revenue to $23,000/year. Thus capital costs incurred from establishing facilities to produce hybrids using options 1, 2 or 3 would pay back in less than 1 growing season. (In WA, specialised hatchery licences would be required for *C. rotundus* and this could increase the cost of hybrid juveniles. Factors will include ease of production of *C. rotundus* broodstock and hybrids and the number of approved "hatchery sites" as this will influence competition between suppliers.)

Risk assessment of translocation

The main concerns regarding the introduction and translocation of any aquatic organism are:

1) Their potential to impact upon genetic diversity;
2) The introduction of pathogens and disease; and
3) Their effect on ecosystems including the establishment of feral populations and their impact on indigenous aquatic species.

These three factors will be considered separately below and an assessment made based on current scientific knowledge.

1) The potential to impact upon genetic diversity

It has been suggested (Lawrence 1993) that if translocated animals were to escape and then breed with the existing population, the genetic integrity of the local population that had developed over time may be compromised. Interbreeding could also result in ÒhybridÓ populations, which, due to heterosis, could prove genetically superior to existing stock (Lawrence 1993). This could result in some existing strains being eliminated due to hybridisation and / or competition. Lawrence (1993) also suggests that another outcome of interbreeding is that the naturally occurring genetic diversity that could prove essential for future selective breeding programmes may be lost.

Determining whether there is any potential for there to be any significant impact upon genetic diversity is not straightforward in relation to yabbies as there is some debate over the taxonomy of *Cherax* from central and eastern Australia.

Briefly, since Reik (1969) categorised the *Cherax* in Australia into five broad groups, Sokol (1988), Campbell *et al*. (1994) and Austin (1996) have all revised the taxonomy of the 'yabby complex', but there is no agreement between the classifications. Sokol (1988) based on morphological traits concludes that *Cherax destructor*, *Cherax albidus* and *Cherax rotundus* should be considered to be separate and distinct species. In contrast, Campbell *et al*. (1994) and Austin (1996) feel that based on electrophoretic evidence that *C. albidus* should be considered to be a subspecies within the '*C. destructor* complex' as should *C. rotundus* (Austin 1996).

While the distinction of *C. albidus* and *C. rotundus* as separate species or as sub species of a greater '*C. destructor* complex' is not agreed upon, their ability to interbreed does mean that should *C. rotundus* escape into the wild, there could be some mixing of genes with the already present *C. albidus*. As mentioned above, the female *C. rotundus* crossed with a male *C. albidus* results in all male progeny, while the reciprocal cross results in a sex ratio of 1:1. However, there is also some doubt as to whether it would matter if *C. rotundus* and *C. albidus* were to interbreed and perhaps alter the genetic diversity in regions where these species are not native (i.e. Western Australia).

2) The introduction of pathogens and disease

In theory, if introduced or translocated animals contain pathogens, they could be detrimental to local stocks. Conversely, translocated animals may succumb to local pathogens or disease if they have not encountered them before.

The main disease issue for yabbies in Australia is *Thelohania*. *Thelohania* is a spore forming protozoan that infects the muscle of freshwater crayfish and is thought to be transmitted when a healthy animal eats the flesh of a diseased individual. The disease had until March of 1999, not been recorded in yabbies from Western Australia, however, it has now been recorded from a number of yabby properties in the state.

Recent extensive testing of yabby and marron properties throughout Western Australia did not identify *Thelohania* in any commercial marron ponds.

As stated in the commercialisation strategy section above, a sample of *C. rotundus* that are being held in quarantine facilities are to be tested for *Thelohania* and if found to be *Thelohania* free, then the risk of introducing pathogens and disease is not considered to be a significant reason to refuse the translocation of *C. rotundus* to commercial yabby farms in Western Australia.

The other diseases of yabbies that have been reported, including bacteria such as *Pseudomonas* and flat worms like *Temnocephalus* are all present in the natural environment in Western Australia and it is only poor management practices in culture conditions that make these diseases more prevalent.

It is anticipated that protocols developed by the Senior Fish Pathologist to minimise the spread of disease will need to be strictly adhered to ensure that the farmers and the surrounding environment are protected from any further spread of diseases.

It should be noted that both *C. rotundus* and the hybrid can return positive readings for *Thelohania* and that natural populations of *C. rotundus* are likely to contain *Thelohania* positive individuals (on the basis of spore digests) although external symptoms may not be evident.

3) The effect on ecosystems and their impact on indigenous aquatic species

It has been suggested that the introduction of some aquatic organisms could affect the composition of the local community either directly through predation and / or competition or by indirectly altering the existing environment (Lawrence 1993). They could also affect the local biodiversity of the environment should they interbreed with native species or strains.

While yabbies have previously been introduced into Western Australia, *Cherax rotundus* is arguably (as discussed above) a different species.

In terms of the potential for *C. rotundus* to have any further or greater impact on the surrounding ecosystem or on other aquatic species, it is believed that this potential would be low. However, at least in WA, burrowing and trophic characteristics should be assessed.

Lawrence *et al*. (1998) compared the environmental and water quality parameters between Western Australian industry farm dams and Merwyn Swamp, Victoria, the location from where the yabbies introduced into Western Australia originated from. What they found was that the important chemical and environmental aspects are very similar in both locations and they suggest that this must be, in part, a major contributing factor to the rapid growth and spread of the original yabbies which now are farmed over a large area of Western Australia.

Given that *C. rotundus* is only found in a small area of NSW (see Figure 1) and the chemical and environmental aspects differ to those found in Western Australia (Lawrence *et al*. 1998, Lawrence *et al*. 2002), it is not likely that it could be as successful in Western Australia as *C. albidus* was. It is also likely that *C. rotundus* would not 'out compete' *C. albidus* in the wild, so even if *C. rotundus* was to escape from commercial properties, it would be unlikely that it could spread further than *C. albidus* already has within the yabby area in Western Australia, nor is it likely to pose any threat to the native freshwater crayfish.

Another reason for the spread of *C. albidus* through Western Australia was due to yabbies being used for bait by recreational anglers who may move animals between river systems and also because the yabby has been misidentified as the native koonac or gilgie. As *C. rotundus* would only be introduced onto commercial yabby farms within the yabby area and only under strict protocols and guidelines, the risk of spreading *C. rotundus* through misidentification or by recreational anglers is virtually eradicated.

Conclusions

Based on the information above, it would appear that there is a low risk from the introduction of *C. albidus x C. rotundus* hybrids to commercial yabby farms in Australia. Strict protocols and guidelines for stocking of *C. rotundus* should be applied to areas where it is not native, to minimise the risk of escape and impact on the environment, while giving farmers access to technology that could increase production by stocking all male hybrids. A plan for commercialisation of the hybrid in WA is included in this report (Chapter 6) and its enactment in this form depends on reproductive performance of *C. rotundus* in ponds, critical assessments during the translocation process by a range of interest groups, particularly the Department of Environmental Protection, and disease clearances.

Note that since this chapter was written a comprehensive Department of Fisheries Management Paper (No. 160) has been prepared by J. Bellanger, the subject matter for which partly overlaps this chapter and may provide alternative estimates for some variables. The major concern expressed by various parties during the public comment period was to ensure that *C. rotundus* does not escape into the wild. There was little concern expressed over farming of hybrids.

Please note that this chapter was written prior to feedback from the Department of Environmental Protection, now the Department of Environment in Western Australia. That feedback makes commercialisation of hybrid yabbies unlikely in Western Australia, unless juvenile hybrids are produced by a government hatchery or obtained from interstate. See Chapter 6.

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COMMERCIALISATION/TRANSLOCATION/IP STRATEGY FOR WA (WITH COMMENTS ON OTHER STATES)

Below is an edited version of the original planning for the commercialisation process. Progress on this plan is included up to July 2004.

Translocation/commercialisation

- The growth trial comparing all-male hybrids with mixed sex *Cherax albidus* in model ponds terminated in April 2000 (see Chapter 2). Sensory evaluation and simulated live transport tests on the all-male hybrids compared to mixed sex *C. albidus* were conducted (see Chapter 4). A draft final report to FRDC and an interim report to the WA Aquaculture Development Council (ADC) were provided in June 2000.
- *Build up broodstock numbers of Cherax rotundus at the University of WA/Department of Fisheries WA quarantine facility at Shenton Park using existing pools.* (These are separate from the pool complex for which funding was obtained by University of WA from the ADC and FRDC for marron research.) This was attempted but juvenile production in the pools was not encouraging. Subsequently, indoor aquaria and an intensive culture system were used successfully prior to transfer to Avondale (see below). However, some breeding stocks have been maintained in pools at Shenton Park. Only *Thelohania*-free stocks are retained in the Shenton Park and Avondale facilities. The build-up in breeding stocks has been severely hampered by the high incidence of *Thelohania* in the only commercial farm that breeds them in NSW. Subsequent attempts, with CSIRO, to collect more stocks from the wild in NSW have been unsuccessful.
- *Subject to individual* C. rotundus *broodstock or juveniles being clear of* Thelohania*, move these to a quarantined section of the Avondale pond complex developed at a Department of Agriculture WA research centre for hosting FRDC funded yabby research.* Department of Fisheries WA facilitated biotechnological research by University of New England and CSIRO to allow identification of *Thelohania* positive individuals non-sacrificially. The PCR technique (Moodie and Le Jambre 2002; Moodie 2003; Moodie et al. 2003) has now been transferred to the Department of Fisheries, Fish Health Unit in Perth by agreement with CSIRO and University of New England. (All of the stocks held indoors were individually isolated and tested by the University of New England. *Thelohania*-free stocks and their progeny were transferred to ponds at Avondale. Pleopod squashes were also examined for confirmation of *Thelohania*-free status.)
- *Install solid fencing around individual yabby ponds and upgrade predator-netting covers at Avondale to allow it to be used as a quarantine facility. Use esfenvalerate to remove residual* C. albidus *from ponds to be used for* C. rotundus *and hybrids.* These activities were completed prior to transferring *Thelohania*-free stocks to Avondale. The Department of Fisheries withdrew from the Avondale site in 2004, apart from minimum site maintenance activities.
- *Breed large numbers of* C. rotundus *in the Avondale ponds there during summer 2000/2001.* This was attempted by providing higher levels of refuge provision, aeration and feeding than normally occurs in yabby ponds. However, reduced run off from rainfall at Avondale over 3 years hampered management of these ponds. This inability to do significant water

exchange, except for pumping from a saline soak, led to a series of management problems.

1. Rising pond salinity.
2. Uncontrollable algal blooms in part due to residual nutrients from multiple FRDC trials in these ponds and lower yabby biomass than predicted leading to overfeeding.

The second problem was addressed by sediment removal from the ponds. The first problem probably affected reproductive performance. Heavy rainfall in July 2003 overcame this problem and may have allowed a substantial build up in *C. rotundus* numbers for transfer to commercial aquaculture facilities, if *Thelohania*-free *C. rotundus* stock had been available from drought affected waterways or farmed stock in NSW. *C. albidus* survived well in a small trial in autumn-Winter 2004, in clean ponds at Avondale, indicating the ongoing suitability of this site for yabby trials, subject to rainfall.

- *Review whether the all-male hybrids interbreed with* C. rotundus *(at Shenton Park)*. A review of breeding records from Shenton Park has confirmed that viable interbreeding has not occurred.
- *Decide on the selection criteria by which 'tenderers' will be selected after an Expression of Interest to receive the* C. rotundus *animals for broodstock*. Those criteria were to be developed by a joint Department of Fisheries WA and Yabby Growers Association Committee with technical advice being provided by Department of Fisheries WA Research Staff. These criteria have been incorporated into Fisheries Management Paper No. 160, prepared by J. Bellanger in 2002.
- *Set any new licensing conditions e.g. those governing transfer of* Cherax rotundus *animals out of an approved farm.* Draft license conditions have been incorporated in the above Management Paper.
- *Getting translocation approval for: (1) releasing* C. rotundus *animals from quarantine so that they can later be transferred to commercial operators, outside of the marron zone, for breeding more broodstock (but not for direct farming), and (2) commercial farming outside of the marron zone, of* C. rotundus *x* C. albidus *juveniles*. The aim was to minimise the likelihood of release of *C. rotundus* into natural waterways. Escape of sterile hybrids is seen as less critical. Subsequently, the Department of Environmental Protection advised that although there were few concerns over farming of hybrid yabbies, approval to move *C. rotundus* out of quarantine was unlikely unless there was agreement to establish a government operated hatchery. As the Department of Fisheries has now terminated its yabby research program, this is unlikely to occur.
- *Complete any final disease testing recommended by the Agency's Fish Health Unit*. This was scheduled for the May 2004 harvesting of Avondale ponds but very poor survival of *C. rotundus*, did not allow for effective disease assessment.
- *Harvest* C. rotundus. *Pond discharge to land disposal. Evaluate the burrowing behaviour of* C. rotundus *animals in those ponds.* Considerable data has already been obtained including casts of individual burrows (see Lawrence *et al.* 2002 for methodology).
- *Transfer of* C. rotundus *animals to the successful 'tenderers' hopefully in 2004.* (Timing depends on progress with the tender process and breeding success in Avondale ponds.) Because of the production and translocation obstacles, the tender did not proceed.
- *Prepare a final report in the Department of Fisheries WA Final Contract Report Series to cover all of the hybrid work funded by FRDC and ADC (for general distribution to*

interested parties in Australia and overseas). This progressed after FRDC reviewed the final report and further information was requested by the ADC.

- At this stage, it is unlikely that hybrid yabbies will be commercialized in Western Australia unless juvenile hybrids are purchased from interstate (subject to disease testing).

Intellectual property issues

The plan envisaged, when the hybrid yabby technology was discovered, was to make this information available to yabby farmers nationally without recovery of IP by Department of Fisheries WA or FRDC. (This may not necessarily be the policy of FRDC or the Western Australian government for future discoveries of this type.) Accordingly, the technology for producing the hybrids was published in the scientific literature (Lawrence *et al*., 2000) It is now highly unlikely that income will be generated from the cost of producing *Thelohania*-free *C. rotundus* (and *C. albidus*) broodstock.

There was an understanding that the IP position of FRDC was to ensure that no one group was able to monopolise the technology without other groups having had the opportunity to use it. Balanced against this is the need to reduce the risk of *C. rotundus* becoming established in natural waterways in Western Australia. Advice from the Department of Environmental Protection, if acted upon, would see a government operated hatchery established. This would have the benefit of ensuring that a suitable brood range of recipients could be supplied with hybrids. However, it is unlikely that funding would be available for such a hatchery. Interstate transfers of *C. rotundus* from WA may not be necessary as it occurs in NSW and stocks may exist in Victoria. It is understood that stocks can be obtained from NSW for SA growers, subject to translocation approval.

In summary, there is no intention to recover IP or to allow it to be monopolised unreasonably. Distribution of *C. rotundus* broodstock (and *C. albidus*, if necessary) would be dependent on translocation approvals and an availability of broodstock.

FRDC funds covered evaluation of the hybrids but not subsequent mass production of broodstock for generating hybrids. State funds have been used to help generate the broodstock and to develop a *Thelohania*-free line. If available, surplus stock could have been made available to interstate agencies, however this is not seen as a primary responsibility for the Department of Fisheries, WA.

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CONCLUSIONS AND RECOMMENDATIONS

It has been shown that hybridising two species of freshwater crayfish, female *Cherax rotundus* x male *Cherax albidus* produces only male progeny. This hybrid offers considerable potential for controlling reproduction by yabbies in aquaculture ponds.

This study has confirmed the previously reported discovery of male hybrids from mating female *Cherax rotundus* x male *Cherax albidus* yabbies. In this study no female hybrids were produced.

The average growth rate of hybrid *C. albidus* x *C. rotundus* yabbies was greater (0.11 ± 0.004 g/day) than that of *C. albidus* yabbies (0.06 ± 0.002 g/day). The growth rates of *C. albidus* x *C. rotundus* hybrids and the *C. albidus* yabbies were similar until they reached sexual maturity, at around 14 g, at which stage the growth rate of *C. albidus* decreased. At the conclusion of the experiment the *C. albidus* x *C. rotundus* hybrids (50.7 ± 2.2 g) were almost twice as large as the *C. albidus* yabbies (27.2 ± 1.7 g).

The greater growth and increased numbers of yabbies in the larger and therefore more valuable size categories resulted in the value of *C. albidus* x *C. rotundus* hybrids being 4.8 times greater than that of the *C. albidus* yabbies at harvest after 424 days.

The male only *C. albidus* x *C. rotundus* hybrids did not produce progeny when mated with *C. albidus* or *C. destructor* females.

Sensory evaluation has shown that the taste of the hybrids is comparable with currently farmed *C. albidus* yabbies. Although the survival of hybrids (63%) in simulated transport trial was lower than *C. albidus* yabbies (90%), this difference was only evident after very extended live storage (11 days). This trial should be repeated when a larger number of animals of similar size are available. Survival of hybrids in this study was greater than that reported previously for transport survival of *C. albidus* (Roe 1994).

There do not appear to be any barriers to the immediate commercialisation of hybrids in NSW as stocks of both *C. albidus* and *C. rotundus* can be found on farms in this state. In SA and Victoria the main limitation is availability of stock which, if approved by state authorities, may be purchased from farmers in NSW. Importantly the lack of reproduction between hybrids and either *C. albidus* or *C. destructor* females shows that they are unlikely to disturb the gene pool of existing yabbies on commercial farms. This should however be monitored by SA and Victorian authorities to protect other species of endemic crayfish populations. In Western Australia, concerns over *Thelohania* necessitated the development of a broodstock population free of this disease. Currently, the translocation assessment by the Department of Environmental Protection (now Department of Environment) would require the establishment of a government operated hatchery to allow commercialisation of hybrid yabbies in Western Australia. This is unlikely to occur and the alternative would be regular transfers of hybrid juveniles from interstate (subject to disease testing and translocation approval).

An economic evaluation of three methods for producing hybrids 1) Stocking breeding ponds, 2) Cage reproduction and 3) Hatchery production, shows that a small farm producing $5,000 of *C. albidus* yabbies/year could expect to increase returns to $23,000/year. Thus capital costs incurred from establishing facilities for hybrid production using any of the three methods proposed would pay back in less than one year.

In a subsequent Department of Fisheries WA Management Paper (No.160 prepared by J. Bellanger), it was estimated that hybrid juveniles could be produced in breeding ponds or farm dams for $0.08 each. If 200 new farm dams were stocked with hybrids, the resultant increase in annual turnover was estimated to be about $500,000. As about 4,000 dams are harvested in years of reasonable farm water storage volumes in WA the potential benefit could be much greater.

SENSORY EVALUATION OF YABBIES

May 2000

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EXECUTIVE SUMMARY

The Food Centre of WA (Inc) conducted a sensory evaluation on Yabbies (Hybrid v WA) on behalf of Fisheries WA.

The evaluation was conducted on Wednesday 19 April 2000 at the premises of The Food Centre and involved a total of 30 self-selected male and female respondents from Central Metropolitan College of TAFE and the general public.

The evaluation required respondents to consume three yabbies (two samples of one type, one of the other) and answer eight questions regarding sensory attributes such as general appearance, flavour pleasantness, eating texture, after-taste and overall acceptability.

Results indicated no significant difference between the two types of Yabbies and it may therefore be inferred that the Hybrid Yabbies are comparable with current commercially available WA Yabbies.

It must be stressed, however that this conclusion is based on a relatively small quantity of sample available to each of the respondents, as well as a relatively small sample population.

It is therefore recommended that

1. Fisheries WA conduct additional evaluation using a larger sample size, to enable a better representation of the population at large.
2. the study be run again using larger sized yabbies or greater quantities of yabbies.
3. the study be conducted with male yabbies only, to avoid potential bias, particularly in the appearance attribute.
4. A question on peelability of the yabbies be included.

1.0 RESEARCH DESIGN: METHODS AND PROCEDURES

1.1 DESCRIPTION OF RESPONDENTS

The respondents were self-selected staff at Central Metropolitan TAFE, and various members of the public. The selection of respondents was non-random and all respondents were shell fish consumers.

The majority of the respondents were untrained in sensory evaluation.

1.2 TEST ENVIRONMENT

Sensory evaluation of the fish was conducted on Wednesday 19 April 2000 between 10.30am and 2.30pm at The Food Centre of Western Australia.

Respondents were allocated a table each and were presented with a cup of water, a questionnaire, a fork and some toast to cleanse their palates. Samples of the yabbies were supplied as required.

Environmental factors such as crowding, lighting, increased temperature and noise were not controlled in this experiment.

1.3 EXPERIMENTAL VARIABLE

Three samples were used in this experiment. The three samples varied depending on the specific combination (triangle test) presented to each of the respondents. If the respondents were given sets A-C, the samples consisted of 1 sample of the WA Yabbies and two samples of Hybrid Yabbies, whereas if they were presented with sets D-E, respondents would have had to evaluate 2 samples of WA Yabbies and 1 sample of Hybrid Yabbies.

The samples were presented in this particular way to prevent positioning bias.

The main difference between the sets of two yabbies was their relative size. The Hybrid Yabbies tended to be larger than the normal WA Yabbies as a result of faster growth.

The Hybrid yabbies were all males, in contrast with the WA Yabbie samples which consisted of males and females.

The samples were evaluated as cold products and were refrigerated until required.

1.4 PREPARATION OF SAMPLES

The Yabbies were cooked by the staff at Fisheries WA. The cooking method involved the cleaning and removal of the yabbies heads and then steaming (100°C) for 5 minutes.

After cooking they were delivered to The Food Centre, where they were cooled in the refrigerator and then peeled and placed into clear 70mL containers.

All products were stored in the refrigerator overnight and served as cold products.

1.5 CODING

Each of the samples were allocated a symbol (+, V, O and ▽), which reduced the probability of coding bias occurring.

The coding system that was employed in the evaluation was a follows:

SETS A-C

Code	**Sample**
+	WA Yabbies
V	Hybrid Yabbies
O	Hybrid Yabbies

SETS D-E

Code	**Sample**
+	WA Yabbies
V	Hybrid Yabbies
▽	WA Yabbies

1.6 QUESTIONNAIRE DESIGN

The questionnaire, consisting of two pages, was self-administered. The first page related to demographic data, whilst the second related to the sensory evaluation.

The questionnaire comprised of eight questions regarding sensory attributes of the yabbies. The sensory attributes included questions regarding general appearance, flavour pleasantness, eating texture, after taste and overall acceptability.

Demographic data regarding the age and gender of the respondents were also collected.

A copy of the questionnaire can be found in Appendix A.

1.7 DATA ANALYSIS

After the sensory evaluation, the questionnaires were coded accordingly with the appropriate values and entered into the SPSS statistical package, where a number of statistical tests were applied.

1.8 STATEMENT OF HYPOTHESIS

The "null hypothesis" states that there is no significant difference between the samples.

When a significant difference is found the null hypothesis is automatically rejected and the alternative hypothesis is accepted.

The level of significance is set at 0.05 (ie in the succeeding tables, "sig" must be greater than 0.05, for the null hypothesis to be accepted)

For this test, the nearer "sig" is to 1.0, the more likely it is that all samples fall in the same "statistical' population.

2.0 RESULTS

2.1 DEMOGRAPHICS

A total of 30 individuals took part in the sensory evaluation of the yabbies. The typical respondent on the day was male (28.8%), between 20-30 years of age (22%) and married (25.4%).

The sampling method involved non-random self-selected respondents, and it must be stressed that the results are not necessarily representative of the whole population at large and thus must be regarded as a pilot study.

2.2 GENERAL APPEARANCE

Very unpleasant to very pleasant

Sets A-C

Sample n=15	**Mean**	**SD**	**F**	**Sig**	**Decision**
+ WA	32.4667	8.364			
V Hybrid	30.9333	11.0419	.294	.747	Accept
O Hybrid	33.6000	9.1792			

Very unpleasant   Very pleasant

Respondents did not find any significant difference between the WA and Hybrid Yabbies. As the mean indicates, there was little difference in terms of appearance between the yabbies samples.

Sets D-E

Sample n=15	**Mean**	**SD**	**F**	**Sig**	**Decision**
+ WA	31.4667	8.6921			
V Hybrid	29.6000	9.2489	1.541	.226	Accept
▽ WA	34.9333	7.2651			

Very unpleasant   Very pleasant

Respondents did not find any significant difference between the WA and Hybrid Yabbies. All yabbies were rated quite high in terms of their appearance.

2.3 FLAVOUR PLEASANTNESS

Very unpleasant to very pleasant

Sets A-C

Sample n=15	Mean	SD	F	Sig	Decision
+ WA	30.5333	10.5415			
V Hybrid	29.2857	8.5524	.108	.898	Accept
O Hybrid	28.9333	10.3127			

Very unpleasant Very pleasant

No significant difference was found between the WA and Hybrid Yabbies. Similar ratings were given by the respondents in terms of the flavour of the yabbies.

Sets D-E

Sample n=15	Mean	SD	F	Sig	Decision
+ WA	32.2000	8.2997			
V Hybrid	32.4667	7.8364	.049	.952	Accept
▽ WA	31.5333	9.0858			

Very unpleasant Very pleasant

Respondents did not find any significant difference between the WA and Hybrid Yabbies. All yabbies were rated quite high in terms of their flavour.

2.4 EATING TEXTURE

Very unpleasant to very pleasant

Sets A-C

Sample n=15	Mean	SD	F	Sig	Decision
+ WA	31.3333	8.7723			
V Hybrid	30.7333	8.2762	.039	.962	Accept
O Hybrid	30.4000	10.7358			

Very unpleasant Very pleasant

Respondents did not find any significant difference between the WA and Hybrid Yabbies. All samples were rated very similarly in terms of eating texture. All samples were quite pleasant.

Sets D-E

Sample n=15	Mean	SD	F	Sig	Decision
+ WA	33.0667	8.9160			
V Hybrid	34.1333	7.6706	.077	.929	Accept
▽ WA	33.0667	9.0984			

Very unpleasant Very pleasant

No significant difference was found between the WA and Hybrid Yabbies. As the means indicate there was little difference between the Yabbies

2.5 TOUGHNESS

Very tough to very tender

Sets A-C

Sample n=15	**Mean**	**SD**	**F**	**Sig**	**Decision**
+ WA	30.8000	8.9459			
V Hybrid	32.4000	8.8463	.125	.883	Accept
O Hybrid	30.9333	11.1833			

Very unpleasant Very pleasant

No significant difference was found between the WA and Hybrid Yabbies. All samples were rated quite tender, as indicated by the results above.

Sets D-E

Sample n=15	**Mean**	**SD**	**F**	**Sig**	**Decision**
+ WA	29.7333	10.4708			
V Hybrid	32.9333	10.1522	1.593	.215	Accept
▽ WA	36.3333	9.7444			

Very unpleasant Very pleasant

The ▽ WA sample was rated more tender then the +WA and V Hybrid Yabbies, although the difference was not significant. All three samples were rated as being quite tender.

2.6 DRYNESS

Very dry to very moist

Sets A-C

Sample n=15	**Mean**	**SD**	**F**	**Sig**	**Decision**
+ WA	35.8667	10.2600			
V Hybrid	36.6667	4.5461	.034	.966	Accept
O Hybrid	36.3333	9.2247			

Very unpleasant Very pleasant

Respondents did not find any significant difference between the WA and Hybrid Yabbies. All samples were rated very similarly in terms of their moisture content.

Sets D-E

Sample n=15	**Mean**	**SD**	**F**	**Sig**	**Decision**
+ WA	34.2857	8.8355			
V Hybrid	32.000	8.5540	.241	.787	Accept
▽ WA	33.6667	7.6314			

Very unpleasant Very pleasant

Respondents did not find any significant difference between the WA and Hybrid Yabbies. As the means indicate, there was little difference between the Yabbies in terms of their moisture content.

2.7 MOUTHFEEL

Very unpleasant to very pleasant

Sets A-C

Sample n=15	**Mean**	**SD**	**F**	**Sig**	**Decision**
+ WA	36.4667	8.5262			
V Hybrid	34.6000	7.1594	1.023	.368	Accept
O Hybrid	31.7333	11.343			

Very unpleasant Very pleasant

Respondents generally preferred the mouthfeel of +WA and V Hybrid. It is not known why the O Hybrid Yabby was rated slightly lower. This variation however, was not significantly different.

Sets D-E

Sample n=15	**Mean**	**SD**	**F**	**Sig**	**Decision**
+ WA	32.7333	8.1193			
V Hybrid	32.9333	9.8522	.015	.986	Accept
▽ WA	32.4000	7.8722			

Very unpleasant Very pleasant

No significant difference was found between the WA and Hybrid Yabbies. All samples were rated very similarly.

2.8 AFTERTASTE

Very unpleasant to very pleasant

Sets A-C

Sample n=15	Mean	SD	F	Sig	Decision
+ WA	33.0000	7.9821			
V Hybrid	31.4667	7.9180	.592	.558	Accept
O Hybrid	29.4000	11.0246			

Very unpleasant Very pleasant

Respondents did not find any significant difference between the WA and Hybrid Yabbies. They did however, feel that the +WA and the V Hybrid Yabbies had a slightly better aftertaste than the O Hybrid Yabby.

Sets D-E

Sample n=15	Mean	SD	F	Sig	Decision
+ WA	33.4667	8.6261			
V Hybrid	33.0667	9.7429	.564	.573	Accept
▽ WA	36.1333	7.2394			

Very unpleasant Very pleasant

Respondents found no significant difference between the WA and Hybrid Yabbies. All samples were rated similarly in terms of the aftertaste.

2.9 OVERALL ACCEPTABILITY

Totally unacceptable to totally acceptable

Sets A-C

Sample n=15	Mean	SD	F	Sig	Decision
+ WA	32.6667	10.1254			
V Hybrid	31.8667	9.7605	.346	.710	Accept
O Hybrid	29.6667	10.7748			

Totally unacceptable Totally acceptable

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Respondents did not find any significant difference between the WA and Hybrid Yabbies. All samples were rated as being quite acceptable.

Sets D-E

Sample n=15	Mean	SD	F	Sig	Decision
+ WA	33.3333	7.3743			
V Hybrid	33.3333	9.7076	.275	.761	Accept
▽ WA	35.2667	7.4303			

Totally unacceptable Totally acceptable

: •••• •••• •••• •••• •••• •••• •••• •••• •••• •••• :

No significant difference was found between the WA and Hybrid Yabbies. Respondents tasting sets D-E found the ▽ WA to be slightly more acceptable than both the +WA and the V Hybrid.

3.0 DISCUSSION AND CONCLUSIONS

The results of the sensory evaluation indicated that the sample population as a whole found the yabbies to be quite acceptable.

The WA Yabbies were rated slightly better than the Hybrid yabbies on most of the attributes tested, and the results were not affected by the triangle test combinations used.

Whilst the WA Yabbies indicated slightly better ratings, no significant differences were found between the Hybrid and the WA Yabbies on any of the attributes tested. All results fell in the 'accept' category. This indicates that the Hybrid Yabbies are quite comparable to current commercially available WA yabbies, relative to those parameters investigated.

Whilst these results are encouraging, it should be remembered a number of factors can increase the probability of a type II error occurring (the false acceptance of the null hypothesis).

One of the major problems that was encounter through the evaluation was the quantum of sample for each respondent. Each respondent was given three yabbies (one for each symbol) and were required to evaluate 8 questions with it.

The yabbies, unfortunately, were very small, weighing on average 5.5 g (WA) and 8.0 g (Hybrid) and therefore, did not allow some respondents to fully evaluate the yabbies.

As suggested in the recommendations, a set of larger yabbies or greater numbers of these yabbies should be used for further sensory work.

As with all evaluations of this size, the results can only be interpreted as a pilot study as the respondents who participated in the evaluation may not be representative of the total population.

4.0 RECOMMENDATIONS

Whilst the data from this sensory evaluation provides useful information for Fisheries WA, it is recommended that additional sensory evaluation be conducted using a larger sample size, to enable a true representation of the population at large.

It is also suggested that the study be run again using larger sized yabbies or that greater numbers of yabbies be made available for each respondent, enabling more comprehensive evaluation.

There was discussion during design of the questionnaire, as to whether a question on the peelability of the yabbies be included. It is felt that future sensory work would benefit from the inclusion of this attribute.

It is noted that the sex of the yabbies may potentially cause a bias, particularly in the general appearance attribute. Therefore, it is suggested that future sensory work be conducted on male yabbies (The Hybrid samples were all males, whereas the WA samples consisted of both male and female) only, especially during breeding season.

Appendix 2

Project staff and acknowledgements

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