Factors required for the successful aquaculture of black bream (*Acanthopagrus butcheri*) in inland water bodies

Sarre G.A., Partridge G.J., Jenkins G.I., Potter I.C. and Tiivel D.J.





CHALLENGER TAFE



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FINAL REPORT

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99/320 Factors required for the successful aquaculture of black bream (<i>Acanthopagrus butcheri</i>) inland water bodies		
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OBJECTIVES:

The objectives of the proposed study were to determine the suite of conditions which, in inland water bodies, are required for rearing black bream to a size that is suitable for recreational angling. Specific objectives:

- 1. Determine the effectiveness of using cages to house young black bream.
- 2. Determine, under controlled laboratory conditions, which of the currently available commercial fish feeds lead to optimal growth of black bream, and then determine the appropriate rate of feeding under field conditions over an extended grow-out period.
- 3. Determine whether yabbies are preyed on by large black bream and if, where appropriate, yabbies can be cultured in polyculture with black bream.
- 4. Determine the effectiveness of introducing under and above water cover to reduce the predation of black bream by cormorants in inland water bodies.
- 5. Determine whether the very different growth rates of black bream in the Swan and Moore River estuaries are paralleled by comparable differences when black bream from these two systems are cultured in the laboratory under identical salinity, temperature and food conditions.
- 6. Determine the relationship between the types of potential food that are naturally present in inland saline water bodies and those that are ingested by different sizes of black bream.
- 7. Determine whether black bream are able to spawn successfully in inland water bodies and, if so, the broad characteristics of those water bodies where spawning occurs.
- 8. To provide information to farmers that will enable them to grow black bream successfully and thus constitute an extra source of revenue through charging for access to fishing on their land.

NON-TECHNICAL SUMMARY:

OUTCOMES ACHIEVED

The results of this project will contribute to the on-going development and success of inland black bream recreational fisheries and the inland saline aquaculture industry in general. Several commercial black bream fishout ventures are now operational in inland Western Australia, one of which utilises saline groundwater. It has been proposed that a pilot project to stock the saline pools of the upper Avon River, which is located in the heart of the W.A. Wheatbelt, should be undertaken to provide recreational fishing opportunities for the inhabitants of a number of rural towns. The continued development of the W.A. inland saline aquaculture industry will result in social and economic benefits to rural areas through increased tourism and opportunities for farm diversification.

During recent years, there has been a great deal of interest in the concept of stocking the inland water bodies of southern Western Australia with fish that would be able to provide recreational angling opportunities, or even possibly a small scale commercial fishery. In 1992, the development of hatchery techniques for black bream (*Acanthopagrus butcheri*), an estuarine species endemic to the estuaries and rivers of southern Australia, enabled large numbers of black bream juveniles to be produced and subsequently sold to property owners and farmers for stocking inland water bodies. A two-year FRDC Project (97/309) entitled "Elucidation of the characteristics of inland fresh and saline water bodies that influence growth and survival of black bream" demonstrated that the majority of water bodies stocked were fresh or low salinity dams and that the survival and growth rates of black bream appeared an excellent candidate for stocking saline inland water bodies. This finding was particularly pertinent to those regions of the WA Wheatbelt affected by rising water tables and the associated increases in groundwater salinity and it has resulted in renewed interest from farmers in establishing black bream in the saline ponds and dams on their properties for private and small scale commercial recreational fishing ventures.

In the absence of information concerning the best methods and techniques to grow black bream in inland saline water bodies, a series of laboratory and pond-based field trials were undertaken to identify the factors required for their optimal survival and growth in an inland aquaculture environment.

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RESULTS

The results from this project have demonstrated the following.

- The use of floating fish cages is a useful technique for culturing juvenile black bream, particularly when stocked in a water body for the first time, as these cages provide protection from avian predators and increase the ease of monitoring fish health and feeding activity and the harvesting of fish.
- While naturally occurring feed within research ponds is adequate to ensure survival of juvenile black bream, additional supplementary feeding is crucial for optimal growth.
- Large black bream will readily feed on adult yabbies when stocked in free-range polyculture. In low salinity water bodies, yabbies may provide a semi-sustainable, in-pond food source for black bream providing that the adult yabbies can avoid predation by having refuge in the form of hides or burrows.
- The types of natural food items found in inland saline water bodies are generally in low abundance and/or inadequate as feed for large black bream (> 200mm T.L.) and thus supplementary feeding is essential for their survival and growth.
- While the provision of in-water cover, such as wire coils and branches, may reduce predation of juvenile black bream in ponds by cormorants, above-water netting of ponds is the only 100% effective method for preventing the loss of juvenile stock through avian predation.
- Poor Food Conversion Ratios (FCRs), presumably due to uneaten feed and an associated reduction in water quality, are likely to occur when large black bream are fed as little as 2% (dry feed: total wet weight of fish) in a single feed event, particularly in cage and small pond culture. This may be partly alleviated by using floating feed.
- Poor water quality is likely to be a major factor in limiting the growth of large black bream, particularly in small and static, *i.e.* zero exchange, water bodies.
- Based on preliminary data, black bream are capable of spawning successfully in inland water bodies, providing the following conditions are present:
 - Water with a salinity level of at least 10ppt.
 - Water temperature $> 20^{\circ}$ C during the spawning period (late spring early summer).
 - The absence of mosquito fish (Gambusia holbrooki).
 - The presence of sufficient and appropriate live feed, usually copepods, in the water body during the spawning period of black bream (September-December).
 - Presence of macrophyte growths as a refuge for larvae and juveniles.
 - Supplementary feed is provided at regular intervals.
 - An absence of algal blooms during the spawning period.
- Based on growth rates, FCRs, lipid content and economic data, the Australian-produced Ridley Agriproducts and Pivot salmon grower diets were the most suitable for growing juvenile (< 50g) and large black bream, respectively.
- Commercial heat-extruded diets are significantly more water stable and produce greater growth rates and lower FCRs than a cheaper, cold-pressed diet, even after the application of a stabilisation process.
- The very marked differences in the growth rates of black bream in the Swan and Moore River estuaries are not replicated when juvenile black bream, cultured from broodstock collected from these two systems, are grown in the laboratory under identical environmental and food conditions.

GENERAL CONCLUSION

This project, and also the previous project (FRDC 97/309), have demonstrated that black bream is ideally suited for stocking inland saline water bodies for recreational fishing. The species is robust, tolerant of a wide range of salinities and temperatures and, in some water bodies, will successfully reproduce and establish a self-sustaining population. Optimal survival and growth can be achieved by using techniques such as cage culture for initial stocking of juveniles, the provision of an appropriate supplementary diet and feeding regime and reducing the predation of juvenile fish by birds.

Further work is required to improve the growth rate of black bream in inland water bodies to reduce the time between the stocking of juveniles and the attainment of a size suitable for angling. A faster growth rate would also increase the potential of black bream for commercial aquaculture production in inland saline waters.

KEYWORDS: Black bream, aquaculture, inland water bodies, *Acanthopagrus butcheri*, farm dams, feeding.

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This report is dedicated to the late David Tiivel.

1.0 GENERAL INTRODUCTION

1.1 BACKGROUND

The majority of the natural and artificial inland water bodies in south-western Australia are saline. Thus, any fish species that is to be considered a candidate for stocking in these water bodies must be euryhaline, *i.e.* physiologically able to live in a wide range of salinities. Furthermore, that species must be hardy and able to tolerate the high temperatures found in many inland regions. One species that obviously falls into this category is the black bream *Acanthopagrus butcheri*, which is one of the most important angling and food fish species in the estuaries of Western Australia and contributes to the commercial fishery in several of those estuaries (Lenanton and Potter, 1987). This species is also found in south-eastern Australia, where it is likewise widely-fished recreationally and contributes to the commercial fishery (Kailola *et al.*, 1993).

Since 1992, the development of hatchery techniques for black bream by the Aquaculture Development Unit of Challenger TAFE Fremantle (ADU) has led to large numbers of juveniles being sold to private property owners in Western Australia for the purpose of stocking the water bodies on their land. Between 1994 and 1996, over 200 000 black bream juveniles were stocked in a large number of inland water bodies, which varied markedly in their physico-chemical characteristics. Despite a rapid increase in the interest shown by farmers and private industry in stocking inland water bodies with black bream for recreational fishing and, in some cases, smallscale commercial aquaculture, there were limited data on either the success of past stockings or the broad characteristics of the water bodies that had been stocked with black bream. A two-year project entitled "Elucidation of the characteristics of inland fresh and saline water bodies that influence growth and survival of black bream" which was funded by the FRDC (Project 97/309) focused mainly on obtaining preliminary data on whether it was feasible to stock black bream in inland water bodies and, if so, to determine the basic conditions likely to be conducive for successful stocking. The project demonstrated that the majority of water bodies stocked were fresh or low salinity dams and that the survival and growth rates of black bream stocked in these water bodies were poor. However, it also demonstrated that black bream were an excellent candidate for stocking saline inland water bodies. This conclusion was based on the fact that, in some saline water bodies, black bream survived well and grew to a size that was sufficiently large to be fished by anglers. This finding was particularly pertinent to those regions of the W.A. Wheatbelt affected by rising water tables and associated increases in groundwater salinity.

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Furthermore, it resulted in renewed interest by farmers in establishing black bream in saline ponds and dams on their properties for both private and potentially commercial recreational fishing ventures.

However, it must be recognised that the conditions identified in the above project as conducive to the survival and growth of black bream were very broad-based and further work was clearly required to determine more precisely the most appropriate combination of conditions for the optimal rearing of black bream in farm dams and other inland water bodies.

1.2 NEED

For the specific reasons below, there is clearly a need to determine the optimal conditions required for the successful aquaculture of black bream in inland water bodies in order to assist in the development of a recreational inland fishery in south-western Australia. These are:

- To provide, for local residents and tourists in rural areas, access to an outstanding angling and food fish species that occurs naturally in Western Australia and which is both hardy and adapted to living in a wide range of salinities and temperatures.
- To increase the potential for tourism in rural areas which, during recent years, have suffered economic decline through land degradation and salinisation.
- To determine whether the very different growth rates recorded for geographicallyisolated natural populations of black bream are likely to be due to genetic differences or differences in the environments in which they live. Such data are important for ascertaining whether it is necessary to select specific populations for use as broodstock.
- To explore the possibility that inland water bodies could be used for producing black bream economically on a limited commercial scale.
- To provide fisheries biologists both in W.A and other states with information on the best methods and conditions for growing black bream in inland water bodies.

1.3 OBJECTIVES

- Determine the effectiveness of using cages to house young black bream.
- Determine, under controlled laboratory conditions, which of the currently available commercial fish feeds are optimal for the growth of black bream, and then determine the appropriate rate of feeding under field conditions over an extended grow-out period.
- Determine whether yabbies are preyed on by large black bream and, where appropriate, if yabbies can be cultured in polyculture with black bream.

- Determine the effectiveness of introducing under and above water cover to reduce the predation of black bream by cormorants in inland water bodies.
- Determine whether the very different growth rates of black bream in the Swan and Moore River estuaries are paralleled by comparable differences when black bream from these two systems are cultured in the laboratory under identical salinity, temperature and food conditions.
- Determine the relationship between the types of potential food that are naturally present in inland saline water bodies and those that are ingested by different sizes of black bream.
- Determine whether black bream are able to spawn successfully in inland water bodies and, if so, the broad characteristics of those water bodies in which spawning occurs.
- To provide information to farmers that will enable them to grow black bream successfully and thus constitute an extra source of revenue through charging for access to fishing on their land.

2.0 GENERAL MATERIALS AND METHODS

2.1 RESEARCH POND FIELD TRIAL STATION

It was initially proposed to undertake the pond field trials at the Avondale Research Station near Beverley. However, due to a very limited supply of saline water of an appropriate quality at this site and the potential for conflict between the proposed work and that of ongoing FRDC yabby research trials, an alternative location was sought.

After negotiation with the property owner, it was decided to undertake research pond trials adjacent to a privately owned fish farm on a farming property situated in the W.A. Wheatbelt town of Northam, 90km east of Perth.

This location was ideally suited for the proposed work due to its following characteristics.

- It was situated on salt-affected land with a history of traditional agriculture and climatic conditions similar to that of many properties in the Wheatbelt region that could potentially be used to farm black bream.
- Security of tenure at no charge for the duration of the project.
- Abundant reserves of saline water from several sources (ground, river, bore, storage reservoir) with proven track record in sustaining black bream, as demonstrated through field surveys conducted during FRDC Project 97/309.
- Resident landowner willing to oversee and assist in project fieldwork.
- Sufficient area for excavating ponds for research purposes.

- Hard clay soils for pond excavation.
- Proximity to the regional town of Northam provided ease of access to infrastructure and resources.

With the in-kind (financial and advisory) assistance of the property owner, a qualified earthworks engineer, the excavation of research ponds commenced in September 1999. Based on those of the Avondale facility, each experimental pond measured 10 x 10m (100m² surface area) and was 2.0m in depth (Fig. 1). The upper 1.0m of each wall was battered at a 3:1 ratio. At an operating depth of 1.5m, each pond held approximately 75KL of water. A 1m² sump was located in the corner of each pond to facilitate the harvesting of animals after draining.

Following construction, each pond was filled with saline groundwater (salinity range 12-14ppt) and left to condition prior to commencing the first experimental trial in summer 2000 (Fig. 2).



Figure 1. Excavation of a 100m² research pond at Northam showing wall batters.



Figure 2. Completed research ponds prior to commencement of first experimental trial.

Water was recycled between an existing storage reservoir of 2000KL capacity and the research ponds using a petrol transfer water pump (1200L/min). Ponds were protected from runoff by a contour bank located in the upper catchment. Each pond was netted to prevent avian predation using black 6", 12-ply multifilament mesh, suspended over paired wire cables located approximately 1.0m above the water surface.

All of the black bream employed in the field trials were produced by the Aquaculture Development Unit of Challenger TAFE, Fremantle using the culture method as described by Partridge *et al.* (2002).

At the conclusion of each experiment, each pond was drained to approximately 0.5m in depth and the fish removed using a 15m seine net. Ponds were subsequently emptied and any remaining fish were collected from the storage sump. The black bream were anaesthetised and their total length. and wet weight recorded. Note that here and subsequently the total length and wet weight are referred to as T.L. and W.W. and where mean values are given, the accompanying + and – values refer to standard errors.

The mean condition factor of black bream from each treatment at the completion of the trial was calculated using the formula, $C.F. = 100 \times W.W./T.L.^3$, where the T.L. and W.W. are recorded to the nearest 1mm and 1g, respectively.

Food conversion ratios (FCR) were calculated by dividing the total biomass gain in treatment by the total food consumption. Biomass gain was calculated by summing the weights of all fish remaining at the end of the trial, including mortalities, then subtracting from this the initial biomass stocked.

Specific growth rates (SGR) were calculated for each treatment replicate using the equation:

SGR =
$$\frac{\ln(wt_1) - \ln(wt_2)}{t_2 - t_1} \times 100$$

 wt_1 = natural logarithm of the wet weight of fish at stocking (t₁) wt_2 = natural logarithm of the wet weight of fish at harvest (t₂)

The mean water quality parameters, final mean W.W., T.L., survival, FCR, SGR and C.F. of black bream were subjected to Analysis of Variance (ANOVA) to determine whether there were any significant differences among the different treatments. (Note that prior to subjecting these data to ANOVA, Cochran's C test was used to determine whether they were heteroscedastic and, if so, the data were log transformed). When ANOVA showed that there were significant

differences, the Scheffé's *a posteriori* test was used to ascertain where those differences between treatments lay. All statements of statistical significance refer to the 0.05 level. Unless otherwise stated, this statistical approach has been adopted, where appropriate, in all subsequent studies.

For each research trial, ponds were randomly assigned to treatments. Detailed methods for each field trial are provided in the subsequent relevant chapters. A summary of the field trial experiments conducted is provided in Table 1.

Table 1.	FRDC back bream experimental pond trials undertaken at Northam between 1999-
	2002.

Field Trial	Trial Period	Treatments	
Trial A	6 months	Effectiveness of use of floating cages to house juvenile black bream.	
		Growth and survival of juvenile black bream provided with supplementary feed <i>vs</i> no supplementary feed.	
Trial B	4 months	Survival and spawning of yabbies (free range <i>vs</i> PVC hides) in polyculture with black bream.	
		Evaluation of submersed cage culture of yabbies within fish ponds.	
Trial C	6 months	Effectiveness of under- and over-water cover in reducing predation of black bream by avian predators.	
Trial D	9 months	Growth and survival of large juvenile black bream under different feed regimes in ponds.	
		Growth and survival of large juvenile black bream in floating fish cages in two Wheatbelt locations (Northam and Tammin).	

3. POND FIELD TRIALS

3.1 EFFECTIVENESS OF USE OF FLOATING CAGES TO HOUSE JUVENILE BLACK BREAM

3.1.1 INTRODUCTION

One of the problems associated with stocking juvenile black bream, that was identified in FRDC Project 97/309 (Sarre *et al.*, 1999), was the difficulty of monitoring the survival, health and feeding of the fish. This problem is mainly due to the bottom-living habit of black bream and the fact that, in many cases, very large inland water bodies were stocked with only small numbers of fish (<100 per ha). Consequently, the newly-stocked black bream were rarely observed after the initial introduction and property owners were thus unclear if fish had survived or grown.

The aquaculture of fish in cages or net pens is widely practiced as it enables stock health, growth and feeding to be monitored and harvesting to be easily undertaken. Furthermore, cages situated within large bodies of water, *e.g.* oceans, rivers and lakes, may permit higher stocking densities at the semi-intensive level, if passive water exchange through the cage is sufficient to remove solid and nitrogenous waste products.

Ingram *et al.* (2000) demonstrated that the survival and growth of silver perch stocked in floating cages in south-eastern Australia were comparable to those of fish reared in conventional aquaculture ponds. Anecdotal evidence from several property owners in Western Australia, who had used cages to house juvenile black bream, indicated that they were useful in overcoming many of the problems associated with free-range culture in large water bodies. The main aim of this trial was to evaluate the effectiveness of floating fish cages for stocking juvenile black bream at the size they were normally purchased by farmers for stocking their inland water bodies. The specific objectives of the trial were as follows:

- (1) Determine the survival and growth rates of juvenile black bream placed in floating cages.
- (2) Determine if the size of the water body in which the floating cages are located influences survival and growth rates
- (3) Determine the effect of supplementary feed on the survival and growth rates of juvenile black bream reared in saline ponds.

3.1.2 MATERIALS AND METHODS

Fish cage construction

A prototype floating fish cage was constructed and successfully tested for two months prior to the commencement of the trial. Each cage consisted of a 2 x 2m square frame made of 50mm PVC

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pipe. A net "bag", with a drop of 1.5m and comprising 9mm x 9mm knotless mesh, was attached to the frame using plastic cable ties. Plastic trellis (10mm squares) was fixed to the top of the floating frame to prevent fish escaping and to protect fish from avian predators.

Nine research ponds were each stocked with 100 juvenile black bream (Mean T.L. = 74 ± 7.9 mm, mean wet weight = 5.5 ± 0.5 g, n = 100) on 3 February 2000. Using a 3 x 3 randomised design, the treatments were assigned as shown in Table 2.

Table 2.Numbers of ponds, stocking density and types of treatment and feeding regime for
the six month field trial (Effectiveness of caging juvenile black bream).

Number of ponds (fish stock)	Effective stocking density (fish/m ³)	Treatment	Feeding regime
3	1.3	Free range	No supplementary feed
3	1.3	Free range	Supplementary feed provided (artificial)
3	16.5	Cages	Supplementary feed provided (artificial)
Saline Lake*	16.5	Cages x 3	Supplementary feed provided (artificial)

*As an additional treatment, 100 juvenile black bream were also stocked in each of three floating cages placed in a large saline lake adjacent to the research ponds and fed using the same regime as the fish in cages in ponds.

Supplementary feed was provided on alternate days at the rate of 2% dry weight feed per total W.W. of fish, *i.e.* 1% per day. The feed was a locally produced, commercial, sinking, 2.5 mm pellet (Glen Forest Stock Feed: Black Bream Grower, 48% protein, 10.5% fat).

Temperature, salinity, pH and dissolved oxygen in each pond and the saline lake were recorded during the early evening (*ca* 6pm) at two weekly intervals. The water level in each pond was maintained at a constant level through periodic "topping up" from the nearby storage reservoir.

N.B. The C.F. for black bream, of comparable T.L. to the trial fish, which were collected by seine netting from the Swan River Estuary during August in 1993, 1994 and 1995 (see Sarre and Potter, 1999) is also presented as a general comparison only. These data have not been included in the statistical analysis.

3.1.3 RESULTS

Water quality parameters

There were no significant differences in water temperature, salinity, dissolved oxygen or pH between the nine ponds during the trial. A summary of the water quality parameters is presented in Table 3. The mean salinity in the ponds gradually increased over the six month trial period

from 9.0ppt at the commencement to 13.9ppt at completion (Fig. 3). In contrast, mean water temperatures decreased from 28.1°C to 14.1°C over the trial period (Fig. 3).

	Researc	ch Ponds	Lake	
Parameter	Mean	Range	Mean	Range
Temperature (°C)	20.9	13.8 - 29.6	18.8	14.3 – 26.4
Salinity (ppt)	11.4	5.4 - 14.8	14.4	13.5 – 16.1
Dissolved Oxygen (mg/L)	6.8	4.7 – 10.7	7.7	6.8 - 10.4
РН	8.6	8.3 – 9.1	8.9	8.8 - 9.0

Table 3.Summary of water quality variables recorded at two weekly intervals at the nine
Northam research ponds and adjacent lake from February to June 2000.



Figure 3. Mean water temperatures and salinities ± 1 SE in the nine research ponds at two weekly intervals between February and June 2000.

Growth and survival

The mean percentage survival of black bream was greater than 90% in all treatments and ranged from 91% for fed fish in cages in lakes to 98% for free range fed fish in ponds (Fig. 4). ANOVA showed that the mean percentage survival among the four different treatments were not significantly different.

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Figure 4. Mean percentage survival +1SE of juvenile black bream at the completion of each of four trial treatments.

The final mean W.W. and T.L. of black bream among the various treatments are shown in Figs 5a & b. ANOVA demonstrated that the final mean W.W.s, T.L.s, condition factors and FCRs of fish subjected to the four treatments were significantly different. The final W.W.s and T.L.s of free range fed fish and free range unfed fish were significantly greater and lower, respectively, than those of fish in the pond and lake cages (Figs 5a & b). There were no significant differences between these variables for caged black bream in research ponds and the adjacent lake.



Figure 5. Mean wet weight (a) and total length (b) +1SE of juvenile black bream at the completion of each of the four trial treatments. Columns sharing the same letter are not significantly different (p > 0.05).

The mean condition factor (C.F.) of unfed, free range fish was significantly less than those for all other treatments, while the C.F. for fed caged fish in ponds was significantly greater (Table 4). The C.F. for wild stock black bream, collected from the Swan River Estuary, was greater than those of fish in all experimental treatments (Table 4). The FCR of fed, free range fish was significantly less than that for the fish fed in both caged treatments (Table 4).

The mean SGR of unfed, free range fish and fed, free range fish was significantly lower and higher respectively, than both cage treatments (Table 4).

Table 4.Mean condition factor (C.F.), food conversion ratio (FCR) and specific growth
rate (SGR) of black bream from each treatment at trial completion. N.B. The C.F.
for black bream collected from the Swan River Estuary is provided for
comparison. Values sharing the same letter are not significantly different (p > 0.05).

Treatment	Condition Factor	FCR	SGR
Free range (unfed):	1.41 ^a	N.A.	0.41 ^d
Free range (fed):	1.70^{b}	1.78^{a}	0.92^{f}
Pond cages (fed):	1.63 ^c	2.48^{b}	0.86^{e}
Lake cages (fed):	1.59 ^c	2.49 ^b	0.86^{e}
Wild population (Swan River) (at similar size/time of capture)	1.85	N.A.	N.A.

3.1.4 DISCUSSION

The results strongly indicate that the stocking of juvenile black bream in cages does not have a significantly adverse effect on their survival relative to free range fish. While the sizes attained by caged fish were marginally lower than those of free range fish in the experimental ponds, the advantages of cage culture, *e.g.* ease of harvest and monitoring of health, make this relatively inexpensive technique beneficial. It will also be useful for farmers who stock juvenile black bream in water bodies that are large or in areas where avian predators are common.

There were no significant differences between either the survival or the growth rate of fish housed in cages located in the research ponds as opposed to the lake. However, it should be recognised that the larger water bodies are likely to be less prone to a fluctuations in water quality, particularly if stocking densities and associated feeding rates are higher and/or cover a longer grow-out period. Ingram *et al.* (2000) noted that survival rates of silver perch *Bidyanus*

bidyanus grown in cages, that were located within small groundwater tanks (35m³), were low due to poor water quality. In the current trial, the cages located within the ponds had more extensive growths of algae on the mesh, which would, over time, reduce the amount of water exchanged through the cage and thus lead to a decline in water quality.

The significantly greater mean T.L., W.W. and SGR of the free-range black bream when they were provided with supplementary feed indicates that, while naturally occurring feed within the ponds was adequate for survival, supplementary feeding is crucial for optimising the growth of juvenile black bream. The advantages of supplementary feed are also reflected in the presence of the higher condition factor (C.F.) recorded for black bream in this treatment than in those where no supplementary feed was provided. The lower C.F. of black bream housed in cages suggests that less natural feed is available since they are unable to forage extensively for feed and thus have to rely on that which moves through the cage mesh. The fact that the C.F. of similarly sized, wild caught black bream from the Swan River Estuary was greater than that of fish in all treatments indicates that the extensive variety of natural foods ingested by black bream in its natural environment (see Sarre *et al.*, 2000) meets more adequately the nutritional requirements of the species. Furthermore, the relatively high FCR recorded for the black bream in cages suggests there is a need to improve the commercially available diets for this species.

3.2 EVALUATION OF THE POLYCULTURE OF BLACK BREAM AND YABBIES

3.2.1 INTRODUCTION

The yabby *Cherax destructor albidus* (Austin, 1996; Campbell *et al.*, 1994) is one of the most common of the large crustaceans that has been introduced into farm dams in south-western Australia. A closely related species will reproduce at salinities up to 8ppt Mills and Geddes (1980) and survive as adults in salinities up to 22ppt (Morrissy *et al.*, 1990). This raised the possibility that yabbies could be used either as a supplementary food source for black bream or as a candidate for polyculture in water bodies with low to moderate salinities.

Data provided in FRDC Project 97/309 (Sarre *et al*, 1999) indicated that black bream were stocked in polyculture with yabbies in *ca* 70% of all the 151 water bodies surveyed. Discussions with property owners indicated that many of these owners were under the impression that the black bream and yabbies would co-exist, while others assumed that yabbies would provide a self-sustaining food source for their black bream, which would thus not require any additional supplementary feed. However, preliminary data gathered during FRDC Project 97/309 suggested that large black bream, in particular, were capable of rapidly reducing the size of a population of yabbies when in free-range polyculture. This conclusion was based on the fact that sampling conducted in water bodies that contained large numbers of yabbies after the introduction of black bream. Furthermore, analyses of the diet of black bream in these systems often identified yabbies in their stomach contents.

Studies on the aquaculture of silver perch (*Bidyanus bidyanus*) and marron in Western Australia provided evidence that the provision of protection in the form of weed or artificial hides might prevent marron stocks from being totally depleted when stocked in polyculture with silver perch (Whisson, 1995). There have not yet been any studies to determine whether this also applies to black bream and yabby polyculture.

The current trial was thus designed with the following objectives:

(1) Determine to what extent a known number of large black bream will deplete the abundance of a known number of large yabbies when stocked in free-range polyculture over a three-month period.

- (2) Determine whether large yabbies, which are protected from predation by large black bream, can survive and reproduce to provide a substantial source of food for large black bream.
- (3) Determine whether the provision of yabby hides significantly reduces the level of predation on yabbies by large black bream.

3.2.2 MATERIALS AND METHODS

Twelve $100m^2$ S.A. research ponds at Northam, that were netted to prevent avian predation, were each stocked with yabbies and/or bream using a 4 x 3 randomised design as shown in Table 5.

Table 5.Numbers of ponds and types of treatment used for the black bream and yabby
polyculture trial conducted at Northam.

	Treatment	
Number of ponds	Yabbies (stocking density #/m ²)	Bream
3	Free range (1)	Free range
3	Caged (50)	Free range
3	PVC hides (1)	Free range
3	Yabbies (1)	None

In one triplicate set of ponds, a total of 25 yabby hides were randomly distributed over the floor of each pond. The "hides" consisted of 300mm lengths of PVC pipe with a diameter of 65mm (Fig. 6a). In a second triplicate set of ponds, yabbies were enclosed within a submersed cage placed in the centre of the pond floor. Each cage was constructed of wire mesh (15mm squares) and measured 1m (length) x 1m (width) x 0.4m (height) (see Fig. 6b).



Figure 6. (a) Partially drained research pond showing PVC yabby hides and (b) the wire cages used to house yabbies on the floor of the research ponds.

Ponds were filled with low salinity water (range 4 - 5ppt) and the yabbies and black bream were both fed on alternate days. A length of PVC pipe was used to direct feed into the submerged cages to ensure that the feed reached the caged yabbies.

A total of 100 adult yabbies $(35 \pm 4.2g)$ were stocked in each pond in mid-September, representing a stocking density of 1 yabby m⁻², and 20 large back bream (T.L. = 250-300mm) were introduced five weeks later. One triplicate set of ponds contained only free-range yabbies and thus acted as a control. A total of 50 yabbies were also stocked into each of three cages located in an additional three ponds representing a stocking density of 50 yabbies m⁻².

Water temperatures and salinities were recorded at two weekly intervals.

At the completion of the trial, all 12 ponds were drained and the numbers of black bream and adult and juvenile yabbies were recorded. The percentage of yabbies found within the PVC hides and the numbers both of yabby burrows in each pond and of adult yabbies carrying eggs and/or juveniles were also determined. The stomach contents of two adult black bream from each pond were analysed to determine whether either adult or juvenile yabbies had been consumed.

N.B. Since reserves of low salinity water became limited six weeks after the introduction of the black bream and the salinities within several of the research ponds had reached levels considered to exceed the upper limit for the survival of juvenile yabbies, *i.e.* 10ppt, the trial was concluded prior to the end of its intended 12 week duration.

3.2.3 RESULTS

One week prior to the completion of the trial, all yabbies in one of the control ponds died when the water level became very low and the salinity exceeded 18ppt. Data from this pond were excluded from analyses.

The mean water temperature and salinity of the research ponds increased from 19.6°C and 4.3ppt at trial commencement to 23.8 °C and 9.8ppt at trial completion (Table 6).

Table 6.Summary of initial and final water temperatures and salinities recorded from
eleven Northam research ponds over the yabby/black bream polyculture trial
conducted between mid-September and late November 2000.

	Initial		Final	
Parameter	Mean Range		Mean	Range
Temperature (°C)	19.6	19.0 – 19.8	23.8	25.4 - 24.1
Salinity (ppt)	4.3	3.8 - 5.1	9.5	6.8 – 10.4

ANOVA showed that survival varied significantly among treatments. The mean percentage survival of yabbies in the control ponds (free-range yabbies only) at harvest was 91.5%, which was significantly greater than in all other treatments (Fig. 7). The lowest mean survival of 20.3% was recorded in ponds containing yabbies and bream in free-range polyculture (Fig. 7). A large number of yabby body parts, consisting predominately of claws, were observed on the floor of drained ponds in the free-range polyculture and polyculture/hide treatments.



Figure 7. Mean percentage survival +1SE of adult yabbies in each treatment. Columns sharing the same letter are not significantly different (p > 0.05).

The mean percentage number of PVC hides ± 1 SE containing at least one adult yabby at harvest was 78.7 $\pm 7.1\%$. On only one occasion were two yabbies found in the same PVC hide.

There were significant differences between the mean number of yabby burrows per pond at the completion of the trial. The mean number of burrows was significantly greater in ponds containing yabbies and black bream in free-range polyculture, *i.e.* 11.0 per pond compared to the control and free-range polyculture ponds *i.e.* 1.5 and 3.5 per pond, respectively (Table 7).

Treatment	Mean number of burrows ±1 SE
Free-range yabbies (control)	1.5 ± 0.41
Bream + caged yabbies	Not applicable
Free-range polyculture	$11.0 \pm 2.52^*$
Free-range polyculture/hides	3.5 ± 0.34

Table 7.Mean number of yabby burrows per pond found in four different treatments.
(* indicates a significant difference)

The mean percentage number of adult yabbies that were berried or carrying juveniles was significantly lower in the caged treatment, with only a single berried individual recorded, than in all other treatments. The highest mean number of such individuals was the 14% recorded in the control ponds containing free-range yabbies only (Fig. 8).



Figure 8. Mean percentage number +1SE of berried/juvenile bearing adult yabbies in each treatment. Columns sharing the same letter are not significantly different (p > 0.05).

Juvenile yabbies were found in 5 of the 12 research ponds.

The estimated number of juvenile yabbies found in particular ponds and their location in those ponds at harvest are shown in Table 8. All juvenile yabbies measured <8mm T.L.

Table 8.Estimated number of juvenile yabbies and their location within the ponds when
found.

Treatment	Estimated # of juveniles (see below)	Location of juvenile yabbies within pond
Pond 4 (Free-range yabbies)	****	Pond floor/walls
Pond 7 (Free-range yabbies)	*	Pond floor/walls
Pond 9 (Polyculture with hides)	***	Hides + burrows
Pond 12 (Polyculture with hides)	**	Hides + burrows
Pond 11 (Free range polyculture)	****	Burrows only
$(\mathbf{V}_{12}, \mathbf{v}_{11}, 1, 1, \mathbf{v}_{12}, \mathbf{v}_{11}, \mathbf{v}_{12}, \mathbf{v}_{1$	100 *** 100 500 ****	E 00)

(Key: * = 1-10, ** = 10-100, *** = 100-500, **** = 500+)

The stomach contents of black bream collected from ponds containing the caged yabbies contained only fish feed. The remains of adult yabbies were found in the stomachs of two and

three out of the six black bream collected from the free-range polyculture and the polyculture/hides treatments, respectively. No juvenile yabbies were found in the stomach contents of black bream.

3.2.4 DISCUSSION

Since adult yabbies were found in the stomach contents of subsamples of large black bream taken from polyculture ponds, large black bream will feed on adult yabbies when they are stocked together. Furthermore, the fact that the survival of yabbies in polyculture ponds (<20%) was significantly lower than in control ponds where survival of yabbies was very high (>91%) provides strong circumstantial evidence that the level of predation can be high. In the case of one polyculture pond, the black bream had apparently eaten all but two of the 100 yabbies that were stocked prior to the six week trial.

Although these data were not statistically significant, the differences between the means were so marked that they suggested that the level of predation of black bream on yabbies was reduced by the provision of PVC hides within polyculture ponds. The survival of yabbies in polyculture ponds with hides, *i.e.* 78%, was far higher than those stocked in ponds without hides, *i.e.* 20%. Since only 25 hides were provided in each pond and, in all but one case, only a single yabby inhabited each hide, the provision of additional hides, *i.e.* one per yabby stocked, may have resulted in a greater number of yabbies being able to avoid predation by the black bream.

The greater number of yabby burrows in free-range polyculture than in polyculture/hide ponds probably reflects a predator avoidance response in the absence of available hides. This view is supported by the fact that very few burrows were found in the control ponds containing only yabbies and no black bream.

The high number of yabby mortalities recorded in the submerged cages demonstrates that the use of such cages is not an option for maintaining yabbies. This may be due to the high stocking density of yabbies created within the cage and an associated increase in cannibalism, lack of food and/or a decline in water quality parameters such as dissolved oxygen. Both of these factors have been shown to have negative effects on the survival, growth and spawning rates of yabbies (Lawrence *et al.*, 1998). This view is supported by the fact that <1% of the yabbies in cages were berried, compared with 14% of yabbies in control ponds. Furthermore, there was no evidence of juvenile yabbies within the ponds outside the cages or in the stomach contents of the free-range black bream within the same ponds.

Juvenile yabbies were recorded in five research ponds, even though the salinities in these ponds had increased to levels considered to be at the upper limit of those in which a related species of yabby were shown to reproduce, *i.e.* 8ppt (Mills and Geddes, 1980). The juvenile yabbies found in polyculture ponds were all located either in hides or within burrows, whereas they were present on the pond floors and walls of the control ponds. The absence of these juveniles in the stomachs of black bream sampled from the polyculture ponds could be a result of their small size, *i.e.* <8 mm, and/or because they were still attached to the females and were only released during the harvesting of the ponds.

In summary, the results strongly suggest that adult yabbies could provide a semi-sustainable, inpond, food source for black bream in low salinity water bodies, providing that the adult yabbies are prevented from being preyed upon by black bream through the provision of a sufficient number of hides. In most cases, property owners will initially stock water bodies with juvenile black bream. Since small black bream are only likely to consume small juvenile yabbies, yabby/bream polyculture may be beneficial at least in the short term.

3.3 EFFECTIVENESS OF INTRODUCING UNDER AND ABOVE WATER COVER TO REDUCE THE PREDATION OF BLACK BREAM BY CORMORANTS

3.3.1 INTRODUCTION

The vast majority of water bodies surveyed during FRDC Project 97/309 did not contain any protection for black bream from avian predation other than that which occurred naturally within the water body. Indeed, comprehensive protection from avian predators was only provided in two water bodies, this being achieved through placing nets over the entire water surface. In almost all cases, cormorants (*Phalacrocorax* spp.) were the major predator of stocked black bream, particularly when the water bodies were shallow and clear and/or devoid of cover. This was also the case in farm dams in New South Wales that had been stocked with silver perch during a study by Barlow (1982).

Large numbers of cormorants were frequently observed on a large adjacent lake at the Northam field trial station during the summer and winter. This high abundance can be attributed to the large number of roosting sites along the banks of the nearby Mortlock River. These cormorants were often observed feeding on small black bream that had been stocked in the lake. Confirmation that black bream are ingested by cormorants was provided by examining the stomach contents of cormorants that had died after becoming entangled in the netting that covered the research ponds (Fig. 9).



Figure 9. Numerous small fish, including juvenile black bream, removed from the stomach of a cormorant that had become entangled in pond bird netting.

The current trial was designed to test three types of under- and over-water cover that could be used to reduce predation of juvenile black bream by birds and specifically cormorants. The types of cover used were chosen because they could be easily sourced from most farms and/or were inexpensive to purchase. They included plastic flag bunting (over water), coils of ring-lock fencing (under water) and tree branches (under water). Sarre et al. Factors required for the successful aquaculture of black bream in inland water bodies

3.3.2 MATERIALS AND METHODS

Twelve $100m^2$ research ponds at Northam were each stocked with 100 juvenile black bream (mean T.L. *ca* 55mm) in late February 2001 and treatments allocated using a 4 x 3 randomised design as shown in Table 9.

Table 9.Numbers of ponds and the types of under- and over-water cover used for the black
bream predation protection trial conducted at Northam.

Number of ponds	Type of Cover	
3	Plastic bunting	
3	Wire coils	
3	Branches	
3	Nil (Control)	

The plastic bunting was criss-crossed about 0.5m above the water surface, while the four wire coils (each 1m high, 0.5m diameter, 150mm squares) were placed on the floor of each of the ponds. The branches were placed at one edge of the ponds and covered about one quarter of the floor area (see Fig. 10 a-c).



Figure 10. The three types of over- and under-water cover used for the predation trial at Northam. (a) Plastic bunting, (b) ring-lock wire coils and (c) tree branches.

The black bream in each pond were fed on alternate days (equivalent of 1% per day) and the ponds inspected daily to determine whether any non-predation related mortalities had occurred. All ponds were drained in late July 2001 (6 months after stocking the black bream) and the number of surviving black bream recorded.

3.3.3 RESULTS

Cormorant droppings were observed around the research ponds during the final month of the trial. On two occasions, cormorants were also observed flying away from the vicinity of the research ponds.

The mean survival of juvenile black bream was high (>87%) in all treatments (Fig. 11). No black bream mortalities were observed during the trial period and nor were any observed after the ponds had been drained at trial harvest. ANOVA showed that there was a significant difference between treatments. The mean survival of black bream in ponds over which plastic bunting was placed (87.7%) was significantly lower than in all other ponds, including the control ponds, where no form of protection was used (Fig. 11).



Figure 11. Mean percentage survival +1SE of black bream in ponds containing the three types of avian predation protection and the control ponds after the six month trial. (* = significant difference)

The final mean W.W. and T.L. of black bream in each of the treatments, which ranged from 23.4 to 24.4g and 112.7 to 115.4mm, respectively, were not significantly different.

3.3.4 DISCUSSION

Since a number of cormorants were observed in the ponds over the six month trial period, the mean survival of juvenile black bream was unexpectedly high in all treatments. The fact that no black bream mortalities were observed either during the trial or after the ponds had been harvested provides strong circumstantial evidence that any losses of black bream were likely to have been be due to predation, particularly since the survival of juvenile black bream in netted ponds in all other trials during this project was typically close to 100%. Since cormorants showed no signs of frequenting the pools until the final month of the trial, it may have taken the birds some time to become accustomed to the area. Any bird-related predation of the black bream may

thus have occurred only over a restricted time period, which would account for the small number of fish that was taken from the trial ponds. Furthermore, cormorants also had access to the adjacent lake, which supported a much larger population of black bream than the trial ponds and within which cormorants were regularly observed feeding during the trial period.

Two factors may have contributed to the significantly lower level of black bream survival in the ponds covered with above water plastic bunting. (1) The cormorants may have been using the wire supports for the plastic bunting as roosts before entering the water and (2) the multi-coloured flags may have been attracting the birds to these ponds.

The fact that only a single black bream was apparently taken from the ponds containing the wire coils strongly indicates that coils provide an effective anti-predation cover for ponds in which juvenile black bream are susceptible to avian predators. The black bream in this trial were almost always observed within the coils and the coils could easily be removed with attached ropes when required.

Nonetheless, over-water netting of ponds is likely to be the only 100% effective method to prevent stock losses through avian predation. Records of the size of the black bream taken from the stomachs of cormorants during the past five years of our research indicates that fish greater than 150mm T.L. are unlikely to be taken by cormorants. This probably reflects an increased tendency for black bream to live around in-water structures as they increase in size and/or that their physical size and the development of sharp fin spines prevent their capture.

3.4 DETERMINATION OF THE OPTIMAL FEEDING REGIME FOR THE GROW OUT OF CAGED AND FREE-RANGE BLACK BREAM IN TWO DIFFERENT WATER BODIES

3.4.1 INTRODUCTION

Data provided in FRDC Project 97/309 demonstrated that black bream in the majority (85%) of water bodies surveyed were not provided with any supplementary feed. Property owners were generally under the impression that natural food within these water bodies would be sufficient for this species. However, preliminary data from our previous study, and that provided in Section 3.5 of the current study, demonstrate that a large proportion of the natural prey in inland water bodies is small and not typically ingested by large black bream. Supplementary feeding is therefore essential to achieve the optimal growth of these larger fish. Since black bream aquaculture is generally a secondary farming activity or a hobby for the majority of property owners, and because water bodies are often located some distance from their residences, it is important to determine which supplementary feeding regime produces optimal growth and survival of stocked black bream. Furthermore, while cage culture of juvenile black bream does not appear to inhibit growth (see Section 3.1), it is not known whether this is also the case for larger black bream.

Laboratory, tank-based feeding trials conducted during the current project have re-identified which of the commercially available feeds produced the best growth and FCR in black bream (See section 4.1). The purpose of this field trial is to use this specific feed type as a supplement in field conditions and determine which of two feeding regimes, *i.e.* daily *vs* alternate days, results in the optimal survival, growth and FCR's of large black bream. Both free-range pond and cage culture trials will be conducted, with the latter replicated in two water bodies that differ markedly in their physico-chemical characteristics.

3.4.2 MATERIALS AND METHODS

A total of 50 black bream (mean T.L. and WW of 138.6 ± 1.0 mm and 66 ± 1.4 g, respectively) were stocked in each of nine $100m^2$ research ponds and each of six 2 m² floating fish cages at Northam in mid-August 2001. A further 50 fish were stocked in each of six $2m^2$ floating cages at Tammin. The cages at Northam were placed in approximately 5m water depth in an approximately 5 acre static water lake and tethered to a floating pontoon for access when feeding (Fig. 12a). The cages at Tammin were placed in a drainage channel which was about 1.5m deep and contained very slow flowing water *ca* 1m min⁻². (Fig. 12b). Water clarity in the drain at Tammin was high (>1.5m secchi depth), but relatively low (< 300mm secchi depth) in the lake at Northam due to tannin stain and algal turbidity.



Figure 12. Floating fish cages used for the black bream grow-out trial in (a) a large lake at Northam and (b) a drainage channel at Tammin.

The black bream in each cage were fed the grower diet identified in the laboratory feeding trials (Section 4.1) that produced the best growth of black bream, *i.e.* Pivot Salmon (sinking) feed (45/25% protein/lipid composition). Feed was initially provided at the rate of 2% dry weight feed per total W.W. of fish. The feeding regime used for each treatment in each location is shown in Table 10).

Table 10.Feeding regime for each treatment used in the black bream grow-out trial in cages
and ponds in Northam and cages in Tammin between August 2001 and April
2002.

Water body	Number of treatments	Location	Supplementary Feeding Frequency (Feed Rate/Day)	Initial stocking density (kg/m ³)	Feed Rate (%/day)
100m ²	3	Northam	Daily	0.05	2
ponds					
$100m^2$	3	66	Alternate days*	"	1
ponds					
$100m^2$	3	"	No Sup. Feed	"	NA
ponds					
2m x 2m	3	"	Daily	0.6	2
cages					
2m x 2m	3	"	Alternate days	"	1
cages					
2m x 2m	3	Tammin	Daily	"	2
cages					
2m x 2m	3	٠٠	Alternate days	"	1
cages					

Twelve black bream were subsampled from each treatment in each location at two monthly intervals. Fish were collected from the ponds using a seine net and from the cages using a long-

handled scoop net. Each fish was anesthetised, weighed and measured to the nearest 1g and 1mm, respectively, and then re-released. Feeding rates for each treatment were recalculated and adjusted accordingly after each subsample. To enable the results for the cage treatments at Northam and Tammin to be compared directly, the same amount of feed was used at the two locations. The amount of feed used was thus based on 2% of the highest mean W.W. of black bream at either locality after each subsample.

The temperature and salinity of each pond and the saline lake were recorded monthly. The water level in each pond was maintained at a constant level through periodic "topping up" from the adjacent lake.

All of the black bream in each pond and cage were harvested after nine months. After draining each pond, its floor was photographed to provide a visual record that could be used to estimate the level of eutrophication reached at the end of the trial period.

3.4.3 RESULTS

Water quality parameters

Research Ponds

Analysis of variance (ANOVA) demonstrated that neither water temperature nor salinity differed significantly among the nine research ponds during the trial. Mean salinity in the ponds gradually increased over the trial period from 15.8ppt in mid-August (trial commencement) to 38.8ppt at completion (Fig. 13). Although the mean water temperature also increased, rising from 16.8°C in mid-August to 27.0°C in January 2002, it subsequently decreased to 22.0°C in April at the completion of the trial (Fig. 13).

Three months after commencing the trial, algal blooms were observed in the research ponds that were receiving supplementary feed, and particularly in those in which food was being added daily. Daytime dissolved oxygen levels in these ponds increased rapidly and ranged from 10.6 - 13.0mg/L, while night-time levels fell to < 3.0mg/L. To avoid further eutrophication of the ponds and potential fish deaths, feeding was suspended for three days and thereafter the amount of feed provided to the fish reduced to 1% dry weight feed per total W.W. of fish. Turbidity within the fed ponds remained relatively constant at about 300mm secchi depth for the remainder of the trial.



Figure 13. Mean monthly salinities and water temperatures ± 1 SE in the nine research ponds at Northam between August 2001 and April 2002.

Cage localities

Water temperature in the irrigation channel at Tammin increased from 15.5° C in August to 27.3° C in January, before declining to 19.9° C in April (Fig. 14), while in the lake at Northam it increased from 16.5° C in August to 28.4° C in January and then decreased to 21.5° C in April. In corresponding months, the difference in water temperature between the two localities was always $<1.5^{\circ}$ C, except in February and October when the temperature in the lake at Northam was 3.4 and 3.1° C higher, respectively (Fig. 14).

Salinity in the irrigation channel increased from 7.0ppt in August to 22.4ppt in March, before falling precipitously to 6.3ppt in April (Fig. 14). At Northam, the salinity increased gradually from 19.5ppt in August to 29.1ppt in March, before decreasing to 21.5 in April (Fig. 14). Salinity in the irrigation channel was significantly lower than in the Northam lake in all months except March.



Figure 14. Monthly salinities and water temperatures in the irrigation channel at Tammin and the lake at Northam in which research cages were placed between August 2001 and April 2002.

As was the case in the Northam research ponds, the turbidity in the lake at Northam, due to the production of algae, also increased during the corresponding period. However, since the dissolved oxygen concentrations at night did not fall below 4.0mgL^{-1} , it was not considered necessary to alter the amount of feed. In contrast to the situation in the research ponds at Northam, the turbidity of the water in the Tammin irrigation channel remained at >1.5m secchi depth throughout the trial.

Pond floor sediment and cage condition

Observation of the pond floors at the completion of the trials showed that the levels of eutrophication, as reflected in the form of unoxidised black mud, were greatest in the ponds that were fed daily, followed by those in which feed was supplied on alternate days (Fig. 15a). In contrast, the floor of unfed ponds contained virtually no evidence of sediment eutrophication (Fig. 15b).

The cages in the lake at Northam became heavily fouled with algae (see insert Fig. 15a) and had to be cleaned on two occasions to maintain water exchange through the mesh. The cages at Tammin showed only minor signs of fouling and did not require cleaning.



Figure 15. Photographs of pond floor sediment at the completion of the trial, with an example of (a) high-level sediment eutrophication in a pond supplied with feed and (b) a negligible level in a pond not supplied with feed. Insert shows a cage taken from the lake at Northam which was subject to extensive algal fouling.

Growth

Northam Pond Trials

During the eight month trial, the mean W.W. of black bream fed no supplementary food decreased from an initial weight of 60g at stocking in August 2001 to 50.5g in April 2002 (Fig. 16a). In contrast, the mean W.W.s of fish in the two treatments in which supplementary feed was supplied increased between each successive subsample, except in February 2002, when the mean W.W. of daily fed black bream was marginally lower than that recorded in the previous subsample in December 2001 (Fig. 16a). The mean W.W.s of fish in the alternate and daily fed groups in each subsample, and at the completion of the trial, were not significantly different, *i.e.* p > 0.05 (Fig. 16a).

The mean T.L. of unfed black bream increased only marginally during the trial, rising from 138.6mm in August 2001 to 143.2mm at its completion in April 2002 (Fig. 16b). Mean lengths of the black bream fed on alternate days and each day increased from 138mm in August 2001, at the commencement of the trial, to maximum values of 169.6 and 169.3mm, respectively, in April 2002 (Fig. 16b). In each subsample, the mean T.L.s of fish fed on alternate days and each day differed by <2.5mm and were not significantly different (p > 0.05).



Figure 16. (a) Mean W.W. (b) and mean T.L. ± 1 SE of black bream grown in research ponds at Northam under different feeding regimes and recorded at two monthly intervals between August 2001 and April 2002. Columns sharing the same letter are not significantly different (p > 0.05).

Cage Trials

In each of the two-monthly subsamples, the mean W.W.s and T.L.s of black bream fed both on alternate days and daily were significantly greater (p < 0.05) in the cages at Tammin than in those at Northam (Figs 17a,b). In the case of the Tammin black bream, there was virtually no difference in either mean between the two feeding treatments throughout the trial, with the greatest difference in weight and length being only 3.5g and 3mm, respectively (Figs 17a,b). While the mean weights and lengths of the black bream fed daily at Northam were usually greater than those fed on alternate days, this difference was also relatively small. The greatest difference was recorded in February, when the mean weights and lengths differed by 11g and 8mm, respectively (Figs 17a,b).

ANOVA demonstrated that the final mean lengths and weights attained by the black bream fed both on alternate days and daily did not differ significantly at either Tammin or Northam. However, the mean lengths and weights of black bream in both feeding treatments in Tammin were significantly greater than those in Northam (p < 0.05) (Figs 17a,b).


Figure 17. (a) Mean W.W.s and (b) mean T.L.s +1 SE of black bream grown under different feeding regimes in floating cages in the lake at Northam and in the irrigation channel at Tammin and recorded at two monthly intervals between August 2001 and April 2002. Columns sharing the same letter are not significantly different (p > 0.05).

Survival rates and feed conversion ratios

Survival of black bream fed on alternate days or daily in cages in the Tammin channel, Northam lake and research ponds were very high, *i.e.* between 96 and 100%, and ranged from $80 \pm 10.5\%$ to 100%, respectively (Table 11). The survival of black bream provided with no supplementary feed was significantly lower than in each of those other treatments (Table 11).

Food conversion ratios ranged widely from 1.84 ± 0.07 to 8.35 ± 1.17 (Table 11). The FCR for black bream fed daily was significantly greater than that of fish fed on alternate days in the case of both the cages at Tammin and the research ponds at Northam, but not in the cages at Northam (Table 11). However, the difference was close to significant in the last case. There was no significant difference between the two lowest FCRs recorded for fish fed on alternate days in the

cages at Tammin and the ponds at Northam. The FCR of black bream that were fed daily in the cages at Northam (8.35 ± 1.17) was significantly higher for both feed treatments in both the Tammin cages and Northam ponds (Table 11).

Specific growth rates for fed treatments ranged from 0.17 ± 0.02 to 0.31 ± 0.01 for fish fed daily in both the cages and ponds in Northam and fish fed daily in cages at Tammin, respectively (Table 11). The unfed fish in the Northam ponds had by far the lowest SGR of -0.06 ± 0.06 . Within each treatment there were no significant differences in the SGR of fish fed daily and on alternate days (Table 11).

Cost of production ranged from a minimum $0.21c \text{ g}^{-1}$ for fish fed on alternate days in the cages at Tammin to a maximum 0.96c g⁻¹ for fish fed daily in the cages at Northam (Table 11). In all cases the cost of production was far lower for fish fed on alternate days than for those fed daily.

Table 11.Survival rates, food conversion ratios (FCRs), specific growth rates (SGRs) and
cost of production of black bream cultured in cages at Northam and Tammin and
in research ponds using different supplementary feeding regimes between August
2001 and April 2002. Values sharing the same letter are not significantly different
(p > 0.05).

Feed	Cages			Cages			Research Ponds					
Regime	((Tammin channel)			(Northam lake)			(Northam)				
. 8	Survival	FCR	SGR	Cost/g	Survival	FCR	SGR	Cost/g	Survival	FCR	SGR	Cost/g
				weight				weight				weight
				gain (¢)				gain (¢)				gain (¢)
Alt.	$96.0 \pm$	$1.84 \pm$	$0.30 \pm$	$0.21 \pm$	100	5.21 ±	$0.18 \pm$	$0.60 \pm$	$98.6 \pm$	$2.52 \pm$	$0.18 \pm$	0.29 ±
	1.1	0.07 ^d	0.01 ^g	0.01		$0.02^{c,e,f}$	0.02 ^h	0.01	1.3 ^a	0.17 ^d	0.01 ^h	0.02
Daily	100	3.78 ±	0.31 ±	0.43 ±	100	8.35 ±	0.17 ±	0.96 ±	97.3 ±	4.73 ±	0.17 ±	0.54 ±
		0.08 ^e	0.01 ^g	0.01		1.17 ^c	0.02 ^h	0.13	2.6 ^a	0.70 ^{e,f}	0.02^{h}	0.08
Nil	NA	NA	NA	NA	NA	NA	NA	NA	$80.0 \pm$	NA	-0.06 ±	NA
									10.5 ^b		0.06 ⁱ	

Evidence of successful spawning

When subsampling the research ponds for black bream in February, a number of small juvenile black bream (mean T.L. = 40.1 ± 5.2 mm) were collected from three of these ponds (Fig. 18) in which feed was supplied on alternate days. The number of juvenile black bream in these three ponds at the completion of the trial ranged from 65 to 271 per pond.



Figure 18. Juvenile black bream found in one of three research ponds in Northam at the completion of the trial in April 2002.

3.4.4 DISCUSSION

The results produced during this trial lead to the conclusion that supplementary feed is essential for optimal survival and growth of large black bream in aquaculture. This conclusion is based on the fact that the mean W.W. of large black bream in research ponds not provided with supplementary feed declined over the trial period. Furthermore, the mean survival rate of these fish, *i.e.* 80%, was significantly less than in treatments in which the fish were fed on either alternate days or daily and in which the mean survival rate was either close to or equal to 100%. Thus, any natural feed in these ponds was inadequate for optimal survival of large black bream.

Since there were no significant differences in either the mean W.W., T.L. or SGR of black bream fed daily or on alternate days, the latter feeding regime is clearly the most desirable option. The high level of "anaerobic mud" in the ponds supplied with food daily and the consistently greater FCRs of black bream fed daily provide strong evidence that not all of the food was consumed by the black bream when that food was provided daily. The feed rate of 2%/day could therefore represent over-feeding for this species. This wastage is both uneconomic, as indicated by the far lower cost of production for bream fed on alternate days, and a major contributor to poor water quality and an associated suboptimal performance by black bream. Furthermore, since juvenile black bream were only recorded from ponds where fish were fed on alternate days, it is possible that excessive feed and poor water quality also had a detrimental impact on spawning success of black bream (also see section 3.5).

The increasing levels of eutrophication that occurred in both the research ponds and the lake at Northam, and the algal fouling of lake cages, indicate that, even at relatively low feeding levels, *i.e.* 2% dry feed per total W.W. of fish, the water quality in static systems can decline over a short

period of time and lead to reduced growth rates. This point is highlighted by the significantly greater weights and lengths attained by black bream stocked in the irrigation channel cages at Tammin only 90km east of Northam. Since there was a slow flow of water through these latter cages, there was a minimal accumulation of wastes and no evidence of algal blooms or cage fouling. Furthermore, the lowest FCR (1.84) was recorded in this channel, *i.e.* by caged black bream fed on alternate days. Ingram *et al.* (2000) also found that poor water quality and fouling of cage mesh contributed to poor growth of cage cultured finfish in inland saline water bodies.

Since water temperatures at Tammin were very similar to those recorded in Northam, the water temperature regime was not considered a contributing factor to the higher growth rate.

Since the amount of feed provided to black bream in ponds was reduced due to poor water quality, direct comparisons with the caged cultured fish were not possible. However, the FCRs of the fish in ponds were significantly lower than those of fish in the corresponding treatments in cages located in the adjacent lake. Furthermore, the pond fish attained slightly greater weights and lengths than those in cages despite receiving less supplementary feed overall. This indicates that the pond cultured fish had access to more natural feed and/or to feed for a greater time period after each feeding. As the feed used in the trial was a sinking formulation, the feed that was not eaten immediately on introduction would have passed through the cage floor within minutes, while pond fish would have had access to any uneaten feed for up to five hours^{*}.

As black bream can be "trained" to take floating feeds (G. Sarre, unpubl. data), it should be used in preference to sinking formulations in order to reduce sediment fouling and to permit an assessment of satiation.

In summary:

- Supplementary feeding is required for optimising the survival and growth of large black bream in inland water bodies.
- The final weight and length attained by black bream fed on alternate days (1%/day) did not differ significantly from that of fish fed daily (2%/day).
- Poor FCRs, presumably due to uneaten feed and an associated reduction in water quality, occur when large black bream are fed daily as little as 2% (dry feed: total W.W. of fish per day) in a single feed event.

^{*} Estimated water stability of feed formulation used in trial (manufacturers specification).

- Poor water quality limits the growth of large black bream, particularly in static, *i.e.* zero exchange, water bodies.
- Cage culture of large black bream is a viable culture technique if water quality is maintained.
- Under favorable water quality and food conditions black bream will spawn naturally in ponds.

3.5 POTENTIAL FOR BLACK BREAM TO SPAWN IN INLAND WATER BODIES

3.5.1 INTRODUCTION

There are obvious benefits to property owners if the black bream that they stock in inland water bodies for recreational fishing are capable of spawning naturally within those systems. Macroscopic and histological examination of the ovaries of black bream collected from ten inland water bodies that were surveyed during FRDC project 97/309 demonstrated that virtually all black bream >170mm T.L. became sexually mature, *i.e.* their ovaries contained mature yolk granule oocytes. However, the yolk granule oocytes in the vast majority of these ovaries were undergoing atresia. Yet, the black bream collected from two of the study dams possessed ovaries containing both hydrated oocytes and/or post-ovulatory follicles, which implies that these fish were about to spawn or had already spawned in these water bodies.

During the current study, the above ten water bodies will be revisited during the spawning period of black bream, *i.e.* late spring/early summer, to investigate whether the black bream in these systems had successfully spawned and, if so, to record the physico-chemical parameters in the spawning area.

N.B. Soon after commencing the current study, black bream spawned successfully in the lake adjacent to the research pond field trial station at Northam and subsequently in the adjacent research ponds (See section 3.4). This provided an ideal opportunity for a more frequent and extended monitoring of the new 0+ cohort of black bream in the former system than would have been possible at the more remote locations where the other water bodies were located.

3.5.2 MATERIALS AND METHODS

The ten inland water bodies, that were used as study dams in Project 97/309, were surveyed at least once during the late spring/early summer of 1999 and 2000.

Each water body was sampled using a plankton tow net, fish traps and a larval seine net. A record was kept of water temperature, salinity, pH and dissolved oxygen and of features such as the amount and coverage of macroalgae (if present) and whether antagonist fish species, such as the mosquito fish *Gambusia holbrooki*, were present.

The T.L. and total W.W. of all juvenile black bream caught were recorded to the nearest 1mm and 0.1g, respectively. Samples of juvenile black bream were collected from the lake in Northam at two-weekly intervals during November and December 1999 and in November of both 2000

and 2001. Up to five black bream juveniles, collected in November 1999, were stored in 70% ethanol for subsequent dietary analyses.

3.5.3 RESULTS

No black bream eggs or larvae were identified in the samples collected by plankton net tows in any of the ten water bodies sampled. However, black bream juveniles were collected from water bodies at Northam (90km east of Perth) and Busselton (229km south of Perth) using fish traps and a seine net.

Saline lake (Northam)

Large schools of juvenile black bream were observed along the banks of the lake on 15 November 1999. The length of this cohort (hereafter referred to as F1s) of fish ranged from 19-35mm, with the majority (>50%) falling within the 26-30mm length class (Fig. 19). By 30 November and 15 December, the length classes had increased to 34-63mm and 41-59mm, respectively (Fig. 19). An analysis of the stomachs of three fish collected in the 15 November sample demonstrated that the fish had been ingesting only calanoid copepods.



Figure 19. Length-frequency distribution of F1 black bream collected from the Northam lake at two weekly intervals in November and December 1999 using fish traps and a larval seine net.

On 23 December, very large numbers of dead F1s were found along the shore of the lake (mean T.L. 65 ± 5.5 mm). Based on these individuals, the estimated density of juveniles in the lake was between 200 and 500 fish/m². Fish trapping after this time failed to collect any fish.

Seine netting in March and July 2000 confirmed that F1 black bream were still present within the lake. Their mean length and weight increased from 196.6 ± 5.4 mm and 124.5 ± 11.2 g in March to

 203.7 ± 4.5 mm and 133.3 ± 10.9 g in July. Black bream were also collected from the lake when they had reached about one and two years of age. Table 12 provides a comparison of the lengths and weights of the F1 stock with those of Swan River fish at corresponding ages.

Table 12.Comparison between the mean lengths and weights of F1 generation black bream
collected from the lake at Northam and from the Swan River Estuary at the end of
their first and second years of life.

Population	First	year	Second year		
	(Nov	2000)	(Nov 2001)		
	Length (mm)	Weight (g)	Length (mm)	Weight (g)	
Swan River (Sarre and Potter, 2000)	125.8	34.5	207.0	142.2	
Northam 153.8 ± 4.5		78.8 ± 8.6	233.9 ± 4.8	235.9 ± 17.8	

Water quality and biological variables

Water chemistry variables from the lake in Northam at the time the F1s were first observed are shown in Table 13.

Table 13.Water chemistry variables recorded from the lake in Northam in mid-November
1999.

Parameter	Value
Temperature (°C)	21.1
Salinity (ppt)	11.7
Dissolved Oxygen (mg/L)	6.9
рН	8.8

Extensive growths of attached algae (*Chara* sp.) were present in shallow (<1m) areas of the lake (see Fig. 20), and these covered about 60% of the total surface area. No antagonistic fish species were recorded.



Figure 20. Shallow region of the Northam lake showing extensive weed coverage

Spawning survey (2000)

Sampling undertaken during November and December 2000 failed to find any evidence of a repeat spawning of black bream. Large numbers of *Gambusia holbrooki* had become established in the lake and several months of high algal turbidity had led to the deaths of much of the attached *Chara* sp.

Research ponds (Northam)

New juvenile black bream recruits were also found in three research ponds at the Northam field research station during subsampling of the final pond trial in February 2001 (see Section 3.4).

Custom-built pond (Busselton)

Juvenile black bream were also caught in the above water body in late November 1999. Water quality parameters recorded during the survey are shown in Table 14. The T.L.s of 20 juveniles sampled from the pond ranged from 38 to 80mm, with a mean of 64.0 ± 15.6 mm. Copepods were the dominant food item of the stomach contents of these juvenile black bream.

Table 14.Water chemistry variables recorded from a pond in Busselton in late
November 1999 in which black bream juveniles were identified.

Parameter	Value
Temperature (°C)	22.3
Salinity (ppt)	17.3
Dissolved Oxygen (mg/L)	6.5
рН	8.0

Weed coverage in the pond was about 30% (Chara sp.). No antagonistic fish species were found.

In June 2000, a major algal bloom probably resulted in heavy mortality of black bream in this system since early morning dissolved oxygen levels were < 1.5 mg/l. Contact with the property owner in November 2001 indicated that, after the pond had been drained, cleaned and restocked with large black bream, a second successful spawning occurred in November 2001, with large numbers of juvenile black bream of *ca* 50mm T.L. observed.

3.5.4 DISCUSSION

The presence of black bream juveniles in two water bodies that had previously contained only the large individuals of this species demonstrates that this species is capable of reproducing successfully in inland water bodies. Furthermore, comparisons between the lengths and weights

attained by the juvenile black bream recruits in the Northam lake with those of comparable age from the Swan River Estuary, indicates that, at least in this system, the growth rate of naturallyspawned black bream is greater than that of this species in that estuary and within which early growth is particularly rapid (see Sarre and Potter, 2000). This may have been due to the presence of higher water temperatures in these water bodies than in the Swan River Estuary.

Black bream are clearly a robust species and capable of successfully spawning in natural environments which differ markedly in their physico-chemical characteristics (Sarre and Potter, 1999). In inland water bodies, the limiting factors for successful spawning and recruitment are likely to be the availability and abundance of both appropriate live feed (copepods) for the larvae and areas of refuge from various predators, *e.g. Gambusia holbrooki*, birds and large black bream. Preliminary data suggest that algal blooms may also be a limiting factor, which could be indirectly related to the associated low dissolved oxygen and/or low pH concentrations typically recorded during these blooms.

Although more information is required to determine precisely the optimal conditions and environmental triggers for the successful spawning and recruitment of black bream in inland water bodies, preliminary data from project 97/309 and the data presented in the current study have identified the following conditions as likely to be prerequisites:

- Water with a salinity of at least 10ppt.
- Water temperature $>20^{\circ}$ C during the spawning period (late spring early summer).
- The absence of mosquito fish *Gambusia holbrooki*.
- A high density of appropriate live feed, usually copepod species, during the spawning period (September-December).
- Presence of weed growths as refuge for larvae and juveniles
- Provision of supplementary feed at regular intervals.
- An absence of algal blooms during the spawning period.

3.6 TYPES AND AVAILABILITY OF NATURAL FOOD SOURCES FOR BLACK BREAM IN INLAND WATER BODIES

3.6.1 INTRODUCTION

Black bream in Western Australian estuaries are highly opportunistic in their feeding habit, ingesting not only crustaceans, molluscs, polychaetes and teleosts in varying amounts, but also detritus and plant material (Sarre *et al.*, 1999). The dietary compositions of black bream in different estuaries vary markedly, presumably reflecting differences in the types and abundance of prey in the various systems (Sarre *et al.*, 1999). The dietary compositions of black bream also undergo size-related changes, with small invertebrates, *e.g.* amphipods, dominating the diet of juvenile fish, whereas larger prey, *e.g.* crabs and teleosts, dominate the diet of large fish (Sarre *et al.*, 1999).

Preliminary data provided in FRDC Project 97/309 indicated that the diet of black bream in inland water bodies likewise undergoontogenetic changes. Smaller prey items, *e.g.* aquatic insect larvae, made a far greater contribution to the diet of small black bream (<150mm) than to those of large black bream. The apparent slow growth rate of black bream >150mm T.L. may reflect a paucity of those larger food items in inland water bodies that would normally be a major component of the diets of large black bream in their natural environment. This is particularly relevant since the majority of landowners surveyed in FRDC Project 97/309 did not provide their black bream supplementary feed which, during the current study, has been shown to be crucial for optimal survival and growth (see Sections 3.1 & 3.4).

The aim of the current study was to determine the relationship between the types and relative abundances of natural food items present in inland saline water bodies and the dietary compositions of the black bream stocked in these systems.

3.6.2 MATERIALS AND METHODS

The potential food items in ten inland water bodies, which had been successfully stocked with black bream, were sampled in spring 1999 and autumn 2000 using a plankton tow net, fish and yabby traps and a substrate corer. The type, extent and form of weed cover (when present) were estimated visually and sweep netting was used to sample any invertebrate fauna present in the weed canopy. At least three samples were collected using each sampling technique in each water body.

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The plankton net was towed over a distance of ca 5m and the resulting sample stored in 70% ethanol for subsequent analysis. Weed (when present) was sampled by towing a weighted plankton net through the weed canopy for a distance of ca 1m. Baited fish and yabby traps were set along the banks of each water body for 30min.

Sediment samples were screened through a stainless steel sieve with a mesh size of 0.5mm. The potential food items, collected by all sampling methods, were sorted and identified under a dissecting microscope. In the case of samples with large numbers of food items, three 10ml subsamples were analysed and the mean percentage contribution by number calculated.

Up to three black bream were collected from each body using a seine net or rod and line. The length and weight of each fish was recorded to the nearest 1mm and 1g, respectively. The stomach was removed from each fish and stored in 70% ethanol. Stomach fullness was estimated visually, using a scale of 1 to 10. Each dietary item was identified and its contribution recorded as a percentage of the total volume and number of all prey present in each gut (Hyslop, 1980).

Data analyses

In order to obtain a general overview of the types and relative abundance of food items present in inland water bodies, data from the ten water bodies and the two sampling occasions were pooled for each of the sampling methods used. The dietary data for black bream collected from each water body were also pooled. Only those potential food items (water body) or dietary items (black bream) contributing >1% to the total numbers was included in the analyses.

3.6.3 RESULTS

Potential food items of black bream recorded from inland water bodies

A total of 11 potential food items for black bream were collected from the ten water bodies surveyed using the plankton net and weed sweep net (Table 15). No macroinvertebrates were identified in sediment core samples. Fish and yabby traps collected only mosquito fish (*Gambusia holbrooki*), which were recorded in six of the ten water bodies surveyed. The number of *G. holbrooki* caught ranged from 1 to 360/trap/per 30 min set.

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Table 15.Natural food items collected sampled from the water column and weed banks in
ten saline inland water bodies. (* Indicates those items also found in the diet of the
large black bream sampled from the same water bodies.)

Potential food items
Teleosts (fish)
Mosquito fish (Gambusia holbrooki)
Crustaceans
Amphipods*
Copepods
Ostracods
Cladocerans
Insects
Chironomid larvae (Midge)*
Dipterans (Fly)
Odonate larvae (Dragonfly nymph)*
Water boatman (Bugs)*
Caddis fly larvae
Eggs
Gastropods
Misc. shells

Copepods were by far the most numerous potential food item in plankton samples, contributing over 77% to the total number of food items in those samples (Fig. 21). The next highest contributor was chironomid larvae with 6.4%. All other food items contributed <5% to the total number (Fig. 21).

Chironomid larvae made the greatest contribution to the total number of potential food items in weed sweeps with 39.9%. Amphipods and insect eggs were the next highest contributors at 22.7 and 22.3%, respectively (Fig. 21).

Dietary analysis

The stomachs were removed from 30 fish caught in eight of the ten water bodies. The T.L. of these black bream ranged from 185 to 326 mm and had a mean of 232 ± 25.6 mm. The mean fullness ± 1 SE of the stomachs containing at least one food item (n=16, 54%) was 1.6 \pm 0.9. A total of seven food items were identified which, excluding the algae (*Chara* sp.), all belonged to the Insecta.

In terms of contribution to the total volume of individual prey items to the diet, larvae of odonates (dragonfly nymphs) and chironomids were the most important, with values of 29.0 and 26.4%, respectively (Fig. 21). *Chara* sp. was the only charophyte weed found in black bream gut contents, to which it contributed 20% to the total volume.

Chironomid larvae and water boatmen (bugs) were numerically the most important dietary items, contributing 64.9 and 26.3% to the diet, respectively (Fig. 21).



Figure 21. Percentage contribution by number of the major potential food items sampled in ten inland water bodies and the percentage contribution by number and volume of food items to the dietary composition of black bream collected from these water bodies. N.B. Only those items contributing >1% to the total number of items have been included.

3.6.4 DISCUSSION

The data collected during the current study provides strong support for the view expressed in FRDC Project 97/309 (Sarre *et al.*, 1999) that the types of biota found in inland saline water bodies would be inappropriate and/or inadequate to act as feed for large black bream. This conclusion is based on the fact that the fauna found to be abundant within the water bodies were either absent or poorly represented in the diet of large black bream in those systems. For example, while copepods contributed over 77% to the total number of potential food items in the water column, they were not found in the diet of the large black bream. This contrasts with that found with small juvenile black bream in both estuarine and inland water bodies, where copepods are a significant contributor to their diet (see Sarre *et al.*, 2000; Section 3.5, FRDC Project 97/309). Furthermore, the high percentage of empty black bream stomachs and the very low fullness value of those containing food suggest that there is an overall paucity of appropriate natural foods for large black bream in inland saline water bodies.

The relatively greater contribution made by large food items, *e.g.* dragonfly nymphs and water boatmen (bugs), to the diets of black bream than to the biota in the water column suggest that the

bream preferentially selected these fauna as food. Sarre *et al.* (2000) demonstrated that large black bream in Western Australian estuaries feed predominately on large prey, such as teleosts, bivalve mussels and decapods. Although *G. holbrooki* was abundant in many of the water bodies sampled, they were never found in the stomach contents of the black bream. This absence may be due to the different habitat occupied by black bream and *Gambusia*, *i.e.* benthic *vs* surface waters, respectively, or possibly due to an unpalatable characteristic of *Gambusia* sp. While yabbies are readily ingested by large black bream, they can only provide an alternative food source in low salinity water bodies and even then will require periodic restocking to maintain numbers (see Section 3.2).

The paucity of suitable large prey items for black bream in inland saline water bodies highlights the need to provide supplementary feed to ensure optimal survival and growth of the black bream (see Sections 3.1 & 3.4).

4.0 LABORATORY TRIALS

4.1 COMPARISON OF COMMERCIALLY AVAILABLE STARTER AND GROWER DIETS FOR JUVENILE BLACK BREAM

4.1.1 INTRODUCTION

A previous FRDC project 97/309 (Sarre *et al.*, 1999) showed that juvenile black bream grew rapidly when they were first stocked in inland water bodies, but that growth declined markedly after they had reached approximately 150mm. As the majority of these fish were not fed, the greatly reduced growth of larger fish probably reflected a paucity of naturally available food for these fish. This hypothesis was tested and proven to be correct in the current project by demonstrating that black bream provided with supplementary feed grew significantly faster than those left to forage for naturally occurring food (see Sections 3.1 & 3.4).

The many different aquaculture diets available in Australia vary significantly in their physical characteristics, composition and price. Most of the few farmers that are currently providing supplementary feed to black bream are utilising the cheapest or most conveniently available feed. The objective of this component of the present study was to assess the efficacy of several commercially available diets with a range of different compositions and prices for growing black bream.

The method proposed to meet this objective was to feed six commercially available diets to juvenile black bream for six months. However, during this period, fish experience a transition from starter diets to grower diets. Starter diets are fed to fish from weaning until they are about 50g in weight, while grower diets are fed to fish that weigh more than 50g. As starter and grower diets differ in their composition, two separate three month trials were conducted to study the effects of these two dietary types on growth. It was originally intended that the current trials would be conducted at the optimal temperature and salinity for black bream growth, as determined during FRDC project 97/309 (Sarre *et al.*, 1999). While the greatest growth was achieved at 24ppt, this was not significantly greater than that obtained at 36ppt (full-strength seawater). For logistical reasons, the current trials were therefore conducted in flow-through tanks with full-strength seawater. A heat exchanger was used to heat this water to 24°C, the optimal temperature for growth, as determined in FRDC project 97/309 (Sarre *et al.*, 1999).

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4.1.2 MATERIALS AND METHODS TRIAL 1 (STARTER DIETS)

The five starter diets investigated in Trial 1 are shown in Table 16, together with their source of origin, composition and cost. Although a sixth, locally-produced trout diet was also included in the original experimental design, its particle size distribution was inadequate and therefore excluded from the experiment. The Glen Forrest diet, which is not specifically a starter diet, was crumbled by the manufacturer for use in this experiment. It was included as a treatment as it was the only other locally-produced diet and the manufacturers indicated that they would be willing to crumble the diet for commercial distribution if it performed well in this trial. All of the diets had a similar particle size and buoyancy.

Thirty juvenile black bream $(1.10 \pm 0.01g)$ were weighed and placed in 140L aquaria, with 4 replicate aquaria per treatment. Fish were fed four times daily to satiety on the treatment diet and the amount of diet consumed recorded. At the end of each month, all fish were anaesthetised (2-phenoxyethanol, 0.5ml/L) and individually weighed. Mortalities were replaced with tagged fish of similar weight to maintain the same stocking density in each aquaria. Any such replacements were not included in the analysis of the data. At the start and end of the experiment (3 months), a subsample of fish were taken for whole-body proximate analyses (*i.e.* protein, lipid, water and ash). Each treatment diet was also subjected to a proximate analysis.

Diet	Diet code	Source	Protein Content (%DW)	Lipid Content (%DW)	Price/Kg \$
Lansy Epac Alpha	EA	European starter diet imported by Primo Aquaculture, NSW.	59	16	40.00
Pivot	PV	Tasmania	54	17	3.10
Ridley Agriproducts	RA	Queensland	58	11	2.50
Kinta	KT	Victoria	55	6	2.20
Glen Forrest	GF	Western Australia	37	9	1.15

Table 16.Source, cost, lipid and protein content of the diets tested in Trial 1.

TRIAL TWO (GROWER DIETS)

The diets investigated in trial 2 are outlined in Table 17. The marron pellet, low in protein and containing no added marine oil, was included as a treatment as many farmers are using it to feed to black bream due to its low cost and availability.

Diet Treatment	Diet Code	Source	Diet Type	Price/kg	Protein Content (%DW)	Lipid Content (%DW)
Wesfeeds (Marron)	WF	Western Australia	Cold pressed; sinking	\$0.61	21%	8.5%
Ridley's (Silver perch)	SP	Queensland	Extruded; floating	\$1.88	35%	8%
Glen Forrest (Black bream)	GF	Western Australia	Cold pressed; sinking	\$1.15	40%	10%
Ridley's (Native fish food)	RN	Queensland	Extruded; sinking	\$1.88	45%	10%
Hunts (Trout)	HT	Western Australia	Extruded sinking	\$1.77	40%	18%
Pivot (Salmon)	PS	Tasmania	Extruded; sinking	\$1.91	45%	25%

Table 17. Source, diet type, cost and proximate composition of the diets tested.

Ten fish (mean weight 60.0 ± 0.1 g) were stocked into each 180L tank (4 replicates per treatment) and allowed to acclimate to the conditions and experimental diets for 1 week. After the acclimation period all fish were removed, weighed and returned to the tanks for the commencement of the trial. Subsequently all fish were removed from each tank every four weeks, anaesthetised (2-phenoxyethanol 0.5mL L⁻¹) and weighed to the nearest 0.1g.

Due to slow feeding by the larger black bream, the method employed in the current trial was different from that used in Trial 1. Tanks operated on a flow-through system with an exchange rate of 2L min⁻¹. of full strength seawater (35ppt). Incoming water provided a circular current that deposited particulate matter under a central standpipe, where it was removed with the outgoing water flow. Fish could not be fed by offering food until satiety was reached, as large black bream are slow feeders, which graze from the tank bottom. In order to be able to quantify the dietary consumption of bream in the current trial, the black bream were offered a fixed ration of 2% of

their body weight (4mm pellets) for a period of 1.5h each day. During this time, the water flow to each tank was turned off to prevent uneaten pellets being removed via the central standpipe. After 1.5h, all uneaten food was collected via siphon onto a 350µm screen. This recovered food was then oven-dried at 90°C for 24h. Water flow was resumed after siphoning, resulting in a clean tank bottom prior to the commencement of the following days feed.

Since differences in water stability between the six diet types led to differential losses of the food through the 350µm screen during collection, tests were conducted to quantify these losses so that a correction factor could be applied. Representative amounts of diet (*ca* 15g) were weighed and placed into each of the trial tanks whilst they contained no fish. The diet was left in the tanks for 1.5h and then collected on the 350µm screen as described above. This recovered diet was then oven dried at 90°C for 24h and weighed. A correction factor was then calculated by dividing the dry weight of diet added to the tank by the dry weight of diet recovered. The dry weights of food recovered from the tanks each day during the trial were multiplied by the corresponding correction factor to account for this loss. Daily consumption of each diet was then calculated by subtracting this corrected recovered weight from the dry weight of pellets offered.

Samples of fish were taken at the completion of the trial for determining their composition (lipid, protein, water, ash and carbohydrate). Unfortunately, due to a technical problem with an autoclave during sample processing, all of the samples were lost.

4.1.3 **RESULTS AND DISCUSSION**

TRIAL 1 (STARTER DIETS)

Survival of fish fed the five different diets, which ranged from $71.2 \pm 6.6\%$ to $92.5 \pm 4.8\%$ (Table 18), did not differ significantly (p > 0.05).

The weight gained and specific growth rates (SGR) of black bream when fed on the various diets, are shown in Table 18. There was no significant difference (p > 0.05) in weight gain between fish fed Lansy Epac Alpha (11.8 ± 0.4g), Ridley Agriproducts (11.7 ± 0.7g) and Pivot (11.3 ± 0.2g). Fish fed on Glen Forrest experienced the least weight gain (8.7 ± 0.4g), and this was significantly less (p < 0.05) than fish fed all other products, except Kinta (9.02 ± 0.72g). Specific growth rates ranged from 2.42 ± 0.07 %/day (Glen Forrest) to 2.73 ± 0.03 %/day (Lansy Epac Alpha). The patterns of SGR between diets were the same as those described above for weight gain. Fish in the Glen Forrest treatment consumed the most food (592 ± 4g), and this was significantly greater (p < 0.05) than that of fish fed all other diets except Pivot, which consumed

556 ± 3g (Table 18). Consumption of Kinta was significantly less than those of all other treatments (492 ± 6g) (p < 0.05). Food conversion ratios (FCR) over the three month period ranged from 1.58 ± 0.08 to 2.34 ± 0.21 (Table 18). The FCR obtained with fish fed Glen Forrest was significantly greater than for all other treatments (p < 0.05). No other significant differences between FCR were detected.

Diet	Survival (%)	SGR (%/day)	Weight Gain (g)	Consumption (g)	FCR	Cost/g gain biomass (¢)
EA	87.5 ± 4.3	$2.73\pm0.03~^a$	$11.8\pm0.4~^a$	550 ± 3 ^b	1.58 ± 0.05 ^a	6.33 ± 0.18
PV	85.0 ± 11.7	$2.68 \pm 0.02^{a,b}$	$11.3 \pm 0.2^{a,b}$	$556\pm3~^{b}$	$1.66 \pm 0.03^{\ a}$	$0.66\pm0.01\ ^a$
RA	92.5 ± 4.8	$2.71\pm0.06~^a$	11.7 ± 0.7 ^a	$531 \pm 17^{a,b}$	1.58 ± 0.08 ^a	0.39 ± 0.02^{b}
KT	71.5 ± 6.6	$2.45 \pm 0.08^{b,c}$	9.0 ± 0.7 ^{b,c}	492 ± 6 ^c	$1.83 \pm 0.10^{\ a}$	$0.46\pm0.02^{\ b}$
GF	87.5 ± 3.2	2.42 ± 0.07 ^c	8.7 ± 0.7 ^c	592 ± 4^{a}	2.31 ± 0.18^{b}	0.27 ± 0.02 ^c

Table 18. Performance of juvenile black bream fed five commercially available starter diets for 3 months. Within each column, values sharing the same letter are not significantly different (p > 0.05).

The whole-body lipid content was positively correlated with the lipid content of the food (Fig. 22). As there was no significant difference in the growth of black bream fed on diets of Pivot, Ridley Agriproducts and Lansy Epac Alpha, the data in Figure 22 suggest that the additional lipid present in Pivot and Lansy Epac Alpha does not lead to increased growth in terms of length but does result in a fatter fish. Unfortunately, the fish used in the current trial were too small to conduct a comparative slaughter to establish whether the additional fat was deposited in the body cavity or within the flesh.



Figure 22. Relationship between dietary lipid content and whole-body lipid content of juvenile black bream fed five commercially available starter diets over a 3 month period. DW, dry weight.

Based on the amount of food consumed during the trial, the cost of each food type and the biomass gain, the cost of food per g of biomass gain was calculated (Table 16). Lansy Epac Alpha was the most expensive diet and yielded no growth benefits. For the purposes of statistical analysis, the Lansy Epac Alpha data was excluded to maintain homogeneity of variance and normality. Glen Forrest was the cheapest diet at $0.26 \pm 0.02 \text{¢ g}^{-1}$ and was significantly cheaper than all other diets. Pivot was significantly more expensive than the other three diets ($0.66 \pm 0.01 \text{ ¢/g}$). There was no significant difference between the cost of Kinta ($0.46 \pm 0.02 \text{¢/g}$) and Ridley Agriproducts ($0.39 \pm 0.02 \text{¢/g}$).

Based on growth rates, lipid content and economics, the Australian produced Ridley Agriproducts appears to be the most suitable for juvenile black bream up to a size of approximately 50g. The high cost of the Lansy EPAC Alpha was not justified, as it did not result in greater growth than the Ridley or Pivot diets. The Pivot diet produced good growth but was more expensive and produced 'fatter' fish than the Ridley Agriproducts diets. The Glen Forrest and Kinta diets were not suitable for juvenile black bream, resulting in significantly lower growth than Pivot and Ridley Agriproducts. As most starter diets contain high levels of lipid, the low lipid content of these diets may have contributed to their poor performance.

TRIAL TWO (GROWER DIETS)

Survival in all treatments was 100% during the 3-month trial period. Black bream fed on Pivot salmon grower gained significantly more weight $(17.9 \pm 1.3g)$ than fish fed all other diets. Fish fed on the Glen Forrest diet performed well, with a weight gain $(13.0 \pm 1.7g)$ significantly greater than those in all other treatments, except Pivot salmon grower and Ridley Agriproducts $(10.3 \pm 0.8g)$. Fish fed on the Wesfeeds marron diet performed poorly, losing weight during the experimental period $(-0.8 \pm 1.0g)$. This demonstrated that the nutritional profile of this diet is insufficient to meet the basal needs of black bream. Despite having a similar nutritional profile to Pivot salmon, the weight gain by black bream fed Hunts trout diet resulted in poor weight gain $(4.0 \pm 0.4g)$ and one that was significantly less than in all other treatments except the Wesfeeds marron diet and the Ridley's silver perch diet. Specific growth rates ranged from $0.27 \pm 0.02\%/day$ (Pivot salmon) to $-0.02 \pm 0.02\%/day$ (Wesfeeds marron). The patterns of SGR between diets were the same as those described above for weight gain.

The consumption of each diet throughout the trial period is shown in Table 19. Fish fed the Glen Forrest diet ate significantly more than in those fed all other diets $(424 \pm 28g)$. Fish consumed significantly less Hunts diet than any other diet $(139 \pm 5g)$, explaining the poor growth performance of fish fed this diet. This diet was very hard and analysis of the moisture content of the diets in the subsequent trial on diet stability revealed that it had a low moisture content, which may have contributed to its poor palatability. Table 18 shows the food conversion ratios (FCR). Due to a negative weight gain, a FCR could not be calculated for the fish fed on the Wesfeeds marron diet. The FCR of fish fed Pivot salmon grower was significantly less than for all other treatments (1.61 ± 0.06) . Although the weight gain of fish fed Glen Forrest was good, the high food consumption of this diet resulted in a FCR that was more than twice that obtained on Pivot salmon grower (3.40 ± 0.37) .

As described for Trial 1 above, the cost of food per g of biomass gain was calculated (Table 19). Based on these calculations, Pivot was the most economical diet to feed to black bream, costing 3.07 ¢/g of biomass gain, which is significantly less than Ridley's silver perch diet ($7.30 \pm 0.56 \text{¢/g}$) and Hunts trout diet ($6.35 \pm 0.57 \text{¢/g}$).

Diet	Survival (%)	Weight Gain (g)	SGR (%/day)	Consumption (g)	FCR	Cost/g (¢)
WF	100 ± 0	-0.8 ± 1.0^{e}	-0.02 ± 0.02^{e}	268 ± 34 ^b	N/A	N/A
SP	100 ± 0	$6.9 \pm 0.4^{c,d}$	$0.12 \pm 0.01^{\ c,d}$	$251\pm17~^{b}$	3.65 ± 0.28 ^a	7.30 ± 0.56^{a}
GF	100 ± 0	13.0 ± 1.7 ^b	$0.20\pm0.02^{\ b}$	$424\pm28~^a$	$3.40 \pm 0.37^{a,b}$	3.91 ± 0.43 ^c
RN	100 ± 0	$10.3 \pm 0.8^{b,c}$	$0.17 \pm 0.01^{b,c}$	$250\pm9~^{b}$	2.45 ± 0.13 ^b	4.89 ± 0.27 ^{b,c}
HT	100 ± 0	4.0 ± 0.4 ^d	$0.07\pm0.01~^{d}$	140 ± 5 ^c	$3.59 \pm 0.32^{\ a}$	$6.35 \pm 0.57^{a,b}$
PS	100 ± 0	17.9 ± 1.3^{a}	$0.27 \pm 0.02^{\ a}$	$286\pm16^{\ b}$	1.61 ± 0.06 ^c	3.07 ± 0.12 ^c

Table 19. Performance of juvenile black bream fed six commercially available grower diets for 3 months. Within each column, values sharing the same letter are not significantly different (p > 0.05).

The growth and FCR data obtained during the current trial suggests that Pivot salmon grower is the most suitable of the diets tested as either a complete or supplementary feed for black bream. This diet resulted in significantly greater weight gain and FCR than all other diets tested and it was also the most economical to use. Due to differences in diet manufacture processes between the different diet types, it is difficult to draw conclusions about the protein and energy requirements of juvenile black bream. However, the best performance was obtained on a diet containing a high protein and energy content. This conclusion contrasts with the previously held perception that black bream would have similar nutritional requirements to silver perch as they are also omnivorous.

4.2 DEVELOPMENT OF A WATER STABLE PELLET FOR LARGE BLACK BREAM

4.2.1 INTRODUCTION

Black bream are being stocked in inland water for recreational fishing purposes. Consequently, artificial diets may be applied infrequently, as fishing may not be the primary income producing activity on the farm. During a previous project (FRDC project number 97/309; Sarre *et al.*, 1999), most of the farmers who provided supplementary feed were found to do so only occasionally (*e.g.* every few days). Many of these farmers provided a few days ration on a single occasion, based on the assumption that this would provide fish with food on those days when they did not provide feed. Unfortunately, many of the diets currently used to feed black bream in inland waters have poor water stability and thus any food that is not eaten within minutes of immersion quickly disintegrates, thus making it unavailable to the fish and likely to lead to pond eutrophication and deoxygenation.

The development of a pellet that will remain stable for an extended period would be likely to reduce the requirement to supply black bream with feed on a regular (daily) basis. Mr Michael Hoxey (M.J. Hoxey and Associates Pty. Ltd.), an animal nutrition consultant who has experience in developing water stable food for aquaculture purposes, was therefore contracted to undertake the development of a water stable pellet. Staff at the Aquaculture Development Unit assessed the stability of the diets manufactured by Mr Michael Hoxey.

The first stage of the project involved comparing the water stabilities of the commerciallyavailable grower trials, followed by the manufacture and assessment of the stabilised pellet.

The initial intention was to increase the water stability of the diet previously identified as producing the best growth of black bream, *i.e.* Pivot salmon grower. It was decided, however, to also carry out the stabilisation procedure on the locally-produced Glen Forrest diet because it is far cheaper than Pivot, locally produced and results in relatively good growth rates of black bream. Since this cold-pressed diet has limited water stability (see below), the development of a successful stabilisation process had the potential to significantly improve its value as a black bream diet.

4.2.2 MATERIALS AND METHODS

Water Stability of Commercially Available Grower Diets

The correction factor for determined diet consumption, as detailed in Trial 2 above, was also used to calculate the gravimetric leaching loss of each diet. As previously described, known weights of each grower diet (*ca* 15g) were placed in replicate 180L tanks containing no fish (4 replicates per treatment). The diets were left in the water for 1.5h and then siphoned on to a 350- μ m screen. The diet retained on the screen was then oven dried at 90°C for 24h and the amount of diet recovered expressed as a percentage of the dry weight of diet added to the tanks. This value represented the gravimetric leaching loss of diet over the 1.5h period.

Manufacture and Assessment of Water Stable Pellets

As the methodology behind the stabilisation process is commercially sensitive, details on the heat treatment procedure are not publicly available. Two stabilised diets were produced from each of the Glen Forrest and Pivot diets at two different heat treatment temperatures to determine the effect of treatment temperature on water stability.

Pivot salmon grower pellets (4mm) were ground down and repelletised under the stabilisation procedure into PV-H (high temperature) and PV-L (lower temperature) diets. The Glen Forrest diet mix was provided to Mr Hoxey and also formulated into 4mm GF-H and GF-L pellets.

Water stability of the four stabilised diets, as well as of the two control diets (PV-C and GF-C), were determined in duplicate according to the procedure described by Evans *et al.* (1998). PVC sieve cups were constructed by securing 2 mm nylon mesh within a short length of 40mm PVC pipe using a piece of 32mm PVC pipe (Fig. 23). Four 20 mm holes in the 40mm pipe allowed water to circulate under the mesh screen.



Figure 23. Screens used for determining water stability of heat-treated diets.

A sample of each diet (*ca* 2.0g) was accurately weighed into each cup. Each cup was then placed in an 800mL glass beaker containing 350mL of seawater at $22^{\circ}C$. Beakers were placed for 2h on a shaker table which operated at 100rpm (Fig. 24). At the end of 2h, the feed remaining on the 2mm screen was rinsed with distilled water on to a 6µm pre-weighed filter paper on a vacuum Buchner funnel. The feed that passed through the 2mm screen was filtered on to another labelled, pre-weighed filter paper. Filter papers were then oven dried at 105°C for 24h, with the subsequent increase in dry weight of each paper corresponding to the dry weight of the diet that had either collected on or passed through the screen.



Figure 24. Shaker table used to determine the water stability of heat-treated diets.

The moisture content of each of the six diet types was determined in duplicate after oven drying accurately known weights (*ca* 4g) of each diet at 105° C for 24h.

4.2.3 RESULTS AND DISCUSSION

Water Stability of commercially available grower diets

Figure 25 shows that the extruded diets, Hunts, Pivot Salmon and Ridley's Native, had the greatest water stability, losing less than 10% of their weight over 1.5h. There were no significant differences in water stability between these diets. Steam extrusion gelatinises carbohydrates within the diet, effectively binding together all of the dietary components and significantly improving the diet's water stability.



Figure 25. Gravimetric leaching loss from 6 commercially available grower diets after 1.5 h soaking in seawater. Columns sharing the same letter are not significantly different (p > 0.05)

Both cold-pressed diets, Glen Forrest and Wesfeeds, had poor water stability. The Glen Forrest diet lost approximately $67 \pm 3\%$ of its weight over the experimental period, which was significantly greater than that of all other diets. As these diets contain no binders, their poor water stability was expected. The Ridley's silver perch diet is a floating extruded diet whose water stability did not differ significantly from that of the cold-pressed Wesfeeds marron diet. The poor stability of this diet, compared with those of the other extruded diets, may be due to ingredients within the diet or processes undertaken during diet manufacture that cause the diet to float.

Manufacture and Assessment of Water Stable Pellets

The moisture contents of the four stabilised and two control diets are shown in Figure 26. Within each diet type, moisture content was highest in the control (unstabilised) diets and decreased with increasing stabilisation temperature. The moisture content was greatest in the Pivot control diet $(10.47 \pm 0.23\%)$ and thus significantly greater than those of all other diets. The moisture content of the Glen Forrest control diet $(7.60 \pm 0.34\%)$ was significantly greater than that of the stabilised Glen Forrest diet produced at high temperature $(4.61 \pm 0.09\%)$. The decrease in moisture content with increasing stabilisation temperature produced a harder, more brittle diet.



Figure 26. Moisture content of water stabilised, heat-treated diets. PV = Pivot salmon grower, GF = Glen Forrest. H & L indicate diets produced under high and lower stabilisation temperatures, respectively. C = control diets. Columns sharing the same letter are not significantly different (p > 0.05).

Figure 27 shows the percentage of diet (on a dry weight basis) retained on the 2mm mesh after immersion for 2h. The water stability of the Pivot control diet was significantly greater than that of all other diets tested. After 2h, $98.4 \pm 0.1\%$ of this diet was still intact and remaining on the 2mm screen. The stabilisation process decreased the water stability of the Pivot diet, with both the low and high temperature Pivot diets being significantly less stable than the control. The Pivot diet produced under the higher temperature was significantly more water stable than that produced under the lower temperature.



Figure 27. Percentage of 4 mm diet retained on a 2 mm screen after two hours immersion in seawater. PV = Pivot salmon grower, GF = Glen Forrest. H & L indicate diets produced under high and lower stabilisation temperatures, respectively. C = control diets. Columns sharing the same letter are not significantly different (p > 0.05).

The stabilisation process significantly improved the water stability of the Glen Forrest diet. After 2 h, $3.6 \pm 0.3\%$ of the Glen Forrest control diet remained on the 2 mm screen, compared with $39.0 \pm 10.5\%$ and $41.7 \pm 1.2\%$ for the low and high temperature stabilised diets, respectively. There was no significant difference in water stability between the high and low temperature stabilised Glen Forrest diets.

Recovery of the diet in all cases was good. The sum of the diet that was retained on the 2mm screen and was washed through the 2 mm screen ranged from 97.8 to 107.6% of the initial weight of diet weighed onto the screen.

Although the stabilisation process significantly improved the water stability of the Glen Forrest diets, these diets were still significantly less stable than the extruded Pivot control. As the Pivot control diet also resulted in significantly greater growth rates and lower FCR than Glen Forrest, this diet is recommended for feeding to black bream. Any further studies on heat stabilised diets should investigate the effect of the heat stabilisation process on diet palatability and digestibility (due to the increased hardness of the stabilised diets) and the loss of unstable dietary components such as vitamins. If farmers are to adopt a feed management practice whereby pellets are only

offered every few days, issues such as the loss of labile dietary ingredients will still need addressing, regardless of whether or not further investigation of diet stabilisation occurs.

4.3 COMPARISON BETWEEN GROWTH RATES OF BLACK BREAM CULTURED FROM SWAN AND MOORE RIVER ESTUARY STOCKS

4.3.1 INTRODUCTION

During recent years, the black bream *Acanthopagrus butcheri*, which is found throughout southern Australia (Kailola *et al.*, 1993), has been the subject of several research projects aimed at developing this species for aquaculture. This research has focused, in particular, on developing the most appropriate hatchery techniques for rearing the young from eggs produced by broodstock and on growing the resultant juveniles in inland saline lakes to provide a "put and take" fishery (Sarre *et al.*, 1999; Partridge and Jenkins, 2002). As a result of the development of successful hatchery techniques, large numbers of juveniles are now able to be cultured to facilitate the restocking of the Blackwood River Estuary, in which the stock of black bream has become severely depleted.

In Western Australia, the black bream is confined to estuaries (Potter and Hyndes, 1999). The populations in the different estuaries are genetically distinct (Chaplin et al., 1998) and exhibit marked differences in certain biological characteristics such as growth rate, size and age at maturity and diets. For example, by the end of their second year of life, black bream have attained total lengths of *ca* 265 mm in the Swan River Estuary, but only *ca* 145mm in the Moore River Estuary (Sarre and Potter, 2000), and maturity is attained at the end of the second year in the former estuary, but not until the end of the fourth year in the latter estuary. Differences in diet amongst estuaries are illustrated by the fact that, during an earlier study (Sarre et al., 2000), the stomach contents of black bream from the Swan River Estuary were found to contain a large volume of bivalve molluscs (64%) and only a small volume of macrophytes (8.3%), whereas those of fish caught in the Wellstead Estuary contained a very substantial volume of macrophytes (56%) and a relatively small volume of bivalve molluscs (2.2%). In contrast to the situation in both of these estuaries, the stomach contents of this sparid in the Moore River Estuary comprised approximately equal volumes of bivalve molluscs (43%) and macrophytes (40%). These implications that black bream is an opportunistic feeder are consistent with the fact that the major prey taxa in the stomach contents of black bream in the Swan River Estuary are abundant components of the invertebrate fauna of that estuary (Kanandjembo et al., 2001) and macrophytes (Cladophora sp.) form very dense growths in the Moore River Estuary (Sarre et al., 2000). However, environmental conditions in the various estuaries in which black bream is found vary markedly. For example, salinities throughout most of the length of the intermittently open Moore River Estuary rarely exceed 7ppt (Young et al., 1997), whereas those in the upper reaches of the

Swan River Estuary, where black bream are particularly abundant and spawn, approach full strength sea water in late summer/early autumn. Far more extreme conditions are found in the Wellstead Estuary, in which black bream spawn in salinities of *ca* 40‰ and have been caught in salinities as great as 70ppt (Young and Potter, 2002).

Although differences in the growth rates and diets of black bream in the various estuaries in which this sparid occur may be related to differences in the abundances of the different types of potential food and environmental conditions among those estuaries, they may also reflect differences in the genetic compositions of the various populations. If the marked differences in growth rate reflect differences in genetic composition, it would be important to select, as broodstock, fish that had been obtained from the estuary in which black bream exhibited the fastest growth. The aim of the present study was therefore to determine whether there were differences in the growth rates of black bream reared under identical conditions from broodstock obtained from the Swan and Moore River estuaries, in which the genetic composition and the growth rate during early life differed.

4.3.2 MATERIALS AND METHODS

Eight males and eight females were collected on separate days from both the Swan and Moore River estuaries in late December 1999. The females were immediately administered an interperitoneal injection of Ovaprim at the rate of 0.1mL kg^{-1} to reduce the effects of stress and thus maintain oocyte integrity. All fish were transported in 1 000L tanks to the Aquaculture Development Unit (ADU) at Fremantle, where they were acclimated to full strength seawater for 5h. They were then transferred to separate 2 500L tanks. All of the males from both estuaries were running ripe. On the day after collection, a biopsy was taken of the ovaries of the eight females from both the Moore and Swan estuaries and the five females in each group, whose ovaries contained oocytes with diameters greater than 400 μ m, were given a further Ovaprim injection at 0.3mL/kg⁻¹ to induce final maturation.

Spawning occurred naturally amongst the groups of fish from both estuaries on 25 December 1999 and fertilised eggs were collected from the tank overflows. Both groups produced approximately 250 000 eggs with >90% viability. Viable eggs from each group were incubated in 1 000 L tanks. On the second day after hatching, 10 000 larvae from each incubation tank were separated into two batches and placed in identical 1 000 L rearing tanks. All four larval batches were reared under the same conditions using the semi-intensive greenwater method described by

Partridge *et al.* (2002). In brief, larvae were reared in flowing seawater ($1Lmin^{-1}$), to which the green microalgae *Nannochloropsis oculata* was continuously supplied via a dosing pump so as to maintain the cell density at 0.5 - 1.0×10^{6} cells mL⁻¹. Rotifers were inoculated into each rearing tank at a concentration of $20mL^{-1}$ and were retained within the culture through the attachment of 53μ m mesh to the outflows. Temperature, dissolved oxygen (DO) and pH were recorded twice daily. When their mean total length had reached *ca* 7mm, the larvae were started to be fed enriched *Artemia metanauplii*. Three days later, the tanks were switched to a flow-through system and the larvae were weaned onto an artificial diet (Nippai ML), a procedure that was complete by day 45.

A subsample of 10 larvae was taken from each tank daily between days 2 and 33, every 3 or 4 days from days 33 to 66 and weekly between days 66 and 87. The total length of larvae <10mm was measured to the nearest 0.1mm using a calibrated ocular graticule under a dissecting microscope, while those of larger larvae and juveniles was measured to the nearest 0.1mm using vernier calipers.

When fish had reached an age of 107 days, their number in each 1 000 L rearing tank was reduced from 1 500 to 150 fish. These fish were then reared for a further five months under the same conditions of water temperature, salinity and photoperiod and were fed to satiety three times daily on a commercially available fish diet (45% protein, 22% lipid). Subsamples of 10 to 20 fish after one, two and three months and of 40 fish after four months and all of the fish after five months, i.e. at the completion of the experiment, were removed, anaesthetised (AQUI-S, 20 mg L^{-1}), rapidly measured to the nearest 1mm total length and returned to the tank from which they had been taken. Few deaths occurred during the above five months.

The following growth equation of Schnute (1981, Eq. 15) was fitted to the length-at-age data for the fish derived from each source.

$$L_{t} = \left[L_{1}^{b} + \left(L_{2}^{b} - L_{1}^{b} \right) \frac{1 - e^{-a(t - \tau_{1})}}{1 - e^{-a(\tau_{2} - \tau_{1})}} \right]^{1/b}$$

where L_1 and L_2 are the estimated lengths at selected reference ages τ_1 and τ_2 years and *a* and *b* are constants (both $\neq 0$).

4.3.3 RESULTS AND DISCUSSION

After fifteen days of variable temperatures, when the larvae were being held in static systems, the mean weekly water temperature gradually rose from ca 21 to 22° C over the next 90 days and then declined slowly by *ca* 0.5°C in each of the next five months to reach a final temperature of just over 19°C. During the larval rearing period (45 days), the mean pH remained close to 7.9 and the mean dissolved oxygen concentration remained well above 90% (Table 20).

Table 20.Water temperatures (°C), pH and dissolved oxygen (percentage saturation) in
tanks used for culturing the two batches of larvae produced from Moore River
and Swan River Estuary broodstock.

	Moore # 1	Moore #2	Swan #1	Swan #2			
Temperature							
Mean:	21.9	21.8	21.9	21.7			
Maximum:	24.8	24.4	24.8	24.5			
Minimum:	20.8	20.5	20.7	20.4			
pH							
Mean:	7.9	7.9	7.9	7.9			
Maximum:	8.1	8.2	8.2	8.2			
Minimum:	7.7	7.7	7.7	7.7			
DO							
Mean:	95	92	92	93			
Maximum:	111	112	111	113			
Minimum:	83	70	80	77			

von Bertalanffy growth curves did not provide a good fit to the lengths at age of black bream in the groups of fish derived from broodstock collected from the Swan River and Moore River estuaries. However, Schnute growth curves did provide a good fit to both sets of length at age data (Figs 28a, b), as is demonstrated by the fact that the coefficients of determination for the growth curves for the above two groups of black bream were as high as 0.978 and 0.974, respectively (Table 21).



- **Figure 28.** Schnute growth curves fitted to the lengths at age of black bream reared from eggs produced by broodstock obtained from (a) the Moore River Estuary and (b) the Swan River Estuary. (c) shows both growth curves on the same plot.
- **Table 21.**Values for the parameters in the Schnute growth equations fitted to length at age
data for black bream reared from broodstock collected from the Swan and Moore
River estuaries.

Parameter	Swan River Estuary	Moore River Estuary
$ au_1(d)$	20	20
$ au_2(d)$	80	80
$a (d^{-1})$	0.024	0.030
В	-0.693	-1.028
L_1 (mm T.L.)	6.701	6.097
$L_2 ({\rm mm \ T.L.})$	31.371	28.515
R^2	0.978	0.974

The plots of these two growth curves on the same figure (Fig. 28c) emphasize that the curves are very similar. Thus, differences in the lengths predicted by the curves for fish at any age between 100 and 267 days (end of the experiment) never exceeded 6% and the lengths derived from the curves for the "Swan River Estuary" and "Moore River Estuary" fish at the end of the experiment, i.e. 114.6 and 115.2mm, respectively, represented a difference of only 0.6mm.

Furthermore, the mean lengths $\pm 95\%$ confidence limits for the above two groups of fish at the end of the experiment, i.e. 113.7 ± 23.8 mm and 115.0 ± 27.7 mm, respectively, were not significantly different (<u>P</u>>0.05).

The above lengths for "Swan River Estuary" and "Moore River Estuary" fish at 267 days are 13 and 57mm greater than those recorded for natural populations of black bream in these two estuaries, respectively (Sarre and Potter, 2000). The fact that the laboratory-reared fish had grown to a greater length at the end of 267 days than was achieved even in the Swan River Estuary, in which the early growth of black bream is the fastest yet recorded in south-western Australia, show that the early growth of black bream under our culture conditions is particularly rapid. However, the trends exhibited by the Schnute growth curves emphasise that growth of juvenile black bream under laboratory conditions does tend to asymptote (Fig. 28). It is thus highly relevant that the growth curves attained their point of inflection, i.e. started to asymptote, when the fish were about 115 days old, i.e. just after the time when the temperature had reached its maxima and begun to undergo its decline of $ca 0.5^{\circ}$ C per month throughout the last five months of the study.

Our results provide overwhelming circumstantial evidence that the very marked differences in the growth rates of A. butcheri in the Swan and Moore River estuaries (Sarre and Potter, 2000) reflect differences in the environmental conditions in these two estuaries, rather than an expression of the genetic differences that exist between the populations of black bream in these two systems. The differences in growth of black bream among these and other estuaries are thus likely to be at least partly related to the marked variations in the types of food ingested in those estuaries and thus of their nutritional value (Sarre et al., 2000). It seems unlikely that the very marked differences in the early growth of black bream in the Swan and Blackwood River estuaries are attributable to the marked differences in salinity that exist between these two systems. This conclusion is based on the fact that the growth of black bream cultured in salinities of 24ppt, as are found in the upper Swan Estuary during the main growth period, did not differ significantly from those of black bream cultured in salinities of 4ppt, as are present in the Moore River Estuary during the same period (Partridge and Jenkins, 2002). However, the possibility cannot be excluded that the particularly slow growth during the first year of life in the Moore River Estuary is a consequence of the high densities in which this species is found along the shallow banks of this estuary (Sarre and Potter, 2000).

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Irrespective of the basis for the marked differences in growth of black bream in the Swan and Moore River Estuaries, our results indicate that the choice of the source of the black bream used for broodstock is not important, at least from the point of view of growth of juveniles under aquaculture conditions. However, since the genetic compositions of the various black bream populations are distinct (Chaplin et al., 1998), these findings will only partly allay the fears of the governmental advocates regarding the translocation of this species from one water body to another or on ensuring that the broodstock used to produce juveniles for restocking an estuary are obtained from that specific estuary (Translocation Policy, Department of Fisheries, Western Australia). Yet, it must be recognised that, although black bream in Western Australia exhibit a low level of genetic diversity in several different regions of the genome, there is genetic evidence that all of the individuals in this large area may have been derived from a common ancestor (small group of individuals) during the last 10,000 years (Chaplin et al., 1998; Santos Yap et al., in prep.). It is thus concluded that the dangers of translocating black bream in Western Australia, through the transport of either members of natural populations or the products of aquaculture, are very small. Although the results and comparisons produced in this paper relate specifically to black bream, they emphasize the importance of taking a number of factors into account when considering the question of which stocks of a species should be used for aquaculture and the appropriatness of introducing the juveniles produced by the broodstock obtained from one water body into another water body.

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6.0 **BENEFITS**

This research has identified the methods and conditions required to optimise the survival and growth of black bream when stocked in inland water bodies, with the aim of producing a source for recreational fishing and possibly also a small scale commercial harvest. The main beneficiaries will be Western Australian farmers and property owners who have access to suitable saline water bodies, particularly in regions such as the eastern wheatbelt where widespread land clearing has resulted in rising ground water and consequently an increase in salinisation.

The adoption of the outcomes of the results of this study will increase the opportunities for recreational fishing in the inland regions of the state initially on a private level, but later on a public and commercial scale and as eco-tourism ventures. This will provide flow-on benefits to property owners through farm diversification. Research workers in other states, who are also investigating the potential of inland saline waters for aquaculture, will also benefit from the results of our research. Furthermore, this work, in conjunction with other research being carried out by Murdoch University, which is aimed at increasing black bream growth rates through selective breeding, will facilitate the development of small scale, commercial aquaculture ventures for providing fish for the table market.

The above benefits and beneficiaries are the same as those identified in the original application.

7.0 FURTHER DEVELOPMENT

Further work is required to improve the growth rate of black bream in inland water bodies so that the time between the stocking of juveniles and the attainment of a size suitable for angling is reduced. Faster growth would also increase the potential for commercial aquaculture production of this highly regarded eating species in inland saline waters.

8.0 PLANNED OUTCOMES

Aspects of the current project have been incorporated into a one day short course for C.Y. O'Connor College of TAFE in Northam, which delivers training for participants from throughout the Western Australian Wheatbelt region. This introductory level short course, entitled "Farming black bream", is designed to provide basic information that can be used by farmers interested in black bream aquaculture. It includes a visit to a commercial black bream farm that uses saline groundwater to grow fish for farmers in the region. The project outputs have also assisted in the development of a draft management paper for the Department of Fisheries, which addresses the issue of black bream translocation within Western Australia. When adopted, this policy will greatly simplify the procedure for farmers applying for translocation permits to stock black bream for recreational fishing. Currently, translocation regulations incorrectly classify black bream in WA as "non-endemic" and there is thus no distinction made between black bream and eastern states fish species such as Murray cod and Australian bass. This regulation has thus far been a major factor in inhibiting farmers establishing regimes on their farms for stocking black bream.

Since the WA inland saline aquaculture industry is still in its infancy, the outputs from the current project will assist in its on-going development in promoting the potential of aquaculture for farm diversification in rural areas (and the benefits as described above), many of which are suffering from economic decline through drought and salinisation.

9.0 GENERAL CONCLUSIONS

This project and the previous project (FRDC 97/309) were designed to determine the factors and conditions required for successfully growing black bream in inland bodies, mainly for the purpose of providing recreational fishing opportunities. These studies have demonstrated that black bream is an ideal species for this purpose due to its tolerance of a wide range of water quality parameters, particularly salinity and temperature.

Laboratory and research pond trials undertaken in the current project have also demonstrated that:

- The use of floating fish cages is a useful technique for growing juvenile black bream, particularly when stocked in a water body for the first time. Its benefits reside in the ease with which it enables fish health and feeding activity to be monitored and for fish to be harvested, and its provision of protection from avian predators.
- While naturally occurring feed within research ponds was adequate to ensure survival of juvenile black bream, supplementary feeding was essential to optimise growth.
- Large black bream will prey heavily on adult yabbies when stocked in free-range polyculture.
- In low salinity water bodies, yabbies may provide a semi-sustainable, in-pond food source for black bream providing that the adult yabbies can avoid predation by black bream by being able to seek refuge in hides or burrows.
- The use of a submerged cage to maintain yabbies within polyculture ponds resulted in high mortality, presumably due to poor water quality and cannibalism.

- While the provision of in-water cover, such as wire coils and branches, reduced predation of juvenile black bream in ponds by cormorants, the netting of ponds was the only method that was 100% effective in preventing the loss of juvenile stock through avian predation.
- Supplementary feeding is required for the optimal survival and growth of large black bream in inland water bodies.
- There was no significant difference in the growth of black bream that were fed on alternate days and daily, either when maintained in floating cages or research ponds.
- Poor FCRs, presumably due to uneaten feed and an associated reduction in water quality, are likely to occur in large black bream fed as little as 2% (dry feed: total W.W. of fish) in a single feed event, particularly in cage and small pond culture if fed sinking feed.
- Poor water quality restricts the growth of large black bream, particularly in static, *i.e.* zero exchange, water bodies.
- Cage culture of large black bream is a viable culture technique if water quality is maintained.
- Based on preliminary data, black bream are capable of successfully spawning in inland water bodies, provided the following conditions are present in a specific water body:
 - Saline water with a salinity of at least 10ppt.
 - Water temperature >20°C during the spawning period (late spring early summer).
 - The absence of mosquito fish Gambusia holbrooki.
 - A high density of appropriate live feed, usually copepod species, during the spawning period (September-December).
 - Weed growths are present as refuge for larvae and juveniles
 - Supplementary feed is provided at regular intervals.
 - Algal blooms are absent during the spawning period.
- The types of natural prey found in inland saline water bodies are generally in low abundance and/or inadequate as feed for large black bream.
- The paucity of suitable large prey items for black bream in inland saline water bodies means that supplementary feed must be added to ensure optimal survival and growth.
- Based on growth rates, lipid content and economic data, the Australian produced Ridley Agriproducts was the most suitable for growing juvenile black bream up to a size of approximately 50g.

- The growth and FCR data show that Pivot salmon grower was the most suitable of the diets tested as either a complete or supplementary feed for black bream.
- Commercial heat extruded diets that are significantly more water stable and produce greater growth rates and lower FCR than a cheaper, cold pressed diet after a stabilisation process.
- Different growth rates of black bream that exist between the Swan and Moore River estuary stocks is not replicated by F1 generation black bream, cultured from broodstock collected from these systems, grown under identical environmental and food conditions.

Sarre et al. Factors required for the successful aquaculture of black bream in inland water bodies

10.0 APPENDICES

APPENDIX 1

INTELLECTUAL PROPERTY

The value of the intellectual property will be 62.19% based on PART C of the FRDC project proposal.

APPENDIX 2

STAFF

<u>Full-time</u> Dr Gavin Sarre (Post-doctoral fellow)

<u>Casual</u>

Gavin Partridge (Challenger TAFE) David Tiivel Stan Malinowski William White Linda Schafer Jordan M^C Creery Kim Smith Darin Carter

Volunteers

Gratitude is also expressed to the many volunteers who assisted over the past three years, particularly Dustin M^CCreery, John Sarre, Trevor Turnock, Richard Nuich and Stan Malinowski.