

Biology, Management and Genetic Stock Structure of Mangrove Jack, (*Lutjanus argentimaculatus*) in Australia

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Non-technical summary

1999/122 Biology, management and genetic stock structure of mangrove jack (*Lutjanus argentimaculatus*) in Australia

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Objectives

Investigate the biology of mangrove jack in coastal rivers including habitat preferences, mortality, reproduction, recruitment, and movements;

Determine the genetic stock structure of mangrove jack in Queensland and other parts of their range.

Non Technical Summary

Outcomes achieved

The project has contributed to the ecologically sustainable management of mangrove jack in Australia by providing information on its biology, habitat requirements, level of exploitation and stock structure. The specific outputs are: an improved understanding of the life history of mangrove jack; the current relatively low levels of exploitation in the fishery; the importance of structural habitat and the likely existence of a single genetic stock throughout Queensland.

Mangrove jack (*Lutjanus argentimaculatus*) is one of 65 species of the genus *Lutjanus* and 103 species of the family Lutjanidae. This species is widely distributed throughout the Indo-west Pacific from Samoa in the east to the African continent and in tropical Australia. In northern Australia, it is a highly prized recreational sport and food fish and is also a component of the commercial reef line fishery on the Great Barrier Reef. Mangrove jack are an incidental catch in offshore trawls, particularly in tropical Western Australia. In Australia, technology for mass-rearing hatchery-produced mangrove jack is being developed and in Queensland three freshwater impoundments have been stocked to create 'put and take' recreational fisheries. There is also interest in the mangrove jack as an aquaculture species.

Mangrove jack has a complex life history with distinct inshore and offshore phases. In rivers and inshore coastal areas, mangrove jack are predominantly juveniles and sub-adults while most fish caught in offshore areas are mature. Most spawning occurs in offshore areas, probably in relatively deep water. Post larvae and juveniles then move inshore to rivers and other coastal habitats. The youngest fish caught inshore in this study was a 20 mm long fish from the mouth of the Russell River in late February. Using the daily growth increments on its otoliths, it was estimated to be 32 days old with a hatch date during the previous January. Similarly, the daily increments on the otoliths of other juvenile fish have been used to back-

calculate their hatch dates to the period between October and March. Variability in recruitment of juvenile fish into rivers occurs from year to year. In the wet tropics streams (Daintree, Mulgrave, Russell and Johnstone Rivers), the catch-per-unit effort of juvenile fish less than 100 mm LCF was generally higher in 2001 than it was in 2000. Recruitment into inshore areas occurred in the first part of the year and early juveniles were usually found close to the river mouth, before moving progressively upstream. While most mangrove jack were sampled in the estuary and lower freshwater reaches of the rivers some fish moved well upstream to a distance of about 100 km. The abundance (catch per unit effort) of mangrove jack in rivers decreased with increasing distance from the river mouth. While mangrove jack were found to be associated with a range of habitat types, they were most commonly found on either rocky substrate or snags. Juveniles that are newly recruited into rivers (20-30 mm LCF) were more commonly associated with rocky habitat. In earlier studies, newly recruited *L. argentimaculatus* were sampled in the same tidal and freshwater nursery swamps that were utilised by juvenile barramundi. Activities such as river desnagging and removal of riparian forests could potentially reduce the availability of complex riverine habitat thereby impacting on riverine stocks of mangrove jack.

Tagging studies showed most (73%) mangrove jack were recaptured within a kilometre of their original release location generally after only a short period of time. Of the fish that moved from their river release location, some made intra-riverine movements both upstream (8%) and downstream (14%) while others moved between rivers (1%), along the coast (1%) or offshore (2%). Fish that were tagged in a river and then moved either to the coast or offshore travelled the largest distances, with some fish moving up to 335 km from the original release location. On the east Queensland coast, tagged fish that had moved from riverine locations to offshore areas were generally recaptured in the vicinity of coral reef areas. However, there are records of fish tagged by recreational fishers that have been recaptured from offshore areas in the Gulf of Carpentaria and in the Pilbara region of Western Australia where reef habitats are not as extensive as on the east Queensland coast. The sizes that the tagged fish moved offshore were between 324 mm and 430 mm LCF however the ageing studies suggest that mangrove jack move offshore at ages of between three and eleven years. A series of tag shedding and mortality experiments showed that tagging mortality was low but the shedding rate for anchor tags and dart tags was 17% and 11% respectively for the 92 day period.

Although a small number of fish with developing or ripe gonads were sampled from inshore habitats, most mature fish were sampled from offshore areas. The maturation of both male and female gonads begins in about October, peaks in December and then declines from January through to March. The length at which 50% of fish are mature was 512.3 mm LCF for females and 449 mm LCF for males.

The level of natural mortality ($M=0.158$) for mangrove jack was comparable to values obtained in other studies for other lutjanids in Great Barrier Reef waters. Total mortality was higher in inshore areas where most fishing for this species occurred, than in offshore areas where the fish were less concentrated and where exploitation was lower. There were no differences in fishing mortality between sexes. The fishery for mangrove jack did not appear to be yet fully exploited with the current level of fishing mortality calculated at about a third of optimal level. Growth increments on the otoliths of mangrove jack are annual. Along the Queensland coast where most samples were taken there was evidence of different growth rates. The length-at-age data suggested that fish in the southern areas reached legal size (350 mm TL) earlier than fish in the northern regions. In this study, the oldest fish was 37+ years but the maximum age for *L. argentimaculatus* is possibly more than 40 years. The current minimum legal size protects a high proportion of the mangrove jack resident in estuaries.

Tissue samples for genetic stock structure analyses were collected from three coastal areas of Queensland; southeast Queensland, north Queensland and the northeast Gulf of Carpentaria.

Additionally, samples were collected from the Pilbara region of Western Australia and from the Indonesian islands of Bali, Java and Sumatra and Samoa. The results suggest that mangrove jack in Queensland, and possibly throughout Australia, experience high levels of gene flow and probably belong to the same genetic stock. Permitted translocations in landlocked impoundments are unlikely to adversely affect the stock structure of the species. Limited and controlled riverine translocations for stock enhancement purposes may be acceptable if donor and recipient populations are from the same area.

Keywords

Mangrove jack, *Lutjanus argentimaculatus*, stock structure, habitat, recruitment, mortality, reproduction, ageing, growth.

Acknowledgements

This project would not have been possible without the assistance of the many commercial and recreational fishers who provided biological samples.

The majority of samples for the reproductive and aging studies were obtained from the catches of MV Loray. Thanks to skipper, Ray Walker, for providing access to samples and for generously giving pertinent information on the location and depths that the fish were captured. Ray also provided comprehensive historical catch data on all fish species caught on the boat. The fish processing premises, 'A Fine Kettle of Fish' gave us access to commercial mangrove jack catches prior to them being processed and provided both working space to the research team and freezer storage for samples. Managing Director, Graeme Hopkirk, further assisted by advising of arrival times of shipments and showed great patience in allowing project staff to work on his premises. Thanks also to the filleting staff, particularly Mark Read, for keeping samples when project staff were not available.

Recreational fishers from throughout Australia generously provided biological samples and information for this project. In particular, thanks to the Australian National Sportfishing Association (ANSA) members that assisted in tagging operations. We gratefully acknowledge the assistance given by Bill and Shirl Sawynok (Suntag and InfoFish services). Bill provided the names of many of the contacts that supplied biological samples and information on potential sampling sites. InfoFish services collated all the tagging information for mangrove jack and routinely provided the project staff with an updated dataset of tag information to analyse. Recreational fishers who generously collected genetic samples from remote locations included Tamlin Little, Marl Cottrell, Mick Volp, Peter Rankine, Tom Winkworth, Gary Prerost, Brett Finger, Ben Shorten. We also thank ANSA members that fished the 'Cooktown Barramundi Competition' and 'Hinchinbrook Barramundi Competition' for providing time to collect genetic samples of mangrove jack. We also wish to thank overseas colleagues who supplied tissue samples including Dr Arnil Emata (Philippines), Dr Haryanti (Indonesia) and Mr Kelvin Passfield (Samoa).

Fishing charter operators that provided samples including Terry Holman, Dave Powell, Graham Vallance, Grahame Dingwell, Gary Wright, Greg ('Slippery') Eels, Leonard Todaro and Geoff Taylor.

Thanks also to the fishing tackle shops that acted as drop-off locations for samples. These included Barra Jacks, The Whitsunday Tackle Shack, Bluefin Sports, Tackle World Strathpine, The Tackle Shop, Swan Boat Hire, Got One - Mackay, Northside Fishing Centre, Jones Tackle, Bills Bait Bar, Yeppoon Tackle & Sport, Tweed River Bait & Tackle, Salty's Tackle Shop, Pro Tackle - Townsville, Pat's Bait and Tackle, Northside Fishing Centre, Mossops Tackle Shop, Lure Shop, Jones Tackle, Erskines Tackle Shop, Davo's Bait & Tackle, Cabarita Bait & Tackle, Bills Bait Bar, Barra Bait & Tackle, Anglers Warehouse, Airlie Bait & Tackle and Proserpine Bait and Tackle.

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Otolith samples were sent to James Aumend (James Cook University) for microchemical analysis, this work assisted in confirmation of aging and the movement of this species. Corey Green of the Central Aging Laboratory provided micrographs of oxytetracycline-injected otoliths for inclusion in this report.

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Background

General

In Australia, *Lutjanus argentimaculatus* (Forsskål, 1775) is most commonly known as mangrove jack but is also known as creek red bream, dog bream, purple sea perch, red bream and red perch (Grant, 1975); in Asia it is generally called red snapper. The species has excellent eating qualities and is a prized sportfish particularly in northern Australia (Grant, 1975). Mangrove jack is distributed widely in the Indo-west Pacific from Samoa and the Line Islands to East Africa and from Australia northwards to Ryukyu Island, Japan (Doi and Singhagraiwan, 1993) (Figure 1). It is also believed to have undertaken Lessepsian migrations from the Red Sea via the Suez Canal (Anderson and Allen, 2001) to the Mediterranean coasts of Israel and Lebanon but is not believed to have become well established in the wild in these areas. In east Africa the species is common in mangrove areas, estuaries and sheltered coastal and reef areas (Talbot, 1960). It is commonly found in Mozambique and extends in decreasing numbers to the Transkei (Day *et al.*, 1981). In northern Australia it is distributed from about northern New South Wales to Shark Bay in Western Australia although there are records of mangrove jack being caught as far south as Sydney (Allen *et al.*, 2002). It is a marine species also occurring in brackish estuaries and in the lower reaches of freshwater streams. *L. argentimaculatus* migrates offshore to deeper reef areas, sometimes penetrating to depths in excess of 100 m (Allen, 1985, 1987; Ludescher, 1997). In Vanuatu it is caught on the outer reef slope of the islands at depths to 260 m (Brouard and Grandperrin, 1984).

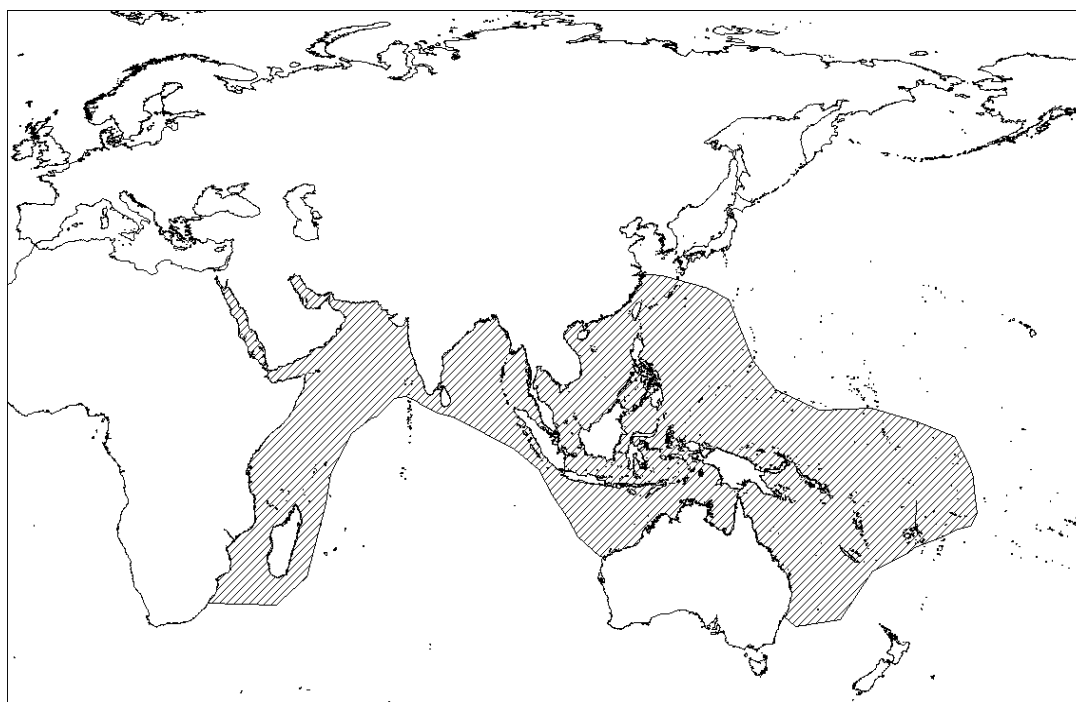


Figure 1. Global distribution of mangrove jack. The species has also translocated into the eastern Mediterranean Sea.

There are 103 species in the family Lutjanidae, of which there are 65 species of the genus *Lutjanus*. Of these 39 species occur in the Indo-Pacific, 9 in the eastern Pacific, 12 in the western Atlantic, and 5 in the eastern Atlantic (Doi and Singhagraiwan, 1993). Of the 17 species of Lutjanids found on the east African coast, the range of all but two extends to the

Australo-Pacific region but in the reverse direction the distribution of the family is not so uniform with many of the species found in the Pacific and eastern Indian Oceans not found in the western Indian Ocean (Talbot, 1960). Talbot (1960) suggests that this indicates a centre of origin, or at least a strong adaptive radiation in the Australo-Pacific region. It is not clear whether the difficulty in dispersal in the east-west direction is due to a paucity of suitable environments in the northern and western Indian Oceans or to some physical barrier (Talbot, 1960).

The spawning grounds for mangrove jack are believed to be offshore (Day *et al.*, 1981; Doi and Singhagraiwan, 1993) and in Palau spawning aggregations were found both in the reef lagoon and on the outer reef slope (Johannes, 1978). Johannes (1978) noted that, in Palau, the timing of spawning activity of *L. argentimaculatus* peaked 14-18 days into the lunar month. Larvae and juveniles subsequently moved inshore and were found in rivers and coastal areas including sea grass beds (Doi and Singhagraiwan, 1993) and also move into freshwater areas (Munro, 1967; Lake, 1971). The extent of their movement into freshwater is generally limited however in northern Australia juveniles and sub-adults have been found 130 km up the Burdekin River and well upstream in the Tully River near its headwaters (Merrick and Schmida, 1984). In northern Australia, juveniles sampled within the estuaries were found to be reproductively immature and smaller than those captured on the reef (Sheaves, 1995). In the Emberly River in the north-eastern Gulf of Carpentaria, *L. argentimaculatus* was more abundant in the middle reaches of the estuary than either upstream or in the lower reaches (Blaber *et al.*, 1989). In their survey of that estuary, Blaber *et al.* (1989) found that mangrove jack was one of 14 species whose juveniles were found only in the estuary and not offshore but they recorded adults both inshore and offshore. In Thailand, *L. argentimaculatus* is an economically important species to coastal aquaculture (Doi and Singhagraiwan, 1994). Juveniles between 16.2 and 31.2 mm TL were sampled in set nets in the estuary of the Prasae River between November and January inclusive (Doi and Singhagraiwan, 1993; Doi *et al.*, 1998). In the same area there is evidence from the occurrence of juveniles that some spawning occurs at other times including March and April (Doi and Singhagraiwan, 1993). In the Philippines, *L. argentimaculatus* were sexually mature from February to December with the peak of maturation from April to October (Emata, 1996). Tagging studies in Thailand have shown that juvenile fish moved inshore towards the coast and into estuaries from March to August and away from the estuaries to coastal or offshore areas from September to February (Doi and Singhagraiwan, 1993). In northern Australia Davis (1988) suggests that while *L. argentimaculatus* is a dominant species in the tidal Leanyer Swamp, they are transient with juveniles using the upper estuary and swamp as a nursery. He found that the numbers of juvenile *L. argentimaculatus* entering the swamp were correlated with the environmental parameters month and tidal height. Higher tides provide greater assistance for the upstream movement of juvenile fish and also enabled them to penetrate further into upstream areas (Davis, 1988). Similarly, in eastern Australia Sheaves (1995) suggests that estuaries are important development grounds for *L. argentimaculatus* and that estuarine populations appear to consist entirely of immature fish. This is supported by Day (1981) who noted that *L. argentimaculatus* seldom attained lengths of more than 400 mm in estuaries.

In a study of the development of swimming and feeding functions of *L. argentimaculatus*, Doi *et al.* (1998) suggest that juveniles larger than 16 mm TL would have acquired swimming or cruising ability strong enough to migrate from offshore spawning grounds to coastal waters and river estuaries. Juvenile fish appeared to move into the estuary after the wet season and they suggest that the upstream movement of *L. argentimaculatus* is governed by freshwater runoff resulting from high seasonal rainfall. They caught no *L. argentimaculatus* larger than 230 mm TL and these authors suggest that by this age fish have moved offshore and into coastal waters. Adults are often found in groups around coral reefs (Anon, 2002). Eventually they migrate further offshore to deeper reef areas, sometimes penetrating to depths in excess of 100 m (Anon, 2002). Mangrove jack appear to be a long-lived species. In northern Australia Sheaves (1995) found that *L. argentimaculatus* (n=22) displayed up to 32 annuli on

their otoliths with growth appearing to slow in older fish. In 276 estuary samples Sheaves (1995) counted between 0 and 8 annuli. *L. argentimaculatus* grows to in excess of 120 cm total length (Doi and Singhagraiwan, 1993) and 8.5 kg but is more commonly found up to 80 cm long (Anderson and Allen, 2001). Numerically, *L. argentimaculatus* do not generally feature significantly in estuarine fish surveys, however they may still make a notable contribution to the total biomass. For example, in a survey of the estuarine fauna of Alligator Creek in north Queensland, mangrove jack was one of six species that, combined, accounted for more than 25% of the total community biomass (Robertson and Duke, 1990a). The diet of *L. argentimaculatus* has generally been accepted to be dominated by mangrove-associated fishes (Robertson and Duke, 1990b). They also found that crabs, particularly those from the genus *Sesarma*, made up more than 40% of the volume of prey items in the stomachs of 10 fish between 79 and 460mm long. In another study of the diet of mangrove jack in three estuarine systems in northeast Australia, sesarimid crabs were the dominant food items occurring in 50% of the stomachs that contained prey and being the most common prey in terms of overall numbers (Sheaves and Molony, 2000). They suggest that a substantial part of the mangrove productivity sequestered by sesarimid crabs may be exported from mangrove ecosystems as a result of offshore migration by mangrove jack. They also suggest that the low incidence of piscivory in these fishes adds support to the theory that reduced predation pressure may enhance the nursery ground value of tropical mangrove systems for fishes. In South Africa, *L. argentimaculatus* are an invertebrate predator commonly feeding on crabs and prawns (Day *et al.*, 1981). *L. argentimaculatus* can occur singly or in shoals of up to 20 fish (Talbot, 1960) although there have been reports of much larger schools being caught in trawls in the Gulf of Carpentaria (Suntag recreational fishing database, 2002).

Aquaculture and stock enhancement

In southeast Asia, *L. argentimaculatus* is an important food fish which is cultured in brackish water ponds and marine cages (Emata *et al.*, 1999). In Thailand, there has been some preliminary work on stock enhancement of *L. argentimaculatus* by releasing fish onto artificial reefs, although little information is available on the success of these releases (Doi and Singhagraiwan, 1993). In Australia the technology for mass production of mangrove jack fingerlings is still being developed and it is expected that both the numbers of fish being stocked and the number of locations will increased as it is refined. Small numbers of hatchery reared mangrove jack have already been stocked into Queensland impoundments to create put and take fisheries. Initial trials were conducted in an impoundment closed to fishing, Lake Morris, to the west of Cairns. The first batch of 2655 mangrove jack with a mean length of 27.5mm were stocked into Lake Morris in December 1998 and a second batch of 1325 fish with a mean length of 86.5 mm was stocked in June 1999 (T. Vallance, QDPI, pers. comm.). While this trial provided information on the survival and growth of mangrove jack in an impoundment it did not address the issue of the creation of mangrove jack recreational fisheries in freshwater impoundments. To address this, permits were issued to community stocking groups to release mangrove jack into two public impoundments; Awoonga Dam near Gladstone (under the Gladstone Area Water Board) and Lake Tinaroo on the Atherton Tablelands. In Lake Tinaroo, three batches of mangrove jack fingerlings have been stocked; one batch of 181 fish in the 1999/2000 season and two batches totalling 4297 fish in the 2001/2002 season (M. Pearce, Queensland Department of Primary Industries, pers. comm.). Up until June 2002, six mangrove jack had been caught in Lake Tinaroo. All of the fish caught were from the first batch of fish released in 1999/2000 and two were longer than the minimum legal length of 350mm. Recent electrofishing surveys in Lake Tinaroo found a number of the 2001/02 stocked fish adjacent to a feeder creek in rock habitat (September 2002) (M. Pearce, Queensland Department of Primary Industries, pers. comm.). Mangrove jack were stocked into Awoonga Dam in central Queensland in 2000/01 (180 fish) and 2001/02 (4013 fish) but there have been no recaptures up to June 2002. Recently another permit has been issued to release up to 6300 mangrove jack per year for three years into Aplin

weir on the Ross River near Townsville but, as yet, no fish have been stocked (M. Pearce, Queensland Department of Primary Industries, pers. comm.).

Fishery

L. argentimaculatus is an important market species throughout the Indo Pacific region (Anderson and Allen, 2001) and is a major recreational species throughout its range in northern Australia and parts of Asia and the Pacific. It is favoured by recreational fishers as both a sports fish and because of its excellent eating qualities (Lake, 1971; Grant, 1975). In northern Australia it is rarely taken in inshore commercial gill nets and arrow head fish traps (Grant, 1975) but is caught by commercial line fishers on the Great Barrier Reef and also occasionally features in otter trawl catches, particularly in the Gulf of Carpentaria. It is never found in large quantities (Anderson and Allen, 2001). In Thailand, because of the high demand for seed for coastal aquaculture, fishers collect the juveniles of *L. argentimaculatus* which are then stocked into coastal fish cages (Doi and Singhagriwan, 1993). Thai fishers use a variety of techniques for capturing these juveniles including push nets, river set nets and scoop nets (Doi and Singhagriwan, 1993). In Vanuatu, bottom fishing for *L. argentimaculatus* is undertaken on the outer reef slope in depths ranging from 100 to 400m. Mangrove jack is abundant in east African coastal waters and is an important market species (Talbot, 1960). In the western central Pacific, the FAO year book of fishery statistics reports that between 1990 and 1995 the yearly catch ranged from around 4,300 to 12,700 tonnes (Anderson and Allen, 2001).

The catch statistics for mangrove jack in Australia are patchy and incomplete. In Queensland the annual commercial production is only 2.3 t (Queensland Fisheries Service, CFISH database) however it is believed that this is an underestimate as considerable quantities are reported under other product categories including mixed reef fish. The commercial catches in the Northern Territory (16.5 t, Steve Wilmore, Primary Industries and Fisheries NT, pers. comm.) and Western Australia (10.6 t, Peta Williamson, Western Australian Department of Fisheries, pers. comm.) are larger in size. In Queensland the recreational catch appears larger than the commercial catch. A 1999 phone survey of recreational fishers estimated the total annual Queensland catch at 52 t (Queensland Fisheries Service, RFISH database). There are no official estimates of recreational catches in the other States although mangrove jack are the 6th most important recreational species in Western Australia (Peta Williamson, Department of Fisheries, Western Australia, pers. comm.). Commercial fishing charter boats in Queensland catch about 1.4 t of mangrove jack per annum (Queensland Fisheries Services, CFISH database).

Currently the State fisheries regulations governing recreational fishing for mangrove jack are not uniform. For example, in Western Australia there is a bag limit of 8 “bread and butter” species including mangrove jack and no minimum size limit (MSL) for mangrove jack. This differs from the Northern Territory, where there is a bag limit of 30 non-regulated species (including mangrove jack) and no MSL and New South Wales, where the bag limit is five mangrove jack and no MSL. In Queensland, there is a MSL of 350 mm total length (TL), with no bag limit on the east coast and a bag limit of 5 fish in the Gulf of Carpentaria.

The MSL for Queensland was introduced in the early 1990’s after concern was expressed by the ReefMac (Reef-fish Management Advisory Committee) about over-exploitation of fish stocks. After reviewing the current state of knowledge of this species, a Technical Working Party recommended an MSL be set at 350 mm TL. The recommended MSL, which was subsequently adopted, was seen as a compromise to balance the perceived high exploitation of mangrove jack in the inshore fishery with under-exploitation in the offshore fishery. The 350 mm TL MSL both protected juvenile fish and allowed recreational fishers the opportunity to catch mangrove jack in the inshore fishery (Geoff McPherson, Queensland Fisheries Service, pers. comm.).

Need

Mangrove jack are an essential component of Queensland recreational and commercial fisheries, being ranked second to barramundi as a target for recreational anglers (Russell and Rimmer, 1997) in tropical rivers and estuaries. Community stocking groups are keen to stock mangrove jack to create recreational fisheries in impoundments and to address perceived declines in natural riverine stocks. Despite this, knowledge of the biology of the species is incomplete and a genetic assessment of population structure has never been attempted. The efficacious management of natural populations of mangrove jack and the future development of effective stocking programs requires information on all aspects of their life cycle, instream habitat requirements, reproduction and maturation, mortality, recruitment into and emigration from river systems.

Managers of wild fish species accept that genetically distinct sub-populations of fish may possess novel genetic, physiological, behavioural and other characters that lead to distinct differences in life-history traits including growth rates, fecundity, disease resistance and abundance (Gold and Richardson, 1998). These differences theoretically contribute to the long-term adaptability, survival and resistance to human-induced or other environmental perturbations and can be jeopardised by inappropriate management. It would be imprudent to proceed with the expansion of stocking and aquaculture programs for this species without comprehensive knowledge of the genetics of the populations. The genetic study will pre-empt potential conflict between management agencies and commercial and recreational interests. Information from the genetic analysis of the species will provide geographic detail for the sustainable use of the resource across State boundaries where differing fishing regulations exist.

Objectives

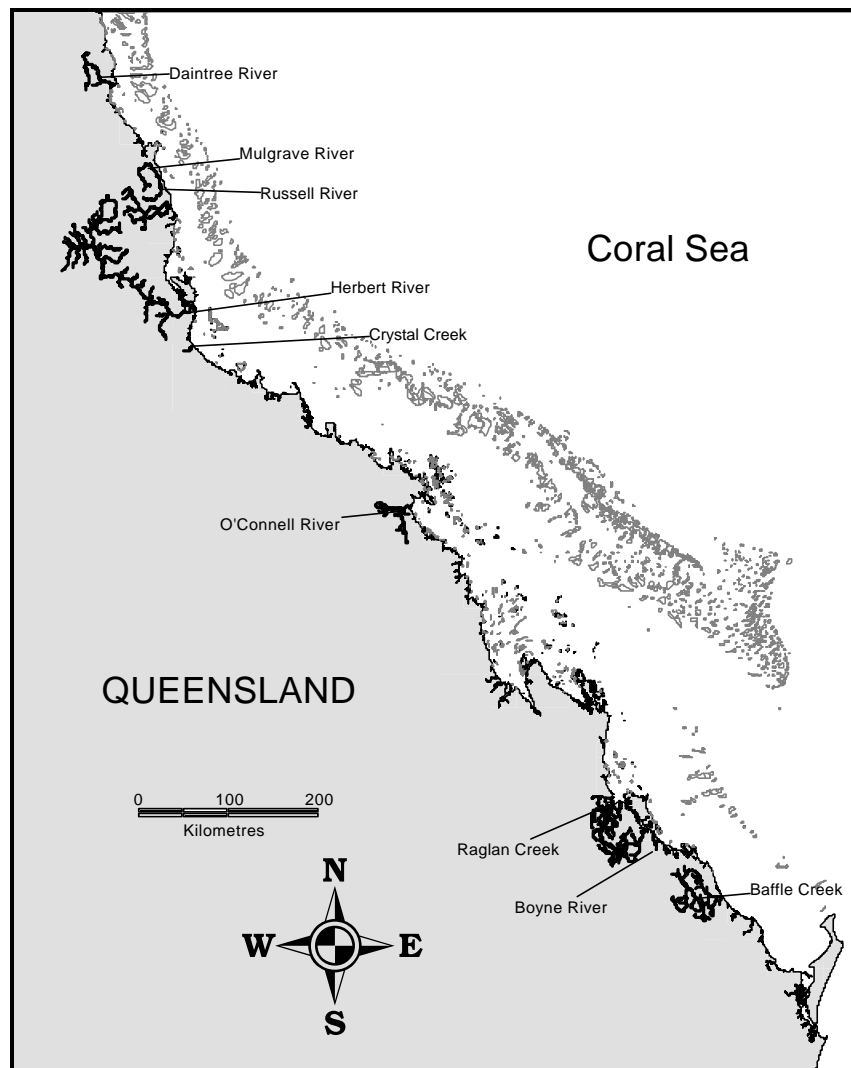
Investigate the biology of mangrove jack in coastal rivers including habitat preferences, mortality, reproduction, recruitment, and movements;

Determine the genetic stock structure of mangrove jack in Queensland and other parts of their range.

Methods

Study Sites

Locations at the tidal influenced brackish-freshwater interface were selected as monthly sampling sites in four north Queensland rivers, and quarterly sampling sites were selected in rivers through Central and Southern Queensland (Map 1). Criteria for these choices included; distance to river mouth, habitat diversity, tidal range and flow, seasonal salinity levels and vessel access. Seasonal river flow levels, and resultant salinities influenced access and electrofishing effectiveness particularly during periods of dry weather. Three replicates at each site, each typically 300m in length were selected. This distance allowed an average 1500 seconds electrofishing time which was found to be sufficient to capture a minimal number of fish. The boundaries of sites were identified by familiar bank side structures that were recorded by GPS for repeat visits. Where possible, a site on the brackish interface and an upstream site in each river were used to investigate within-river movement.

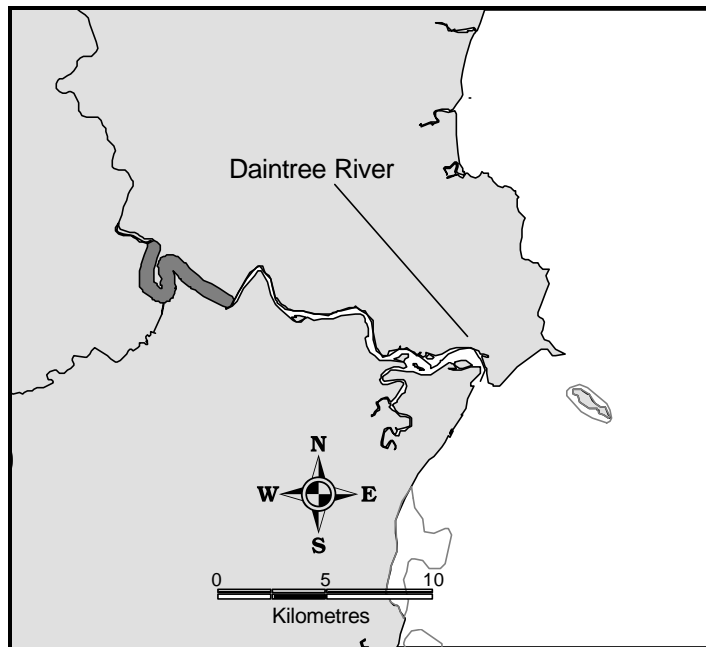


Map 1. Location of study areas in Queensland.

Map 1 shows the locations of the main study locations where mangrove jack were sampled either monthly or quarterly using electrofishing techniques. Detailed descriptions of these locations follow:

Daintree River

The Daintree River, which is the northern most catchment, has its source in the Main Coast Range and is surrounded by the Thornton and McDowall Ranges and drains a small coastal plain before discharging into the Coral Sea (Map 2). These ranges are mostly rainforest covered, although there are some areas of dry sclerophyll in the west of the catchment. The total catchment area is about 1342 km² and the township of Daintree (estimated population of 100) is its only significant population centre. The mean annual river discharge is about 907 000 ML with an average annual rainfall runoff of 997 mm (N. Searle, Department of Natural Resources and Mines, pers. comm.). Most of the catchment is included in the Wet Tropics World Heritage area (Russell *et al.*, 1998).



Map 2. Daintree River.

Cross-hatched area shows location of study sites in.

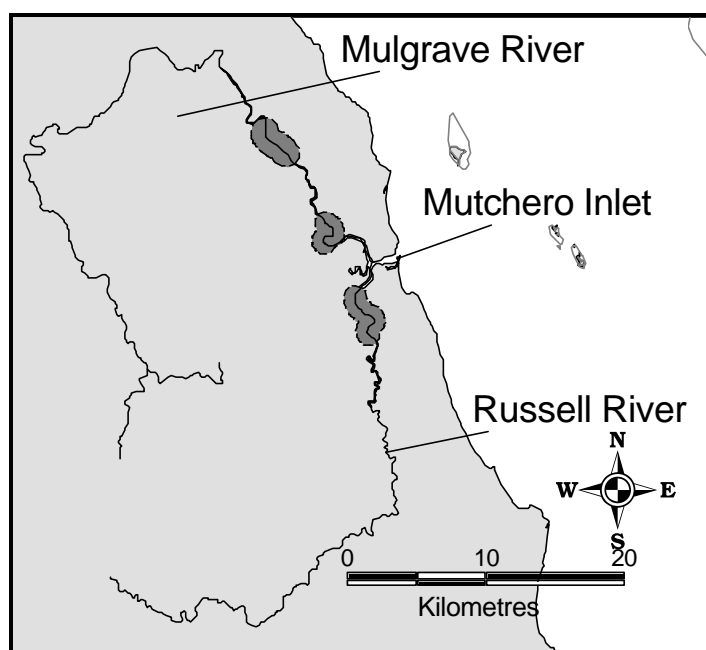
Russell and Mulgrave Rivers

These are perennial waterways that drain the coastal ranges and alluvial flood plains to the south of Cairns. The two rivers flow into Mutchero Inlet before discharging into the Coral Sea through a common mouth (Map 3). The alluvial plain varies in width from about 3 to 13 km and is bounded on the west by the Bellenden Ker Range, to the north and west by the Lamb Range, to the east by the Malbon Thomson and Graham Ranges and to the south by the Francis Range. The Bellenden Ker Range contains Mt Bartle Frere and Mt. Bellenden Ker, the two highest mountains in Queensland. Total catchment area, for both rivers, is approximately 1,370 km². The Russell River is approximately 65 km long and has a total catchment area of 560 km². It rises in the Bellenden Ker Ranges and on the Atherton Tableland and has a mean annual discharge of about 940,000 ML (Department of Natural Resources and Mines, pers. comm.). The Mulgrave River also rises in the Bellenden Ker

Ranges and has a total catchment area of 810 km². Mean annual average discharge of the Mulgrave River is about 770,000 ML (Department of Natural Resources and Mines, pers. comm.). The majority of the catchment is rugged mountain range and remains in a relatively pristine state. Approximately 66% of the total area of the catchment is protected as part of the World Heritage Estate. Much of the coastal plain and some tableland areas have been developed for agriculture. While sugar cane farming is the major agricultural activity on the coastal belt, bananas, pawpaws and exotic tropical fruits are also grown. Tea, pastures and grazing are the predominant agricultural activities on the Tablelands. The catchment has one of the wettest climates in Australia with most of the rain falling during the warm summer months. Rainfall statistics for the two major townships in the catchment, Babinda (17°21'S, 145°55'E) and Gordonvale (17°06'S, 145°47'E) are given in Table 1.

Table 1. Annual rainfall statistics for Babinda and Gordonvale Post Offices.

<i>Location</i>	<i>Period</i>	<i>Average (mm)</i>	<i>Maximum (mm)</i>
Babinda (Russell Catchment)	1911-1994	4,175	6,714
Gordonvale (Mulgrave Catchment)	1963-1994	1,921	3,219



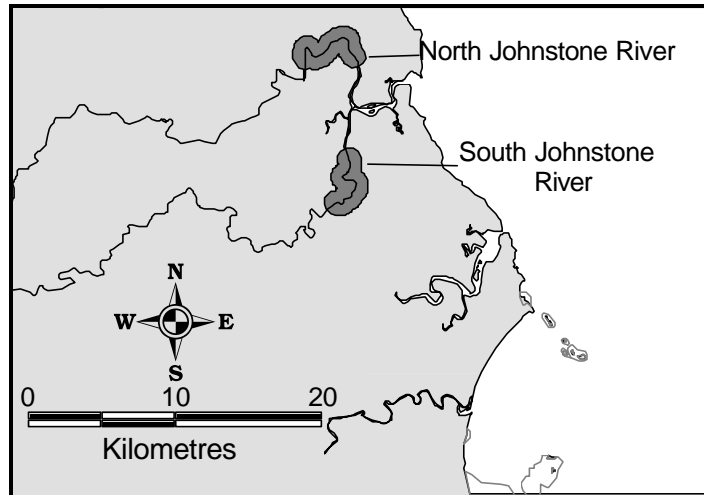
Map 3. Russell and Mulgrave River sites.

Cross-hatched area shows location of study sites.

Johnstone Rivers

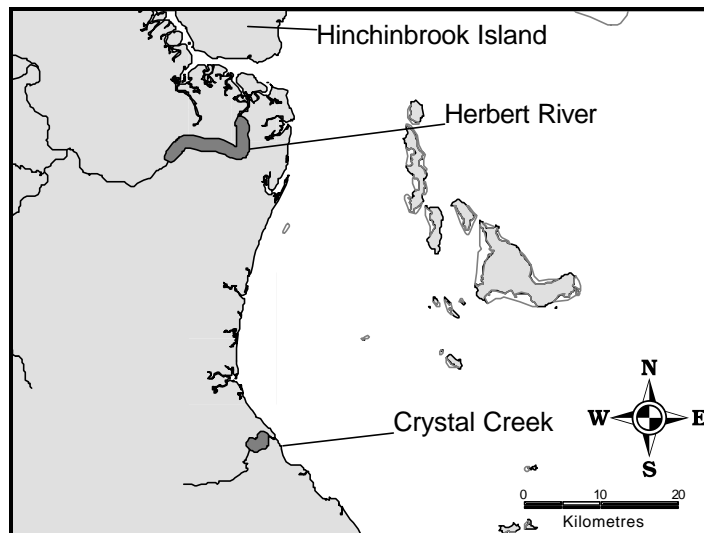
The Johnstone River flows into the Coral Sea near the sugar town of Innisfail (17° 32'S, 146° 02'E). The river bifurcates about 5 km from the mouth into the North and South Johnstone Rivers, both of which have their sources on the Atherton Tablelands (Map 4). The highest part of the catchment is on the Atherton Tablelands and is 1385 m above sea level. The Atherton Tablelands and the coastal plain are separated by steep, heavily forested uplands

where the river flows through deep gorges and is characterised by the presence of rapids and waterfalls. The Johnstone catchment, which has an area of approximately 1634 km², has an annual average rainfall of between 1690 and 3970 mm. Average annual runoff was 2.7 million ML (Department of Natural Resources and Mines, pers. comm.). Land use is varied throughout the catchment. On the Tablelands dairying is the predominant landuse while sugar and banana farming are the main agricultural activities on the coastal belt. About 43% of the catchment, mainly the coastal range, is protected under World Heritage listing. In 1991/92 agricultural production in the catchment was estimated to be worth \$170 million.



Map 4. Johnstone River sites.

Cross-hatched area shows location of study sites.



Map 5. Herbert River and Crystal Creek sites.

Cross-hatched area shows location of study sites.

Herbert River

The Herbert River flows into the Coral Sea near the southern end of Hinchinbrook Island near the small communities of Halifax and Lucinda (Map 5). The total catchment area is 9843 km²

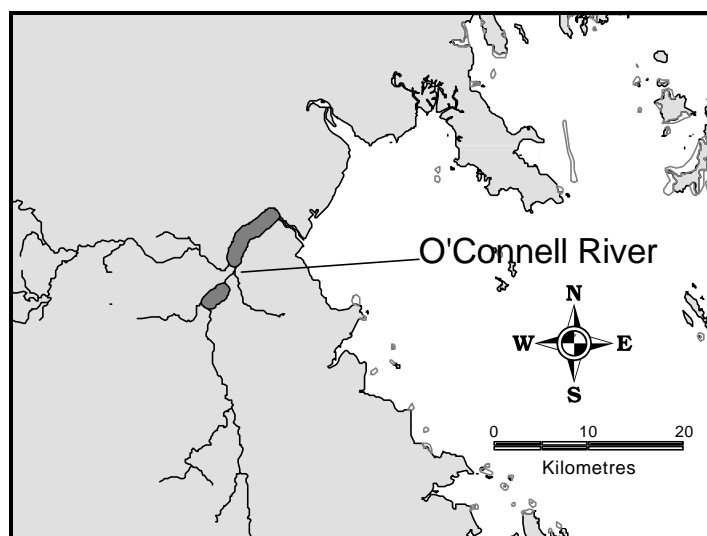
with an average annual runoff of 407 mm/m² and a mean annual discharge of 4 km³ (GBRMPA, 2001). The nearest large population centre is the sugar town of Ingham (18°39'S, 146°11'E) which has an average annual rainfall of 2122.2 mm. The major landuse in the catchment is grazing (7330 km²) with most of the narrow coastal plain under intensive agriculture predominantly sugar cane farming (691 km²)(GBRMPA, 2001). Electrofishing activities were carried out around the upper limit of tidal influence.

Crystal Creek

This small coastal creek drains the Paluma Ranges into Halifax Bay (Map 5). Flows are considerably reduced or cease during the dry winter and spring months. Reduced freshwater flows during these periods resulted in increased salinity and either reduced the effectiveness of, or prohibited the use of electrofishing apparatus at the designated sampling sites.

O'Connell River

The O'Connell River drains into Repulse Bay about 22 km to the south of the sugar town of Proserpine (20°27'S, 148°35'E) in central Queensland (Map 6). The river has a mean annual discharge of 1.5 km³ and an annual runoff of 645 mm/m² (GBRMPA, 2001). The catchment is relatively small (2387 km²) and is 51% cleared. Grazing is the major landuse (1904 km²) followed by sugar cane farming (264 km²). The river is shallow and sandy and during the dry season a series of temporary sand dams are constructed in its lower reaches to provide irrigation water and also limit the penetration of tidal waters.



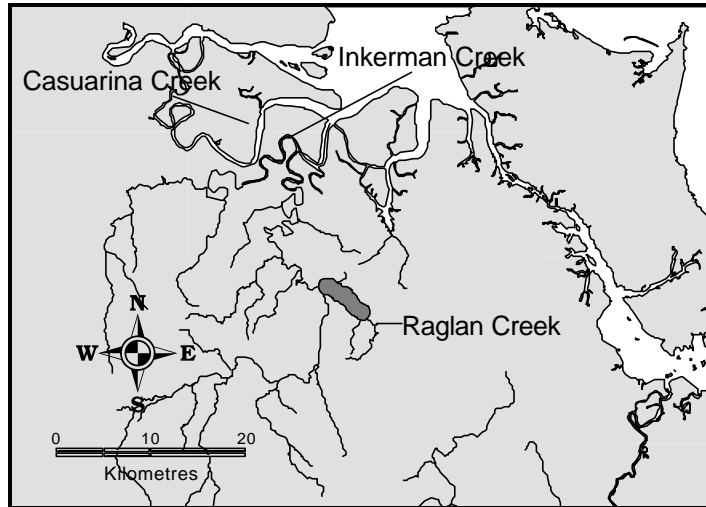
Map 6. O'Connell River sites.

Cross-hatched area shows location of study sites.

Upper Raglan Creek

Raglan Creek flows into the delta of the Fitzroy River at Port Alma to the south of Rockhampton (Map 7). The Fitzroy River, with a catchment of 140,000 km², is the largest river draining central Queensland and has an average annual discharge of 5220 million cubic metres (Australian Water Resources Council, 1976). A barrage has been constructed across the river close to the city of Rockhampton, about 60 km upstream from the mouth. Much of the tidal reach of Raglan Creek has a thin fringe of mangroves that backs onto extensive salt

pans. The substrate is mainly mud and maximum tidal amplitude is around 5 m. Sampling was undertaken in a water hole in the upper reaches which is only connected to the main creek in large high tides or during periods when there is freshwater discharge.

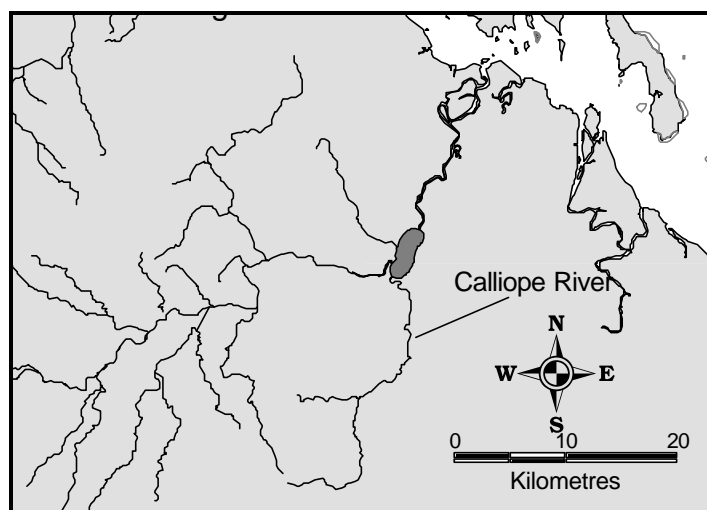


Map 7. Raglan Creek sites.

Cross-hatched area shows location of study sites

Calliope River

The catchment area of the Calliope River is approximately 2150 km² of which 83% has been cleared for agriculture (GBRMPA, 2001). Grazing is the major landuse covering about 2032 km² (95%) of the total catchment area. The river has a mean annual discharge of 0.3 km³ and a runoff of 134 mm/m² (GBRMPA, 2001). It originates in the Calliope Range approximately 100 km to the east of its mouth near the port city of Gladstone (Map 8). The river has no artificial stream barriers that would inhibit fish movements (McKinnon *et al.*, 1995).

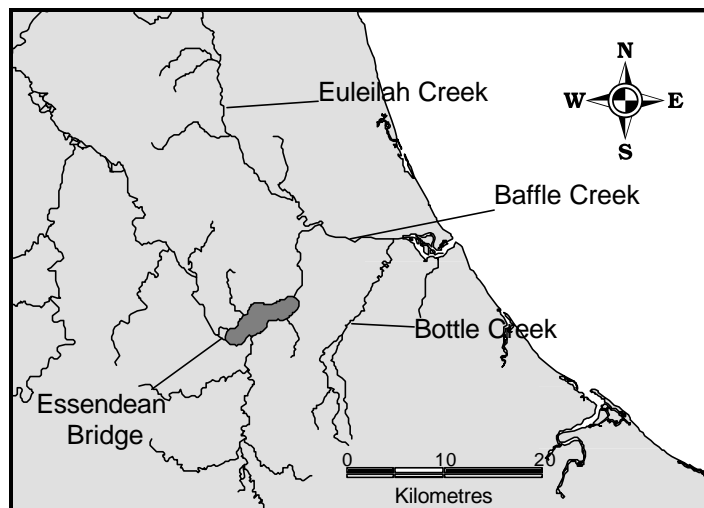


Map 8. Calliope River.

Cross-hatched area shows location of study sites.

Baffle Creek

Baffle Creek drains into the Pacific Ocean on the central Queensland coast about 44 km north of the sugar city of Bundaberg (24°52'S, 152°21'E) (Map 9). Two small fishing communities, Boaga and Winfield, are established on the southern side of the estuary. The catchment has an area of about 3996 km², a mean annual discharge of 0.8 km³ and an average runoff of 195 mm/m². The predominant landuse in the catchment is grazing (87% of total catchment area) but other minor landuses include sugar cane farming (14 km²) and horticulture (8 km²) (GBRMPA, 2001). State Forests and Timber Reserves occupy 477 km² and protected areas cover 214 km² of the catchment. It has a wide sandy estuary which narrows to a small mouth but the upper estuarine and brackish reaches contain isolated rock bars and the channels are deeper and slower moving than the lower estuarine areas. The bar of this creek is exposed to south-east trade winds and also experiences strong tidal currents. The maximum tidal amplitude is about 3m and tidal influence extends 32 km upstream to the Essendean road bridge (24°31.06'S, 152°03.23'E). This is also the upper limit of where sampling with the electrofisher was undertaken and, depending on seasonal variations in the salinity of the river, extended up to 12 km downstream. The Baffle system is one of the few remaining waterways in the Wide Bay-Burnett region that has no weirs, barrages or other artificial structures that disrupt fish migrations, regulate flows downstream or interfere with tidal flow (Lupton and Heidenreich, 1996).



Map 9. Baffle Creek.

Cross-hatched area shows location of study sites.

Field sampling techniques

Habitat Assessment

Analysis of in-stream and riparian habitat for each site was conducted according to a proforma used by Russell (2000), as presented in Appendix 2. Percentages of habitat categorised as grass, macrophyte, overhanging vegetation, rock, root and snag were estimated for each replicate. In addition other factors such as stream width, depth, width of riparian vegetation, presence of mangroves and level of disturbance and sedimentation were also recorded. GPS co-ordinates for replicate start and finish locations were recorded using a Garmin™ GPS12 unit.

Water Quality

Recordings of ambient water quality parameters at the time of electrofishing surveys were taken at a depth of 0.5m with a Horiba @U-10 meter. Parameters recorded were pH, Conductivity (mS/cm), Temperature (°C), salinity, Turbidity (NTU) and Dissolved Oxygen (DO mg L⁻¹). During periods of higher salinities, additional readings were taken at the bottom of the water column to confirm if electrofishing would be efficient.

Electrofishing

A 4.3m electrofishing boat equipped with a Smith-Root® Model 7.5 GPP generator was used as the survey vessel, using a pulsed direct current. Conductivity at the site determined the settings required for efficient operations, with voltages ranging from 135-1 000 V DC. At each site, the vessel was manoeuvred upstream slowly covering the area adjacent to the bank.. The effective stunning radius and depth were approximately 5 metres and 3 metres respectively, although these distances decreased with increasing salinity. The boat was manoeuvred such that the anodes were always in close proximity to suitable habitat. All *L. argentimaculatus* were netted with 3 metre-long handle dipnets, and placed in a 100L recirculating live tank onboard the vessel. Some stunned mangrove jack were carried by any stream flow and may have moved out of the effective stunning range before they could be netted. These fish were recorded on the datasheet with an approximate size. Captured fish were measured and assigned to one of the following niche types; Snag, overhanging vegetation, roots, rock, macrophyte or grass. At the completion of each replicate, all fish were measured, weighed, tagged and released.

Tagging

For *L. argentimaculatus* between 120 and 300mm, a Hallprint type TBF-2 (45mm) fine anchor T-Bar tag was inserted between the pterygiophores of the secondary soft dorsal fin rays using an Avery Dennison Mark III tag applicator. For fish greater than 300mm a Hallprint type PDT (85mm) dart tag was inserted between the posterior pterygiophores of the first dorsal fin spines using a hollow tag needle. All tagging was done on the left-hand side. In all tagging operations, a scale was lifted from the insertion point, the needle was depressed through the flesh until the point passed through the pterygiophores of the dorsal rays and the needle was withdrawn. Tags were then pushed into the flesh and then withdrawn slightly to ensure correct application. Where the first tag was either not placed correctly or was broken, a second tag was inserted to the rear of this location.

To help to determine the rate of tag loss, tag-shedding experiments were conducted in ponds in Townsville and Walkamin. Some double-tagging of wild fish was conducted in Crystal Creek and the Barron River to attempt to compare shedding rates with the pond experiments.

Measuring and Weighing

Fish were placed on an on-board measuring board associated with the live tank. This board was kept continuously moist to ensure minimal loss of slime on fish. Total fish length (TL) and fork length (LCF) in millimetres was measured for the first 1500 fish captured to produce a correlation to be used to correct recreational fisher recapture lengths. After this time only LCF was recorded.

Whole wet weight of live specimens was measured to 1gm using an Arlec (Model DS102) portable digital balance with a custom-built PVC measuring pan which included a moistened towel to facilitate fish health by reducing slime loss. Where fish were larger than 2 kg, 5 kg Salter® hanging spring balances were used.

Scales

On the first occasion a *L. argentimaculatus* was captured and tagged, a couple of scales were removed by fine forceps from the left-hand side of the body, from an area covered by the pectoral fin. In recaptured fish, scales were removed from the right hand side of the fish and a check was made that these were not regrowth scales. Samples were also obtained from reef size fish collected by commercial fishers. All scales were stored in small paper envelopes, marked with tag numbers or specimen numbers, air-dried and stored.

Otoliths

Otoliths were removed from most fish that were sampled from the commercial and recreational fishery or were sacrificed in the research program. A full description of the process of sampling, sectioning and reading otoliths is given in the Age and Growth chapter. Otoliths were also supplied to a post graduate student at James Cook University to determine if microchemical analyses could be used to identify inshore juvenile habitats (see Appendix 3).

Euthanasia and Anaesthetics

Mangrove jack specimens required for laboratory studies were pithed via a knife wound through the roof of the buccal cavity penetrating the brain cavity followed by severance of the thoracic artery. Fish were then placed in an ice slurry to reduce metabolism until they could be dissected. Handling of fish during transport or injection was aided by use of AquaS™ or Benzocaine solution at recommended rates.

Animal ethics

Animal ethics approvals were obtained for all field-sampling activities.

Laboratory techniques

Samples

Specimens of *L. argentimaculatus* were collected by researchers, commercial and recreational fishers from various locations within Australia and overseas. Material supplied included, whole fish, frames, heads or fin clips for genetic analysis. Fish frames were generally frozen and stored at NFC at -21°C. Weekly samples of fresh frozen *L. argentimaculatus* from the GBR adjacent to Cairns were supplied by the seafood processor, “A Fine Kettle of Fish” (FKOF).

Fish frames and heads were defrosted in running water and dissected at NFC. Total fish length (TL) and fork length (LCF) Opercular length (OL) was recorded and the sample dissected to retrieve otoliths, gonads, scales and genetic material.

Reproduction

Gonad tissue, including the associated fat body attaching the gonad to the swim bladder and liver, were dissected from the body cavity. A stage of development was assigned to each gonad using characters according to Davis (1982). Fat and gonad were separated and weighed on a Mettler PC 4400 balance to nearest 0.01gm. Gonad tissue was fixed whole in 10% formal saline (100ml 38% formalin, 450ml distilled water, 450ml 30µm filtered seawater) prior to sectioning. Larger gonads were cut or punctured to allow influx of fixative. Medial sections (1cm² by 3mm thick) of gonads were cut with a scalpel and stored in a labelled histocassette in 10% formal saline fixative.

Staining and mounting of 3µm sections using Lillies-Mayers Haematoxylin and Eosin with Phloxine was completed by DPI Animal & Plant Health, Oonoonba Veterinary Laboratory and Queensland Health Pathology and Scientific Services, Cairns Base Hospital (QHPSS).

Medial, dorsal and proximal sections for a number of gonads were initially sectioned for comparison of development within a gonad but medial sections were deemed suitable for further investigations. Staff at NFC analysed these sections using a Leica™ DMLB bright field-illuminated microscope. The quality of section, percentage of cell types and ranking of maturity was recorded in a MS Access database. For mature stage females, a picture of approximately 15 developed oocytes was taken using a Leica™ digital camera. Measurement of maximum oocyte diameter for late stage MNS and hydrated oocytes was done using UTHSCA® *Image Tool* for Windows Ver.2.

Genetic samples

A 5mm long section of a soft fin ray of the secondary dorsal fin was removed with a pair of fine point scissors and stored in a numbered vial containing DSMO fixative. Samples were kept cool and refrigerated when returned to the laboratory. Samples were collected from both live specimens and from dead/frozen samples dissected in the laboratory. These samples were then sent to the Genetics laboratory at the Southern Fisheries Centre (SFC) for analysis. A field kit containing instructions, scissors, forceps and datasheet was given to recreational fishers helping to collect samples from isolated or estuarine habitats.

Data Analysis

Field and laboratory data were collated in a MS Access® database, with graphical representation of data plotted using MS Excel® and SigmaPlot®. All tagging data was incorporated into the Suntag database. Summaries of catch information for mangrove jack at each site are given in Appendix 4.

Movements

Introduction

Movement studies of snappers have been confined largely to a few species that inhabit tropical reefs although there have recently been a number of studies on red snapper (*Lutjanus campechanus*) in the south eastern United States. On the central Great Barrier Reef, Cappo *et al.* (2000) used a tagging study to validate the periodicity and timing of the formation of opaque bands on the otoliths of 11 species of *Lutjanus*, including a small number of *L. argentimaculatus*. A variety of tag types, including acoustic tags (Eristhee *et al.*, 2001), coded wire and visual elastomer tags (Brennan *et al.*, 2001) have been successfully used to track the movements of a number of species of *Lutjanus*. The acoustic tags were used to track *L. mahogany* over coral reefs in the Caribbean Sea. In the southern United States, Fable (1980) tagged and released 299 *L. campechanus*, of which 17 (5.6%) were subsequently returned. He noted that only a small number had moved and it was usually to adjacent banks or snags. In a larger study of the movements of *L. campechanus*, Patterson *et al.* (2001) found fish moved up to 352 km from their release location on artificial reefs. Of the nearly 3000 fish released, 561 were subsequently recaptured with liberty times of up to 1501 days. They noted that these movements were much greater than what has previously been recorded and speculated that these may facilitate stock mixing in the northern Gulf of Mexico. Similarly, another study Watterson *et al.* (1998) documented movements of red snapper on a spatial scale that would facilitate stock mixing and implicated the occurrence of large scale climatic events such as hurricanes in the stock mixing dynamics. Experiments have also been conducted releasing hatchery-produced red snapper in waters off the southern United States (Ogle *et al.*, 2001). Studies in northern Australia have documented the distribution patterns of a number of inshore fish including *L. russelli* and found that the movements of this species were restricted to within 40 km of the release location (Sheaves, 1993; Sheaves, 1995). Movement studies involving tagged juvenile *L. argentimaculatus* have been undertaken in Thailand (Doi *et al.*, 1992; Doi and Singhagraiwan, 1993). These studies have shown that juvenile fish moved inshore towards the coast and into estuaries from March to August and away from the coast or offshore from September to February (Doi and Singhagraiwan, 1993).

While tagging studies are the most common and convenient way of determining fish movements, unacceptably high levels of tag shedding or tag mortality can have negative implications both for the movement studies and for attempts to use these data to determine population parameters (Muoneke, 1992). Consequently, it is prudent to estimate rates of tag shedding and mortality so that these can be factored into estimates of population parameters. Beverton and Holt (1957) recognized two types of tag shedding; type I shedding occurs immediately after tagging while type II shedding occurs over an extended period. Type I shedding affects estimates of fishing rate, but not mortality and survival rates and it includes losses due to mortality, improper insertion of tags and nonreporting of tags by fishers (Muoneke, 1992). Type II shedding affects estimates of total mortality rate and it results from tag losses and differential mortality over an extended period (Muoneke, 1992).

One of the most common reasons for tag shedding is the improper placement of tags. Specifically for T-Bar and dart tags, the failure to engage the anchor between and behind the interneural bones is a major cause of tag loss. Other authors have also emphasized the importance of engagement of the tags with the interneural bones (Carline and Brynildson, 1972; Davis and Reid, 1982; Tranquilli, 1982). Anchor tag migration out of muscle has been documented in a number of

species, for example in channel catfish (*Ictalurus punctatus*) where a tag loss of 90% of tags anchored in the flesh has been reported (Waldman *et al.*, 1990).

Other means of tag loss include snagging, equipment failure, fouling with filamentous algae, and fish behaviour (Muoneke, 1992). Growth of filamentous algae may promote shedding through snagging and impaired hydrodynamics. Ebener and Copes (1982) suggest that the growth of the filamentous algae *Oscillatoria* on the tags of Lake Whitefish (*Coregonus clupeaformis*) may have contributed to high rates of tag loss.

In this study, we use tag and release methodologies to determine spatial distribution patterns of mangrove jack over a geographically large area of northern and central Queensland. We also undertake a series of experiments to determine tag shedding and mortality for both anchor and dart tags.

Methods

Movement studies

Movement studies were undertaken using information from fish tagged with both anchor and dart tags by recreational fishers throughout Queensland and subsequently recorded on the Suntag recreational fish-tagging program database. Additionally, most fish caught in QDPI research electrofishing activities in the upper tidal or lower freshwater reaches of the Daintree River, Russell and Mulgrave Rivers, Johnstone River, Herbert River, Crystal Creek, O'Connell River, Raglan Creek, Calliope River and Baffle Creek were tagged and subsequently released. To prevent the confusion of multiple tagging databases, all QDPI information was provided to Suntag who in turn assumed responsibility for notifying fishers of relevant recapture details. In the QDPI tagging program, all fish captured using electrofishing were immediately placed in an onboard tank with aeration and flow-through water where they were kept prior to tagging. All mangrove jack were measured and weighed and fish less than 300 LCF were tagged with Hallprint anchor tags, while fish more than 300 LCF were tagged with Hallprint[®] 85mm dart tags. All fish that were not recaptured were examined for evidence of tag wounds before being released. The tag number of all recaptured fish was recorded, and the fish was weighed and measured and the placement of the tag was checked before being released. Where the tag was not properly anchored or damaged it was removed and replaced with another tag. The flag of the tag contained a message requesting anglers to measure the fish and report the recapture to the Suntag freecall number. See General Methods sections for further details on tagging and capture methods.

Tag shedding and mortality experiments

Experiment 1 - Oonoonba Pond Trial

One hundred and twenty five mangrove jack, 200-300 mm LCF were collected from the upper tidal reaches of the Russell and Mulgrave Rivers using a boat-mounted electrofisher. The fish were transported to the Northern Fisheries Centre in Cairns where they were held in 1.2 tonne conical bottom holding tanks with flow through seawater. The fish were kept for 48 hours prior to the commencement of tagging on 6 June 2000, with no mortalities observed. The fish were anaesthetised using clove oil and all were weighed and measured before being tagged; the fish were randomly assigned to either be tagged with a dart tag, an anchor tag or be kept as a control. Forty nine fish were tagged with Hall Print Dart tags, 49 were tagged with Hall Print anchor tag and 27 were kept as controls. To discriminate control fish from fish that had shed their tags at the end of the experiment, a Northwest Marine hand-tagger was used to insert a coded wire tag (microtag) into the cheek muscles. Previous studies had shown that mangrove jack, when tagged

in the cheek muscle with coded wire tags, had very high tag retention rates (>95%) (Russell, unpublished data). The fish were not given any prophylactic treatments (eg. antibiotics) after the tagging operations. Forty-eight hours after tagging (8 June 2000), following checking for tag loss or mortality (Type I loss), the fish were transported in a salinity of 1.5‰ to a 0.5 ha estuarine pond at Townsville, approximately 5 hours by road to the south of Cairns. At the time of stocking the pond was 2‰ salinity and was 16°C. The pond was regularly used as a nursery pond for barramundi and was routinely fertilized to create phytoplankton and zooplankton blooms. However, on this occasion it was not fertilized prior to being filled with water for this experiment. In the pond the fish were fed daily with dry fish food pellets broadcast manually from the bank. After 159 days, the pond was harvested and 116 fish recovered and weighed and measured and their tag number, if any, recorded. All of the mangrove jack recovered from the pond were then transported in 1.5‰ salinity seawater to the Department of Primary Industries' Freshwater Research Centre at Walkamin to prepare for the second tag shedding and mortality experiment.

Experiment 2 - Walkamin Pond Trial

The remaining 116 fish recovered from the estuarine pond were stocked directly into a 0.5 ha freshwater pond at the Walkamin Research Station on the 14 November 2000. The original plan was to allow a seven-day acclimation period before commencing the second tag shedding and mortality experiment and to reduce the algal growth on the existing tags. However, two days after the fish were stocked, there were mortalities, with up to 7 dying each day. Initially, it was thought that the mortalities might have been the combined result of stress from their recovery from the Townsville pond, their conversion to freshwater and the 5.5 hour road trip from Townsville to Walkamin. However, fish continued to die and on 4 December 2000 the pond was drained and the 61 remaining fish were put into 2 tonne flow-through tanks for observation. Biopsies on these fish showed an acute Monogenean fluke infection (*Haliotrema* sp.) and the fish were first treated unsuccessfully with recommended doses of Néguvon® and formalin before being successfully treated using hyper-saline baths on two consecutive days. The fish were double tagged on the 11 December 2000 and released back into the same pond on the 13 December, 2000. Unfortunately, more mortalities occurred, this time due to attacks by cormorants and the fish were again recovered from the pond and eventually 28 fish were stocked into another 0.5 ha pond which was protected with bird netting.

A combination of 27 double tagged, single tagged and control fish were released on the 20 December 2000. Fish which had no existing tags were single tagged with either a dart (3 fish) or anchor tag (2 fish) while fish with existing tags had a second tag inserted. Of the double tagged fish, 8 had 2 dart tags and another 8 had both dart and anchor tags. Of these, only 13 fish with dart (6) and anchor (7) tags first applied at the beginning of Experiment 1 (8/6/01) were used. The remaining tags were all inserted in the period between the end of the first experiment (14/11/01) and the beginning of the second experiment (20/12/01). Six of the original fish tagged with coded wire tags were kept as controls. In the pond the fish were fed daily with dry fish food pellets broadcast manually from the bank. This pond had never had fertilizers applied to it and compared to the Townsville pond, was relatively clear and unproductive. The fish were kept in the pond for 92 days until 22 March 2001, when 25 fish were recovered

Analyses

The differences in fish length and weight from the beginning to the end of the experiments were analysed using a single factor ANOVA. In addition, paired t tests were used to analyse the differences in length and weight of individual fish that could be identified by using the fish tags. The total shedding for both tag types was determined by dividing the total number of tags at the end of the experiment by the total number applied at the beginning of the experiment. Daily

tagging mortality was determined for each tag type by dividing the total number of tags recovered by the total number of tags released and then dividing this figure by the total number of days that the experiment was conducted. Mortality (Z) was determined by calculating the slope of the plot of the natural log of the frequency of individuals in each age group against time (Ricker, 1975).

Results

Tag shedding and mortality experiments

Experiment 1 - Ooonooba Pond Trial

In the period from when the fish were caught in the wild up until they were released into the Ooonooba ponds on the 8 June 2000, none of the fish had died and none had shed their tags. On some of the fish there was evidence of bruising on the opposite side of the fish to where the tag was inserted but otherwise the fish appeared to be in good health. All of the control fish had retained their coded wire tags and previous experience in the use of this tag type with mangrove jack suggested that after the initial 48 hours, tag shedding would be negligible (DJ Russell, unpublished data).

Assuming that the coded-wire tags, once inserted, have no significant ill-effects on the fish, the natural mortality of the control fish in the pond plus any effects of stress, transport and handling of the fish was 0.073 for the whole period of the experiment or 0.0004 fish per day. The estimate of natural mortality of these fish using Ricker's method (Ricker, 1975) was 0.154 compared to 0.158 for the wild fishery (see Mortality and Fishing Yields section). The tagging mortality of fish in the pond is Total Mortality – Natural Mortality. Applying this formula in the experiment, the number of fish expected to survive at the end of the experiment irrespective of the mortality due to the application of the tags was 45.5 fish with dart tags and 45.5 fish with anchor tags. Therefore, the total number of fish that would have survived the experiment without the effects of tagging would be 116 (90 tagged fish + 26 controls), which is exactly the number that were recovered. This suggests that effects on survival of both anchor and dart tags during this experiment were negligible.

Assuming negligible tagging mortality, the shedding rate for anchor tags and dart tags was 51% and 59.2% respectively for the 183 day period. If the shedding rate is assumed to be linear, then this is equivalent to a daily rate of 0.0028 for anchor tags and 0.0032 for dart tags.

Table 2. Analyses of the impacts of tagging on growth of mangrove jack in Experiment 1.

LSD groups at the 5% level displayed by different letters. Groups with similar letters are not significantly different.

Tag Type	Mean Weight Difference	Mean LCF Difference
Control (Microtagged)	247.62	54.89 ^b
Anchor	255	50.2 ^{ab}
Dart	224	43 ^a

There were no significant differences in the effects of either dart tags or anchor tags on either weight difference ($p=0.356$) or LCF difference ($p=0.11$) (Table 2). There was no difference between the weight of control (microtagged) fish and the groups with the other tag types but there appears to be a difference between the lengths of the control fish and the microtagged fish.

Experiment 2 - Walkamin Pond Trial

After 92 days in the pond, 26 of the original 27 fish released into the pond were recovered. These fish included all 6 of the controls and 18 of the original 21 tagged fish. Another untagged fish was also recovered and this fish had two tag wounds clearly visible where tags had originally been placed. Two fish were presumed to have died. Table 3 shows the shedding of both tag types. Nine tags (2 anchors and 7 darts) were lost during this experiment of which one of the anchors and three of the dart tags were on dead fish. Excluding these mortalities, 83% of the dart tags and 89% of the anchor tags were retained. About 7.4% (n=2) of the tagged fish died during the trial and none of the control fish. If only tags applied into fish after the end of the first experiment were considered then the retention rates for the dart and anchor tags was 84% and 100% respectively.

Table 3. Tag shedding from Walkamin pond trial.

Number of fish in various tagging categories are shown. Tagging categories are: D/D double tagged with dart tags, D/A is double tagged with dart and anchor tags, D is single dart tags, A is single anchor tags and M is double tagged fish with one or both tags missing. Control fish are not included in totals.

	D/D	D/A	D	A	M	Total Fish	Total Tags
Released	8	8	3	2		21	37
Recovered	5	5	3	2	4	19	28
Difference	3	3	0	0	-4	2	9

Assuming negligible tagging mortality, the overall shedding rate for dart tags and anchor tags was 0.17 and 0.11 respectively for the 92 day period. If the shedding rate is assumed to be linear, then this is equivalent to a daily rate of 0.0018 for dart tags and 0.0012 for anchor tags.

Pond fish recaptured from the wild

After completion of the pond trials, the remaining fish were released back into the Russell River at a location close to where they were originally caught. Of the 26 fish that were released, all but three were double tagged. Five of these fish have subsequently been recaptured after periods of between 5 and 269 days at liberty (Table 4) with one fish recaptured twice. All fish moved downstream with three fish recaptured in the Russell River or the nearby Mutchero Inlet while the other two fish had moved offshore. The fish that was recaptured twice had only the stub remaining of the second tag when recaptured on the last occasion. It was unclear whether this tag

Table 4. Time at liberty, distances moved and recapture locations for double tagged fish released into the Russell River.

Tag Number	Time at Liberty (days)	Distance (km)	Recapture Location
J72865	5	<3	Russell River
J72884	5	<3	Russell River
J74003	63	60	Thetford Reef
J74010	163	17	High Island
J74004	221	4	Russell River
J74004	269	5	Mutchero Inlet

was lost or damaged on the first occasion that the fish was recaptured or shed for other reasons.

Fish double tagged by recreational fishers

In the Suntag database there are records of 440 mangrove jack that have been double tagged with either dart or anchor tags and then released into the wild with 33 (7.5%) subsequently being recaptured. These fish were at liberty between 16 and 1020 days. Of the 66 tags that were inserted into the recaptured fish, 10 (15.2%) were missing. No information is available on fish that had shed both tags.

Movements of tagged fish in the wild

Table 5 shows the numbers and sizes of recaptured fish and the types of movements of all fish in the Sunfish and QDPI databases. Of the mangrove jack released in rivers, over 73% of all fish recaptured were caught in the same river within a kilometre of their original release location. About 8% made up river movements and 14 % made down river movements but were still recaptured in the same river or creek system. Fish tagged in rivers also made coastal (1%), offshore (2%) or inter-riverine (1%) movements. A smaller number of fish were tagged in coastal situations, but while most (95%) were recaptured close to their release location some moved up into rivers (2%) or to other coastal areas (3%).

Table 5. Numbers and average \pm SD, maximum and minimum sizes of mangrove jack making riverine, coastal or offshore movements.

Included are movements data supplied by Suntag.

Movement	Number	Average LCF(mm)	Minimum(mm)	Maximum(mm)
Coast recapture with no movement	156	240.7 \pm 62.7	184	501
Coast to river movement	3	384.6 \pm 37.3	343	415
Coast to coast movement	5	443.6 \pm 98.7	271	520
River recapture with no movement	891	328.8 \pm 83.2	131	607
Up river movement	98	331.6 \pm 75.3	150	540
Down river movement	173	351.5 \pm 86.6	144	530
River to river movement	16	382.9 \pm 49.8	290	468
River to coast movement	17	435.1 \pm 105.8	240	626
River to offshore movement	30	443.6 \pm 105.8	348	607

The average size of the fish that were both tagged and recaptured in the same river system were smaller than the average sizes of fish that had made river to coast or river to offshore movements. Similarly, the sizes of fish that were both tagged and recaptured in the same coastal location were smaller than those fish that had moved up into a river or along the coast.

While the majority recaptured fish did not make any large movements, these were also the fish which been at liberty for the least time. Figure 2 shows the time at liberty for mangrove jack tagged in Queensland waters. Fish tagged in a river and recaptured at the same location were generally at liberty for an average of 195.5 \pm 6.8 days while those fish which were tagged in a river and moved to a coastal area (RC) or offshore (RO) were at liberty for 537.5 \pm 130.4 and 696.0 \pm 127.1 days respectively. While some fish did move up-river, both from river and coastal

tagging locations, the general trend was for tagged fish to move downstream to lower reaches of the river, or to coastal areas or offshore.

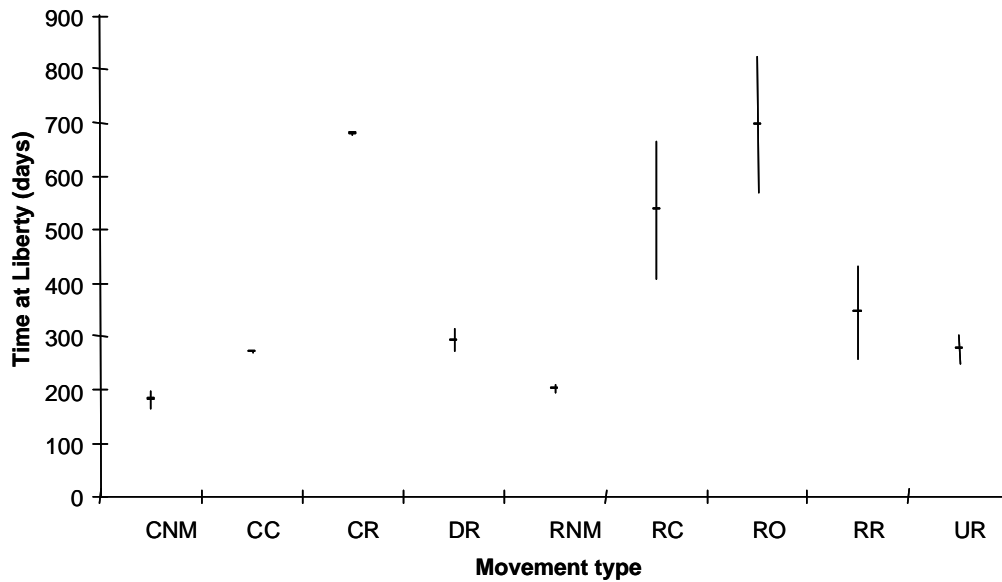


Figure 2. Time at liberty (\pm SE) for tagged mangrove jack recaptured in eastern Queensland.

The different movement types are: CNM – tagged and recaptured in same coastal area; CC- tagged in a coastal area and moved to another coastal area; CR tagged in a coastal area and recaptured in a river; DR – tagged in a river and moved downstream; RNM – tagged in a river and made no movement; RC-tagged in a river and moved to the coast; RO- tagged in a river and moved offshore; RR- tagged in one river and moved to another river; UR tagged in a river and moved upstream. Fish recaptured on the same day as they were released were excluded from the analyses. SE only computed where sufficient data available.

Table 6. Percentage of fish in each movement group and the distances moved.

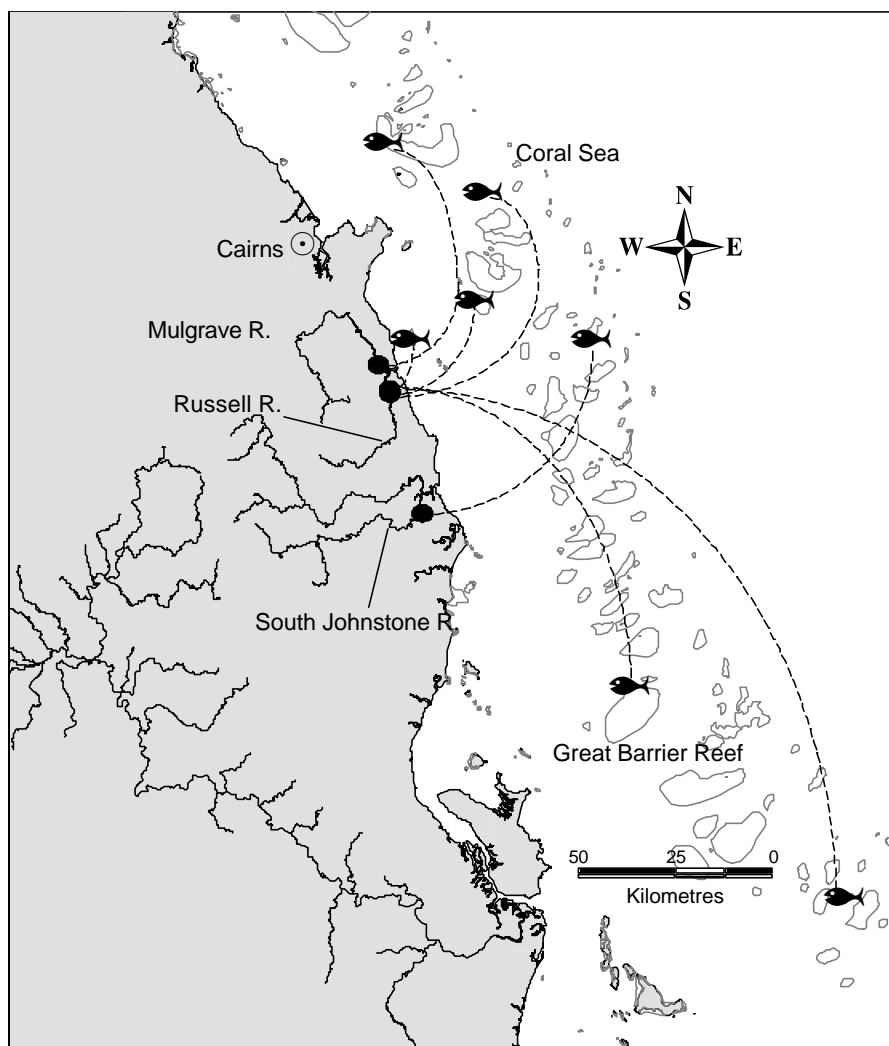
Codes for the movement groups are given in the caption in Figure 2

Distance moved	CC	CNM	CR	RC	RD	RNM	RO	RR	RU
0-1		100.0				99.5			
1-5	60.0				72.8	0.5		18.8	86.7
5-10			33.3	17.6	12.7		3.3		9.2
20-50	20.0			23.5	1.7		13.3	25.0	
10-20	20.0		66.7	35.3	12.7		3.3	25.0	4.1
50-100				17.6			33.3	31.3	
100-500				5.9			46.7		

Distances moved

Tagged fish that moved about within the same river as they were released moved distances of between 5 and 10 km either upstream or downstream. Fish that were tagged in a river and moved either to the coast (RC) or offshore (RO) moved the largest distances (Table 6) with some fish moving up to 335 km from the original release location. Some fish that moved between river systems (RR) also travelled considerable distances, up to 85 km.

Offshore Movements

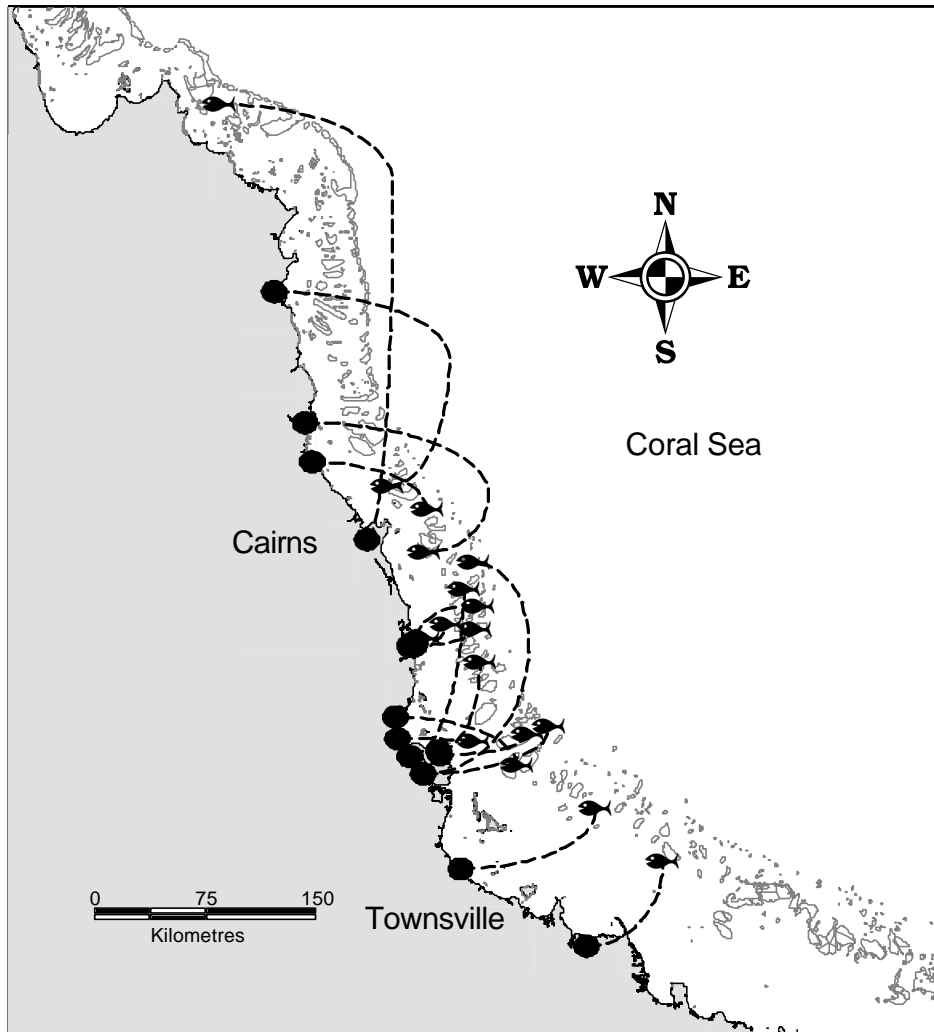


Map 10. Offshore movements by mangrove jack during the current study.

● represents release site and 🐟 represents recapture location.

Map 10 shows offshore movements by mangrove jack tagged during this study in Queensland wet tropics catchments. Of the seven fish recaptured, six were released in either the Russell or Mulgrave Rivers and the other fish was tagged in the South Johnstone River. One of the fish, which was originally tagged in the upper tidal reaches of the Russell River, moved offshore to High Island while the remainders were recaptured on the Great Barrier Reef. The largest movement was a fish that moved more than 200 km from the Russell River south to Fore and

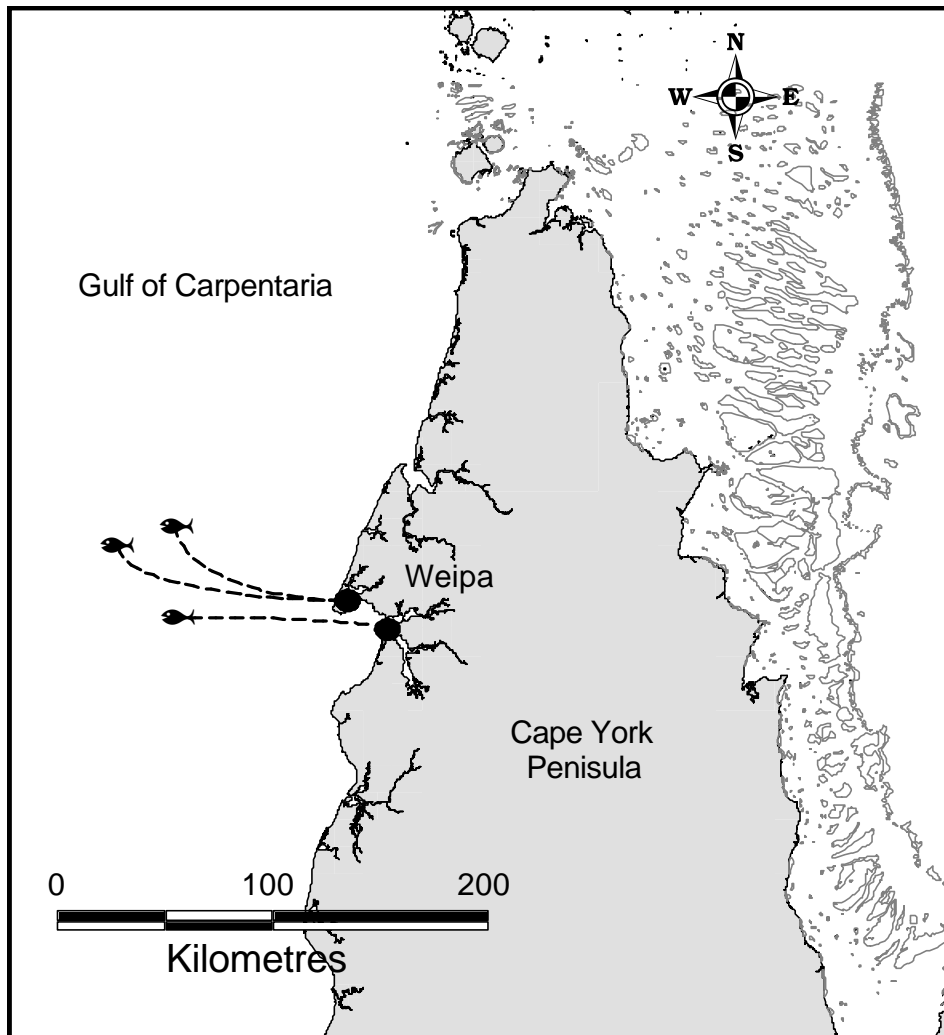
After Reef. This fish was tagged on the 12 December 2001 and was at liberty for 155 days. Information is available on offshore movements of a further 24 fish that are recorded in the Suntag database (see Map 11). All of these fish moved into the Great Barrier Reef province with one fish travelling 335 km from the release location in Trinity Inlet, Cairns to Stapleton Reef just south of Princess Charlotte Bay.



Map 11. Offshore movements of mangrove jack in north Queensland.

● represents release site and 🐟 represents recapture location. Data courtesy of the Suntag database.

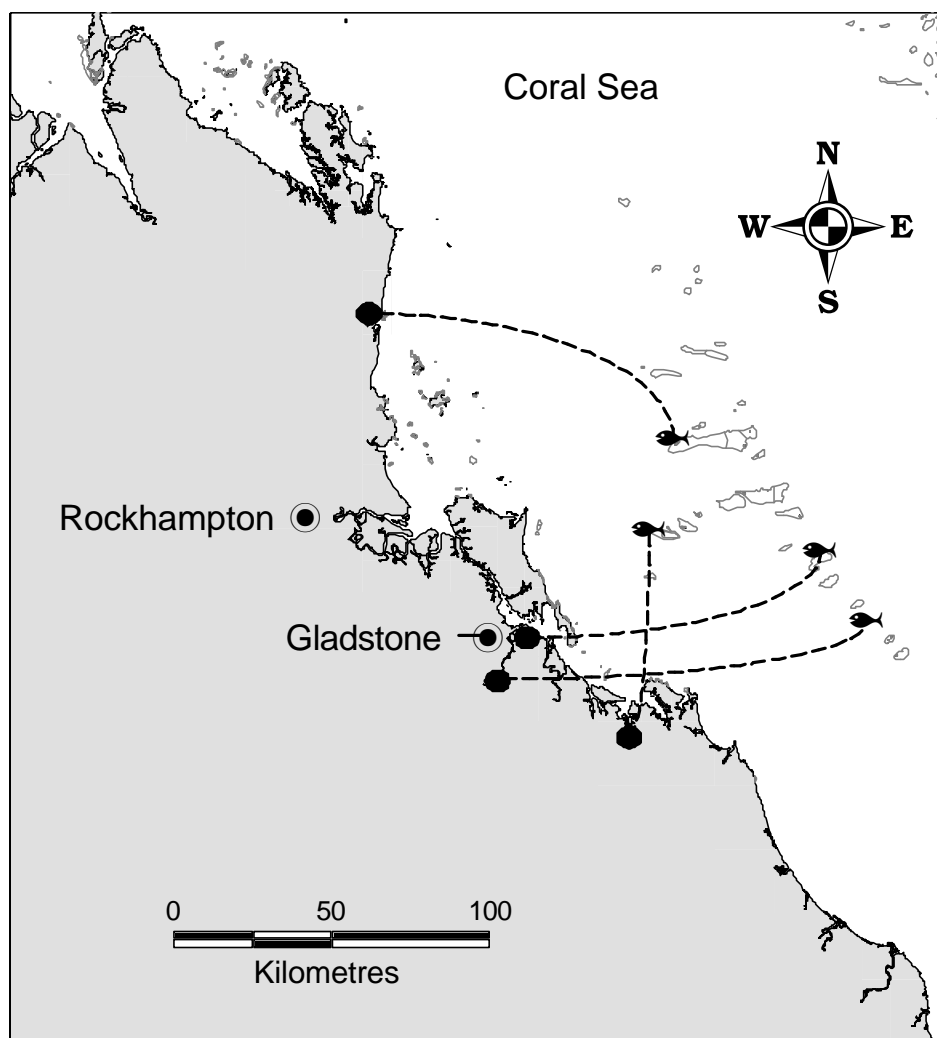
In the records of recapture from the Suntag database, three fish were tagged in the Weipa area in the northeastern Gulf of Carpentaria that were subsequently recaptured considerable distances offshore. The Gulf of Carpentaria does not have offshore coral reefs like on the eastern Queensland coast but these fish may have been congregating over shoals or rocky reefs (Map 12).



Map 12. Offshore movements of mangrove jack in the Gulf of Carpentaria.

● represents release site and 🐟 represents recapture location. Data courtesy of Suntag.

The Suntag database also contained records of mangrove jack moving from estuaries to offshore reefs in central Queensland (Map 13). In this area the Great Barrier Reef is considerably further offshore than it is in north Queensland and some fish have travelled more than 120 km from their original release location.



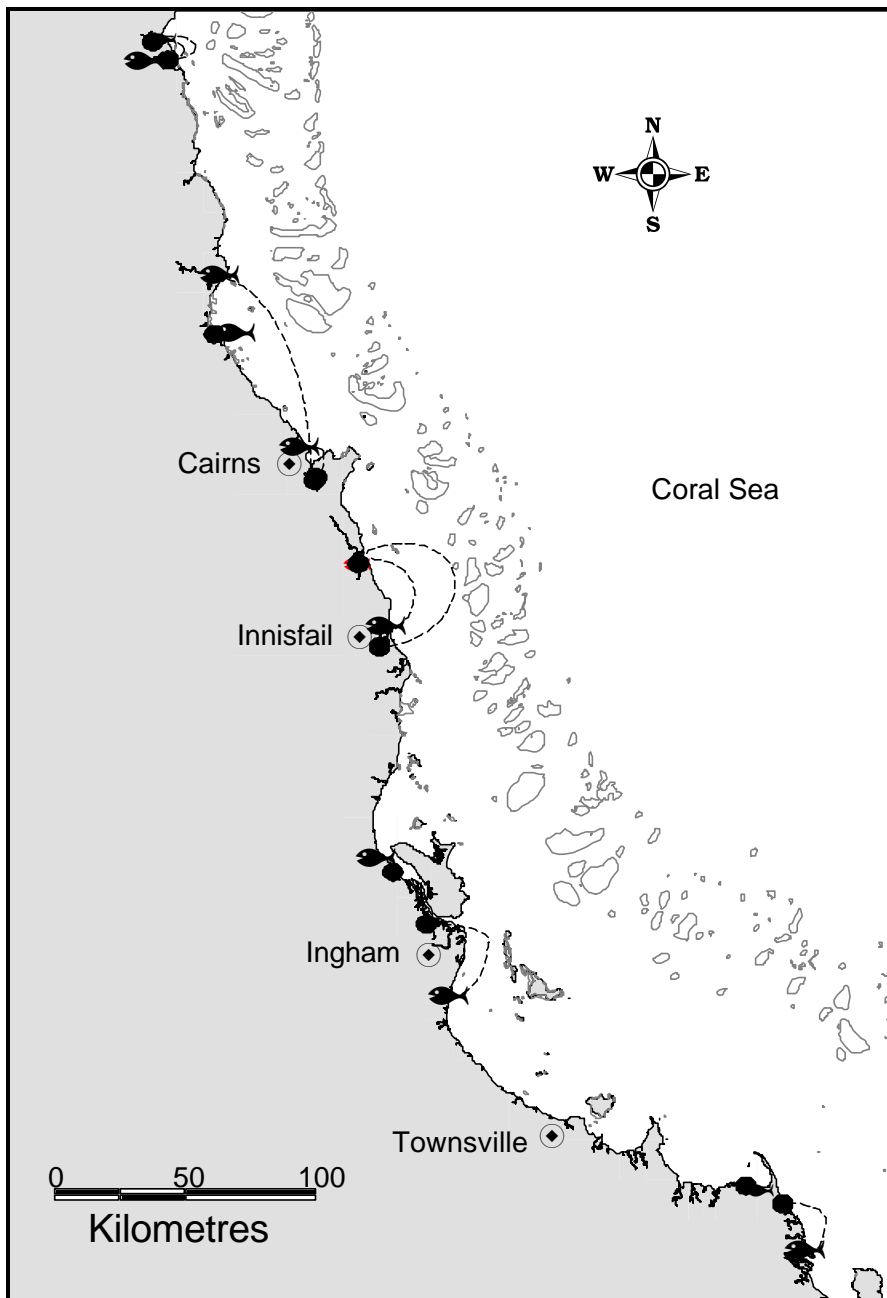
Map 13. Offshore movements of mangrove jack released in central Queensland.

● represents release site and 🐟 represents recapture location. Data courtesy of Suntag.

There are also records of offshore movements of tagged mangrove jack in Western Australia. One 360 mm TL fish was tagged in freshwater at Maree Pool, in the Pilbara region on the 3 March 2000 and recaptured offshore by a trawler at least 50 nautical miles from its release location in about 50 m of water. In the 160 days it was at liberty it increased its length by 25 mm and it was a stage 1 female. Another fish, which was tagged in Yule River in the Pilbara region on the 6 December 2000, was recaptured about 75 nautical miles from where it was released in about 100 m of water. In the 715 days that it was at liberty it increased in size from 330mm TL to 495 mm TL.

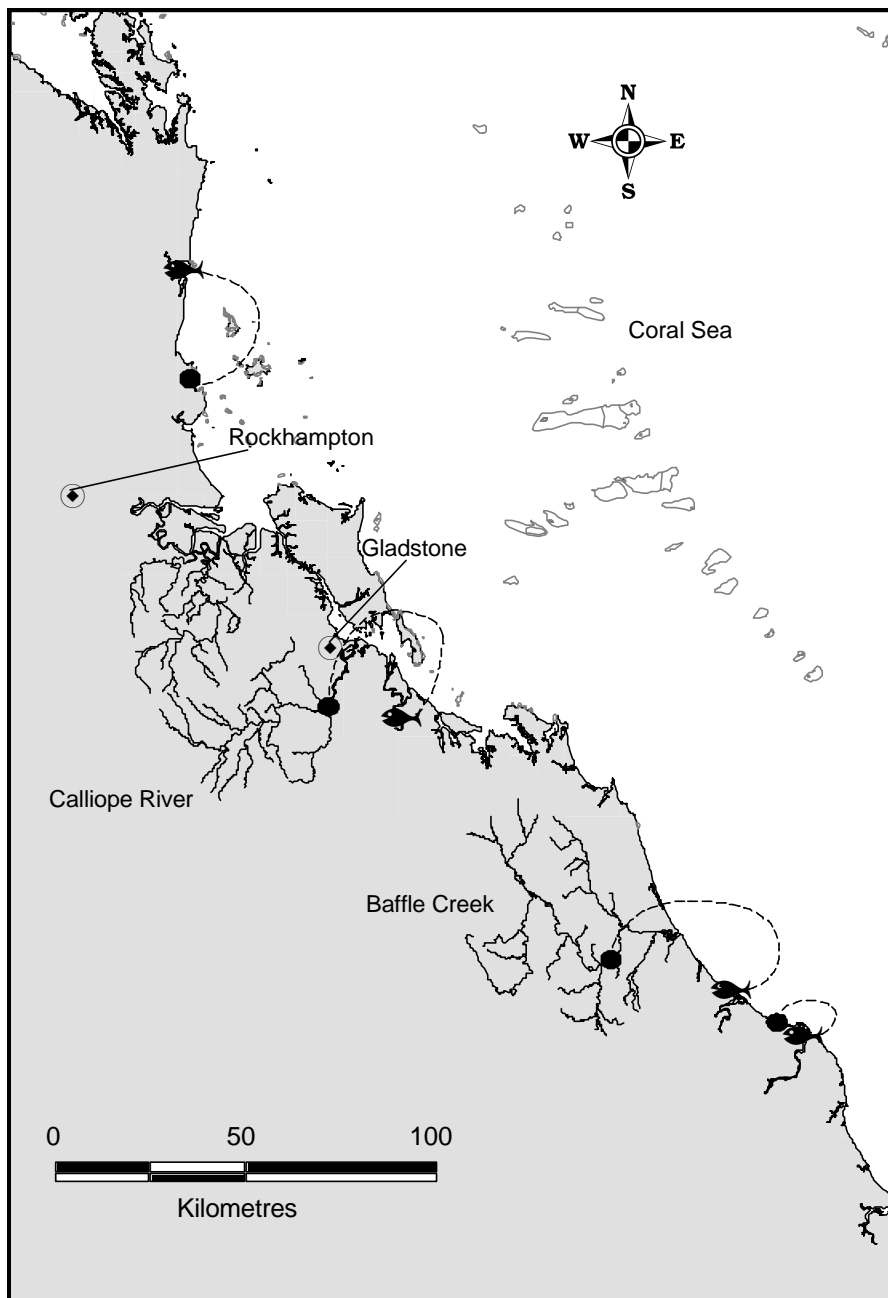
River to Coast Movements

Seventeen fish were tagged in rivers and subsequently moved to coastal habitats. All of these movements were from fish tagged by recreational fishers and not from the current study.



Map 14. Inter-riverine movements by mangrove jack in north Queensland.

● represents release site and 🐟 represents recapture location. Some data courtesy of Suntag.



Map 15. Inter-riverine movements by mangrove jack in central Queensland.

● represents release site and 🐟 represents recapture location. Some data courtesy of Suntag

River to River Movements

Of the 541 mangrove jack that were tagged during this project and subsequently recaptured, only 2 fish made inter-riverine movements. One of those fish moved from the South Johnstone

River near Innisfail, north approximately 50km to the Russell River where it was recaptured after 66 days and then released (Map 14). It was then recaptured again after 210 days back in the South Johnstone River. The other fish was tagged in Baffle Creek in central Queensland and moved about 50 km south to the Kolan River where it was recaptured after 648 days (Map 15). Using the records of mangrove jack recaptures in the Suntag database, there were 16 fish tagged by recreational anglers that made movements between rivers. Most of these movements were less than 50 km, however one fish moved nearly 90 km from Trinity Inlet near Cairns to the Daintree River in 204 days (Map 14). Another fish that was tagged by a recreational angler in the Calliope River in central Queensland was recaptured after 796 days in the nearby Boyne River, approximately 55 km to the south (Map 15).

Timing of offshore movements

Five fish made offshore movements and were recaptured less than 100 days after they were released (Table 7). Four of these fish moved offshore between about November and early February while the remaining fish moved between late March and May.

Table 7. Mangrove jack that made offshore movements and were recaptured within 100 days of release.

Tag	Release Date	Recapture Date	Liberty (Days)
J74003	22-03-01	24-05-01	63
J91338	10-11-00	09-01-01	60
N27312	07-11-01	16-12-01	39
Z31198	31-10-90	10-01-91	71
Z81496	05-11-93	07-02-94	94

Size and age of fish moving offshore

Of the 30 fish that were recaptured after having moved offshore, 11 were recaptured less than a year after being tagged (Table 8). The size-at-release of these fish ranged from between 324 mm and 430mm LCF with an average size (\pm SE) of 374.8 \pm 6.91mm. In two cases the recapture length was less than the release length indicating errors in the measurements.

Table 8. Release and recapture lengths and time at liberty for mangrove jack that moved offshore.

** indicates that recapture length was less than release length. na indicates no measurement was available.*

Tag	Release LCF (mm)	Recapture. LCF(mm)	Days at liberty
Z91091	324	376	224
N27312	348	348	39
Z81496	376	405	94
J74003	380	396	63
J24757	391	386*	232
SM14994	396	450	228
J74014	410	367*	254
J72692	416	453	271
J91338	424	444	60
J74010	430	444	163
Z31198	396	na	71

Otoliths for aging were only obtained from one of these fish (tag number J72692) which was found to be 6+ years old indicating that it moved offshore at an age of 5+ (tagged in June the previous year).

Discussion

During their life cycle, *L. argentimaculatus* undertake at least two major movements; an inshore migration as postlarvae or early juveniles from offshore spawning grounds and a subsequent offshore migration of sub-adult or mature fish. While a number of authors (Day *et al.*, 1981; Blaber *et al.*, 1989; Sheaves, 1995) have noted that juvenile *L. argentimaculatus* are associated mainly with estuaries and that adults are found offshore, little has been investigated and documented for these and other types of movements made by mangrove jack. The recruitment of mangrove jack into inshore nursery grounds is covered in a separate section of this report (see Recruitment section).

All of the mangrove jack tagged during this study and most of the fish tagged by recreational fishers in the Suntag database were released in rivers or estuaries. While most of these tagged fish were subsequently recaptured close to where they were released the majority of those that moved traveled in a general downstream direction to other parts of the river, coastal areas or offshore. In Thailand, Doi and Singhagruiwan (1993) also documented the movements of tagged juvenile *L. argentimaculatus* from an estuary, to the coastal zone and then to a more offshore area. In making these offshore movements some fish move considerable distances in a short period of time. For example one fish that was released in the Russell River was recaptured on an offshore reef some 200 km to the south after only 155 days at liberty (ie. 1.3 km/day). In Western Australia, a trawler recaptured a mangrove jack tagged by a recreational fisher 160 days earlier. In that time, the fish had moved offshore and was recaptured some 50 nautical miles from the release location. While in eastern Australia adult mangrove jack are commonly caught by line fishers in offshore areas associated with the Great Barrier Reef, in the Gulf of Carpentaria and the Pilbara region of Western Australia, they are caught by fish trawlers. The offshore trawl grounds in the Pilbara are a mixture of sandy areas with some sponge/coral relief areas (S. Newman, Western Australian Department of Fisheries, pers. comm.). The Western Australia trawlers regularly work in deep water where they catch *L. argentimaculatus* at depths of up to 100 m or more.

The longer a tagged fish was at liberty, the more likely it was to have moved to a coastal area or offshore. Although some large mangrove jack, up to 610 mm LCF, were caught in river systems during this study fish as small as 348mm long had moved offshore. In Thailand, fish as small as 260 mm TL migrate to coastal and offshore areas (Doi *et al.*, 1992). It was often difficult to get the otoliths from fish recaptured by recreational fishers however it is known that one fish was 5+ years old when it moved offshore. While many of the fish that move offshore are recaptured on offshore reefs adjacent to the river where they were released, some also made substantial coastal movements; one fish moved to an offshore reef over 300 km to the north of where it was originally tagged in 1051 days. The capacity to undertake relatively large coastal movements also has implications for the stock structure of the species and could, at least partially, explain the lack of gross genetic subdivision across the Australian population of *L. argentimaculatus*. In the Gulf of Mexico, a number of tagging studies have documented between reef movements of *L. campechanus* over similar distances (Watterson *et al.*, 1998; Patterson *et al.*, 2001).

The cues for these offshore movements are not known. In Thailand, *L. argentimaculatus* move to offshore zones at an earlier size in winter but environmental cues for such movements are yet to be determined but occur at a time when the water temperature is falling and during the wet season (Doi *et al.*, 1992). In northern Australia, although the data is limited, it appears that most offshore movements occur in summer from November to February a period that also corresponds with the summer monsoon season.

There was evidence that, during their period of residence in inshore areas, mangrove jack do make some local movements. Some fish released into rivers moved both upstream and downstream in those rivers while other fish released outside rivers in coastal areas moved to other coastal areas or up rivers. There was also some inter-riverine movement mostly between adjacent river systems. For example, one fish which was tagged and released in Trinity Inlet near Cairns moved about 75 km to the north to the Daintree River while another fish moved from the South Johnstone River to the nearby Russell River where it was caught and released and then recaptured again in the South Johnstone River.

While mangrove jack can move considerable distances, tracking them over long periods using conventional marking methods like anchor and dart tags may prove difficult because of the rate of tag shedding. In the experiments conducted during this study, initial tagging mortality was negligible although the tagging was undertaken in the relatively controlled environment of a fish hatchery rather than in the field. In the literature, tagging mortality varies from species to species and can be related to local environmental conditions at the time of tagging. For example, Winner *et al.* (1999) found that tagging did not adversely affect red drum survival or growth and that all mortalities occurred in the first 40 days of the experiment. However in *L. carponatus* mortality in tagged fish was significantly higher than in the controls and tag induced mortality peaked during the first week after tagging and greatly reduced thereafter (Whitelaw and Sainsbury, 1986).

In this study, tagging does not appear to affect growth as there were no significant differences in weights or survival of the control fish and the weights or survival of fish tagged with either dart or anchor tags. In other species tagging has impacted on growth and Carline and Brynildson (1972) found that control groups of fish stocked into a 0.7 ha lake grew faster than the tagged groups.

In both experiments, but particularly in Experiment 1 in the aquaculture ponds at Oonoonba, the shedding rates of both dart and anchor tags were high at 59.2% (0.0032 tags /day) and 51% (0.0028 tags /day) respectively. In the Walkamin experiment (Experiment 2), which was for a shorter duration, the shedding rates for both dart and anchor tags was considerably lower at 17% (0.0018 tags/day) and 11% (0.0012 tags/day) respectively. About a 35% of the tags used in this experiment were applied at the start of Experiment 1 with the remainder, including the double tags, being applied before the commencement of Experiment 2. As a result of the inclusion of fish tagged for Experiment 1, it would be expected that shedding rates would be lower because of a higher rate of tag loss in the period immediately after tagging (Type I loss). If tags applied at the beginning of Experiment 1 are excluded from the analyses, then the shedding rates for dart tags was 15.8% (0.17 tags/100 fish/day, assuming a 92 day release period) and 0% for anchor tags. While these data should be viewed with caution because of small sample size, they do suggest that, at least for the dart tags, the use of the fish tagged for Experiment 1 may not have impacted greatly on the overall result in Experiment 2. Reports of high tag shedding rates are not unusual in the literature. In juvenile *Acipenser brevirostrum* anchor tags had higher long term retention rates (92% at 306 DAT) than dart tags (50% at 154 DAT) (Collins *et al.*, 1994). In a tagging study of tailor in marine waters in Western Australia, the average tag loss in undersize and sub-adult fish was 25% and 38% per year respectively (Young *et al.*, 1999). In *L.*

carponatus, the shedding rate of anchor tags was also significantly lower than for dart tags (Whitelaw and Sainsbury, 1986).

There are many factors that influence the rate of tag shedding and retention rates vary from species to species and even within the same species depending what tag type is used. Some authors contend that environmental conditions are instrumental in the shedding of tags. A study of tag retention rates in striped bass in the Hudson River found that in the first year the relative retention rate was 98% for internal anchor tags and 42% for anchor tags and they suggest that rigorous environmental conditions may have contributed to the high loss of anchor tags (Dunning *et al.*, 1987). Evidence presented by Carline and Brynildson (1972) suggests that eutrophic or fertile waters may cause algal growth on the tag that may result in tag loss. Similarly, Ebener and Copes (1982) maintained that growth of the blue green filamentous algae *Oscillatoria* sp. contributed to shedding of anchor tags from lake whitefish (*Coregonus clupeaformis*). Muoneke (1992) also suggest that growth of filamentous algae may promote shedding through snagging and impaired hydrodynamics. Algae present on external tags may increase drag in the water and irritate the tissue surrounding the tag causing eventual loss. Many of the anchor and dart tags on fish recovered from the first pond trial at Ooononba had a heavy growth of filamentous algae (see Plate 1) that may have contributed to the extremely poor tag retention rates. When the fish were transferred to the freshwater ponds at Walkamin for the second experiment the marine algae died and the tags were relatively clean at the end of the experiment. Coincidentally, the rate of tag shedding was lower during this second experiment. The tags of fish recaptured in the wild are rarely fouled with algae to the same extent as those recovered from fish used in the Ooononba pond experiment.



Plate 1. Algal growth on a mangrove jack dart tag.

Note the wound where the tag has been inserted.

Another contributing factor to the high rate of tag shedding could have been a failure to properly engage the tags with the interneural bones. In *Ictalurus punctatus*, 90% of tags anchored in flesh

were lost but this was reduced to 19% for tags inserted behind the dorsal pterygiophores (Greenland and Bryan, 1974). In school shark the proportion of less securely attached tags decreased with increased time at liberty (Xiao *et al.*, 1999). In the current study there was also some evidence of equipment failure (loss of coloured flag from the tag) and some tags were severed, possibly by another fish. On the Great Barrier Reef, divers had observed toad fish picking at tags on *L. johnii* (M. Cappo, Australian Institute of Marine Science, pers. comm.). Muoneke (1992) reported that some fish species pulled the tags from each other and also noted that bright tags could increase a fishes vulnerability to bird predation. An independent field tagging study of *Lutjanus johnii*, *L. sebae* and *L. argentimaculatus* on the Great Barrier Reef showed evidence of high tag shedding and low recoveries (Cappo *et al.*, 2000). They found that 60% of field recoveries of double tagged fish had lost one tag and three others that were not included in their study had an oxytetrachlorine induced mark on their otoliths but no tags. A combination of the bright tag, clear water and lack of suitable cover were probably responsible for the high predation of the captive mangrove jack by cormorants prior to the commencement of the second pond experiment at Walkamin. In contrast, the Townsville pond, while not covered was very turbid and there was no observed bird predation. Other possible causes of tags loss in the wild include being caught on snags or in commercial gill nets (Ebener and Copes, 1982).

Tag mortality does not appear to be a major issue with mangrove jack, however the experiments conducted at both Walkamin and Oonoonba showed that the shedding rates of both tag types were high. This was particularly so in productive waters where excessive algal growth accumulated on tags, thus increasing the drag on the tag through the water and increasing the likelihood that it will be shed. There also appears to be some problem with tag shedding in situations where algal growth was not a problem suggesting that other factors such as tag placement and tag removal by other fish may also be contributing factors. Other marking methods may need to be considered if longer term studies of the movements of mangrove jack are to be made.

Reproduction

Introduction

The Lutjanidae is a gonochoristic (Grimes, 1987), marine and coastal family that has a pan-tropical distribution across much of the Indo-west Pacific from east Africa to Samoa and the Line Islands in the central Pacific (Anderson and Allen, 2001). In general, lutjanids have two patterns of reproductive seasonality: continental populations and species have a spawning season that is typically centred around summer while populations and species associated with small oceanic islands reproduce year round with pulses of activity in spring and autumn (Grimes, 1987).

Spawning for most tropical snappers seems to be over a considerable part of the year and may take place year round in a number of species (Thresher, 1984). Spawning peaks generally coincide with periods of warm water temperature but not necessarily the warmest time of the year (Thresher, 1984). In Australia, three lutjanid species from the Great Barrier Reef were found to have a spawning season of up to eight months during spring and summer (McPherson *et al.*, 1992). In Thailand, the spawning season of *L. argentimaculatus*, has a peak during late September to November corresponding with periods of high rainfall and decreasing water temperatures (Doi and Singhagruiwan, 1993). These authors also suggest that, based on the occurrence of juveniles, there is also some spawning in March and April. However in the lower latitudes spawning may occur throughout the year (Anderson and Allen, 2001). The limited data that are available also suggest a pronounced lunar component to the timing of lutjanid reproduction (Thresher, 1984). In Palau, there is evidence that *L. argentimaculatus* spawns around the full moon (Johannes, 1978).

In the wild, lutjanids spawn at night near open water and spawning is sometimes timed to coincide with spring tides at new and full moons (Grimes, 1987). Similarly in aquaculture situations, spawning in tanks occurs at night usually between 0100 and 0400 (Doi and Singhagruiwan, 1993). The lutjanids have a distinct courtship behaviour which culminates in an upward spiral swim towards the surface when gametes are released (Grimes, 1987). Spawning locations for *L. argentimaculatus* are unknown but are assumed to be offshore (Day *et al.*, 1981; Doi and Singhagruiwan, 1993) and in Palua spawning aggregations are found in both the reef lagoon and the outer reef slope (Johannes, 1978). Many lutjanids larvae are found distributed in open continental shelf waters (Leis, 1987). The embryonic and larval development of *L. argentimaculatus* from the wild has not been documented, however information is available from aquaculture research (Doi and Singhagruiwan, 1993). Fertilized eggs are about 0.8 mm in diameter with a single oil globule around 0.15-0.16 mm diameter present at the vegetal pole (Doi *et al.*, 1998). At water temperatures between 27.8 and 29.7°C the eggs hatch at between 15 and 17.5 hours after fertilization, with newly hatched larvae measuring between 1.56 and 1.87mm TL (Doi and Singhagruiwan, 1993). The larvae are initially planktonic but, as the fin rays develop, they become free swimming by about 12mm TL.

Lutjanids, like most broadcast spawners, are highly fecund with large females producing up to 50.7 million ova, but because they are batch spawners with several size modes of developing ova present in the ovaries it is difficult to realistically estimate total egg production for one spawning season (Grimes, 1987). There is little information in the literature on the length at first maturity for *L. argentimaculatus*, although in east African waters males mature at 350mm Standard Length (SL) (Talbot, 1960).

In this section we outline on the reproductive biology of *L. argentimaculatus* in northeastern Australia including gonadal maturation, reproductive seasonality, fecundity, and size-at-first maturity.

Methods

In this study we obtained samples of gonad tissue from a number of sources including commercial and recreational fishers and from the research sampling. Anglers were asked to refrigerate, but not freeze, the frames of mangrove jack that they had caught. Commercial samples were taken during regular visits (often weekly) to fish processors where the catch of mangrove jack was weighed and measured and the gonads dissected out, weighed to the nearest 0.1 of a gram, given a macroscopic assessment before being preserved in 5% buffered formalin. Samples were taken from both inshore (freshwater and estuarine) and offshore habitats. The majority of the inshore samples came from research electrofishing operations in primary sampling sites in the Queensland wet tropics with the remainder from river systems and coastal areas south to the New South Wales border (See Background for research site locations). The majority of the offshore samples came from a commercial fishing boat working year round, mostly between 16°S and 19°S.

Fecundity

Fish fecundities can be determined either by counting total numbers of eggs or by sub-sampling and then using gravimetric or volumetric methods (Bagenal and Braum, 1978). While total counts for highly fecund fish can be made using electronic particle counters (Davis, 1984), estimates using sub-sampling are more common. Gravimetric determination, which was used in this study, involved counting of ripe eggs in sub-samples of a known weight from mature ovaries and then extrapolating the results to give an estimate of total fecundity (Moore, 1982). Some authors suggest that the wet gravimetric method is accurate to 1% (Bagenal and Braum, 1978). Five sub-samples of approximately 0.5g wet weight were taken from each gonad and the number of eggs in each sub-sample were counted. The total weight of the ovaries, excluding the ovary wall, was determined and then the egg count of the sub samples was used to give five estimates of the total fecundity which were then averaged to give a final estimate. As *L. argentimaculatus* is a batch spawner, the estimates are unlikely to reflect total seasonal egg production; rather, they would estimate production during each spawning event.

Size at maturity

The cumulative percentage of mature fish in each 50 mm size class was determined for both male and female fish and these data were used to calculate a logistics curve using the software program SigmaPlot[®]. Only fish whose maturity stage was determined by microscopic examination of gonad sections were used in these analyses. Mature fish were defined as stage 2b or greater for females and Stage3 for males (Table 9 & Table 10). The length L_m was estimated from the point on the curve where 50% of the fish were mature.

Gonadal development

The reproductive status of all fish was initially assessed by macroscopically assigning the gonads an index of maturity based on a six point gonad classification scheme (Davis, 1982). To confirm these classifications, sub-samples were preserved in 10% formal saline for later examination using standard histological techniques. After sectioning, the tissue samples were stained regressively using Harris's haematoxylin and water-soluble eosin prior to being mounted in Depex[®]. Additionally, gonosomatic indices (GSIs) were used to determine reproductive seasonality. Mean monthly GSIs were calculated as the ratio of the ovary or testis weight to the total weight of the fish.

Macroscopic gonadal staging

The maturity stages of gonads taken from mangrove jack were first assessed macroscopically and later, histologically. A six stage macroscopic classification scheme similar to the one previously described by Davis (1982) was used to describe gonad development (Table 9).

Table 9. Macroscopic characteristics used to classify the maturity stages of mangrove jack gonads. (after (Davis, 1982).

Stage	Males	Females
1-immature	Testes thin, strap like, semi-transparent and often bordered by fat.	Ovary cylindrical with a rounded end and about half the length of testis.
2- developing	Sperm cannot be squeezed from cut testes; opaque and straplike; lateral edges are thin, not rounded.	Ovaries are compact, thick-walled, pink-red and well vascularized.
3- maturing	Testes thicker and more wedge shaped; often surrounded by fat; some spermatozoa can be squeezed from testes when cut.	Oocytes not visible to naked eye; increased size from stage 2.
4-mature	Testes are thick and wedge shaped; spermatozoa flow freely from a cut testes.	Oocytes visible to naked eye; ovaries creamy yellow and walls thinner.
5-ripe	Testes are large and white with rounded lateral margins; direct pressure will cause spermatozoa to be extruded from testes.	Ovaries distended and occupy most of the body cavity; yellow, mature oocytes.
6-spent	Testes become flaccid; some spermatozoa can be squeezed from cut testes; decrease in size from stage 5.	Ovaries are flaccid, elongated and narrow; Some residual oocytes present.

Microscopic gonadal staging

After histological processing, the sections of the 473 female and 389 male mangrove jack gonads were examined and assigned a maturity stage using the criteria in Table 10. Medial histological sections were examined microscopically and the percent total area for each cell type in the field was estimated. These data were used to assign an overall histological stage (Table 10) to each section.

Table 10. Microscopic characteristics used to describe maturity stages in mangrove jack gonads.

n/a indicates that this stage was not assigned.

Stage	Males	Females
1 - Immature	Small testes; honeycombed appearance; vacuolated lumina; testes generally attached to fat tissue.	Small ovaries; thick walled; chromatin nucleus oocytes.
2a - Developing	Spermatogonia and primary spermatocytes present; no brown bodies.	Mostly chromatin nucleolus oocytes but some perinucleolus stage oocytes; lamellae packed.
2b - Resting	Spermatogonia and brown bodies present; thick stromal walls.	Brown bodies, chromatin nucleolus oocytes and some perinucleolus stage oocytes present; Possibly some atretic stage oocytes present.
2c- Late Developing	Spermatogonia, primary and secondary spermatocytes, occasional spermatids but little or no spermatozoa present.	n/a
3 - Maturing	Primary and secondary spermatocytes, spermatids and spermatozoa present; lobule walls thick and few spermatogonia.	Ovaries increasing in size; perinucleolus stage oocytes dominant and yolk vesicle stage oocytes present.
3b - Recovering	n/a	Dominated by yolk vesicle and yolk globule stage oocytes; alpha atretic hydrated or migratory nucleus stage oocytes.
4- Mature	No spermatogonia; nests of spermatocytes and spermatids present; gonad wall thinning; spermatozoa area increasing.	Yolk globule stage and migratory nucleus stage oocytes dominant; gonad wall thinning and gonad increasing in size.
5- Ripe	Few primary and secondary spermatocytes and spermatids; spermatozoa dominant; lobules thin; gonad large; lumina distended with spermatozoa.	Migratory nucleus stage oocytes dominant; hydrated oocytes may be present; gonad wall thin.
6- Spent	Vacuoles collapsed; little residual spermatozoa; Few primary and secondary spermatocytes and spermatids.	Alpha atretic oocytes present; some residual migratory nucleus stage oocytes; post ovulatory follicles present.

Results

Gonadal development

The percentage occurrence of each of the cell type for each of the microscopic male and female maturity stages is given in Figure 3 and Figure 4. These graphs show, for example, that an undeveloped (Stage 1) female fish will have a high percentage of early developmental cells (oogonia and chromatin nucleus oocytes) and a low percentage or absence of latter cell types (migratory nucleus stage and atretic). However, in maturing and spent fish (stages 3-6) the presence of early cell stages provides evidence for batch spawning.

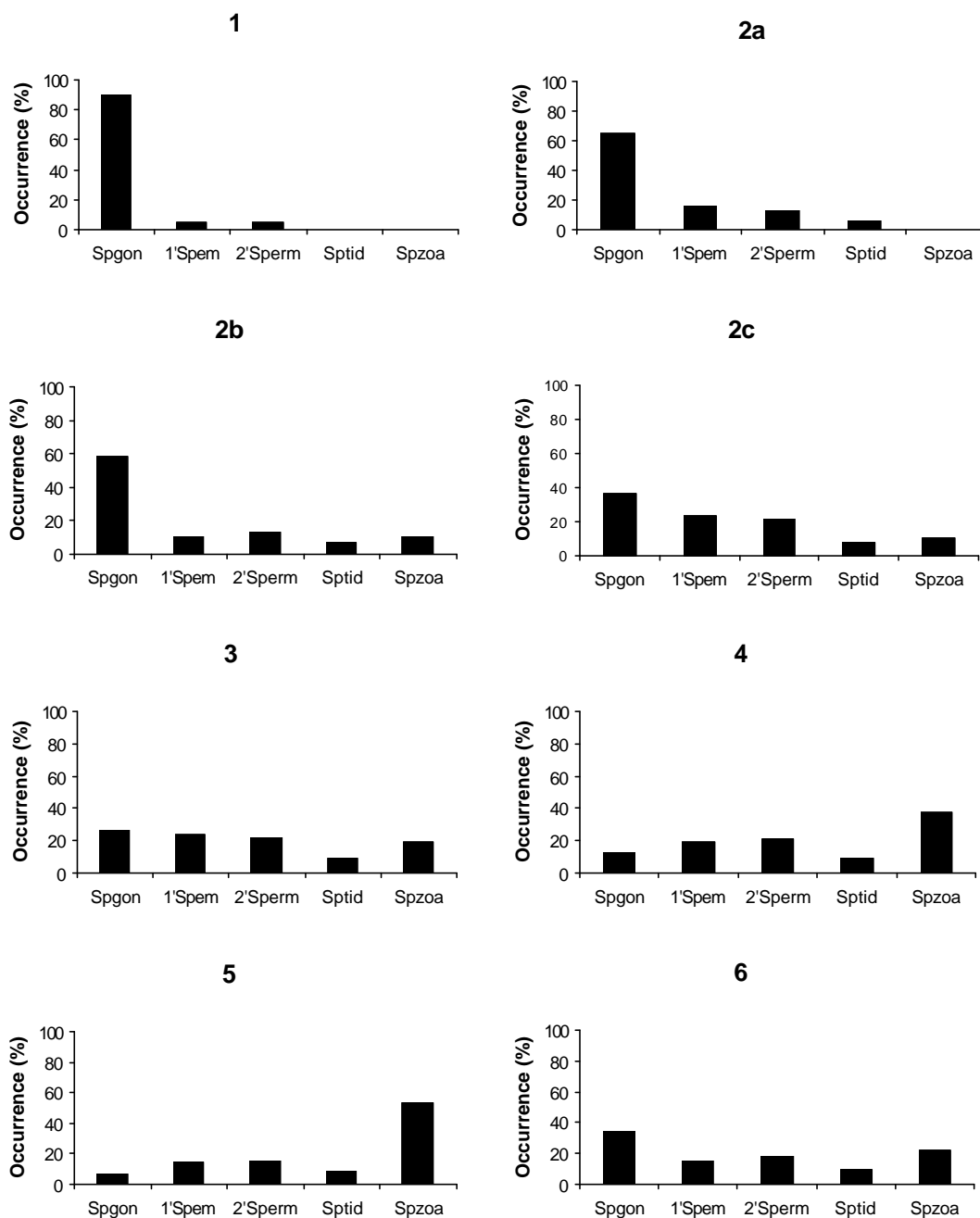


Figure 3. Occurrence of different cells in the maturity stages of male mangrove jack.

Spgon is spermatogonia; 1'spem is primary spermatocyte; 2'spem is secondary spermatocyte; sptid is spermatid and spzoa is spermatozoa.

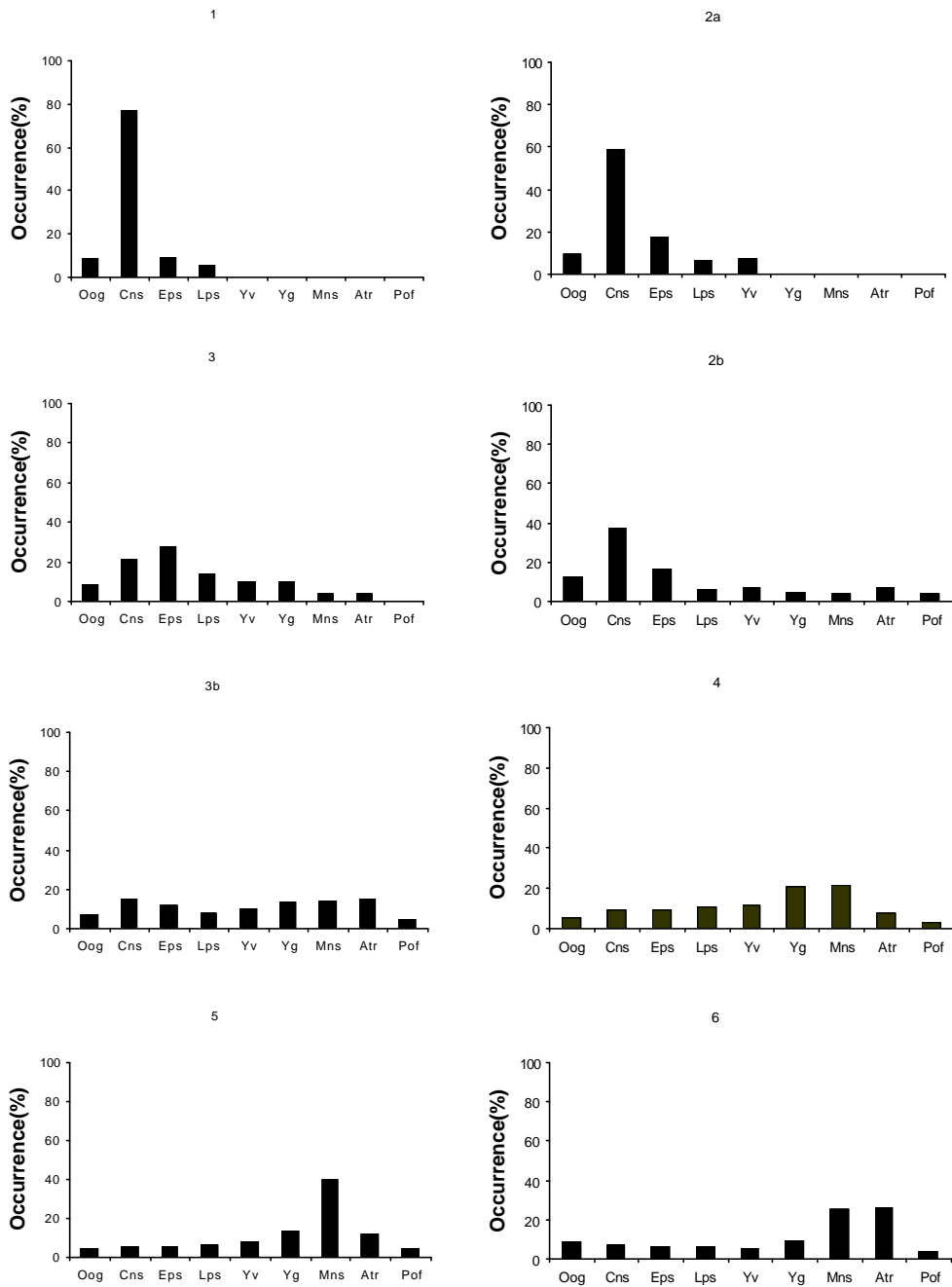


Figure 4. The occurrence of various oocyte types in the maturity stages of female mangrove jack.

Oog is oogonia; *cns* is chromatin nucleolus stage; *eps* is early perinucleolus; *lps* is late perinucleolus; *yv* is yolk vesicle; *yg* is yolk globule; *mns* is migratory nucleus; *atr* is atretic; and *pof* is post ovulatory follicle.

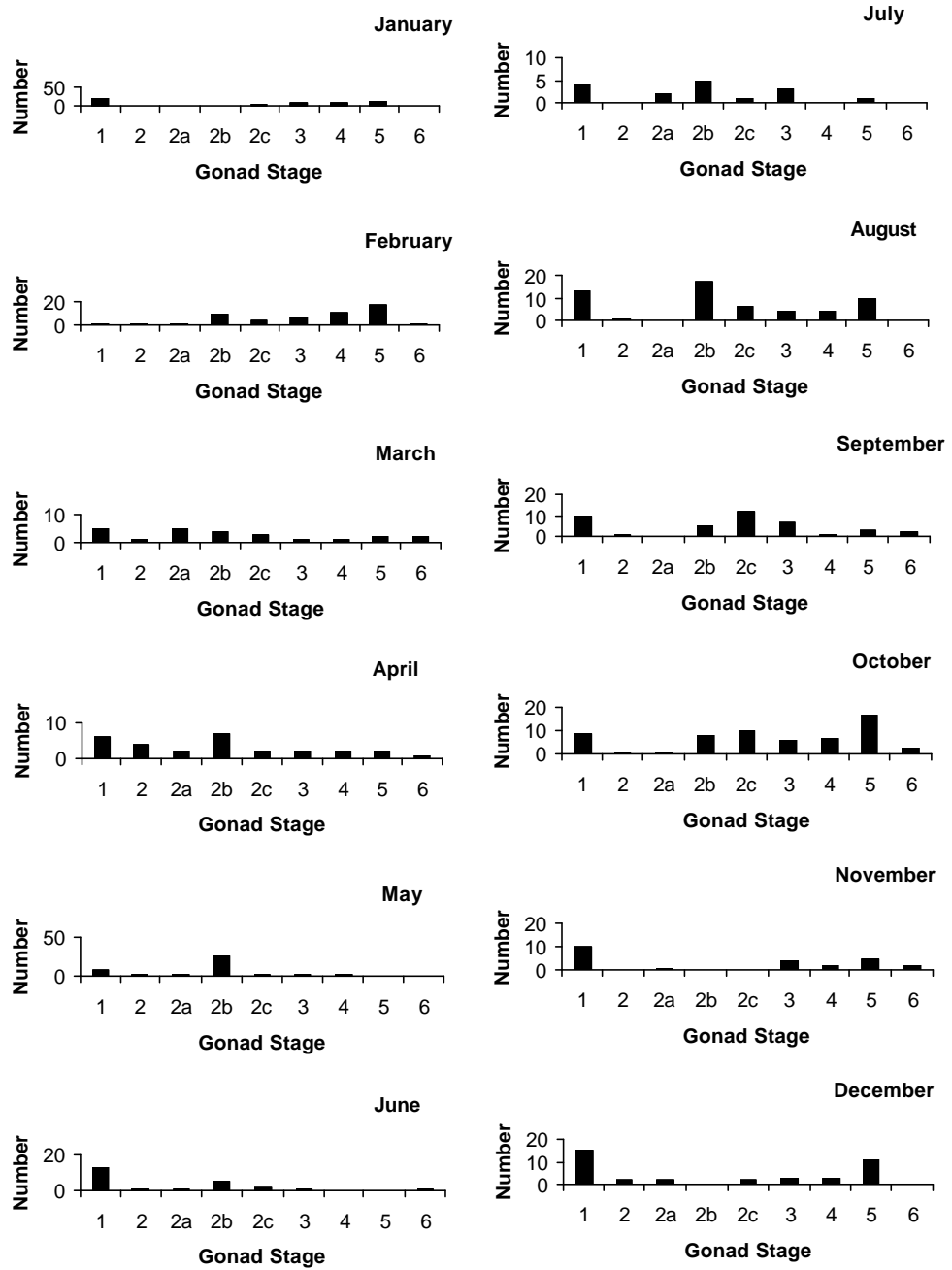


Figure 5 Gonad stages of male mangrove jack by month.

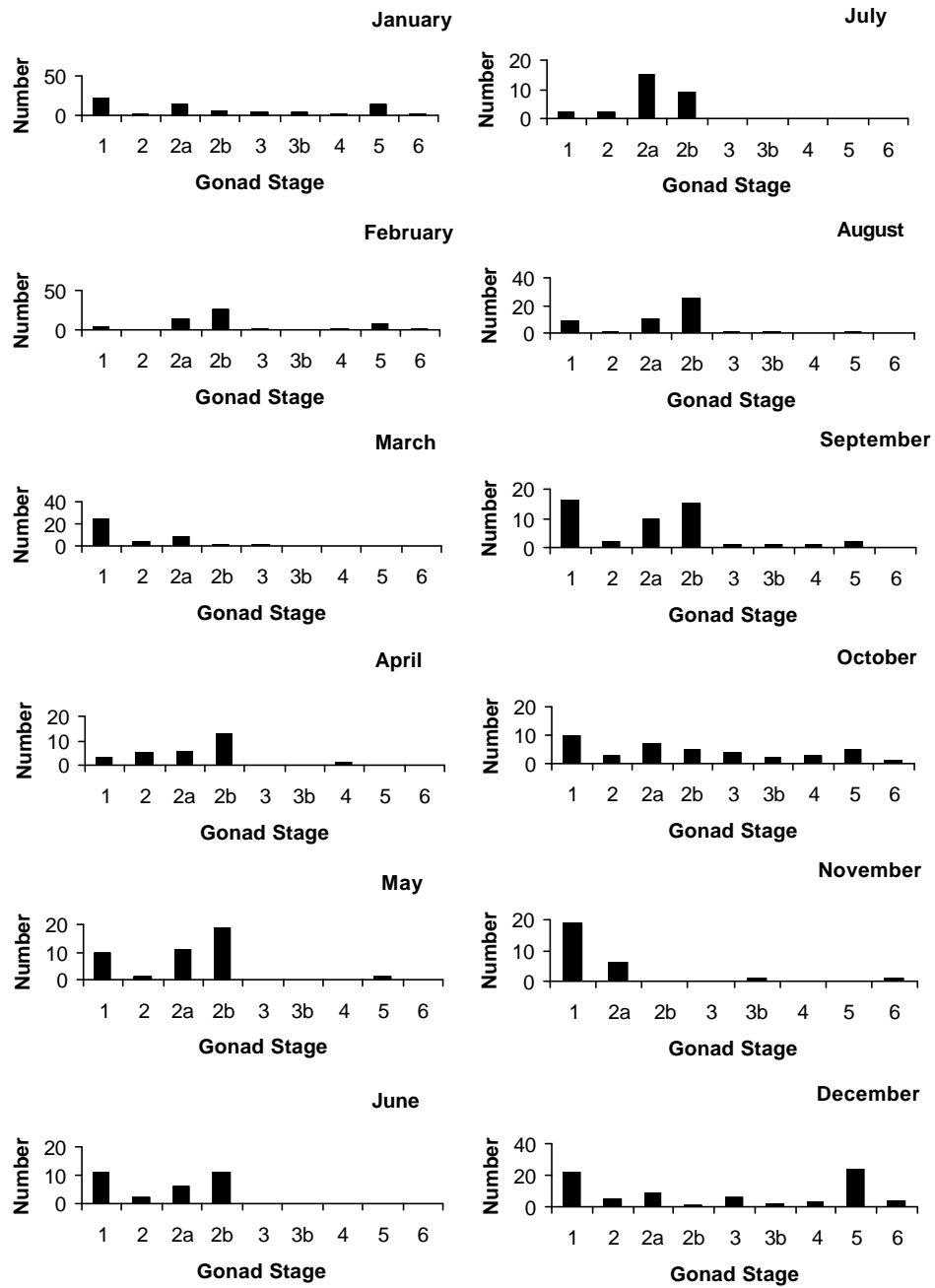


Figure 6. Gonad stages of female mangrove jack by month.

Fecundity

Figure 7 shows fecundity estimates from a mangrove jack between 545 and 730 mm LCF caught in Great Barrier Reef waters off north eastern Australia. These estimates varied from nearly four million eggs down to 250,000 eggs. When all the samples are pooled there is little relationship between fish length and fecundity. However, when the samples are split into early season and late season groups there is a stronger positive correlation between length and fecundity, particularly for the samples taken in December (Figure 7). Although the sample sizes are low, these data suggest that, for similar sized fish, more oocytes are released in spawnings that occur earlier in the season (December) than those that occur later around February.

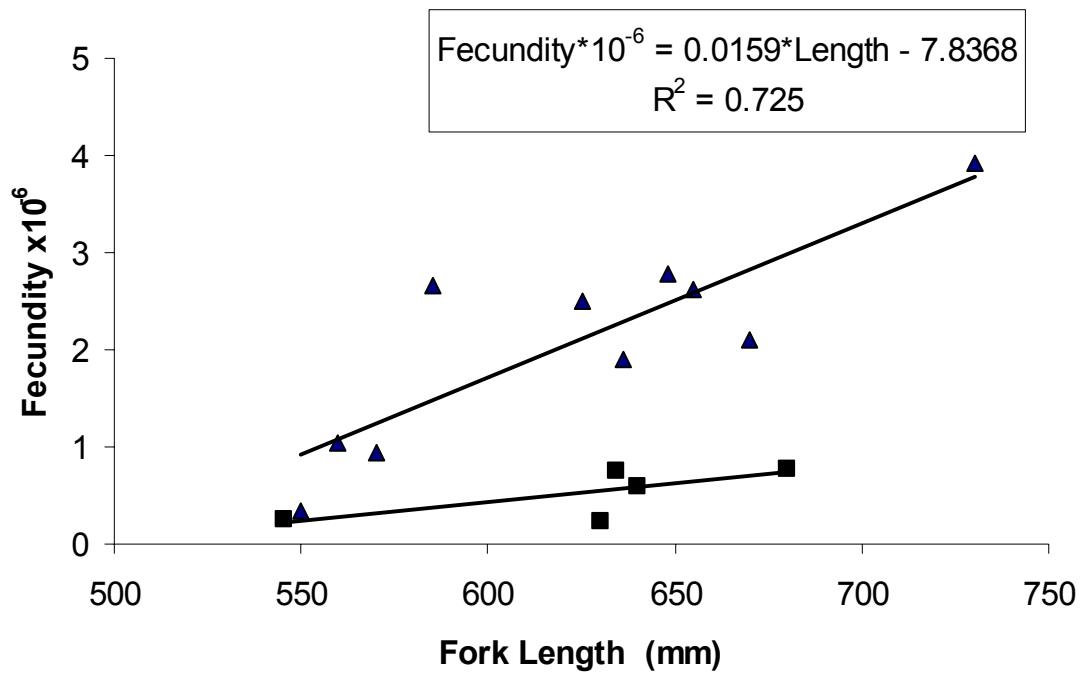


Figure 7. Fecundity estimates of 15 mature mangrove jack from north east Queensland.

▲ markers are fecundity estimates of fish caught early in the season (around December) while ■ markers are from fish caught in the later part of the season (around February).

Spawning season

Pooled average monthly (April 2000-February 2000) gonadosomatic indices for mature (development stage 2 or greater) male and female mangrove jack are shown in Figure 8. Reproductive development in both sexes commences from about October, peaks in December and then declines from January through March. The apparent decline in GSIs in November for female fish and the increase in March for male fish are most likely the result of insufficient data for those periods rather than reflecting bimodal spawning peaks.

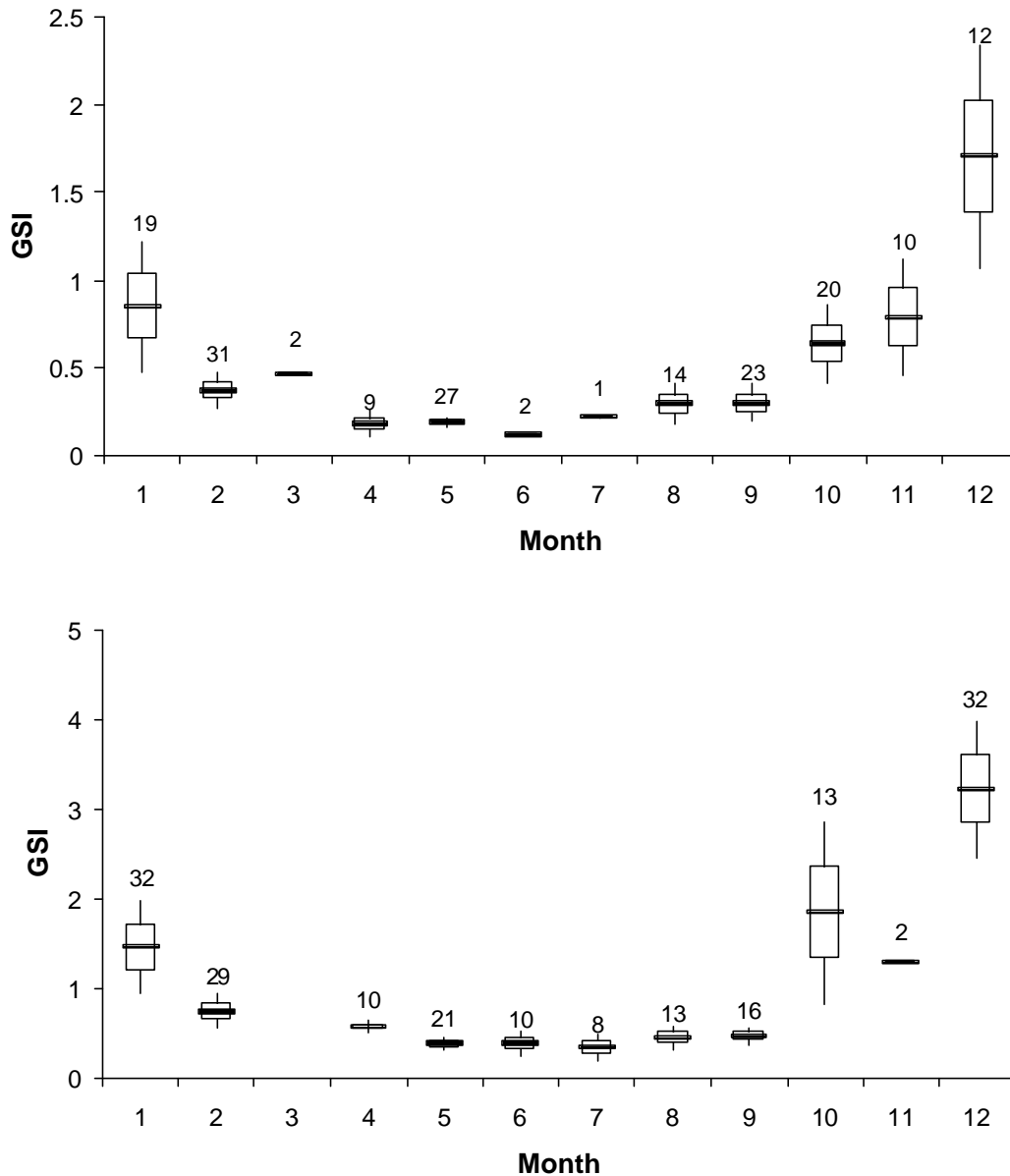


Figure 8. Monthly GSI for mature male (top) and female (bottom) *L. argentimaculatus*.

The horizontal bar represents the mean GSI, the open box shows the standard error around the mean and the vertical bars show the 95% confidence interval. Sample numbers are shown for each month above the box and whisker plots.

This pattern of reproductive seasonality is supported by the histological examination the

development stages of mature fish sampled between April 2000 and February 2002 (Figure 5 and Figure 6). About 92% of developing female gonads (between stages 3 and 6) were from fish sampled between October and February with the remainder sampled in August and September. The pattern for male fish was similar although there was a larger number of stage 4 and 5 males present outside the October to February period, particularly in August.

Gametogenesis in male mangrove jack commenced in about August and continued through to April with peak spawning activity from around October to February (Figure 5). Similarly there are stage 5 females from August through until February and in May, with most activity in December. There is some evidence of very late spawning activity with a small number of stage 4 and stage 5 female fish being sampled in April and May respectively (Figure 6)

Spawning location

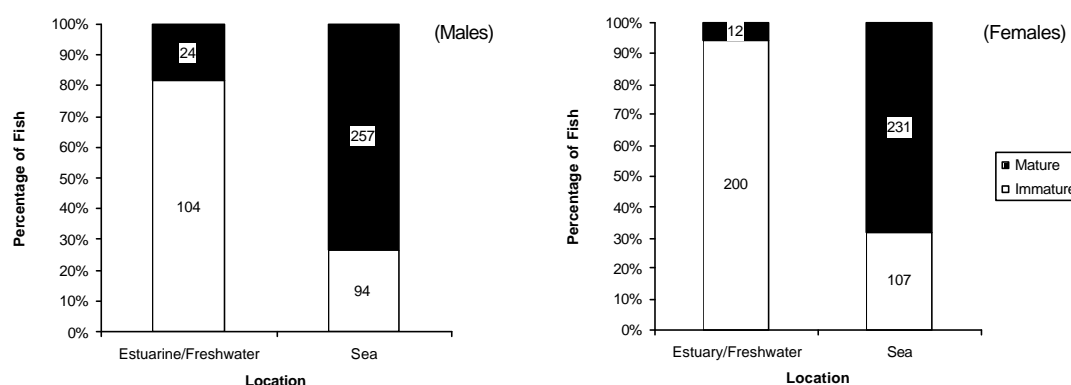


Figure 9. Percentage of mature and immature male and females fish from inshore and offshore areas. Sample numbers are shown inside bars.

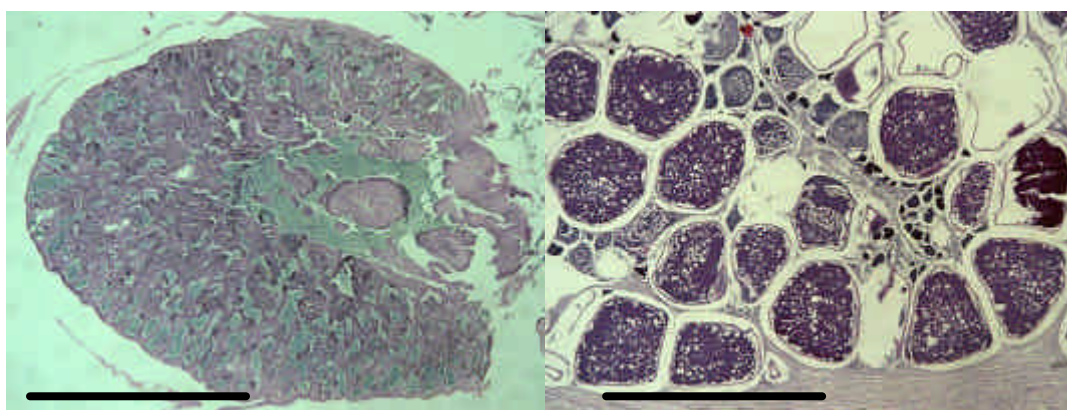


Figure 10. Sections of the gonads of mature fish sampled from estuaries or freshwater.

The testis (left) was from a stage 3, 450mm LCF fish caught in the freshwater reaches of the Mulgrave River in October 2000. The ovary (right) was from a stage 5, 390mm LCF fish caught in Bongaree Canal on Bribie Island in December 2001. Scale bar in both images is 1 mm.

Figure 9 shows the percentages of immature and mature males and females in samples taken from inshore (freshwater and estuarine habitats) and offshore locations. Most fish sampled from either the estuary or freshwater were immature while most of the fish sampled offshore were mature (maturing or resting gonads). About 19% of the males sampled from estuarine areas were mature while only about 6% of the females. These percentages are likely to be an

underestimate as juvenile fish less than 200 mm LCF were assumed to be immature and few were sampled. About 71% of the mature males and 17% of the mature females were in the early stages of maturation (stage 2). Figure 10 shows sections of mature fish caught in estuarine or freshwater areas. During the study nine female and 20 male fish of stage 3 development or higher were sampled, with milt expressed under gentle pressure from at least 7 of the males.

Size at maturity

The cumulative percentage of mature female and male fish by size is shown in Figure 11. The length at which 50% of the 217 female fish were mature (L_m) was 512.3 mm LCF while the L_m of the 177 male fish was 449.4 mm LCF. No mature fish were found up to and including the legal size (350 mm TL). The equation for the logistic curve for female fish was:

$$\sum Percentage = \frac{99.6}{(1 + Length / 512.3)^{-20.06}}$$

For male fish it was:

$$\sum Percentage = \frac{92.56}{(1 + Length / 449.4)^{-22.82}}$$

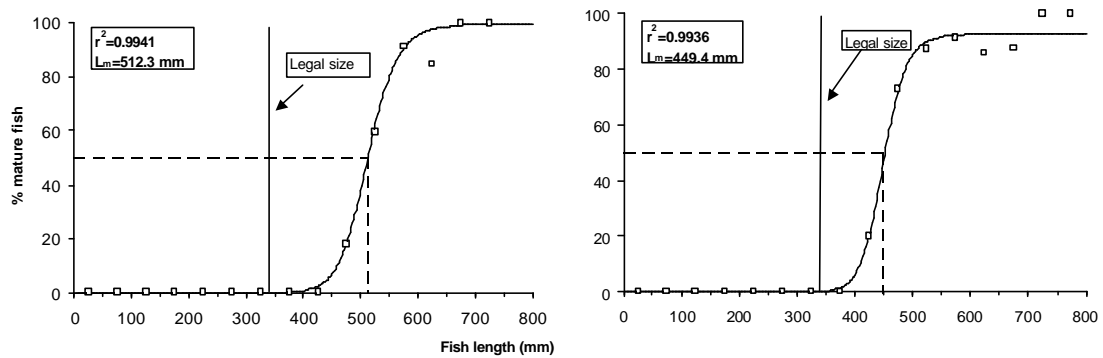


Figure 11 Logistic curves for both female and male mangrove jack.

The solid vertical line shows the legal size (converted to LCF from TL) while the dashed line shows the L_m values (LCF).

Sex ratios

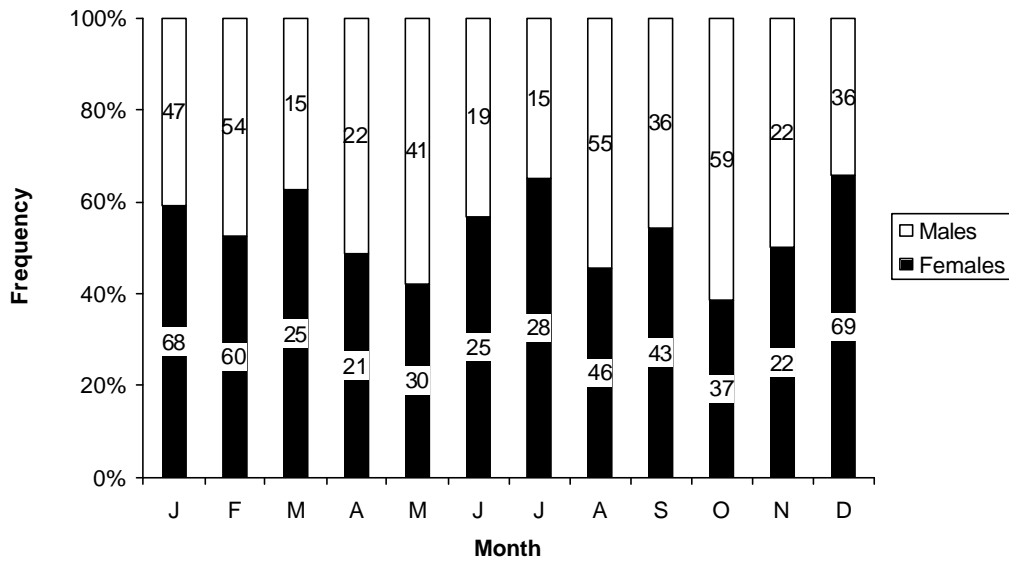


Figure 12 Percent frequency of males and females sampled each month.

The number inside the bars is the number of fish sampled.

The monthly sex ratios of fish caught in the recreational and commercial fisheries are shown in Figure 12. A Chi Square test of the monthly data shows that the relative monthly proportions of males and females are not constant ($\chi^2=26.92, df = 11, p=0.00047$). There appears to be no pattern to these sex ratios which vary from between 1 male : 1.92 females in December to 1 male : 0.63 females in October. The 500-600 mm LCF size classes have the highest percentages of females, while males dominate in the remaining size classes (Figure 13).

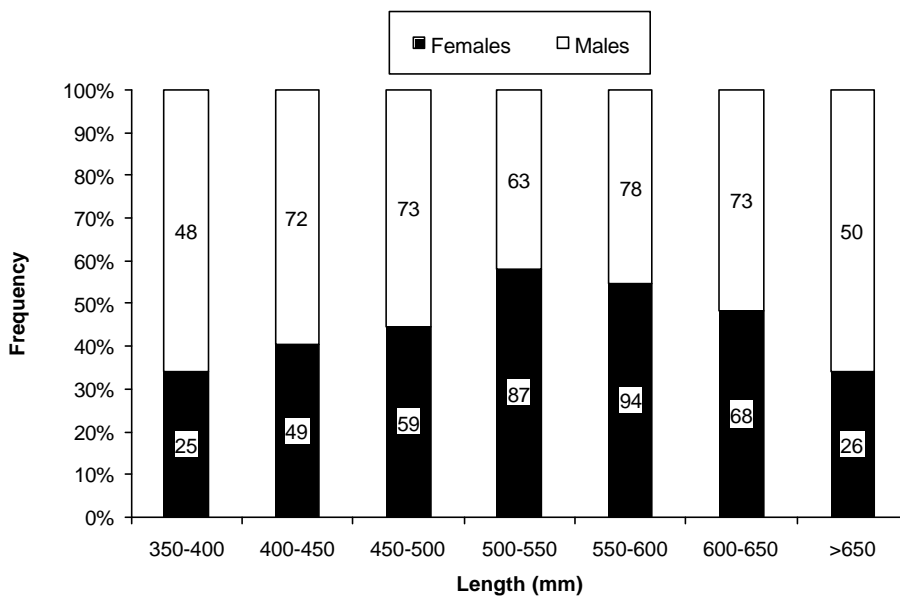


Figure 13. Percent frequency of males and females sampled by recreational and commercial fishers in each 50 mm length class (LCF).

The numbers inside the bars is the number of fish sampled

Discussion

Fecundity in mangrove jack is relatively high, with counts of up to nearly 4 million eggs per fish. The fecundity of mangrove jack is not as high as for some other larger marine teleosts; for example, in barramundi (*Lates calcarifer*) egg counts of nearly 46×10^6 eggs have been made for a ~20 kg fish (Davis, 1984). This was approximately 2.3×10^6 eggs per kg body weight while in this present study, mangrove jack average about 526,000 egg per kg body weight. Histological evidence from this present study suggests that mangrove jack are batch spawners and the egg counts probably more accurately reflect the number of eggs released in a single spawning event. Evidence from the egg counts of fish caught earlier in the season suggests that the magnitude of egg releases for individual spawning events differ throughout the spawning season. The fecundity estimates given in Figure 7 show that the number of oocytes shed earlier (around December) is larger than the numbers released from similar sized fish later in the season (February).

In lower latitudes or on some continental islands *Lutjanus argentimaculatus* spawn more or less all year round (Anderson and Allen, 2001) and in north-eastern Australia there is evidence that spawning does occur over an extended period with stage 5 females present from August to February. However, most spawning appears to be between October and February. In Thailand, the peak spawning period for *L. argentimaculatus* is approximately during the same period (November to January), but there is also evidence of spawning outside the peak period around May (Doi and Singhagruiwan, 1993). Similarly in north Queensland, histological examination of mangrove jack gonads suggests that some limited spawning occurs at other times of the year. Ripe (stages 4-6) males were found in most months and a stage 5 female was sampled in May.

In north Queensland, gametogenesis begins around September/October, a time when both the water temperatures and photoperiod are increasing. The spawning season extends through the warm summer months from about October through to March. In Thailand, falling water temperatures and high rainfall have been identified as key environmental cues (Doi and Singhagruiwan, 1993).

Many of the features of reproductive biology of lutjanids appear to be a strategy to introduce gametes into an environment where predation is relatively low but the strategy must also assure that larvae and young juveniles are returned to suitable habitat for settlement (Grimes, 1987). Most of the fish sampled from estuarine and freshwater areas were immature while most mature fish were sampled offshore in reef areas. There would appear to be an offshore migration of immature mangrove jack from inshore to offshore areas where they subsequently mature. No evidence was found of small, immature juvenile mangrove jack in offshore areas. The smallest fish sampled from offshore areas was a 360 mm LCF immature female fish from a reef off Cairns. Sheaves (1995) reported that the most advanced spermatid tissue present from mangrove jack he sampled in estuaries were primary and secondary spermatocytes but also noted that the testes of three estuarine fish sampled in October had some spermatids and spermatozoa present. In the testes of these three fish the spermatids and spermatozoa were confined to the proximal part of the gonad and their overall size was small suggesting that they were from fish in the early stages of reproductive development (Sheaves, 1995). In this present study only about 19% of the males sampled in estuaries and freshwater showed some spermatid activity, generally early stages with primary or secondary spermatocytes present although some were stage 4 or stage 5. Of the 130 male fish sampled from freshwater or estuaries, only 20 were stage 3 or greater. Milt could be expressed from six of these fish by gently squeezing on their abdomens. Nine female fish with gonad stages of 3 or greater were sampled from estuarine and freshwater areas including one stage 4 fish and one stage 5 fish. Sheaves suggests that a group of three males with spermatid tissue and four females that he sampled may be preparing to migrate from the estuary. Also, while some mangrove jack from inshore areas showed signs of maturation this does not mean that they successfully

spawned in those areas. It is possible that the gonads were resorbed or as suggested by Sheaves, they could have been caught just prior to them undertaking offshore movement. It would appear likely that there is little or no spawning in rivers. Evidence from tagging studies suggests that offshore movement occurs during the period November to February (see Movements section). There is little doubt that the quantity of eggs released in inshore areas would be insignificant compared to that spawned in offshore habitats.

No evidence was found of hermaphroditism and the size-at-maturity data suggests that mangrove jack do not mature until a relatively large size and that the males begin to mature before the females. In a South African study all mature *L. argentimaculatus* sampled were 460 mm or longer (Talbot, 1960). In Queensland the minimum legal size for mangrove jack is 350 mm TL that is well below the L_m for both male (449.4 mm LCF) and female (512.3 mm LCF) mangrove jack. The smallest mature or maturing fish (stage 2 or more) caught were a 420 mm LCF male and a 370 mm LCF female. In a management context, the minimum legal size is unrelated to size at maturity and immature fish are targeted by both recreational and commercial fishers, particularly in inshore areas.

Age and growth

Introduction

An accurate and reliable technique to age fish is an essential requirement for assessing the health of a fish population (Francis, 1990). Over time, there have been numerous techniques developed to estimate the age of fish, mostly by interpreting banding on bony structures. This banding is the result of seasonal growth patterns related to environmental parameters such as water temperature (Akamine, 1993). The types of bony structures used include scales (Davis and Kirkwood, 1984; Marriott and Cappo, 2000), whole otoliths ((McPherson and Squire, 1992; Milton *et al.*, 1995), sectioned otoliths (Anderson *et al.*, 1992; Francis *et al.*, 1992; Rocha-Olivares, 1997; Cappo *et al.*, 2000; Masuda *et al.*, 2000; Burton, 2001, 2002) or whole and sectioned vertebrae (Marriott and Cappo, 2000). Other researchers have used radiochemistry as an alternative to physically age fish by measuring radioactive decay of elements present in the bony structures, with often quite conflicting results (Campana, 1990b; West and Gauldie, 1994; Milton *et al.*, 1995).

Since attempts at ageing of fish began, there has been a need to validate whether the growth increments seen are in fact annual in nature or follow some other cycle. Most validation studies have involved injection of tagged fish with a chemical dye (typically oxytetracycline) that then penetrates the bony structures of the fish shortly after injection. These fish are then released and if recaptured at a later date, the time period between when the chemical mark was laid down and the edge of the bony structure is known. When the bony structures from the fish are sectioned and viewed using ultraviolet light the chemically induced band fluoresces. Validation studies have been undertaken in the past with some lutjanids. Cappo *et al.* (2000) has recently documented the annual nature of the growth increments of 11 species of *Lutjanus* from the Great Barrier Reef using this technique, with an average mean periodicity of formation of 0.96 ± 0.32 cycles/yr. The use of sectioned otoliths for ageing mangrove jack has been validated in this study and by (Sheaves, 1995), however these were all sub-adult fish.

Ageing of lutjanids worldwide has attracted significant research including a number of Australian studies (McPherson and Squire, 1992; Milton *et al.*, 1995; Sheaves, 1995; Newman *et al.*, 1996b; Cappo *et al.*, 2000; Marriott and Cappo, 2000; Newman *et al.*, 2000c, b). The Australia studies are mainly concerned with species from the Great Barrier Reef (GBR) region, however Milton *et al.* (1995) aged three species from the Gulf of Carpentaria, where he obtained different age estimates to the east coast of Queensland. While there are some conflicting results, the consensus now seems to rest on the use of sectioned otoliths for more precise age estimates (Marriott and Cappo, 2000). Recent studies of lutjanids suggest that they are a long-lived family, with age estimates of the gray and mutton snapper (*L. griseus* and *L. analis*) to 24 and 29 years (Burton, 2001, 2002), pacific red snapper (*L. peru*) to 31 years (Rocha-Olivares, 1997) and tropical red snappers from the GBR (*L. erythropterus*, *L. malabaricus* and *L. sebae*) to 32, 20 and 22 years respectively (Newman *et al.*, 2000c).

Tagging of wild fish has also been used as a technique to estimate growth, and hence the age of fish by extrapolating annual growth rates. It is often used as a technique to support ageing data (McPherson and Squire, 1992; Sheaves, 1995), however tagging estimates growth as a function of fish length whereas ageing estimates growth as a function of age (Francis, 1990).

Sheaves (1995) used both sectioned otoliths and tagging to estimate growth rates of inshore mangrove jack in north Queensland. He aged 298 fish with between 0 and 32 increments. The average growth rate obtained from tagging was found to approximate that obtained from growth parameters in fish less than 6 years old. He attempted further validations using oxytetracycline but only had two recaptures of marked fish.

In this section we interpret the banding on otoliths to provide estimates of length-at-age for mangrove jack stocks in eastern Queensland. These data, when used in conjunction with other biological and population parameters, provides useful information pertinent to the management of the fishery

Methods

AGE validation

Broodstock fish

Broodstock mangrove jack, that are kept at the Northern Fisheries Centre as part of the ongoing aquaculture program have a known and well-documented history. During the period of this study, a number of these fish died and subsequently their otoliths were removed and the fish were aged.

It was noted that some of these broodstock fish that had died over the course of the project had, at known times in the past, been treated with oxytetracycline as an antibiotic (dose of 50mg/kg) and therefore should have laid down an OTC band. These otoliths were processed in the same way as the other otoliths (see Otolith procedures) except that the sections were viewed under a combination of ultraviolet and transmitted light to determine the position of the OTC mark relative to any growth increments. The number of increments and their distance from edge of otolith was then noted to compare against the time elapsed since they were treated with the OTC antibiotic. The estimated calendar closing date (CCD) (or the estimated date of completion of the increment) was calculated using the direct method outlined in Cappo *et al.* (2000). The mean calendar closing date (MCCD) is the average of all the calculated calendar closing date values.

Impoundment F1 fish

Stocked mangrove jack are now being recaptured by anglers in freshwater impoundments (see Background section). These fish are useful in validating ageing techniques because they are of a known age. The otoliths from one mangrove jack that was stocked into Tinaroo Dam and then recaptured on a known date was also used to validate the ageing technique.

Recaptured wild fish

A number of tagged wild fish that were subsequently recaptured were sacrificed to provide an estimate of age at recapture. These fish were immediately pithed with a sharp implement, the gills were severed and the animal was placed in an ice slurry to slow metabolism. Other measurements and biological samples taken from these fish are described in the General methods section.

Marginal increment analysis

The use of marginal increment analysis (MIA) can also offer another means of validation. A major assumption in ageing of fish structures is that the increments are formed at the same time of year (annually) in response to a change in catabolic rate. (Everhart *et al.*, 1975). The timing of increment formation for mangrove jack has been previously validated using a small number of samples by Cappo *et al.* (2000).

Otolith procedures

Two methods were employed to remove the sagittal otoliths from the base of the skull of the fish. The first method involved a cut with an 18 tpi hacksaw blade in a line from the top of the eye to the dorsal attachment point of the operculum, allowing dorsal access to the rear brain cavity. Secondly, for fish that had been gilled and gutted, a pair of wire cutters was used to make two cuts in the anterior section of the spine at the top of the buccal cavity. Otoliths

were then removed with a pair of forceps, rinsed and scrubbed with a nylon brush in distilled water, then air-dried for 48 hours in acid-washed labelled vials.

Length, width, thickness and weight of each otolith was measured using Mitutoyo CD-6" C digital callipers (± 0.001 mm) and an AND HR-200 balance (± 0.0001 grams). Otolith samples were set and orientated in clear casting resin (2% MEKP hardener) using custom-built moulds constructed of Silastic 301[®] (individual moulds were either 9 x 7 x 24 mm or 15 x 8 x 24 mm). Initially, approximately 3 mm of resin was poured into all molds and allowed to set for 20 minutes. Further resin was added to fill the mould, with the otolith then correctly orientated for sectioning, with a label finally added to the mold. The primodium was first marked with a fine marker pen and then a number of 300-350 μ m medial sections were cut through the block using a diamond bladed Struers Minitom (model # 04436133). Sections were immediately viewed using a Nikon SMZ-2B dissecting microscope to determine the sections closest to primordium. If required, the thin sections were polished using 1 000 grade sandpaper and Struers lapping paper (4 000 grade) to improve visibility. Sections that required further treatment were temporarily immersed in 5% HCL to enhance anuli by accentuating increments through unequal chemical reaction with the acid. Appropriate sections were mounted in Crystalbond[™] 509 transparent adhesive on a microscope slide. Difficult (unclear) samples were heated to further increase the contrast of the increments through burning of the concentrated proteins in the increments (Gauldie, 1988).

Counts of anuli were made using a Leica MZ6 dissecting microscope (1-4 x) and a Leica DMLB bright field microscope (4-25x). A combination of transmitted and reflected light was used to identify the increments. Images were recorded with a Leica digital camera, and were stored as JPEG files. Otolith radius, increment width and marginal increment width were measured using UTHSCA[®] *Image Tool* for Windows Ver.3 (Wilcox *et al.*, 2002). Counts, measurements and clarity of section were recorded in an Access database. A number of randomly chosen samples (n=821) were sent to the Central Ageing Facility (CAF) (Department of Natural Resources and Environment, Victoria) for independent comparison to the ages obtained by NFC staff, with 25% of these samples subsequently re-read by CAF.

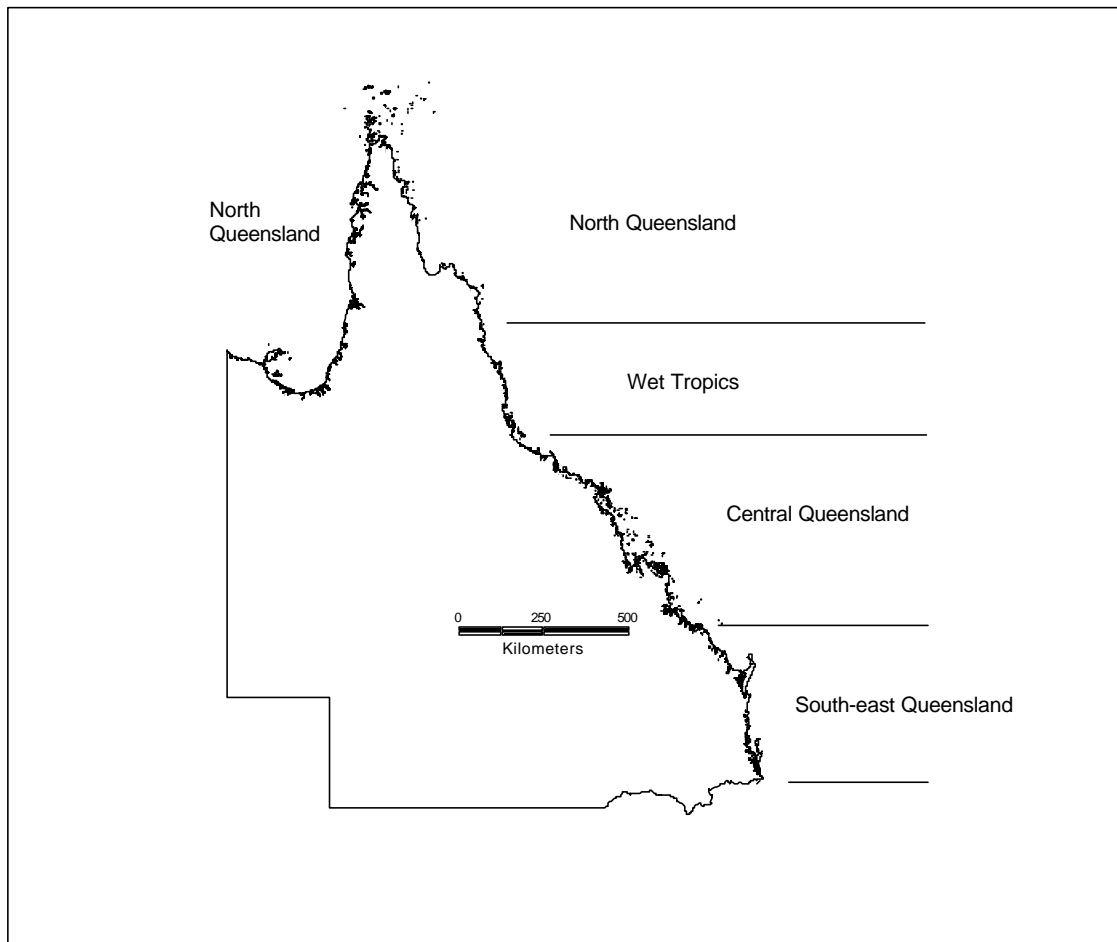
Growth

The growth of recaptured individuals was calculated by subtracting the length (LCF, mm) at tagging from length (LCF, mm) at recapture. Time of liberty was also calculated in the same manner using tagging and recapture dates. Growth rate was calculated for each fish by dividing fish growth by time at liberty and expressed as mm/day.

Analysis

Otolith morphometrics and fish lengths were analysed with Genstat[®] ver. 5.0 statistical software package using non-linear regression models. Back-calculated length-at-age data were produced using a regression of the plot of otolith radius and fish length (Burton, 2001). This was used to produce von Bertalanffy growth parameters initially using VONBIT software (Stamatopoulos, 1999) and then the Genstat[®] ver. 5.0 package. Comparison of growth equations was made using non-linear regression models in Genstat. Analysis of differences in back-calculation equations and von Bertalanffy equations were conducted on fish collected from the four Queensland east coast zones: North, Wet tropics, Central and South-east Queensland (see Figure 14). Back-calculated length at age was estimated by testing the applicability of several equations such as (Escot and Granado-Lorencio, 1999). Otolith radius and fish length regression differences were tested between sexes and locations. Growth rates for separate regions and between fish caught in research sampling and those caught by recreational fishers (ANSA) were tested using linear regression and an accumulated ANOVA.

Percentage error of ageing estimates was calculated using average percent error (APE) and an age bias plot used to determine bias.



*Figure 14. Location of discreet zones in eastern Queensland.
These were used to determine geographical growth differences.*

Results

A total of 1 325 otoliths were collected from mangrove jack between November 1999 and March 2002. Of these, 1147 otoliths were sectioned (97%), 79 otoliths were kept for microchemistry (some of these were also sectioned) and 56 otoliths were retained for daily increment analyses (10%). There were only 69 otolith sets that were found to be unusable either because of deep cracking or breakage. The breakages were mainly caused by the fisher using a knife to pith the fish through the cranium, or through breakage during extraction from the fish (3% total loss of samples).

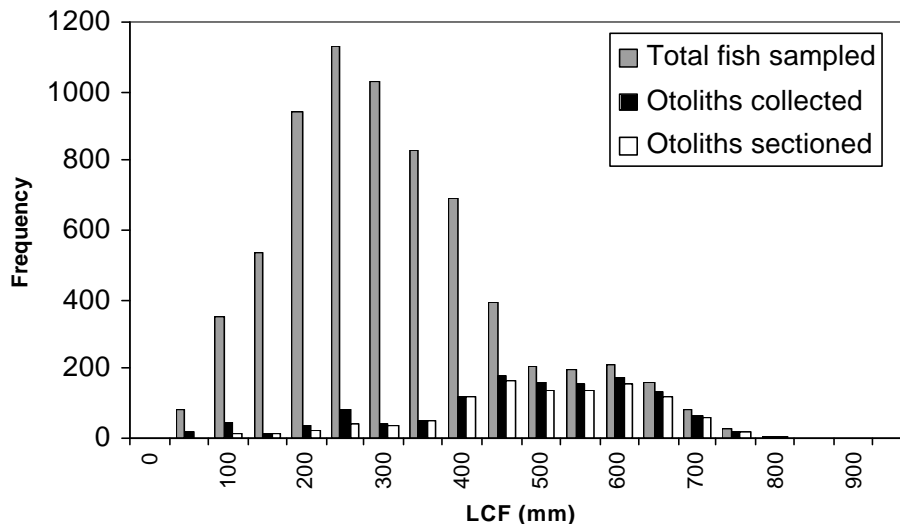


Figure 15. Proportion of fish sampled, otoliths collected and sectioned in each fish size class.

As shown in Figure 15, while most of the fish sampled in the larger size classes have had their otoliths recovered, the majority of smaller fish (≤ 450 mm LCF) sampled from electrofishing operations (5 607 individuals) were not euthanased but were tagged and released. The sampling protocol required that only about 5% of the individuals captured were to be destructively sampled. This was to minimise adverse impacts on stocks at the sampling locations and to ensure that there was sufficient numbers of tagged fish for the movement studies. Otoliths were collected from nearly 80% of the fish above 450 mm LCF that were sampled.

Back-calculation

The relationship of fork length and otolith radius was used to back-calculate the fish length at last increment formation (L_i). This was done by first finding a relationship between the otolith radius and the length at capture (L_c) (Burton, 2001)(Equation 1). Fish with less than one increment were excluded from the analysis as these fish were not fully recruited to the 1+ year class and did not have any increment where the age could be back-calculated.

Equation 1

$$L_c = a + b (R_c)$$

Although the relationship for all mangrove jack samples produced a reasonable linear fit ($r^2=0.8841$), a plot of the residuals revealed that the relationship would be better described by a curve-linear relationship (Zar, 1984). Plots of quadratic, log-log and asymptotic had higher

Table 11. Various equations used to describe the relationship between fish length and otolith radius.

Information from 123 samples were used to calculate these equations.[@] ($P<0.001$). [§] Constants and SE in full: $-124.0\pm 13.6^@$, $165.20\pm 5.97^@$, $-7.298\pm 0.633^@$

Equation type	Equation	Coefficient of determination (r^2)	Shape of residual plot
Linear	LCF=22.215+97.336 (RAD)	0.8841	Curvilinear
Quadratic [§]	LCF=-124.0+165.2(RAD)-7.298(RAD ²)	0.897	Linear
Exponential	LCF=1144.1-1296.4*0.8643 ^(RAD)	0.896	Curvilinear
Log-log	LCF _{log} =1.0306(RAD _{log})+1.9868	0.9065	Curvilinear

r^2 values, with the plots of residuals equally spaced around zero only for the quadratic equation but this also produced increased variances with otolith radius (Table 11). It was decided that the quadratic equation was the most parsimonious model because of the reduced number of constants and increased fit of data. This model also appeared to show some uncoupling of otolith width with fish length similar to that described for other species such as Murray Cod (Anderson *et al.*, 1992). In comparison, Cappo *et al.* (2000) found significantly increased growth rate along the ventral axis of sectioned otoliths of captive *L. johnii*, but this increase did not translate to increased otolith weight when compared to fish length. The inflection point of the quadratic relationship generally falls in the range where mangrove jack begin to sexually mature ($L_m= 449$ mm for males and 512 mm for females – see Reproduction section). This may explain the reason for the change in otolith growth with respect to fish length. It is also at this point that the somatic growth of the fish is reduced (see later this chapter).

In a study of the European barbel (*Barbus sclateri*), Escot and Granado-Lorencio (1999) found that they had to log-transform their regression to stabilise the increasing variances associated with the larger otolith radius. However, this did not work for mangrove jack as the plot of residuals was still curvilinear. Analysis of covariance (ANCOVA) was also used by these authors to demonstrate that this single regression was suitable for both sexes of European barbel, but separate equations were needed to suitably explain the relationship from different locations.

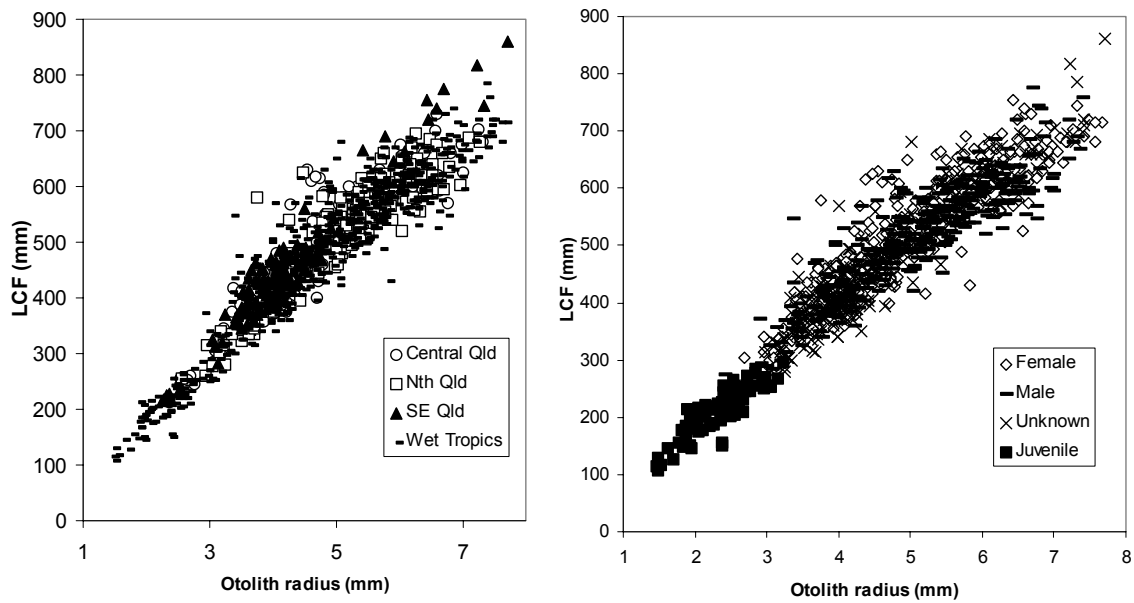


Figure 16. Scatter plot of LCF and otolith radius for separate zones (left) and for separate sexes (right).

The plot of LCF versus otolith radius in Figure 16 shows one apparent anomaly. The fish from southeastern Queensland (triangular symbols) appear to fit a separate regression to the fish from all other areas (mostly collected from the North Queensland and the Wet Tropics zones (n=1028, 91%)). To determine if there were separate regressions for each sex, the unknown and juvenile fish were excluded from the analysis. An accumulated ANOVA of the pooled sex dataset revealed that although the “shapes”(or quadratic function) of the regressions were similar (P=0.420, 1 df, F=0.65), the level of displacement between the two curves was significant (P=0.010, 1df, F=6.73) (Table 12). Plots of the respective regressions for males and females show some interesting trends. Up until about 436 mm LCF, the radius of the male otoliths is larger than the radius for the female otoliths after which the reverse occurs with female radii larger for the same sized fish. The point at which the two curves cross is approximately the point where the logistic curve for male size-at-maturity becomes linear (see Reproduction section).

Table 12. Parameters for quadratic equations for separate sex regressions for LCF and otolith radius comparisons.

$$LCF = \text{Intercept} + RAD (\text{radius}) + RADSQ (\text{Radius}^2). \quad n=769. \quad ** (P < 0.001)$$

Sex	Intercept	RAD	RADSQ±SE
Female	-107.2**	161.7**	-7.08±1.22**
Male	-76.1**	154.18**	

There were significant differences between the regressions (Figure 16) in otolith radius and LCF for the various zones ($P=0.003$, 3df, $F=4.64$, $n=1028$)(Table 13). The differences are due to the South-east Queensland fish when compared with fish from the other zones. All parameter estimates for central, wet tropics and north Queensland are not significantly different. The geographical and sex differences described above have been noted in other migratory species such as spotted mackerel (Cameron and Begg, 2002). It is important to note that the sample sizes from both Central and South-east Queensland were small (<100 fish), and hence caution needs to be used when examining the significance of the relationships.

Table 13. Regression parameters for separate zone equations for the LCF and otolith radius relationship.

Explanation of terms is same as table above. $n=1028$. * $P<0.05$, ** $P<0.001$.

Zone	Intercept \pm SE	RAD \pm SE	RADSQ \pm SE
South-eastern Queensland	-67.2 \pm 83.4*	128.4 \pm 35.2*	-1.08 \pm 3.56*
Central Queensland	-242.1 \pm 64.7**	216.1 \pm 27.2**	-12.28 \pm 2.73**
Wet Tropics	-123.61 \pm 66.3	168.0 \pm 28.0	-7.68 \pm 2.82
North Queensland	-234.6 \pm 93.4	211.9 \pm 38.6	-12.23 \pm 3.86

Because there were differences in the regressions using pooled data for male and female fish it was decided to extend the analyses to examine the regressions for male and female fish from the zones on the eastern coast of Queensland (Figure 14). An accumulated ANOVA was used, first on linear and then on the quadratic regressions to determine if differences in the relationship between LCF and otolith radius occurred between sexes within each zone (Table 14). For south-eastern Queensland ($r^2=0.914$)($P<0.001$, 1df, $F=709.91$, $n=67$) and north Queensland ($r^2=0.732$)($p=0.015$, 1df, $F=6.10$, $n=146$) zones, linear regressions suitably explained the relationship between LCF and otolith radius. In the case of south-eastern Queensland, the data were mainly restricted to smaller fish with the resulting residuals being highly variable and not normally distributed. Because of these anomalies with the data, no significant differences in the length-otolith radius relationship could be found between the sexes in south-eastern Queensland.

In contrast, there were significant differences between sexes for the expressions calculated for fish from the wet tropics ($r^2=0.809$)($p<0.001$, 1df, $F=17.54$, $n=480$) and central Queensland ($r^2=0.819$)($p<0.001$, 1df, $F=14.24$, $n=73$).

Table 14. Regression parameters for equations explaining the relationship between length and otolith radius in each of the east coast zones.

*($P<0.001$) **($P<0.05$), n.a is not applicable)

Zone	Sex	Intercept \pm SE	RAD \pm SE	RADSQ \pm SE
SE Queensland	Both	-31.5 \pm 19.2	115.49 \pm 4.33**	n.a.
Central Queensland	M	-318.5 \pm 55	255.2 \pm 11.1	-17.12 \pm 4.54*
Central Queensland	F	-371 \pm 114**	269.7 \pm 46.2*	
Wet Tropics	M	-55.4 \pm 19.6	147.68 \pm 3.76	-6.59 \pm 1.57*
Wet Tropics	F	-88.6 \pm 42.4**	154.6 \pm 16.5*	
North Queensland	M	185.6 \pm 46.4**	66.14 \pm 8.5**	n.a.
North Queensland	F	85.2 \pm 26.3**	87.13 \pm 4.97*	n.a.

Once the relationship between otolith radius and fork length of the fish was established, then the next step was to determine the best method for back-calculation. The biological intercept method, (Equation 2)(Campana, 1990a) was found to best describe this relationship because it gives very small deviations to expected values (Escot and Granado-Lorencio, 1999). Two

parameters were needed as a prerequisite for establishing this regression; the length of the fish at hatching (L_o) and otolith radius (R_o) at hatching (Campana, 1990a). Unfortunately, these parameters were not available for wild fish but estimated values were used to compare this equation with the linear correlation between otolith radius and the length at capture (Equation 1). This equation was also compared with the quadratic form of the body-proportional equation (Equation 3)(Francis, 1990).

Equation 2

$$\text{Log } L_i = \text{Log } L_c + (\text{Log } R_i - \text{log } R_c)(\text{log } L_c - \text{Log } L_o) / (\text{Log } R_c - \text{Log } R_o)$$

Equation 3

$$L_i = [(\text{regression } (R_i)) / (\text{regression } (R_c))] * L_c$$

where: L_i = Length at age i

L_c = Length at capture

L_o = Length at hatching

R_i = Otolith radius at increment i

R_c = Otolith radius at capture

R_o = Otolith radius at hatching

regression = suitable regression from equations above

To improve the precision of the predicted values, only fish with an increment that was $\geq 95\%$ of the total radius of the otolith was used in the test for the best fit of predicted and observed values (Escot and Granado-Lorencio, 1999).

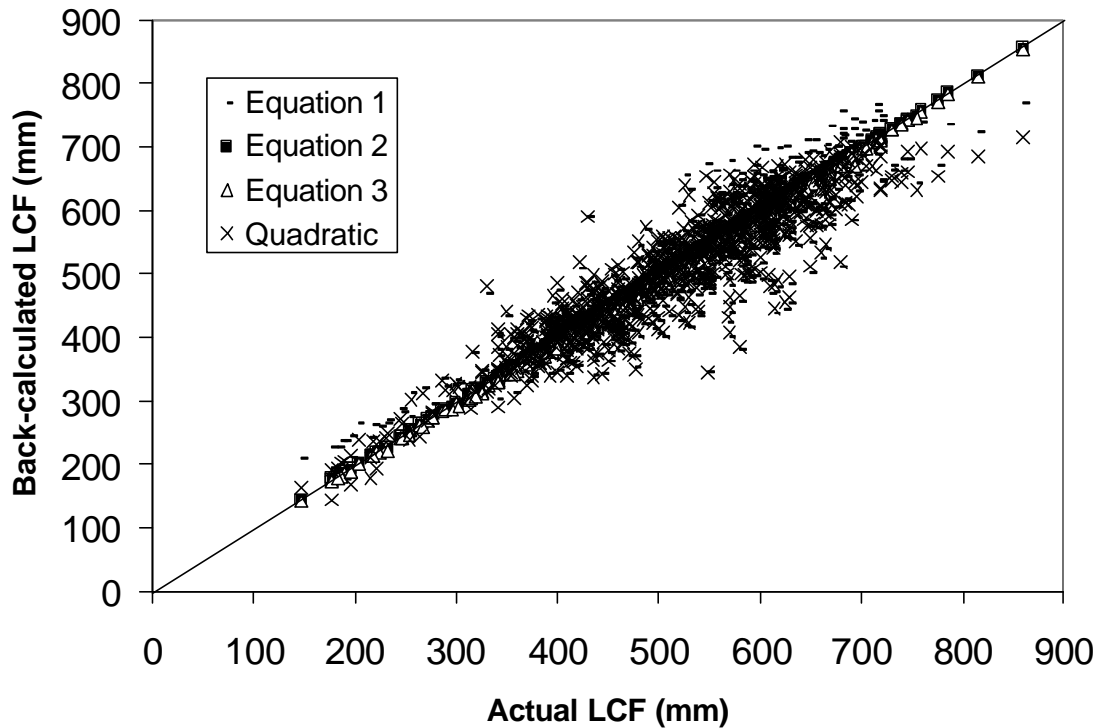


Figure 17. Back-calculated length vs observed length for mangrove jack (combined sexes)

Both the body-proportional equation and the biological intercept method produced low deviances from observed values, with all fish within 95-100% of the observed values and an average deviance of 1.65% and 0.98% respectively (Equation 2, Equation 3) (Figure 17). The regression equation (Equation 1) (Figure 17) produced results that were highly scattered with only 22.8% of values within the accepted range, and an average deviation of 7.24%. The regression model also underestimated values for smaller lengths and overestimated the values for larger lengths. Because the first two equations used the predicted value relative to the observed value in the equation, the fitted values fell within the accepted range in more than 95% of observed values. Based on the observation that there was very little difference in estimation between the first two equations, and the fact that the constants for the biological intercept method were only estimated values, the body-proportional equation (Equation 3) was used for correcting for time of capture.

Age validation

Age validation is an essential requirement for the use of any ageing technique (Beamish and McFarlane, 1983). Two of the most commonly used techniques in the literature are to mark otoliths (and other calcareous structures) with a dye or to use marginal increment analysis. Both of these techniques have been used successfully previously in studies with small numbers of mangrove jack in north Queensland. (Sheaves, 1995; Cappo *et al.*, 2000).

Broodstock fish

Fish with a known history

Table 15 shows the records of seven broodstock mangrove jack which died during the period of the project. These fish were held either in tanks at the Northern Fisheries Centre in Cairns or in ponds at the Oonoonba Veterinary Laboratory in Townsville. The ages of these fish were estimated from sectioned otoliths and all estimates were found to be greater than the time period that the fish were in captivity. The lengths back-calculated for the year of capture using the Wet Tropics zone otolith equation (Table 14) and the body-proportional equation provides further evidence that the increments are formed annually. The estimated lengths from this method are imprecise, varying from 13 mm to 166 mm from actual fish length. This demonstrates that the increments are formed annually and that, for most fish, this method provides a reasonable estimate of length at capture. The previous studies using oxytetracycline to validate ageing techniques used only juvenile or sub-adult fish (Sheaves, 1995; Cappo *et al.*, 2000), whereas these data provide evidence of validation for adult fish.

Table 15. Details of mangrove jack broodstock including length at capture and death, estimated age and back-calculated length at capture.

Back-calculated length is derived from the standard curves for each sex. ^a identified from internal pit tag number; LCF ^b of capture from measurement of annual increment radius; ^cbold numbers are length at closest increment to time of capture.

Tag	Sex	Date of death	LCF death (mm)	Time in captivity (yr)	Age (yr)	LCF ^b (mm)	Back-calculated lengths (mm) ^c
A165	F	29/1/01	585	2.72	9+	412	400- 425
00400	M	4/4/01	580	3.95	8+	475	443- 490
A398	F	4/4/01	665	7.11	13+	478	432- 509
A239	F	4/4/01	625	5.89	12+	449	491- 534
----- ^a	F	2/11/01	705	7.97	14+	405	429- 475
A008	M	20/12/01	590	4.20	9+	351	491- 517
A395	M	1/1/02	665	6.12	16+	500	465- 518

Oxytetracycline injection

In their respective studies both Cappo *et al.* (2000) and Sheaves (1995) observed the expected number of growth increments on recaptured fish that had been previously injected with OTC. These recaptures were from wild fish and represented only 8-10% (Cappo *et al.*, 2000) and 1% (Sheaves, 1995) of the total fish marked in their experiments. During this current study an experiment was initiated to use OTC in wild fish to determine if otoliths increments were formed annually. The experiment was done on fish resident in Lake Placid on the Barron River in January 2002. Lake Placid was chosen because it is a relatively confined area where it was thought that the probability of recapturing the fish would be higher than in another open river system. To date there has only been one recapture by a recreational angler and the fish frame was not kept so no otoliths were recovered.

In the NFC aquaculture program broodstock mangrove jack were regularly injected with the antibiotic Terramycin® (OTC) at the recommended dose of 50mg/kg of body weight for treatment of external ulcers or lesions (Julian O'Brien, Qld Department of Primary Industry, pers. comm.)(Table 16). The otoliths were obtained from four of these fish that had been in captivity for between one and two years from the time they were injected with OTC to when they died.

Table 16. Details of OTC injected broodstock and the associated correlation between time at liberty and increment formation in the otoliths.

CD is the annual Closing Date. **Tag lost in tank, identification deduced from other fish present in tank.

Tag	Sex	Time at liberty (yr)	No. increments formed	Final LCF (mm)	Age	CD
A559	M	1.05	1.09	660	16	26 th Aug.
A301	---	1.76	1.88	540	7	6 th Sep.
A053	---	1.76	1.8	502	8	12 th Aug.
**	---	1.76	1.81	512	7	19 th Aug.

The last three fish in Table 16 were all injected with OTC on the same day and all died on the same day in the same tank due to an oxygen shortage. All three fish showed very clear increment formation and a very clear OTC mark at a short distance after the closing of the previous increment (0-3.3% of next increment width) (Plate 2). Because these fish were injected on the same day (15th September 1997), and the percentage increment values are similar, this strongly suggests that the previous increment probably formed up to about a month earlier. The number of increments after the OTC band and the width of the marginal increment also agree closely with the known time at liberty. In all fish an increment was beginning to form at about the time of death (20th June 1999), which is consistent with the estimated time the increment was laid down before the OTC band formed.

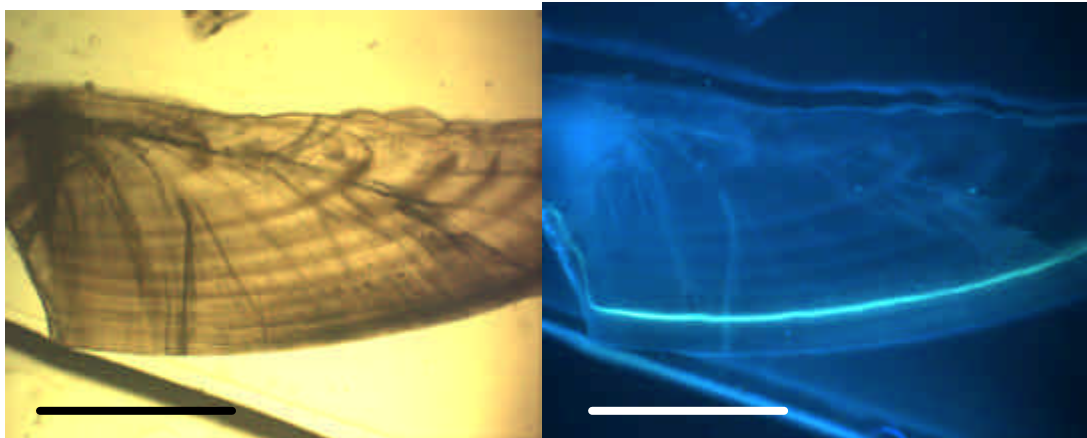


Plate 2. Photograph of sectioned otolith from fish A301 using transmitted (left) and then UV light (right) sources.

Note clear OTC mark throughout otolith. Scalebar in each image is 1 mm long. Photo: Corey Green (CAF).

The remaining fish in Table 16 (A559) was injected with OTC after it was purchased from a live fish trader on 14th December 1999. It was subsequently held at NFC for just over a year before it died. The OTC mark on its otolith was proportionally further away from the previous increment (27% of next increment width) but the closing date, calculated for the following year, was similar to that calculated for the other fish.

All four fish were kept in flow-through 53 tonne tanks during the time that they were held at NFC. The tank water was pumped from Trinity Inlet and the water temperatures in the tanks followed a similar pattern to the temperature regime in the Inlet.

Impoundment F1 fish

Otoliths were obtained from one of 181 mangrove jack stocked into Tinaroo Dam, on the Atherton Tablelands west of Cairns (Table 17). Because this fish was spawned and reared at the NFC, its exact hatch-date and age were known to the day.

Table 17. Details on growth of the recaptured mangrove jack from Tinaroo Dam.

LCF start (mm)	LCF final (mm)	Growth rate at NFC (mm/day)	Growth rate in dam (mm/day)	Age (known) (yr)	Age estimated and marginal increment
105	340	0.42	0.36	2.18	2.30

Table 17 shows the estimated age determined from the otoliths and the actual known age. Both of these ages are similar; the otolith displayed the correct number of increments and the width of the marginal increment was about what was expected. In otolith section, the increments were clearly identifiable and may add further evidence of the importance of significant seasonal water temperature differences to provide distinct annual increments. Lake Tinaroo, where the fish was caught is at a higher altitude than the coastal rivers where most sub-adult and juvenile samples were taken, and therefore has cooler winter water temperatures (MacKinnon and Herbert, 1996). The general quadratic regression (length vs otolith radius) for the Wet Tropics and body-proportional equation were used to estimate length at the second increment. By substituting the otolith radius into this equation the estimated length-at-capture of this fish was 340 mm LCF, which corresponded to the exact length at capture. The estimated length at the completion of the second increment was 291 mm, which is 126 mm larger than the mean length at age calculated for wet tropics fish using the von Bertalanffy equation (see Age estimates section later this chapter) and shows that the impoundment fish may grow at an appreciably faster rate.

Recaptured wild fish

Recaptured wild fish may also be used to add weight to the validation techniques outlined above as these fish are subject to a range of environmental influences including food availability, temperature, salinity changes and other stresses, that may have impacts on the formation of increments (Romanek, 1996). The otoliths of four recaptured mangrove jack of different sizes and from different habitats were kept for age determination (Table 18).

Table 18. Sectioned otolith ages and back-calculated sizes (LCF) at tagging for recaptured mangrove jack.

See Table 15 for explanatory notes.

Recapture location	Release Date	Release size (mm)	Back-calculated length (mm)	Recapture Date	Recapture size (mm)	Age
Offshore	23/06/00	416	386 - 436	21/03/01	445	6+
River	20/09/00	408	392 - 411	28/07/01	435	11+
River	17/04/00	290	261 - 290	18/07/01	303	5+
Freshwater lagoon	31/12/94	415	380 - 442	11/12/99	590	9+

Note that the first two fish were at liberty for less than a year, with one of the fish moving from the release location in the Russell River system to an offshore reef. The last fish in the table was tagged and recaptured in a freshwater lagoon and had showed significant growth (175 mm) in the five years it was at liberty. The back-calculated lengths were determined

using the body proportional equation and show its utility in providing a reasonable estimate of the length at the previous increment. For both of the river fish, the back-calculated lengths were very close to the observed length-at-release, however the fish recaptured offshore showed a slightly higher back-calculated length (possibly due to measurement error because the fish was gilled and gutted). The estimate of the length at the 4th year increment for the freshwater fish was 35 mm less than the observed value. The freshwater fish provides evidence that growth rates may vary according to environmental conditions and that fish stocked in lentic habitats may exhibit faster growth rates than those in estuaries or rivers. This freshwater fish was large (590mm) for its age when compared to the mean length of 515 mm of a nine year old fish from Central Queensland (estimated using the von Bertalanffy equation - see Age estimates section later this chapter). Access into the waterhole was restricted to years when there were reasonable flow events therefore this fish would probably have been recruited in the 1990/91 wet season.

Marginal increment analysis

Marginal increment analysis is another technique used to validate the annual formation of increments. (Beamish and McFarlane, 1983). Although this technique could be used across all age classes to confirm the annual nature of increment formation, the difficulty in interpreting very small increment widths in larger fish results in an increased coefficient of variation. For example, the coefficient of determination doubled from approximately 4% (ages 1-9) to more than 8% thereafter. This was associated with a 50% decrease in increment width (Figure 18). For this reason, only fish with fewer than ten increments were used in the marginal increment analysis.

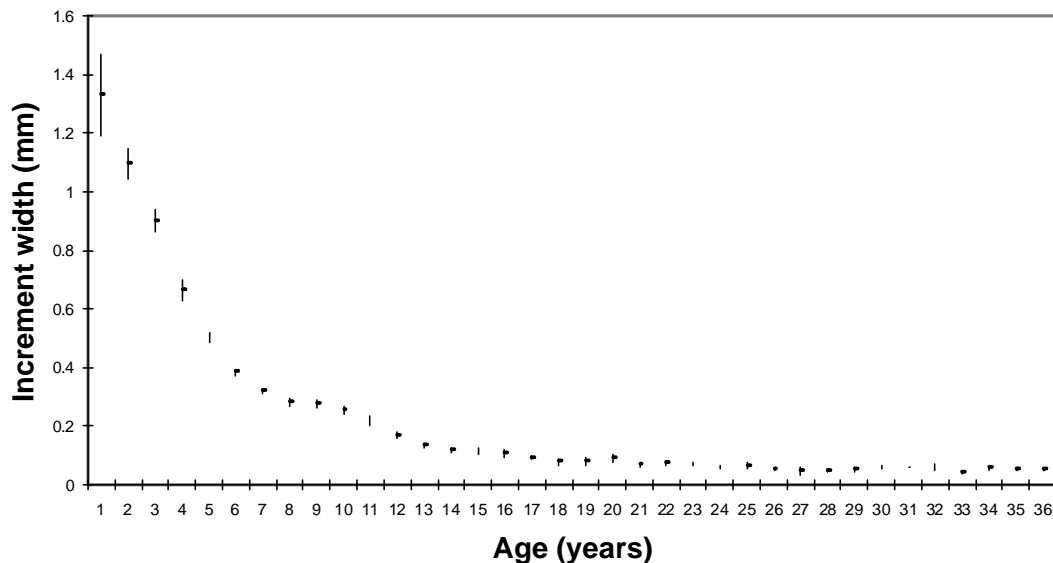


Figure 18. Increment width (\pm SE) in sectioned mangrove jack otoliths from fish of different ages. Note that increment width decreases with increasing age due to declining somatic growth..

As shown in Figure 19, the mean monthly marginal increment percentage is at a minimum in November (late spring) and a peak value in July (mid-winter). This is consistent with the results of another study of mangrove jack (Cappo *et al.*, 2000) that, using OTC marked individuals, estimated October 26 as the mean calendar closing date (MCCD) for annulus formation. This compares to a MCCD of 10th August (average of values from Table 16) for the broodstock kept at the NFC on flow-through seawater. The two and a half month

differences in these two estimates of the MCCD could be due to a number of factors. The fish used in this comparison came from two spatially separate locations more than 300 km apart. Further, changes in temperature of the water from the estuarine system that supplied the broodstock tanks probably occurred more quickly than in the oceanic water of the GBR. Also, the fish used in the NFC study were captive broodstock whereas the fish used in the Cappo study were all wild tag-and-release fish. In the literature there is some variation in the timing of the completion of the increment in other lutjanids on the GBR, however all species appear to complete formation between late winter and late spring (August to November)(Newman *et al.*, 1996a; Cappo *et al.*, 2000). The timing may also be affected by changes in other environmental conditions such as turbidity, sea conditions and salinity. Cappo *et al.* (2000) found that extra increments were formed during the cyclone season, with the effect of significantly changing the estimated closing dates.

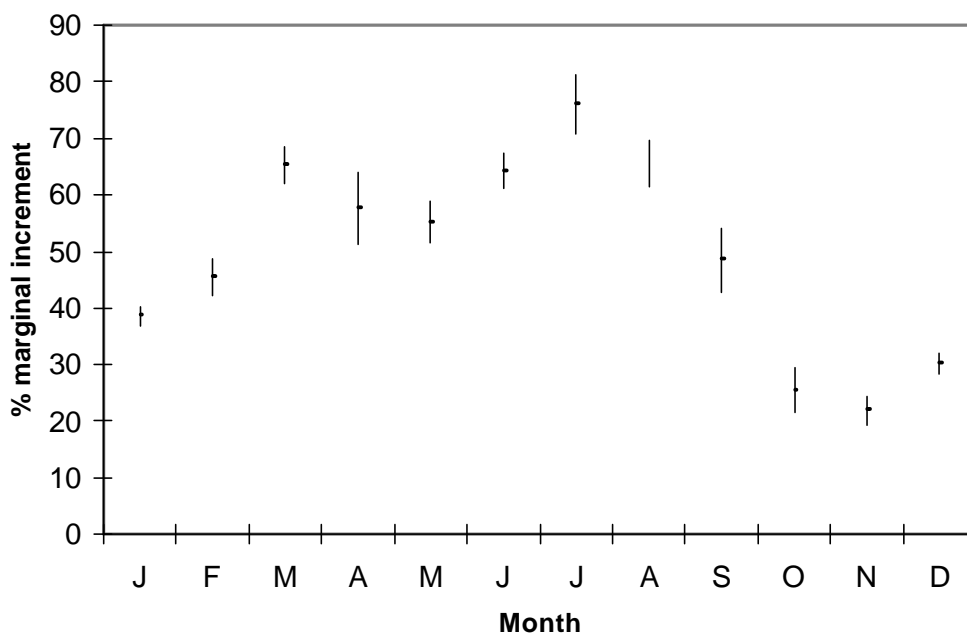


Figure 19. Mean monthly marginal increment percentages (\pm SE) for 651 mangrove jack otoliths. To allow the marginal increment to be accurately measured, only fish with less than 10 whole increments were included in these analyses.

Because the mangrove jack in this study were collected from a wide geographical range throughout Australia covering more than 19° of latitude and 37° longitude, an attempt was made to investigate what affects geographical influences have on the timing of increment formation. However, with the exception of the south-east Queensland and Wet Tropics regions, there were insufficient data available to plot monthly marginal increments (Figure 20).

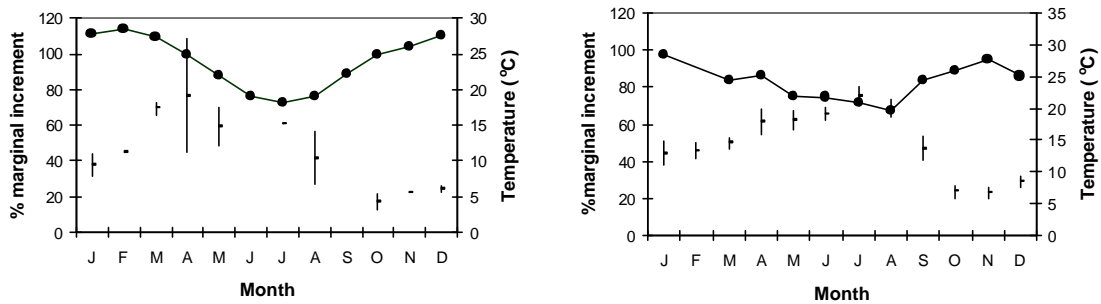


Figure 20. Marginal increment percentages for mangrove jack collected in Baffle Creek (left) in the south-eastern zone of Queensland and the Russell River (right) in the wet tropics zone.

Ambient water temperature (?) are plotted as separate series.

The graph of the Russell River temperatures (Figure 20) represents the trend that occurs in all wet tropics streams and, when compared with the marginal increment percentages, suggest an inverse relationship between percentage marginal increment and temperature. As the temperature decreases after summer and the onset of the wet season (January –March), there is a gradual decrease in the growth of the fish and the marginal increment slowly increases. As winter progresses, and the temperature reaches a minimum (July/August), the marginal increment percentage stalls as fish reach their lowest growth rate for the year. As water temperatures begin to climb throughout spring, fish begin to rapidly grow and produce growth on the outside of the increment. Although the temperature information is sparse for Baffle Creek, the pattern is consistent, with a minimum percentage increment also occurring in October (Figure 20).

One minimum in the marginal increment percentage for each age class is a key requirement for complete validation (Beamish and McFarlane, 1983). The plots of mean marginal increment percent for each year increment class of 2+ to 9+ fish are shown (Figure 21). In all age classes the minimal value is found between September and November. In the younger fish (2+ year class), which are from inshore areas, the minimum value is formed earlier in September but later in November in the older (7+ to 9+) offshore fish. The fish with fewer than two full increments have been excluded from this graph due to very small sample sizes.

Age estimates

Age estimates were standardised to the length at the formation of the last annulus to reduce the error associated with age-at-length estimates from different times of capture throughout the year (Ricker, 1975; Escot and Granado-Lorencio, 1999; Burton, 2001). The back-calculated length-at-age data were plotted for separate sexes and for separate areas (see Figure 14) to determine if there was any significant relationship (Figure 22 and Figure 23).

Using pooled data from all east Queensland sampling locations, there was a significant difference between the sexes in the von Bertalanffy growth estimates ($r^2=0.77$) ($P<0.001$, 1df, $F=12.30$, $n=769$). Some data were excluded from this analysis for the same reasons given in the back-calculation procedures. Although the shapes of the regressions were the same, the estimates for t_0 and L_8 were significantly different ($P<0.001$)(Figure 23). The differences between the estimates for t_0 should be disregarded as the data were skewed by the disproportionately small number of juvenile fish represented.

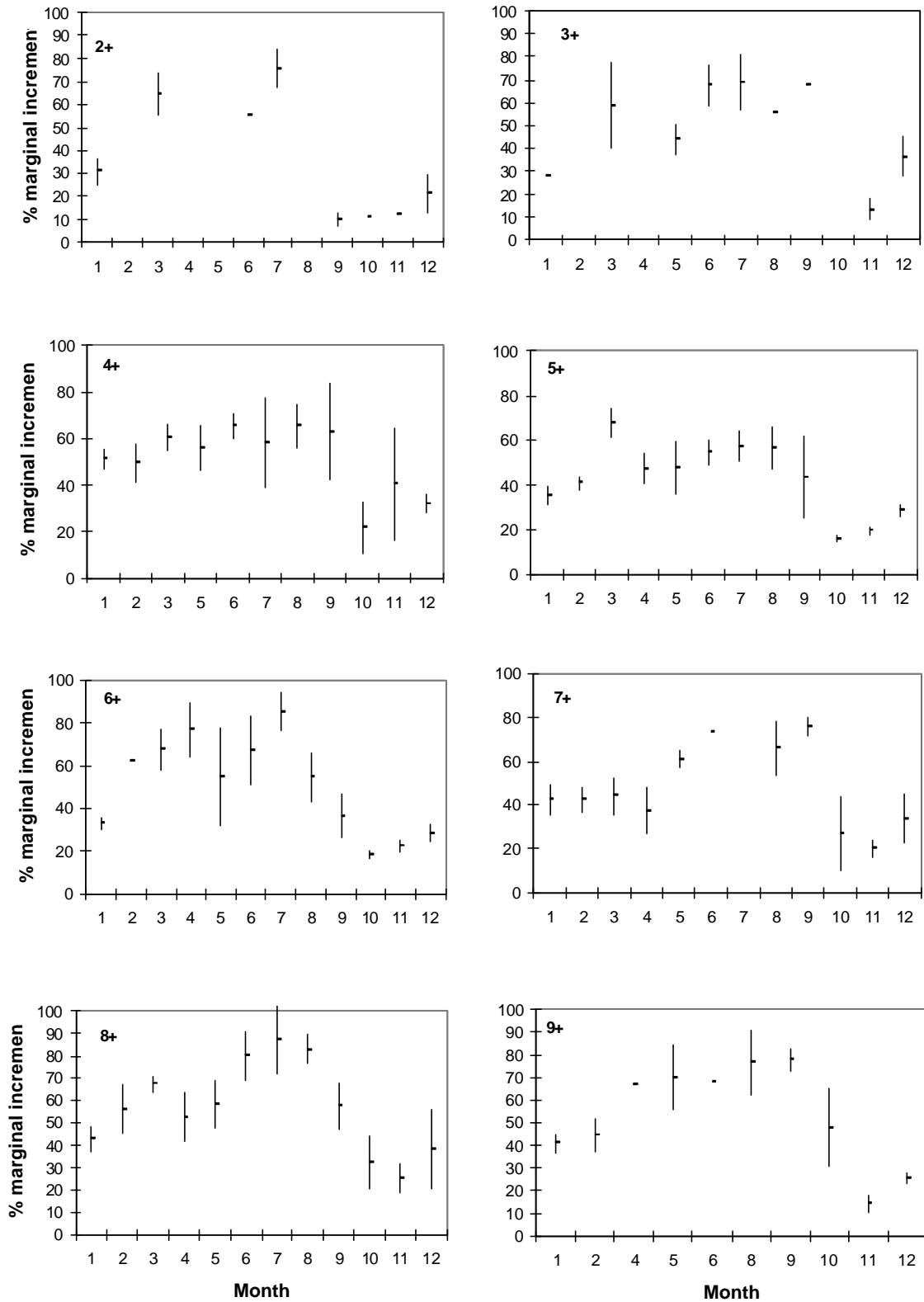


Figure 21. Mean monthly marginal increment percentages for age classes 2+ to 9+.

To determine if there were differences in growth parameters between fish caught in different areas, fish were assigned to zones based on similarities in temperature, rainfall and latitude (Figure 14). The analyses were undertaken using an accumulated ANOVA design and showed significant differences in parameters for the eastern Queensland zones ($r^2=0.876$) ($P=0.002$, 3 df, $F=4.89$, $n=1028$)(Table 19). Figure 22 illustrates that, for the pooled sex data, the predicted K value decreases while L_8 increases with increasing latitude. However, due to limited sample size from the more remote zones (WA, NT, Gulf and NSW), this analysis could only be performed for the eastern zones (Figure 22). Further, the sample sizes in both central and south-eastern Queensland are small (<100 fish) and apparent differences in these zones may be attributed simply to the small sample sizes or differences in age structure of samples.

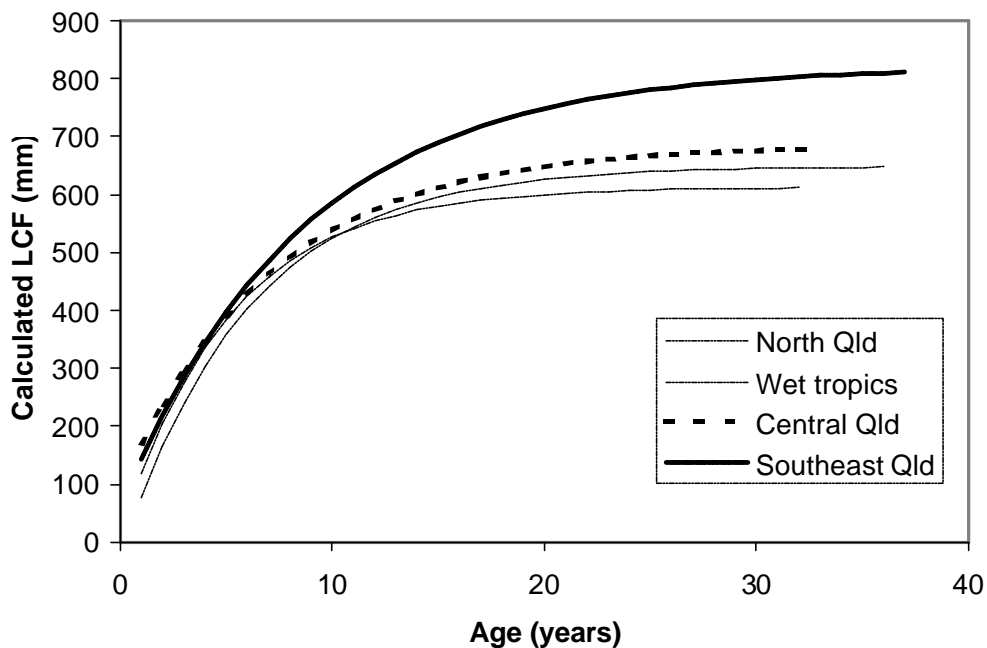


Figure 22. von Bertalanffy curves for each of the four eastern Queensland geographic zones (north, wet tropics, central and south-eastern Queensland).

The accumulated ANOVA showed differences in growth parameters for male and female fish within zones (Table 19). Again small sample sizes limited the analyses that could be undertaken but there were significant differences in the north Queensland and wet tropics zones.

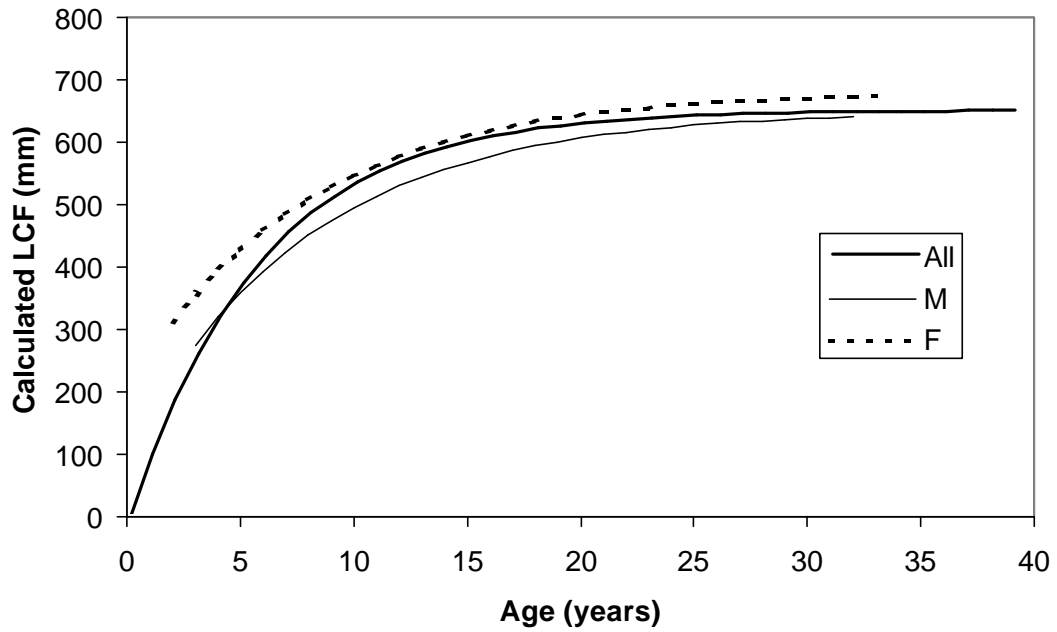


Figure 23. von Bertalanffy length-at-age growth curves for pooled, male and female mangrove jack from the east coast of Queensland.

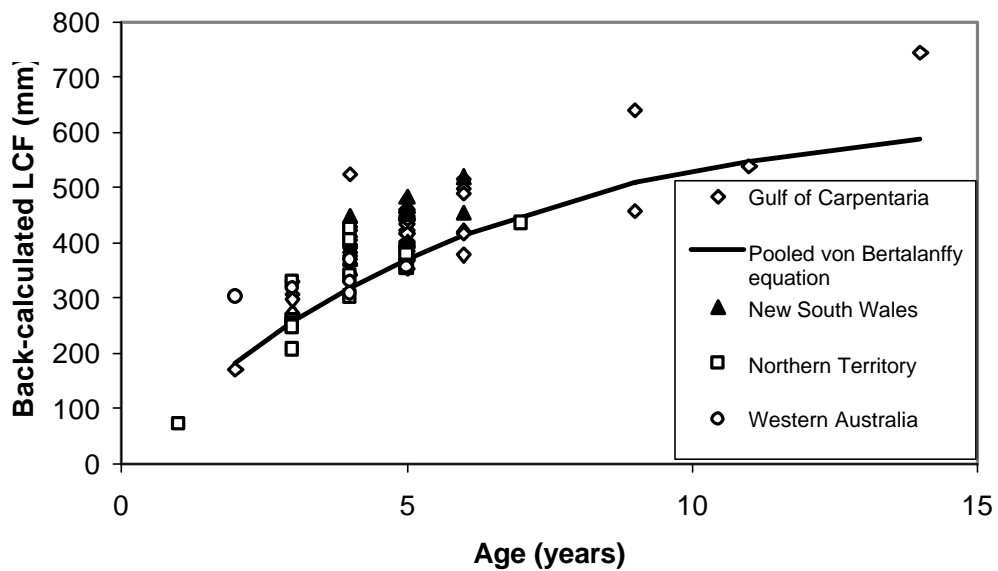


Figure 24. von Bertalanffy curve for pooled data and samples collected from other areas within Australia and overseas.

Note that New South Wales and Western Australian fish appear to have a relatively fast growth rate when compared to the pooled data.

Where separate von Bertalanffy parameters could not be estimated for a zone, the back-calculated lengths-at-age were plotted against the pooled von Bertalanffy curve as illustrated in Figure 24. It appears that, assuming equal ageing errors between zones, there are some trends not explained by the general equation. The slope of the fitted regression for the fish from the Gulf of Carpentaria is greater than the general equation (lower K value), even though the general equation fits within the range of values. The fish from the Northern Territory and Western Australia seemed to be described adequately by the equation except the one 300 mm fish (age 2 years) from Western Australia. This fish was recaptured 50 n.mi. offshore only 160 days after being tagged in a freshwater area, perhaps further evidence of faster somatic growth in freshwater sites for this species. The growth of fish from New South Wales also appeared to be greater than the general growth rate and may be better described by the south-eastern Queensland equation.

Table 19. The von Bertalanffy equation parameters (\pm SE) for all analyses of fish from the east coast of Queensland.

* $p < 0.05$

Zone	Sex	r^2	n	t_0	SE	K	SE	L_8	SE
North (all data)	n.a	0.876	1028	0.68	0.30	0.193	0.021	612.8	11.6
Wet tropics (all data)				0.05	0.09	0.170	0.006	649.08	5.77
Central (all data)				1.07	0.35	0.139	0.018	685.3	20.6
South-east (all data)				-0.90	0.34	0.117	0.013	821.4	30.7
North	M	0.66	146	1.77	0.49	0.164	0.017	616.2*	-
	F							632.7*	-
Wet tropics	M	0.77	480	2.364	0.381	0.136	0.007	644.2	12.6
	F			1.051				0.369	673.7
All Zones	M	0.77	769	1.761	0.298	0.126	0.006	650.6	10.4
	F			2.893				0.254	681.2

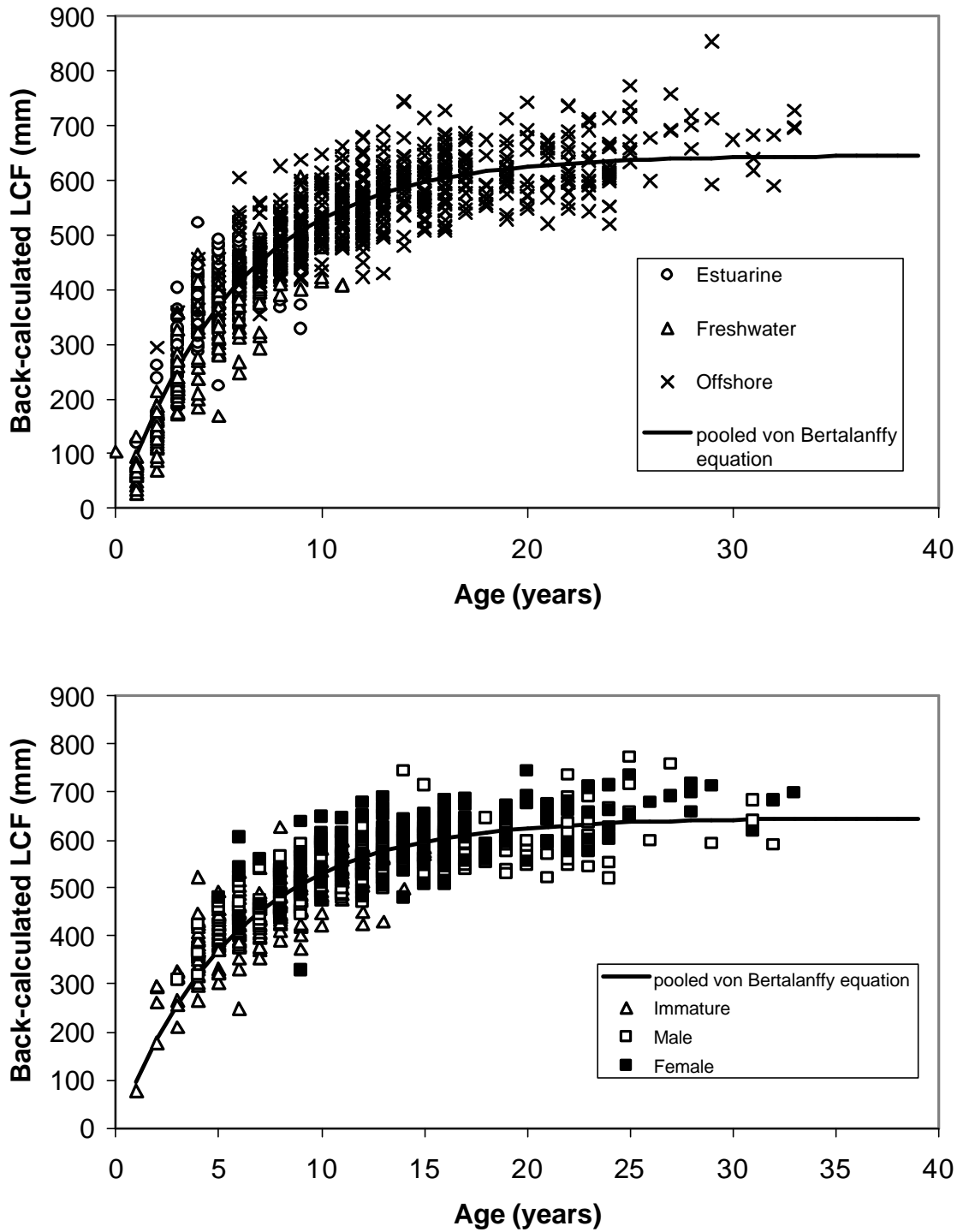


Figure 25. von Bertalanffy curve showing pooled data split into estuarine, freshwater and offshore habitats (top) and male, female and immature fish (bottom).

The two graphics in Figure 25 illustrates that generally most immature fish are less than about 10 years old and that most fish from these age classes were sampled inshore while the older, mature fish of both sexes were generally found offshore. There is some overlap where offshore fish are still immature.

Table 20. Average length-at-age data for mangrove jack from each of the zones within Australia.

Standard error and sample size shown in brackets. Where brackets are missing, only one sample available.

Age	Zone							
	WA	NT	Gulf	North Qld	Wet Tropics	Cental Qld	SE Qld	NSW
1		72			79 (9,11)			
2	302	-	170	200(31,3)	147(6,34)	209(29,2)	158(7,6)	
3	317	258(14,7)	301 12,4)	251(9,7)	241(11,3)	281(20,7)	299(18,6)	
4	336(18,3)	368(29,4)	412(14,10)	351(13,12)	326(10,38)	372(16,14)	375(8,16)	397(16,6)
5	355	367(12,2)	414(7,22)	383(32,3)	368(6,59)	378(5,13)	412(6,31)	450(17,4)
6		-	453(23,6)	442(33,7)	403(7,64)	416(17,9)	438(8,28)	487(33,2)
7		435	-	499(17,5)	430(8,45)	468(34,2)	418(12,7)	
8			-	485(17,9)	470(7,51)	509(31,3)	493(6,2)	
9			548(91,2)	479(10,11)	500(8,41)	524(30,5)	495(15,2)	
10			-	522(15,10)	518(10,27)	551(30,5)	648	
11			538	538(14,14)	548(9,37)	551(16,3)	-	
12			-	551(12,15)	555(9,34)	530(36,4)	638(10,2)	
13			-	555(12,13)	576(9,28)	605(11,3)	690	
14			744	578(14,10)	597(10,17)	-	745	
15				564(7,11)	593(11,18)	611(7,2)	711	
16				597(13,14)	604(13,21)	632(7,4)	-	
17				631(30,4)	608(9,16)	-	-	
18				574(12,3)	586(16,5)	675	-	
19				600(22,4)	612(15,11)	623(25,3)	-	
20				642(44,3)	603(18,7)	588	741	
21				586(9,3)	634(18,8)	-	-	
22				597(20,2)	630(13,16)	635	-	
23				-	629(14,14)	-	-	
24				553	616(13,13)	660	-	
25				658	679(17,5)	-	753(18,2)	
26				-	688(54,3)	-	-	
27				-	714(22,3)	-	-	
28				-	719	680(21,2)	-	
29				-	654(60,2)	-	854	
30				-	-	674	-	
31				640	651(32,2)	-	-	
32				590	674(8,2)	-	-	
33					696	714(15,2)	-	
34					-		-	
35					-		-	
36					710		-	
37							811	
38								
39								

Length at age

The length-at-age estimates for all eight zones within Australia are given in Table 20.

Growth

The tag and recapture data for both the Suntag database and the data collected from this project were analysed to determine growth rates (Figure 26). The scatter plot in Figure 26 shows some apparent anomalies with the recreational data where some fish at liberty for more than 500 days have shown negative growth. However, while there may be some problems with measurements, the recreational data does provide valuable growth information for tagged fish at liberty for nearly seven years. However, the research data included only about two years of tagging and therefore recaptures were restricted to fish at liberty for less than 800 days

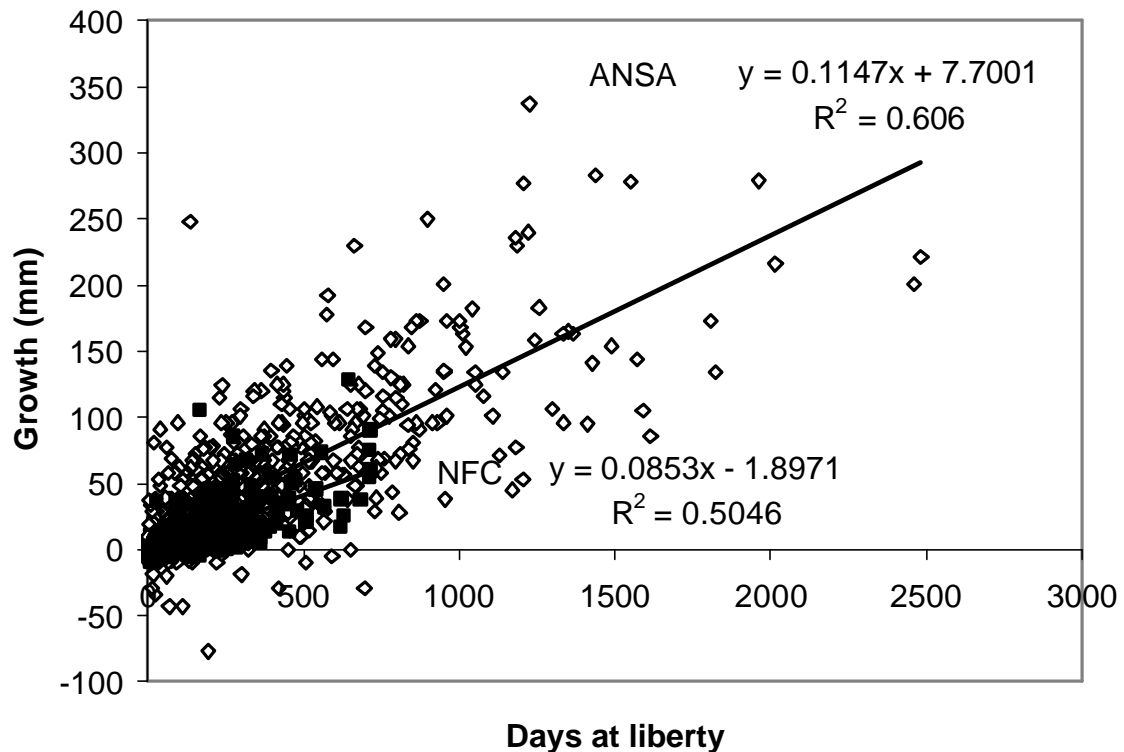


Figure 26. Growth data for tagged mangrove jack.

The square markers are data from the research program and the diamond markers are data extracted from the Suntag recreational fishing database.

The use of the tagging data for estimation of von Bertalanffy parameters was not possible because of the small size of the fish that were tagged and recaptured and because there was little tagging done in offshore areas.

There was a significant difference in growth rates between the Suntag data and NFC research, with the former having a larger estimated growth rate and increased variability (41.8 mm/yr for Suntag vs 31.1 mm/yr for NFC)($p=0.002$, $F=10.07$, 1 d.f., $n=1321$). When the datasets were split between the different Queensland zones (Table 21), there were significant differences in the regressions (NFC: $p<0.001$, $F=25.17$, 2.d.f., $n=462$ and Suntag: $p<0.001$, $F=16.34$, 4 d.f., $n=859$). Using the Suntag data, fish from south-eastern Queensland were found to have the fastest growth rate, whereas the NFC data suggested that central Queensland mangrove jack had faster growth rates. The higher water temperatures of the

northern latitudes may not be optimal for mangrove jack growth. In both data sets the growth rate of fish was faster in the southern and central Queensland than the more northern latitudes.

Table 21. Regression equations for growth of tagged mangrove jack in each zone by tagger type (NFC and Suntag). * $p < 0.05$, ** $p < 0.01$, n.s. (not significant).

Zone	Constant	Slope
NFC - Wet tropics	-8.19 ^{n.s.}	0.0683 ^{**}
Central Qld	-4.50 [*]	0.1507 ^{**}
South-eastern Queensland	-5.29 ^{n.s.}	0.1156 [*]
Suntag – Gulf of Carpentaria	10.24 [*]	0.10078 [*]
North Qld	5.38 ^{n.s.}	0.10718 [*]
Wet tropics	7.16 [*]	0.09106 ^{**}
Central Qld	9.78 ^{**}	0.13248 ^{**}
South-eastern Queensland	5.18 ^{n.s.}	0.16548 [*]

Part of the explanation for the faster growth rate for the central Queensland fish when compared to the south-eastern Queensland fish was due to inclusion of two impounded freshwater sites in the Central Queensland data. Mangrove jack from both these sites (Raglan Creek and in sand dam in the O’Connell River) have displayed particularly rapid growth. An accumulated ANOVA was used to determine if there were differences in the growth rate between fish in the sand dam in the O’Connell River and fish caught at a tidal location approximately two kilometres downstream in the same river. The regressions describing the growth rates at the respective sites were significantly different, with fish from the impounded area having a faster growth ($p < 0.001$, $F = 18.32$, 1 d.f., $n = 32$). The growth rate obtained from the impounded area is approximately 50-100% greater than the rate calculated for other zones (Table 21).

Reader bias and precision of estimates

The most accepted method to determine reader bias is to use an age-bias plot, as it is sensitive to both linear and non-linear biases (Campana, 1995). Of the 813 otolith sections were sent to the Central Ageing Facility (CAF) to receive a second and independent reading. Of these, the CAF selected 769 (94.6%) as suitable for ageing. A plot of the age estimate differences between readers showed that there was a 52.5% agreement between readers and in 86.5% of fish the difference in age estimated was no more than one year (Figure 27). The plot does appear to be slightly skewed, with perhaps some bias towards underestimation by reader 2 (CAF) relative to reader 1 (NFC).

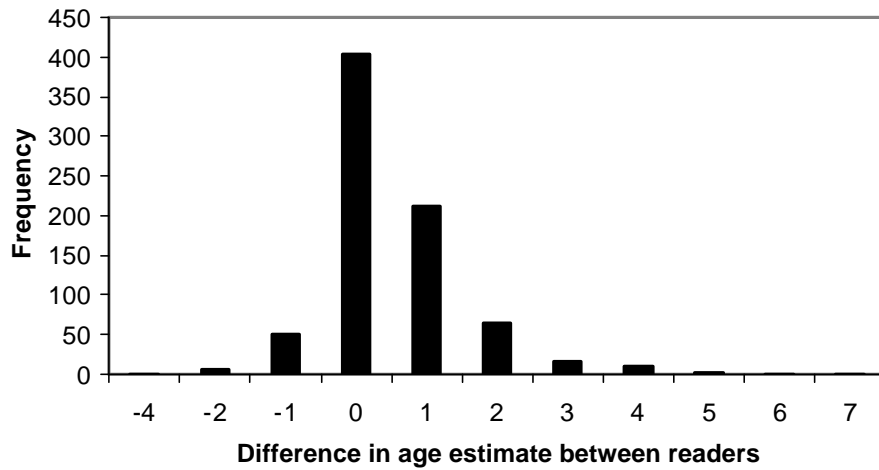


Figure 27. Age estimate differences between readers for sectioned mangrove jack otoliths.

When the estimates are placed on an age-bias plot, differences between the readers do not appear until an age estimate of 12 years after which the ages estimated by reader 2 are generally less than those given by reader 1(NFC)(Figure 28).

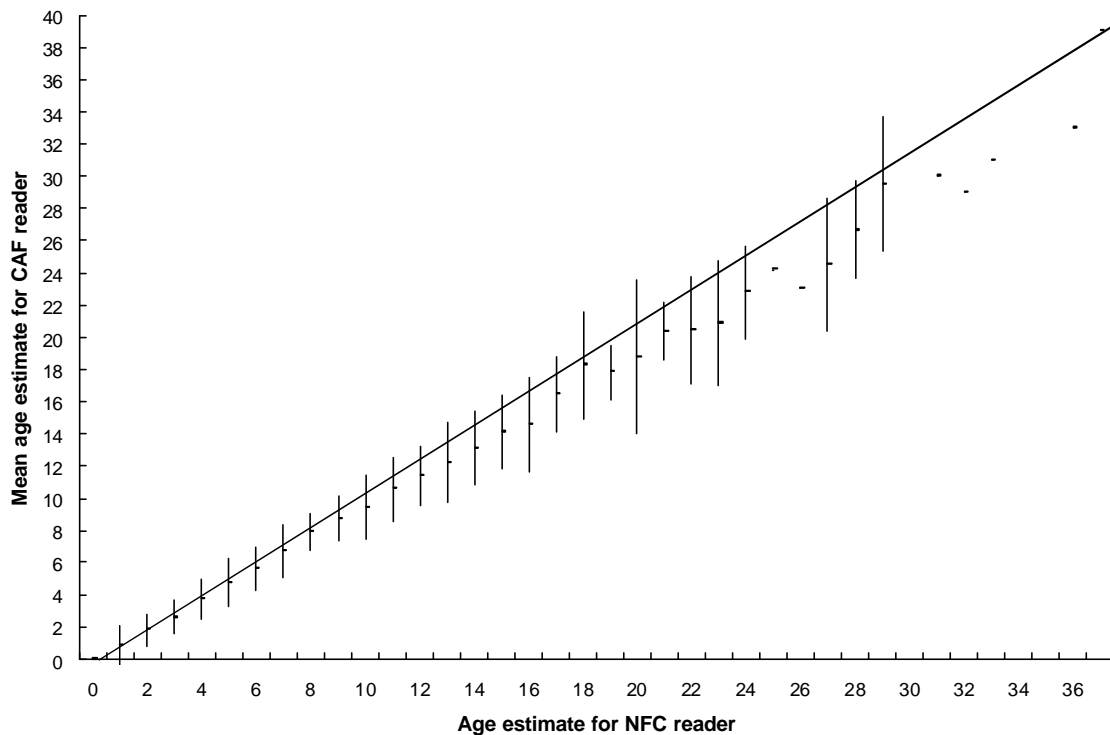


Figure 28. Age bias plot of sectioned mangrove jack otoliths comparing readers.

Error bars are 95% Confidence Intervals for the mean, shown only when $n > 2$. Line through the origin represents $Y=X$.

The age estimates in older fish are not consistently under or over-estimated, with all age classes containing more than 2 fish within the 95% confidence limits.

The average percent error (APE) between readers was calculated to be 4.99% on the 768 fish that were read twice. Twenty-five percent of these fish were re-read by the CAF, with an APE of 1.28%. Because there were only two readings made of the majority of the samples, the Coefficient of Variation (that is also used as an indicator of precision) produced the same value of 4.99%.

Discussion

Sampled fish

Approximately 1300 fish were collected from Australian waters to be used for the ageing of mangrove jack. These fish were mainly collected from the Wet Tropics and North Queensland, with lesser numbers from other areas. Although more than 5 600 fish were collected during the project using electrofishing, most were tagged and released with fewer than 5% kept for ageing and reproductive investigations. Most of the otolith samples used in this study came from material supplied by commercial and recreational fishers.

Back-calculation

Regressions of fish length and otolith radius were used to calculate an estimated length at the time of the formation of the previous increment. This technique has the advantage of avoiding bias due to time of sampling (Campana, 1990a). The plot of the fish length and otolith radius for different sexes and geographic zones (Figure 16) clearly shows that there were differences in regressions between males and females and for South-east Queensland fish compared to fish from the other areas. Attempts were made to fit a linear equation to these data but subsequent examination of the residuals suggested that a curve would give a better fit. It was found that a quadratic relationship best fitted the pooled data for all Australian samples. In other studies, linear regressions were fitted to describe the relationship between fish length and otolith radius, perhaps based upon the assumption of an integral correlation between these two body measurements. For example, Burton (2002) used a simple linear regression to describe a pooled dataset for *L. analis* where there appeared to be outliers to the linear regression at the larger fish sizes. In a separate publication, he used a linear regression to explain differences in otolith radius and fish length of *L. griseus* in two geographic locations (North and South Florida), without investigating if there were significant differences between these regressions (Burton, 2001). After fitting a linear regression, Newman *et al.* (1996a) suggested that otolith breadth was erratic in its use as a predictor of fish length in *L. adetii* ($r^2=0.568$) and *L. quinquelineatus* ($r^2=0.793$). However, this author did not appear to have tried to fit curves to these data. In another case, Escot and Granado-Lorencio (1999) tested for differences between linear relationships established for two geographically different sampling locations, but also did not try and fit standard non-linear regressions even though the plot presented in his publication suggested a non-linear fit to the data.

The relationship between otolith radius and fish length changes because, as the fish grows, the otolith depth increases and the shape tends to become more concave. The ratio of the fish length to otolith length began to decrease at approximately the point where gonad development begins (~260 mm) and then became linear. The growth of the width of the otolith relative to fish length begins to slow slightly after the fish reaches approximately 130 mm LCF after which the relationship tends to become linear. There is a different slope for the south-eastern Queensland fish, with these fish having narrower otoliths at a given LCF when compared to fish from other zones. The relationship between fish length and otolith

radius changes from approximately linear to curvilinear when the fish reach about 130 mm LCF and approaches asymptotic for larger fish. The fish from south-eastern Queensland can be clearly separated in this plot and it appears that the somatic growth of these fish follows a different trend to fish of comparable sizes from other zones. In the south-eastern Queensland fish, the otolith continues to increase in depth even though somatic growth slows; this phenomenon has also been described in the Murray cod (Anderson *et al.*, 1992). The overall effect of these changes is that otolith weight increases more rapidly in larger fish than it does in smaller fish. As a consequence, the fish from south-eastern Queensland generally have lighter otoliths than fish of comparable sizes from other zones but appear to have a higher growth rate.

The implications of these differences in otolith morphometrics were investigated by comparing the relationships between fish length and otolith radius for different sexes and geographic locations (Figure 16 and Table 14). Significant differences were obtained for the intercepts and displacements, however both sexes had the same “shape” term. The regression equations for males and females crossed at a length of approximately 460 mm LCF, which is close to the length at maturity for male mangrove jack (see Reproduction section). Increased energy load on the breeding fish could conceivably be causing the uncoupling in the somatic and otolith growth in larger fish.

Further analysis of differences in regression estimates between zones and sexes on the east coast of Queensland suggested a more complex explanation was required. Firstly, it was found that there were significant differences in parameter estimates for the otolith radius versus length regressions for the various zones. Further, the south-eastern Queensland fish had a significantly different “shape” (squared term) regression when compared to the other three zones, all of which were similar. Significant zonal differences in the relationship between otolith radius and fish length have also been demonstrated for the European barbel (*Barbus sclateri*) (Escot and Granado-Lorencio, 1999). Cameron and Begg (2002) also found significant differences in otolith radius and fish length between sexes and zones in spotted mackerel (*Scomberomorus munroi*) on the east coast of Queensland. Of the three small mackerel species they investigated, it was only the more migratory spotted mackerel that displayed the differences in the otolith radius/fish length relationship. More samples of both large and small fish are required for south-eastern Queensland to unequivocally confirm these differences in the sex and zonal relationships for mangrove jack.

Three techniques, a standard linear regression, a body-proportional regression and the biological intercept method, were compared to determine the best equation to back-calculate fish length at a previous increment. Only samples where the previous increment radius was more than 95% of the otolith radius were used to minimize errors (Escot and Granado-Lorencio, 1999). The standard linear regression (Equation 1) produced the highest average variability from the actual LCF of 7.24%. Only 22.8% of the calculated values fell within the accepted size range. Out of the two other methods tested (Equation 2 and Equation 3), the body-proportional equation gave the best fit because it not only produced very low deviance from expected values (1.65%), but all back-calculated values fell within the expected range of predicted LCF. The biological intercept method produced an even lower average deviance (0.98%), however the constants used in this equation for the radius of the otolith at birth are not known and were only estimated values.

Back-calculation correction is essential to attempt to reduce ageing errors, especially in determining the parameters for the von Bertalanffy equation. An alternative technique is to provide an estimate of age at the time of capture relative to a single uniform birth date. Unfortunately, the breeding season for this species appears to extend from October to March (6 months), with fish spawning a number of times throughout the season (see Reproduction section). Using this technique, a suitable birth date of 1st January would be appropriate, since the peak in the GSI is late December for both years of sampling. Timing of the increment

completion is more variable, occurring between about late winter and summer (mean closing date of 10th August for this report and 30th October by Cappo *et al.* (2000) (see validation section of discussion). Use of the latter method could potentially double the error associated with age estimates as the errors are additive and there are problems assigning an age class after the nominated mean closing date.

Validation

Comparing the back-calculated length-at-increment closest to year of capture to the known length provided a means of indirectly validating of the age of seven mangrove jack broodstock held at the NFC for a range of periods (Table 15). Firstly, it was determined that, in all cases, the age at death (9+ to 16+ age classes) was greater than the time in captivity (2+ to 7+ years). Back-calculated lengths at the closest increment to time of initial capture were relatively similar to actual lengths at capture (13 to 85 mm difference), with all size predictions slightly larger than actual size. Four of the fish were found to have an actual length within the range calculated, while the sizes of the remaining three fish were below the estimated size range. This variation from the actual value could simply be explained by Lee's phenomenon that suggests that there may be differences of the lengths at previous ages due to selective mortality or differences in annual growth (Ricker, 1975). In his study, Escot and Granado-Lorencio (1999) found that the lengths calculated for previous increments were larger than the expected values in all but the penultimate increment. In studies of two other lutjanid species, the use of average back-calculated length-at-age data resulted in the average deviation from penultimate values to vary by up to 100 mm for the first seven increments for fish of 7 to 16 years of age (Burton, 2001, 2002). The other possible explanation for the variation in the calculated length may be due to the fish growing at a greater rate in captivity and thereby having a different otolith radius to fish length regression to that established wild fish. This was demonstrated by (Cappo *et al.*, 2000), who showed that captive *L. sebae* showed significantly different otolith growth and somatic growth when compared to wild fish. However, this current investigation of the mangrove jack with a known history suggests that an accurate back-calculation can provide useful length estimates at times of capture over numerous years.

Examination of the otoliths of the four OTC marked broodstock further reinforced the validation of the ageing technique, with all four fish demonstrating at least one full increment cycle over a year at liberty (Table 16). As shown in this table, the number of increments and the time at liberty are very similar thereby suggesting that the periodicity estimate is very close to one cycle per year. This is in contrast to the results obtained by Cappo *et al.* (2000) that indicate that the number of increment cycles per year for this species varied from approximately 0.4 – 0.85 cycles/yr. This difference in estimated cycle frequency between these data which both used the same "direct" method could explain the difference in the mean calendar closing date (MCCD) of 10th August for this report and 30th October as calculated by Cappo *et al.* (2000).

OTC marking and tagging studies of other wild fish in inshore areas confirmed that the number of observed increments on the otoliths of two recapture fish corresponded closely with the time they were at liberty (Sheaves, 1995). A further combined OTC/tagging study to validate the otolith ageing technique for wild fish was initialised in January 2002 in the Lake Placid on the Barron River near Cairns. To date, only one fish is known to have been recaptured and the angler released the fish rather than keeping it so that its otoliths could be extracted.

The recapture of a single stocked mangrove jack in Tinaroo Dam proved useful in validating the otolith ageing technique for mangrove jack. The growth rate of this fish was faster than wild fish in coastal streams and similar to growth rates observed in mangrove jack that were experimentally stocked into another impoundment, Lake Morris near Cairns (Alf Hogan, Queensland Department of Primary Industries, unpublished data). Mangrove jack appears to

survive readily in tropical impoundments and can grow to the minimum legal size (350 mm TL) within two years (Alf Hogan, Queensland Department of Primary Industries, unpublished data).

A number of tagged mangrove jack were euthanased for ageing after being subsequently recaptured and these fish provided useful information on both when and at what age mangrove jack make offshore movements. For example, one fish was estimated to be six years old when recaptured and its actual length at tagging was within the range of back-calculated lengths for the year it was tagged. Two other tagged fish recaptured 10 months and 15 months after release also had estimated size ranges that included the actual tagging size. Another fish was at liberty for 5 years in a freshwater pool on a stream in Central Queensland. This fish was the largest fish caught by the research team using the electrofishing sampling technique, and was one of two large fish of approximately 600 mm LCF caught in this area. The estimated age of this mangrove jack was 9 years. Even though this fish was at liberty for an extended period, the actual size at tagging was within the range of estimated sizes that were back-calculated for the year that the fish was tagged.

Marginal increment analysis (MIA) was also used to validate the formation of annual growth increments. Beamish and McFarlane (1983) suggest that the percentage monthly increment over a period of a year needs to be plotted for each age class to completely validate the annual formation of increments throughout a species' life. There was a unimodal minimum percent marginal increment in all age classes, which fell between September and November. In the older age classes, the minimum percent increment may occur slightly later. These data confirm that increments are formed annually for fish with less than 10 increments and, despite geographical differences, the increment completion occurs at approximately the same time (Figure 20).

Figure 18 shows the average increment width reduces with increasing age and also suggests that initial growth of this species is relatively rapid but then slows to a near constant rate as the curve becomes near asymptotic. There were some difficulties associated with the MIA including errors associated with measuring the small increment sizes in the larger fish and small sample sizes in the older cohorts. As a result, only fish less with less than 10 increments were selected for MIA. Figure 19 illustrates that the distribution of percent of marginal increment is unimodal for mangrove jack with less than ten increments. This graphic also demonstrates that the increment appears to be completed in fish as early as August with the lowest percentage marginal increment in November. This is consistent with the estimated mean calendar closing date of 10th August (this study) and close to the other estimate of 30th October (Cappo *et al.*, 2000). Overseas studies with other lutjanids also suggest that the increments are completed in late spring or summer. Burton (2001) found that the increment for gray snapper (*Lutjanus griseus*) in Florida formed in the summer months. He also concluded that mutton snapper, (*Lutjanus analis*), from the same general location, completed forming an annulus in late spring. Rocha-Olivares (1997) found that the increment for the Pacific red snapper, (*Lutjanus peru*), on the southeast coast of Mexico, was completed in the summer months.

Because zonal differences were found in the relationships between fish length and otolith radius, MIA was undertaken for two geographically separated zones (Figure 20). Both graphs show a unimodal distribution, with a significant decrease in percent marginal increment in October in both areas. The interesting difference in the graphs is the slope of the increase in percent marginal increment in late summer. In south-eastern Queensland, the slope is greater than in the Wet Tropics zone, and the percentage does not change from April to August. In contrast, the percent marginal increment steadily increases in the Wet Tropics to a maximum in July, a pattern that mirrors the changes in water temperatures.

Age estimates

In the literature there appear to be some considerable differences in the estimates of the values of growth parameters for mangrove jack. Using length-frequency analysis, Ambak *et al.* (1985) suggested that a sample of 80 fish obtained from trawling off the east coast of Malaysia were composed of fish up to 7 years old (734 mm LCF). From this study, the lengths of 2-7 year old fish were 327.9 mm, 444.5 mm, 531.7 mm, 611.3 mm, 688.5 mm and 734.1 mm respectively. The L_8 calculated was 996.9 mm, which is approximately 12 years old (from Figure 5 in Ambak *et al.*, 1985). These estimates are very different to what was obtained in this current study and the other Australian study by Sheaves (see below). There appears to be a number of deficiencies in Ambak's investigation including small sample sizes and a lack of data for fish less than 300 mm long that, in Malaysia, presumably also reside inshore.

Sheaves (1995) completed the only other documented ageing of this species in Australia, with samples collected from both the estuarine and offshore habitats. Out of the 276 specimens analysed from estuarine areas, the age estimates ranged from 0-8 years. He collected 22 larger specimens from offshore areas that had up to 32 growth increments.

In this current study, the ages of the freshwater and estuarine fish ranged from 0+ to 11+ years old and the offshore fish were between 2+ years and 39+ years old. Similar age estimates were also obtained in an independent study that was investigating the microchemistry of mangrove jack otoliths (see Appendix 3). The ages of immature fish ranged from 0+ to 15 years old. Using histological techniques, it was possible to discriminate the sex of most fish that were more than 2 years of age. The smallest mature male fish was 3 years, however most male mangrove jack did not mature before they were about 7+ years old. Female fish matured from 5+ years, with most fish specimens not maturing until they were 8+ years. Similarly, Emata *et al.* (1999) found that mangrove jack captured from the wild at approximately 96 mm long (approximately 1 year old using size estimates generated in our study) and then reared in captivity did not reach maturity as males for another four years (mean length 496 mm) and as females, after five years in captivity (mean length 570 mm).

There appears to be differential growth between the sexes with the data suggesting that female mangrove jack are generally larger than males of the same age (Figure 23). This is the reverse of what occurs in some other lutjanids from the Great Barrier Reef and overseas where the male fish have been shown to grow faster than the females (McPherson and Squire, 1992; Newman *et al.*, 1996a; Rocha-Olivares, 1997; Newman *et al.*, 2000a). However Burton (2001) suggests that *L. griseus* grow at the same rate regardless of sex. He also found no difference in growth parameters of male and female *L. analis*.

There appeared to be significant differences in the growth parameters of mangrove jack from the four east coast zones and also for males and females in north Queensland and the wet tropics. In both these northern zones, the female fish were larger than males but the growth coefficients were similar. The value of the growth coefficient (K) appears to decrease with increasing latitude while L_8 increases with increasing latitude (Figure 22, Figure 24, Table 19). Although the increase in L_8 has not been previously shown for lutjanids in Australia, it has been hypothesized for lutjanid species on the Great Barrier Reef (M. Cappel, Australian Institute of Marine Science, pers. comm.). This effect has been demonstrated for some lutjanids in Florida where it was suggested that the cause was over fishing (Burton, 2001, 2002). This trend has also been shown in spotted mackerel where female fish are larger than males of the same age and values of K decreases as the distance south increased (Cameron and Begg, 2002). There are a number of explanations for the differences in these parameters between zones including unequal sample sizes and variations in modal, minimum and maximum sizes of fish in the samples. However, the mean length-at-age data for each zone,

along with the differences in growth rates from tagging and the maximum size of fish sampled in each zone support the differential growth hypothesis.

Examination of the length-at-age data from the other areas around Australia (Figure 24, Table 20) suggests that not all areas are adequately described by the general growth equation that was calculated from the pooled data. The data points from New South Wales fish are all above the von Bertalanffy regression for the pooled data and, perhaps not surprisingly, they appear to be similar to data obtained from south-eastern Queensland fish. Coincidentally, the two largest fish caught during this project (estimated to be approximately 1 030 mm long and 14 kg weight) were caught in the south-eastern Queensland and New South Wales zones. The south-eastern Queensland fish was estimated to be approximately 39 years old although caution needs to be exercised because vertebrae, rather than otoliths, were used to determine its age. Vertebrae may be a less reliable index of age than otoliths (Marriott and Cappo, 2000).

Growth

In his study Sheaves (1995) found that, from 10 recaptures of 120 mangrove jack tagged (7.5%) in tropical estuaries in North Queensland, there was a maximum growth of 66 mm from a fish at liberty for 395 days. The average growth rate in his study was 0.1780 ± 0.007 mm/day ($r^2=0.9896$, 5df) but, probably because of the low sample size, he found that the correlation between growth rate and mean length for time at liberty was poor ($r^2=0.1944$). This translates to a growth rate of approximately 65 mm/yr for the fish between the 150 mm and 541 mm size range. Slightly lower estimates of growth rates of between 0.0683-0.1507 mm/day (24.9 – 55.0 mm/year) were observed in this current study.

Local habitat also appears to have a considerable influence on growth rates. There was a significant difference in the growth rates of fish trapped in a sand dam on the O'Connell River when compared the fish down stream in the tidal reaches of the same river. Similarly, fish in impoundments in other areas also appeared to grow faster than fish resident in nearby tidal streams.

Bias and precision

The level of precision in determining of the age of fish from sectioned otoliths appears dependant on a number of factors including the relative visibility of the increments on different types of equipment and recognition of what is the first increment. Some sections were clear and very easy to read while others were less distinct. Capture location appeared, to some extent, to influence how easy a section was to interpret. Generally, the otoliths of fish from more southern latitudes and those from inshore locations were easier to interpret. This may be because cooler temperatures in the more southern latitudes during winter are likely to affect otolith growth more than in the northern parts of Queensland. This is despite evidence presented earlier that overall average growth rate in southern latitudes is higher than in northern latitudes. Experience and skill of the reader is important in reducing error when using otoliths in ageing studies. Age estimates by two independent readers produced an Average Percentage Error (APE) of 4.99% (n=768), compared to an APE of 1.28 % by the Central Ageing Facility (CAF) from a sub-set (n=208) of the same otoliths (Green and Talman, 2002). These values are comparable to results obtained from other lutjanid species such as *L.johnii* (APE=9.5%) (Marriott and Cappo, 2000); *L. erythropterus* (APE=2.85%), *L. malabaricus* (APE=4.47%), and *L. sebae* (APE=6.73%)(Newman *et al.*, 2000c); *L. carponatatus* (APE=2.33%), and *L. vitta* (APE=13.05%)(Newman *et al.*, 2000b); *L. adetti* (APE=0.72%), and *L. quinquelineatus* (APE=1.73%)(Newman *et al.*, 1996a); and *L. peru* (APE=2.13%) (Rocha-Olivares, 1997). The reduced APE obtained by CAF for re-reading indicates another source of error, bias measurements.

Initially, the first increment was not counted in age estimates as this increment was thought to occur before the fish were actually 365 days old. However, the use of daily growth estimates

suggested that the estimated hatch date for some fish may be as early as October/November so that these fish would be a year old at the first increment. Once this difference in interpretation was corrected, there still appeared to be a minor difference in estimation of ages between readers, with a slightly skewed age distribution difference graph (Figure 27). There was 52.5% agreement in age estimation between readers, with 86.5% of samples within a year of estimation. When the samples were re-read by the same reader, 78% of samples were in agreement with 99% and 100% of samples within one and two years respectively (Green and Talman, 2002). The age distribution difference graph produced for the re-read data appeared to be normal in distribution.

The age bias plot (Figure 28) confirmed that, where more than two samples were available for each increment, all increments were within the 95% confidence intervals (Campana, 1995). In addition, this plot demonstrates that there does not appear to be any linear bias (consistent differential readings were not parallel to 1:1 line) or non-linear bias (means and 95% confidence intervals outside 1:1 line randomly). There were fewer samples of older fish resulting in larger 95% confidence intervals for these older fish. Also some age cohorts only had a small number of samples, so confidence intervals could not be calculated. When the readings obtained from NFC and the CAF were plotted against each other the mean values deviated significantly from the 1:1 line for fish with 12 or more increments suggesting that the older fish are more difficult to age.

Conclusions

The body-proportional equation was found to give the best relationship of length at previous increments to the length at death. Quadratic equations, rather than linear equations, provided the best fit when describing the relationship between fish length and otolith radius. There were significant differences in the regressions for otolith radius and fish length both between zones and between sexes.

Validation that the increments on mangrove jack otoliths were annual was established using both marginal increment analysis and OTC marking. The mean calendar closing date was calculated to be the 10th August. The back-calculated lengths for fish that had been at liberty for known time intervals were within acceptable limits.

The von Bertalanffy growth equations that were calculated for each of the zones on the east coast of Queensland appeared to be significantly different. Fish from southern Queensland gave a higher estimate for L_{∞} and a lower estimate for K . There was a significant difference in the parameter estimates between the sexes for north Queensland and the wet tropics areas. The von Bertalanffy parameters for the data pooled for both areas and sexes did not appear to suitably describe the growth of fish from the Gulf of Carpentaria and New South Wales. More samples are required for the other states and from south-east Queensland to confirm these trends.

The length-at-age data suggests that the southern fish reach the current legal size at a faster rate than fish from the northern zones. Fish begin to move offshore at an age of 2 years, with fish fully recruited to offshore habitats by age 12. This is a long-lived species, with the maximum age within the population possibly in excess of 40 years. Growth rates slow substantially after the fish mature.

Growth rates obtained by tagging suggest that fish older than 2 years found in inshore areas grow at a rate of between 25 – 50 mm/year. There appears to be differences in growth rates between zones in Queensland, with fish from southern zones tending to grow at a faster rate, which reinforces the conclusions from the ageing data.

The precision of the age estimates was 4.99%, which compares favourably with other studies. There does not appear to be any substantial bias in age estimates between readers, however the age estimates made by the CAF were slightly lower than those made at the NFC.

Habitat Preferences

Introduction

In estuarine and riverine environments, physical structures often provide habitat for a wide variety of fish and other aquatic biota. These habitats include large woody debris (snags), rocks, overhanging vegetation, roots and pneumatophores, undercut banks and grasses and aquatic plants. Association with these types of habitat has a number of advantages, primarily protection from predation (Primavera, 1997). The adaptive significance of fish associations with snags may be related to protection from current, food availability, or camouflage from predators or prey (Angermeier and Karr, 1984). Snags have habitat values similar to highly heterogeneous reef habitats providing refuges from predators and sites from which to ambush prey (Bell *et al.*, 1984; Sheaves, 1996). Robertson and Duke (1987) also suggested that the structural complexity provided by prop roots, pneumatophores and fallen snags in mangrove forests was likely to play an important role in determining the dependency of some species of juvenile fish on this habitat type.

The relative importance of snags as fish habitat probably varies with environmental conditions; for example the role of providing fish with cover from predators is probably greatest during low flow periods when habitat availability and diversity are reduced and the streams become relatively more assessable to birds and other predators (Angermeier and Karr, 1984). As well as the benefits described above, other types of habitat provide additional advantages to aquatic biota. For example, overhanging vegetation, as well as providing cover also assists in a number of other functions including providing food (eg. leaf, insects and fruit) for aquatic biota, reducing water temperature and raising oxygen saturation through shading and increasing bank stability (Mahoney and Erman, 1984).

A number of studies have included habitat preferences of mangrove jack in the wild. In the Emberly river estuary in northern Australia, Blaber *et al.* (1989) noted that catch rates of most fish species declined from the lower to the upper reaches of the estuary. One of the exceptions to this pattern was for mangrove jack which were most abundant in the middle reaches of the estuary. In a study of habitat-specific distributions of some fishes in a tropical estuary in northern Australia, Sheaves (1996) found the highest number of species were caught in snaggy habitats. He found that the probability of occurrence of *Lutjanus russelli* in structurally complex snag habitat to be more than three times that for any of the other habitats that he sampled. In an experimental study of the effects of habitat size on habitat association, young of the year red snapper (*L. campechanus*) were found to have a strong affinity for habitat with greater structural complexity (Bailey *et al.*, 2001). As the size of artificial reef habitat increased, then the mean distance of juvenile red snapper from the blocks decreased significantly and the time spent near the structures increased significantly. After the introduction of larger conspecifics, the time spent near the structures by the young-of-the-year fish decreased significantly as the larger fish actively defended the structure from occupation by the juveniles.

In a laboratory experiment assessing the role of structures and substrate on fish predation of mangrove-associated penaeids, Primavera (1997) found that habitat complexity regulates predation on some invertebrate prey species. Predation of shrimp by *L. argentimaculatus* was significantly lower amongst medium density pneumatophores than that for *Lates calcarifer* (Primavera, 1997). In estuaries, freshwater and inshore areas mangrove jack are often associated with snags (Grant, 1997), pneumatophores and prop roots (Primavera, 1997) and rocky areas (Primavera, 1997) and coastal reefs (Allen, 1991, 1997). In coastal areas of southern Africa, juvenile mangrove jack from Morumbene estuary to the Mngazana, are mainly found in rocky areas (Day *et al.*, 1981). Small numbers of mangrove jack were among juveniles of 38 species that used a tidal swamp in northern Australia as a nursery area (Davis,

1988). Once offshore, older fish are known to be associated with reef habitats and are also found in deeper offshore waters (Allen, 1991, 1997).

In this present study we investigate the habitat preferences of juvenile and sub-adult mangrove jack in estuaries and upper tidal and lower freshwater reaches of rivers in north eastern Australia.

Methods

The Genstat[®] statistical package was used to develop a multiple forward stepwise regression model to relate geographical and habitat parameters to relative abundance (CPUE). Only sites where the total electrofishing time was in excess of 10,000 seconds were included in the analyses. These sites, the number of replicates and their length and distance upstream from the river mouth are given in Table 22. The environmental and geographical parameters were: distance from river mouth, latitude, average depth, percentage of stream with undercut banks, percentage of stream banks with snags, percentage of stream banks with associated aquatic macrophytes, percentage of stream banks covered with grasses, percentage of stream banks with rocks, percentage of stream banks with overhangs. The percentage values of each habitat type at each site were estimated using the methodology for habitat assessment given in (Russell *et al.*, 2000).

CPUE was calculated for each replicate at each site by dividing the total number of mangrove jack caught by the total electrofishing time and multiplying the result by 1000. This gave number of fish caught per 1000 seconds of electrofishing as the measure of relative abundance. The sites were all sampled with a generator powered Smith-Root[®] Model 7.5 GPP electrofisher fitted to a 4.3 m vessel. A pulsed direct current was applied to the areas of likely fish habitat, usually along the banks but also including midstream structures such as snags or rock bars. Mangrove jack were captured using pole nets and the habitat type from which they were taken, their length and weight were recorded. Most fish were tagged before being returned to the water in the general vicinity of capture. A small number of mangrove jack were kept for related studies. The wet tropics sites (Daintree, Mulgrave, Russell and North and South Johnstone Rivers) were generally sampled monthly from late 1999 to February 2002. Sampling at the Crystal Creek and Herbert River sites commenced in December 2000 and December 1999 respectively and these were sampled quarterly until March 2002. Periodic local conditions at these sites including high salinities and high river flows precluded sampling on some occasions.

Table 22. Details of sites used in habitat analyses

Site	Code	Replicate	Distance upstream(km)	Latitude (°S)	Site Length (m)
Crystal Creek	CR1	1	2	-18.93	250
Crystal Creek	CR1	2	2.5	-18.93	300
Crystal Creek	CR1	3	3	-18.93	300
Herbert River	HE2	4	7.2	-18.58	300
Herbert River	HE1	1	19	-18.53	350
Herbert River	HE1	2	19.8	-18.53	500
Herbert River	HE1	3	20.7	-18.53	500
South Johnstone River	SJ1	4	8.2	-17.56	400
South Johnstone River	SJ1	2	10.2	-17.56	300
South Johnstone River	SJ1	3	9	-17.56	400
North Johnstone River	NJ1	1	7	-17.5	400
North Johnstone River	NJ1	2	8.5	-17.5	400
North Johnstone River	NJ1	3	10	-17.5	500
Russell River	RU1	1	8	-17.26	400
Russell River	RU1	2	7.5	-17.26	550
Russell River	RU1	3	7	-17.26	200
Mulgrave River	MU4	1	15	-17.24	200
Mulgrave River	MU4	2	16	-17.24	200
Mulgrave River	MU4	3	16.5	-17.24	300
Mulgrave River	MU6	1	8.5	-17.18	300
Mulgrave River	MU6	2	7	-17.18	400
Mulgrave River	MU6	3	7.5	-17.18	400
Daintree River	DA1	1	21	-16.3	300
Daintree River	DA1	2	22	-16.3	500
Daintree River	DA1	3	23	-16.3	400
Daintree River	DA2	1	16	-16.3	300
Daintree River	DA2	2	16.7	-16.3	300
Daintree River	DA2	3	17.3	-16.3	400

Results

Instream habitat preferences

The habitat preferences of mangrove jack in the upper tidal and lower freshwater sections of rivers and creeks in north east Queensland are shown in Figure 29. The majority of mangrove jack were sampled in either rocks or in snags with only small percentages of fish caught in other habitat types including grass, aquatic macrophytes, tree roots, under undercut banks or overhanging vegetation or in open water. More than 50% of the fish in size classes larger than 150 mm, with the exception of the 550-600 mm cohort, were caught in amongst snags. Most of the fish sampled in the 0-50 and 50-100 mm size classes were caught in amongst rocks. As well as naturally occurring rock bars, boulders, coffee rock and other naturally occurring rock features, artificial rock structures also appear to be suitable as mangrove jack habitat. In many of the wet tropics streams, the banks have been armoured with rocks to minimize erosion during periods of high flow (Plate 3). Juvenile mangrove jack make use of many of these structures which have been placed in estuarine or lower freshwater reaches of rivers by utilizing the crevices between the rocks as temporary refuges. Other habitat types such as undercut banks, overhanging vegetation, roots and grasses are utilized to a lesser degree and predominantly by the smaller sized fish. Very few fish (<0.001% of the total number sampled) were caught in open water where there was no cover.



Plate 3. Rock wall constructed on the Russell River to minimize erosion.

Juvenile mangrove jack use spaces between the rocks as refuges.

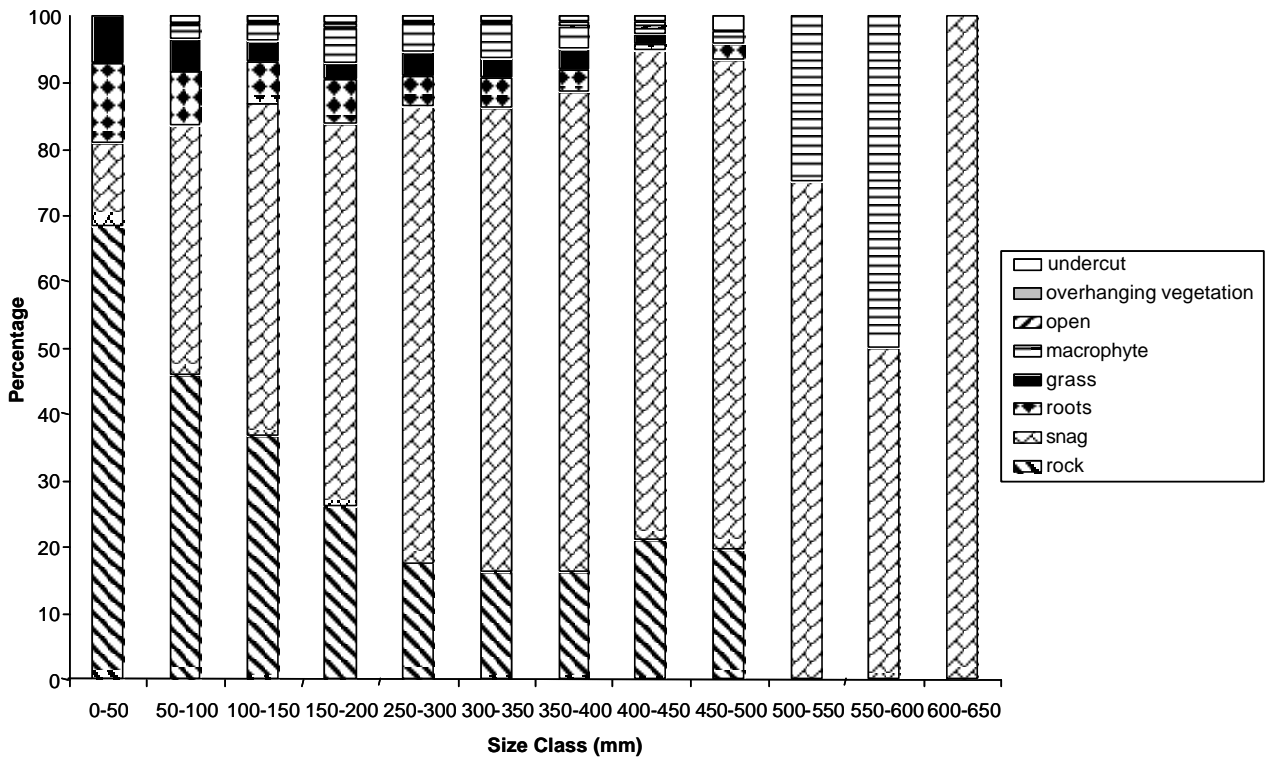


Figure 29. Habitat preferences of different 50 mm size classes (LCF) of mangrove jack.

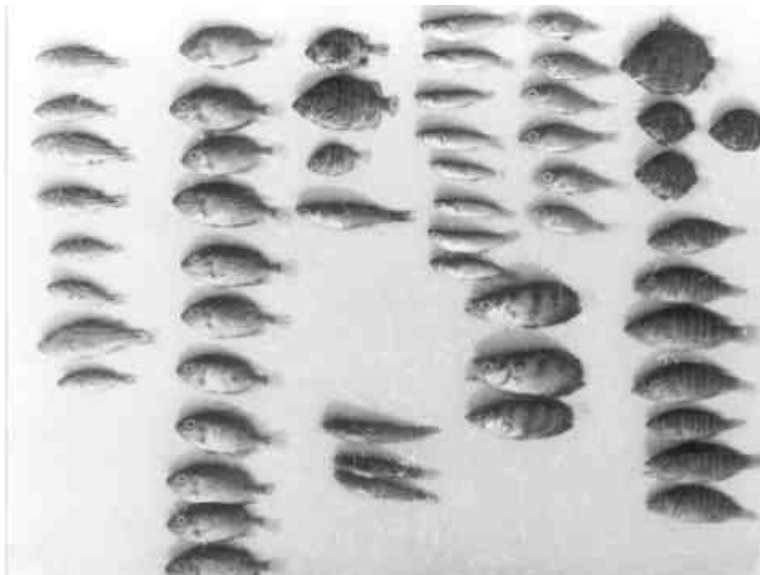


Plate 4. Juvenile fish taken from a supra-littoral nursery swamp in north Queensland.

Tidal Nursery swamps

As well as in riverine situations, juvenile mangrove jack, like barramundi, utilize tidal swamps as nursery areas. These nursery areas may be brackish or freshwater and form a mosaic of different ecotypes from saltpan pools to mangrove swamps and brackish lagoons. Plate 5 shows a typical fish nursery swamp adjacent to a saltpan near Princess Charlotte Bay in north Queensland. Apart from mangrove jack, these swamps can act as nurseries for a

range of different fish species including barramundi (*Lates calcarifer*), archer fish (*Toxotes sp.*), scats (*Selenotoca multifaciata* and *Scatophagus argus*), rabbit fish (*Siganus sp.*), blue eyes (*Pseudomugil sp.*), Oxeye herring (*Megalops cyprinoids*), mullet (Mugilidae) and Teraponids. Plate 4 shows a range of juvenile fish sampled from a mangrove nursery swamp in north eastern Queensland. The juvenile mangrove jack shown on the right of the photo were between about 15mm and 25 mm long. Many of these swamps are ephemeral and the residence time by juvenile fish can be comparatively short, often less than three months.

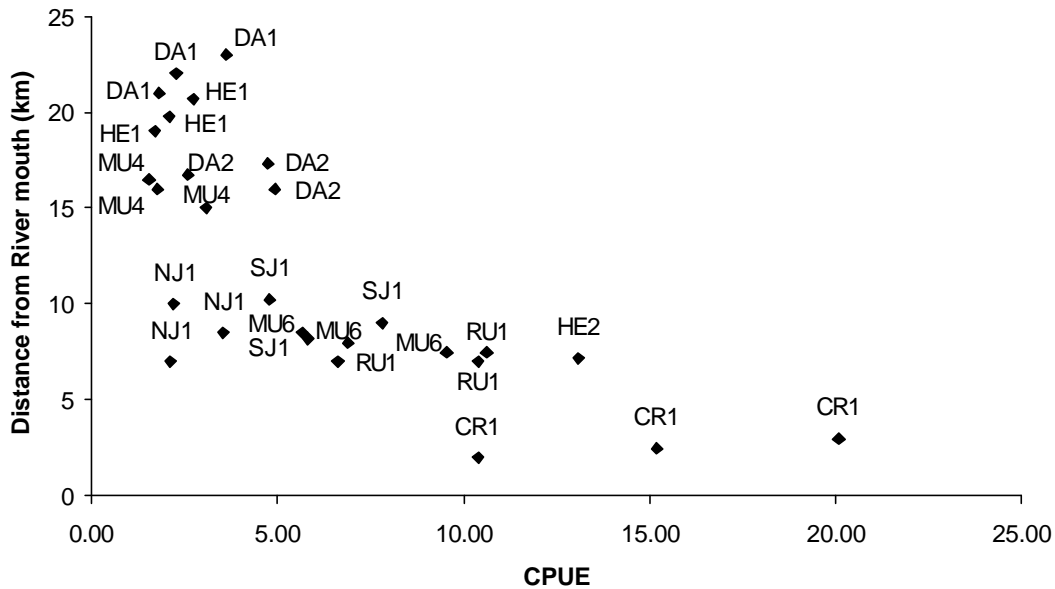


Figure 30. Relationship between CPUE and distance from the sea.

The codes for the sites are: CR Crystal Creek, DA Daintree River, HE Herbert River, MU Mulgrave River, NJ North Johnstone River, RU Russell River, SJ South Johnstone River.

Intra-riverine distribution

Figure 30 shows the relative abundance (CPUE) of mangrove jack as a function of distance from the river mouth. As the distance from the river mouth increases the CPUE declines with relatively low CPUEs (< 5 fish per 1000 seconds of electrofishing time) at distances between about 15 and 23 km upstream from the mouth. However, at a distance of less than 5 km from the mouth the CPUE had increased to more than 10 fish per 1000 seconds fished. While mangrove jack are capable of moving large distances upstream into freshwater (Merrick and Schmida, 1984), most juveniles and sub adults are confined to the estuaries and lower freshwater reaches of coastal rivers (See Recruitment section).

Table 23. Summary of the effects of the main parameters in the stepwise multiple regression analyses of mangrove jack CPUE.

None of the other parameters improved the result.

Parameter	% of variation explained	P
% of rocks	55.2	<0.001
Distance from mouth	68.9	0.002
Average depth	70.7	0.123
Latitude	70.8	0.308

Environmental factors affecting abundance

Even when the location of the sampling site within the river is taken into account, the relative abundance of mangrove jack varies from river to river (Figure 30). The effect on relative abundance of mangrove jack in coastal river systems of a range of habitat and geographical variables measured at each site was tested by developing a multiple, step-wise regression model (Table 23). In the model there was a significant correlation ($P < 0.001$) between CPUE and the percentage of the stream banks with a rocky cover. This model explained 55.2% of the variation but was significantly improved ($P = 0.002$) by including the distance from the sea parameter. With the inclusion of these two factors, the model explained 68.9% of the variation but was not significantly improved by the inclusion of any of the other parameters (Table 23).



Plate 5. Juvenile fish nursery area in northern Australia.

While barramundi were most commonly found in this type of habitat some juvenile mangrove jack were also sampled

Discussion

Mangrove jack that are resident in rivers and estuaries are mostly associated with structural habitat including rocks, snags, undercut banks, aquatic macrophyte beds, grass growing on the stream verges and tree roots including mangrove prop roots. Very few fish were caught in open water where there was no cover available. In the upper tidal, freshwater sections of the estuaries, where most sampling was undertaken, snags and rocks were consistently the preferred habitat type of mangrove jack. There would appear to be habitat segregation of fish resident in the rivers and between fish resident in the rivers and those resident offshore. Within the rivers, most fish less than 100mm long were sampled primarily from rocky substrates, mostly naturally occurring habitat but also on rock walls with an abundance of interstitial spaces. As the fish increased in size, snags became the dominant habitat type with fewer being caught in rocks and in the other habitat types. Similarly, in their study of habitat preferences of red snapper in the Gulf of Mexico, Bailey *et al.* (2001) found that young snapper and adults or bigger snapper were rarely found on the same structures. They also concluded that the young-of-the-year snapper had a strong affinity for habitat with some vertical relief and /or refuge space. Habitat types which are characteristically found in

shallow water along stream banks such as grass, roots and undercut banks appear to be less suited to larger fish which prefer cover in deeper water such as snags (see Plate 6) or dense beds of aquatic plants.

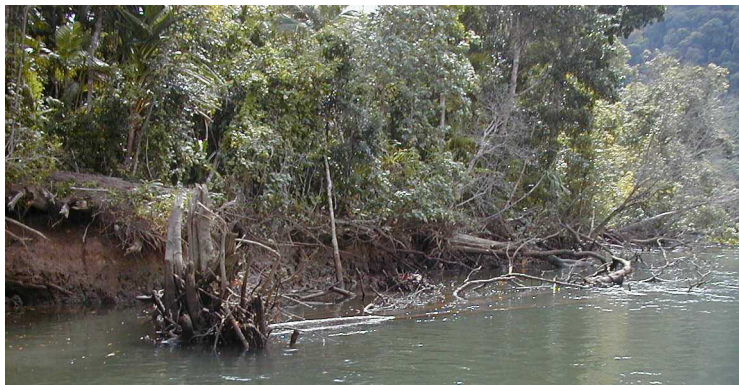


Plate 6. Snag habitat in the Russell River.

This is typical of the type of snag that makes ideal habitat for mangrove jack. Note the structural complexity of this habitat particularly around the roots at the base of the tree.

While the diet of mangrove jack in estuaries consists largely of sesarmid crabs (Sheaves and Molony, 2000), this species is also an ambush predator of mangrove-associated fish (Robertson and Duke, 1990b). The cover provided by the snags, rocks, aquatic plants and the other habitat types allows protection from the current, enhances food availability and provides camouflage from predators or prey. While roots were not a significant habitat type for mangrove jack in the sites sampled in this study, these may assume a greater importance in the lower sections of the estuary. In tropical estuaries much of the structure in snaggy areas is derived from mangrove prop roots or fallen timber (Marshall, 1994; Sheaves, 1996).

As well as being found in riverine habitats, juvenile mangrove jack are also found in coastal nursery swamps like the areas that support juvenile barramundi (Moore, 1982; Russell and Garrett, 1985; Russell, 1987; Davis, 1988). While this species may coexist with barramundi in these coastal nursery swamps (Davis, 1988), in some areas the two species are not found together. For example, in the south eastern Gulf of Carpentaria, supralittoral nursery swamps adjacent to the Norman River contain barramundi and other fishes but not mangrove jack. Similarly in Papua New Guinea Moore (1982) found juvenile barramundi and a number of other species in coastal nursery swamps but did not record the presence of mangrove jack. These nursery swamps may be permanent but are generally ephemeral and are filled each year by seasonal rains and high tides that occur in late spring and summer. The environments in these nursery swamps are conducive to rapid growth and increased survival of these juvenile fish because of an abundance of food in the form of prawns, insects and other fish, minimal predation and ample cover (Moore, 1982; Russell and Garrett, 1985).

Within individual river systems, the relative abundance of mangrove jack is likely to be higher closer to the river mouth than in the lower freshwater reaches of the rivers. However, in the Emberly River system in northern Australia the abundance of mangrove jack was found to be greatest in the middle reaches of the estuary (Blaber *et al.*, 1989). Only small numbers of mangrove jack were sampled during this study and some of the sampling techniques used, for example, because of their association with habitat structures like snags, traditional fishing techniques like gill nets and seine nets are inefficient in catching mangrove jack. In the present study, because of the limitations of electrofishing in brackish and saline waters, no sampling was undertaken in lower estuaries except on a small number of occasions when high

flow events reduced the estuarine salinity. On those occasions, mangrove jack in the lower estuaries were sampled in similar numbers to those from upstream sites. However there is no doubt that considerable numbers of *L. argentimaculatus* do inhabit the lower estuaries of coastal rivers and tidal creeks (Lake, 1971). During the present study, large numbers of mangrove jacks samples were obtained from recreational fishers from lower estuarine areas. While individual mangrove jack have been caught considerable distances upstream, e.g. up to 130 km (Lake, 1971; Merrick and Schmida, 1984; Herbert and Peeters, 1995), most juvenile and sub-adult mangrove jack remain resident in the tidal reaches of the river.

Analyses of the relationships between mangrove jack abundance and the various habitat and geographical parameters suggests that the abundance is higher at rocky sites and also that abundance increases at sites closer to the river mouth. It may be hypothesised, that the apparent preference of mangrove jack for rock structures may explain why numbers of this species are fewer in some areas than others. For example, numbers of mangrove jack in the large, muddy rivers of the southeast Gulf of Carpentaria, and the Fitzroy and Burdekin Rivers on the east coast are apparently lower. While this may be because of the lack of rock structures suitable as juvenile habitat the numbers may also reflect other factors or combinations of factors.

Sediment depositions in these rivers may have covered much of the suitable juvenile habitat thus effectively creating habitat-dependent limitations on the riverine stocks. This proposition is supported in an experiment with habitat preferences of young-of-the-year red snapper where Bailey *et al.* (2001) found that by removing small shelters from artificial reefs the number of juvenile fish associated with those reefs was significantly reduced. However, this hypothesis does not explain why, if lack of rocky substratum for use as juvenile habitat is depressing the numbers of juveniles, the smaller fish at least, do not use the numerous available supralittoral nursery areas that are utilised by barramundi particularly in the south-east Gulf of Carpentaria (Russell and Garrett, 1983).

Although a large number of fish appeared to be associated with snags (Figure 29) the number of snags present at sites in this study did not appear to influence the abundance of mangrove jack. Indeed observations during the study suggest that mangrove jack may preferentially choose snags that they use for habitat. Many snags were observed to have no fish associated with them while others had considerable numbers. For example, in May 2000, 30 fish were caught in one snag in Baffle Creek, which was about 79% of all fish caught in that replicate during that particular sampling session. Similarly, in the Russell River in May 2000, a single snag (see Plate 6) yielded about 80 of the 99 fish caught in the 550 m of riverbank sampled. The average sizes of mangrove jack caught in the Russell River (316.6 mm FL) and Baffle Creek (301.4 mm FL) sites on those occasions were relatively large. The analyses done during this study did not test what effects that the absence of snags would have on mangrove jack abundance. All of the sites had at least 5% snag cover and about 72% of sites had more than 20% cover. Factors that may be important in determining if a snag is suitable for residence by fish is its location with respect to strong tidal or river currents and whether it is located in deeper water rather than in shallow water in the intertidal zone where fish may need to move regularly and the complexity of the structure.

As most mangrove jack were caught in amongst snags, the removal of this type of habitat through river desnagging programs to facilitate or redirect stream flow is likely to effect populations of mangrove jack in the river. Inappropriate development in estuaries is also likely to impact on mangrove jack and other fish stocks (Robertson and Duke, 1990a). In estuarine areas much of the structure in snaggy areas is derived from mangrove prop roots or fallen timber and it is these habitats that will be most directly affected by any loss of mangrove wetlands due to coastal development (Marshall, 1994; Sheaves, 1996).

In the Queensland wet tropics, Pusey and Kennard (1996) described a strong latitudinal gradient in freshwater fish assemblage structure. This latitudinal gradient reflected a greater representation by eleotrid and oxyeleotrid gudgeons in the north and a greater abundance of *Hephaestus fuliginosus* and *Melanotaenia splendida* in the south. In this present study no evidence was found of any latitudinal gradient in abundance of mangrove jack in seven streams over more than 320 km of coast from the Daintree River in the north to Crystal Creek in the south. However, on a larger scale, there is a latitudinal gradient with northern New South Wales being the effective southern limit of the distribution of mangrove jack on the east Australian coast (Merrick and Schmida, 1984; Grant, 1997)

There are parallels between the life history of red snapper in the Gulf of Mexico and mangrove jack. Adult and juvenile red snapper in natural populations are generally segregated with young fish mostly found in shallow refuges in inshore waters whereas larger juveniles begin to recruit to offshore reefs once they have obtained a certain size (Bailey *et al.*, 2001). Similarly the adults and juveniles and sub-adults of mangrove jack populations are also segregated with the younger fish found in inshore and riverine habitats before moving offshore. There may be many reasons for this offshore movement, but like for red snapper it may be a mechanism to reduce competition for the limited riverine habitat and thereby reducing the probability of predation and cannibalism.

Anthropogenic activities have the potential to impact on the habitats of riverine and coastal stocks of mangrove jack. These activities include snag removal, loss of nursery swamps and refuge areas through coastal development and increased sedimentation and loss of riparian forest as a result of inappropriate land use practices. The upper tidal and lower freshwater reaches of many rivers are important habitat for mangrove jack and the construction of tidal weirs without provision for fish passage can effectively diminish the area of available riverine habitat

Recruitment

Introduction

In tropical Australia the most common life-history pattern amongst estuarine nekton is saltwater spawning followed by recruitment of larvae, post-larvae or small juveniles into estuaries where they remain for some time before emigrating to join adult stocks (Robertson and Duke, 1990b). One well documented example of this pattern is the life history of barramundi, *Lates calcarifer* in Australian tropical waters (eg. (Davis, 1982; Russell and Garrett, 1983, 1985; Davis, 1987; Griffin, 1987; Russell, 1987; Davis, 1988; Russell and Garrett, 1988; Griffin, 1995; Pender and Griffin, 1996). Adult barramundi migrate to high saline, coastal areas to spawn and the post-larvae and juveniles then recruit into coastal nursery swamps and later upstream into the freshwater reaches of rivers and lagoons (Russell and Garrett, 1983; Davis, 1985; Russell and Garrett, 1985; Griffin, 1987; Davis, 1988; Griffin, 1995).

This pattern is reflected in the life history of some of the tropical snappers. For example, in southern Africa, *L. fulviflamma* juveniles between 50 and 160 cm are commonly found in estuaries but adults breed at sea probably among inshore reefs (Day *et al.*, 1981). Also recent studies have reported that *L. argentimaculatus* juveniles are commonly found inshore but adults are mostly caught offshore. In a survey of the Emberley River in the north-eastern Gulf of Carpentaria, Blaber *et al.* (1989) found that mangrove jack was one of 14 species whose juveniles were found only in the estuary and not offshore but they recorded adults both inshore and offshore. Similarly, in north-eastern Australia Sheaves (1995) suggests that estuaries are important development grounds for *L. argentimaculatus* and that estuarine populations appear to consist entirely of immature fish. In Thailand, *L. argentimaculatus* juveniles between 16.2 and 31.2 mm TL were sampled in set nets in the Prasae River estuary between November and January inclusive (Doi and Singhgraiwan, 1993). Tagging studies in Thailand have also shown that juvenile fish moved inshore towards the coast and into estuaries from March to August and offshore from September to February (Doi and Singhgraiwan, 1993). As well as being recruited into rivers, there is evidence that juveniles recruit into tidal swamps and lagoons. In northern Australia Davis (1988) suggested that while *L. argentimaculatus* is a dominant species in the tidal Leanyer Swamp, they are transient with only juveniles remaining in the upper estuary and swamp using it as a nursery. He found that the numbers of juvenile *L. argentimaculatus* entering the swamp were correlated with the environmental parameters month and tidal height. Higher tides provided greater assistance for the upstream movement of juvenile fish and also enable them to penetrate further into upstream areas (Davis, 1988). The contention that only juvenile fish are found in inshore areas is supported by Day *et al.* (1981) who noted that *L. argentimaculatus* seldom attained lengths of more than 400 mm in estuaries. The timing and size at which juvenile mangrove jack begin to be recruited into estuaries is not well documented but in a study of the development of swimming and feeding functions of *L. argentimaculatus*, Doi *et al.* (1998) suggest that juveniles larger than 16 mm TL would have acquired a swimming or cruising ability strong enough to migrate to coastal waters and river estuaries. They also note that by this size, the enlargement of the stomach would be sufficient to allow the increased food storage capacity necessary to facilitate the shift between habitats. They also observed that juvenile fish appeared to recruit into the estuary after the wet season and they conjecture that the upstream movement of *L. argentimaculatus* is governed by freshwater runoff resulting from high seasonal rainfall.

Movement of juvenile fish that are initially recruited to estuaries and later migrate offshore to adult habitats may also assist in exporting mangrove primary productivity from estuarine systems offshore (Sheaves and Molony, 2000). In northern Australia, estuarine populations of the groupers *Epinephelus coioides* and *E. malabaricus* and *L. argentimaculatus* consist of

juveniles that eventually migrate to adult habitats offshore (Sheaves, 1995). The dominant prey of these fish are sesarmid crabs, and the primary mangrove productivity that they sequestered may be exported from mangrove ecosystems as a result of their offshore movements. Similarly, in the estuaries of the Gulf of Mexico as much as 5-10% of total primary production is transported offshore during the migration of *Brevoortia patronus* from estuarine nursery grounds (Deegan, 1993).

In this chapter we document the age, size and timing of the recruitment of mangrove jack juveniles into estuaries and inshore areas and examine factors affecting recruitment variability.

Methods

In this section, juvenile mangrove jack were regarded as fish less than 100mm LCF. The primary sampling methodology was a generator powered Smith-Root® Model 7.5 GPP electrofisher fitted to a 4.3 m vessel. A pulsed direct current was applied to the areas of likely fish habitat usually along the banks but also including midstream structures such as snags or rock bars. Mangrove jack were captured using pole nets and the habitat type from which they were taken, their length and weight were recorded. Most fish were tagged before being returned to the water in the general vicinity of capture. A small number of mangrove jack were kept for related studies. Primary sampling sites were wet tropics streams (Daintree, Mulgrave, Russell and North and South Johnstone Rivers) that, where practical, were sampled monthly from late 1999 to February 2002. Secondary sampling locations included sites along the Queensland coast south to Baffle Creek and these sites were sampled quarterly. At each site, a number of defined sections of the riverbank were electrofished. Details of sampling times are in the General Methods section and maps showing the location of the sampling sites are in the Background. Apart from the regular sampling locations, a number of other sites were sampled opportunistically when conditions allowed. For example, in February 2001 seasonal flooding in the Russell River depressed the salinity in the lower estuary allowing electrofishing in areas where, under normal conditions, this sampling method is not feasible.

CPUE was calculated for each section fished at each site by dividing the total number of mangrove jack caught by the total electrofishing time and multiplying the result by 1000. This gave number of fish caught per 1000 seconds of electrofishing as the measure of relative abundance. Periodic local conditions at these sites, including high salinities and high river flows, precluded sampling on some occasions.

Results

Size and age at recruitment

The smallest mangrove jack caught during the study were two 20 mm fish caught at Mutchero Inlet at the mouth of the Russell River during a flood event on the 26 February 2001. The otoliths of these fish were extracted and, using daily growth increments, their age was calculated at 32 days, giving a hatch date of 25 January 2001.

Figure 31 shows the month of hatching of 25 juvenile mangrove jack back-calculated using daily growth increments on their otoliths. Most of the fish hatched between December and March however small numbers were also hatched in October and November. This is consistent with the peak spawning times for mangrove jack as determined through gonosomatic indices (see Reproduction section).

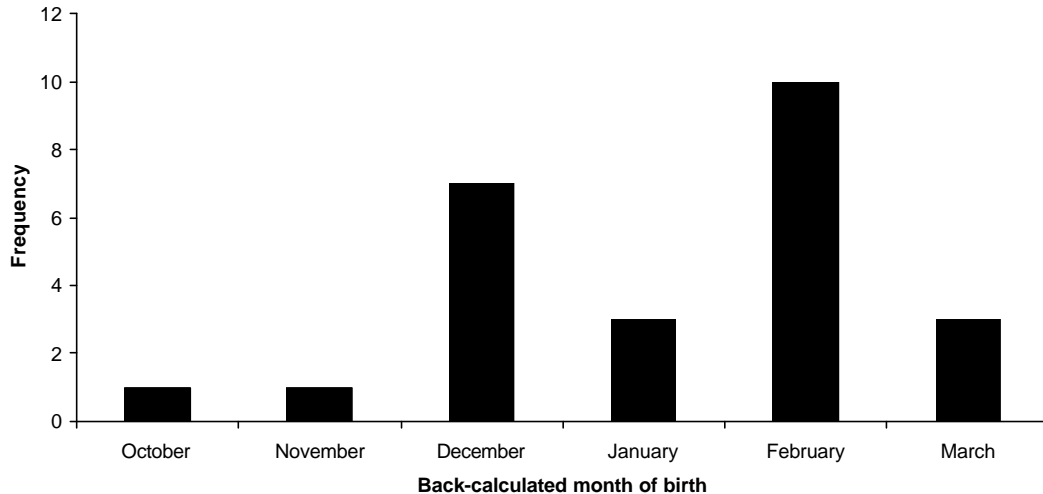


Figure 31. The back-calculated birth month of 25 juvenile mangrove jack using daily growth increments.

Seasonality in recruitment

There is evidence for recruitment of juvenile mangrove jack into estuaries and rivers during the first half of each year (Figure 32). As all of the sampling sites are more than 5km from the river mouths it is possible that recruitment may have commenced as early as November or December. However the juvenile fish were resident in the lower estuary and did not penetrate upstream to the sampling sites until about February. The two 20mm LCF mangrove jack, the smallest caught during the study, were sampled at the mouth of the Russell River in late February when a flood event depressed estuarine salinities and allowed electrofishing to within 300 m of the river mouth. During the same event a 22 mm fish was also sampled as well as a four others up to 50 mm long. Flood events and increased sequential discharges during the wet season may impede upstream movements of juvenile fish. Fish less than 50 mm long were sampled in all months between February and July suggesting that juveniles were recruiting into freshwater riverine habitats over an extended period. Towards the end of the year none of the smaller size classes were sampled.

In central Queensland, juvenile mangrove jack appear to move up into the freshwater riverine habitats in late summer or early autumn. At the Ben Anderson tidal barrage on the Burnett River 10 juvenile mangrove jack between 24 and 36 mm LCF were captured moving upstream through the vertical slot fishway in April 1999 (A. Berghuis, QDPI, pers. comm.). The barrage is 24 km upstream from the river mouth. In subsequent monitoring no other juvenile mangrove jack have been caught moving through the fishway.

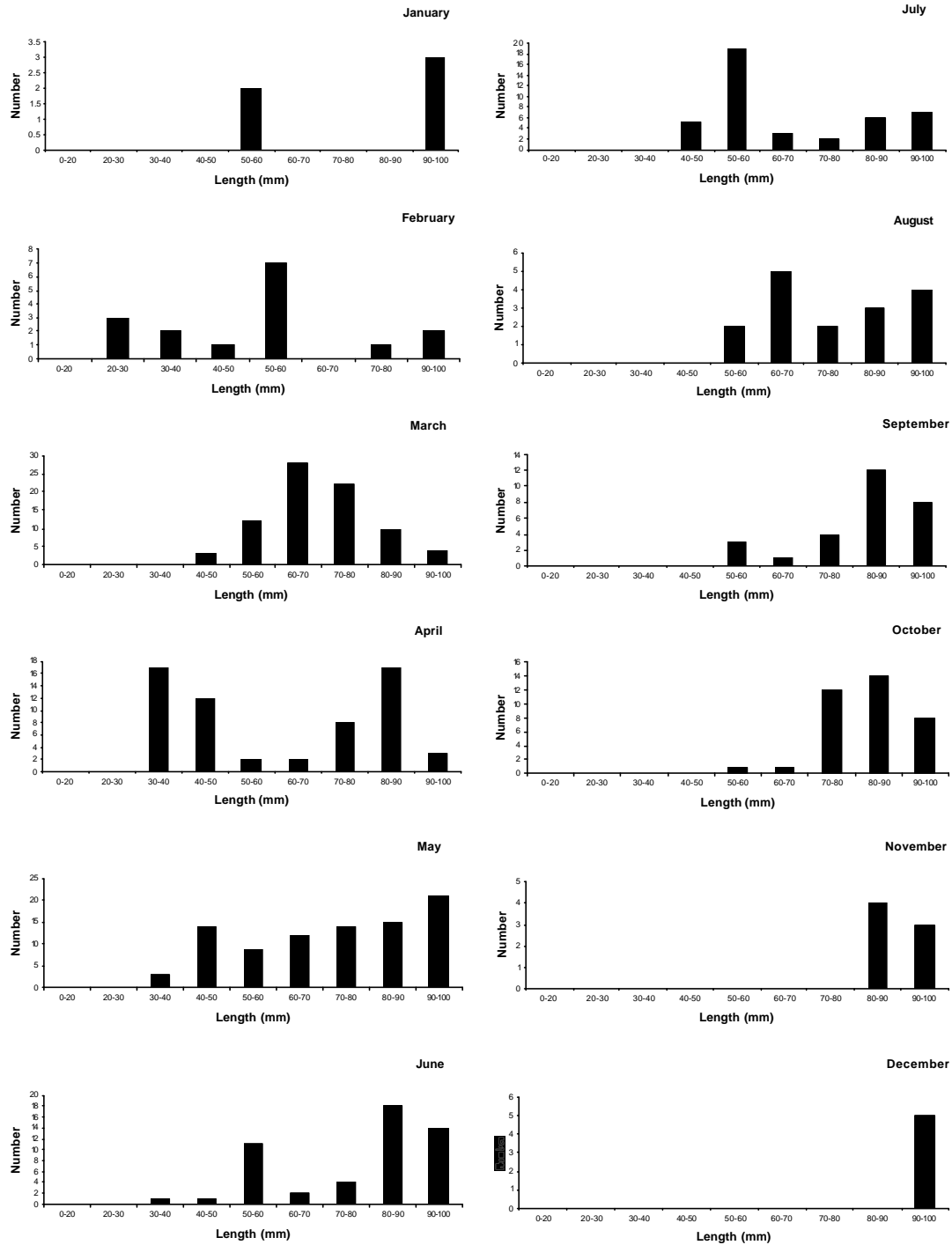


Figure 32. Monthly abundance of juvenile mangrove jack by 10 mm size classes (LCF) pooled for all sites and all years.

Recruitment variability

Variability in recruitment of juvenile fish into rivers occurs from year to year. In the wet tropics streams (Daintree, Mulgrave, Russell and Johnstone Rivers), the catch-per-unit effort of juvenile fish less than 100 mm LCF was generally higher in 2001 than it was in 2000 (Figure 33). However, this is not the trend in the Herbert River where CPUE progressively declined from a high at the commencement of sampling in May 2000 to a low in December

2001. In Baffle Creek, in 2001, lack of flow due to dry weather conditions allowed saltwater to penetrate much further upstream than normal, thus precluding use of electrofishing to sample sites. Consequently the Baffle Creek sites were only sampled once in 2001 (March) and no juvenile mangrove jack were caught on that occasion.

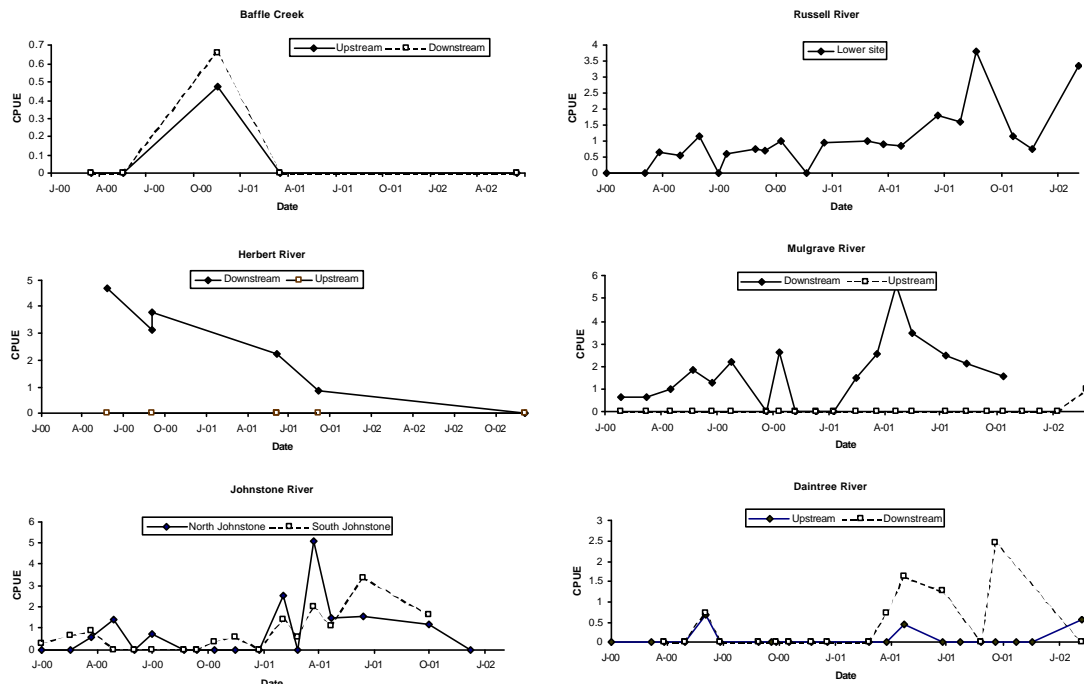


Figure 33. Catch per unit effort of juvenile mangrove jack (< 100 mm LCF) at study locations along the Queensland coast.

As the distance from the river mouth increased, the relative abundance of juvenile mangrove jack decreased (Figure 34). With the notable exception of sites in the Herbert River and Baffle Creek, the relative abundances of mangrove jack at most sites was lower in 2000 than 2001 (Figure 34).

Table 24. Average sizes (mm LCF) of juvenile fish caught in 2000 and 2001 with sample number in parentheses.

The codes are BA, Baffle Creek; CA, Calliope River; DA, Daintree River; HE, Herbert River; MU, Mulgrave River; NJ, North Johnstone River; OC, O'Connell River; RU, Russell River; SJ South Johnstone River.

Site Code	2000	2001
CR1	97.75 (4)	65.36 (14)
DA2	50 (1)	81.42 (12)
HE2	81.73 (15)	89.23 (13)
MU6	78.49 (51)	59.37 (91)
NJ1	80.29 (7)	62.26 (68)
OC1	87.44(9)	0
RU1	91.25(12)	77.68 (28)
SJ1	78.3 (3)	81.60 (28)

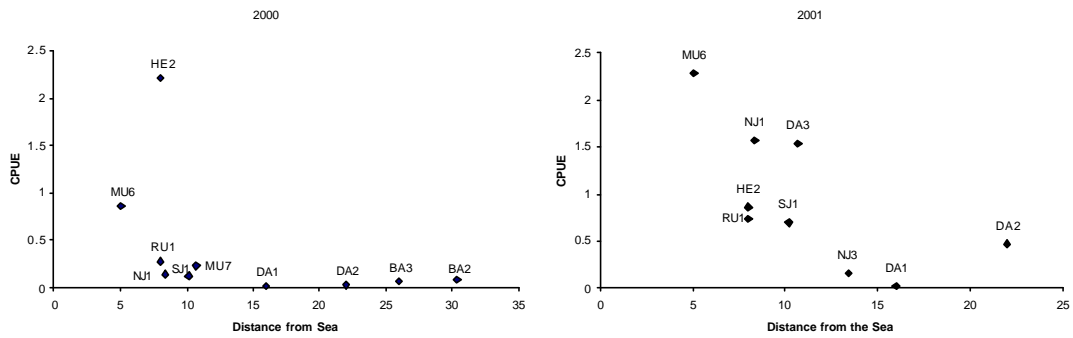


Figure 34. CPUE in 2000 (left) and 2001 (right) at study sites for juvenile mangrove jack plotted against distance (km) from the river mouth.

The codes are BA, Baffle Creek; DA, Daintree River; HE, Herbert River; MU, Mulgrave River; NJ, North Johnstone River; RU, Russell River; SJ South Johnstone River.

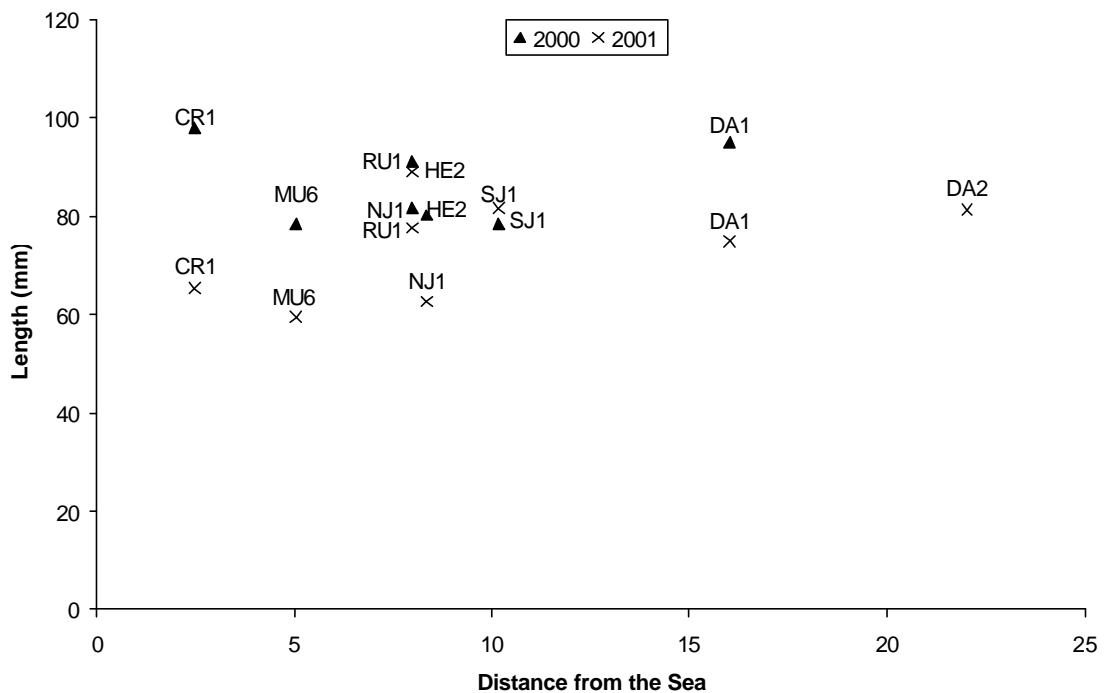


Figure 35. Average length of juvenile fish (<100mm LCF) versus distance from sea.

Sites codes as for Figure 34.

The average sizes of juvenile fish (<100 mm) caught in the North Johnstone, Mulgrave and Russell Rivers were higher in 2000 than those juveniles sampled in 2001 (Table 24). Given

that the relative abundance in those river systems was also high in 2001 than in 2000 these data suggest the presence of larger numbers of smaller fish in those systems in 2001.

The relationship between the average size of juvenile fish and distance from the river mouth is shown in Figure 35. For fish sampled in 2000 there is a very weak ($R^2 = 0.22$) positive linear relationship between average fish length and distance of the site from the sea. However there is no linear relationship for the fish caught in 2001 ($R^2 = 0.002$).

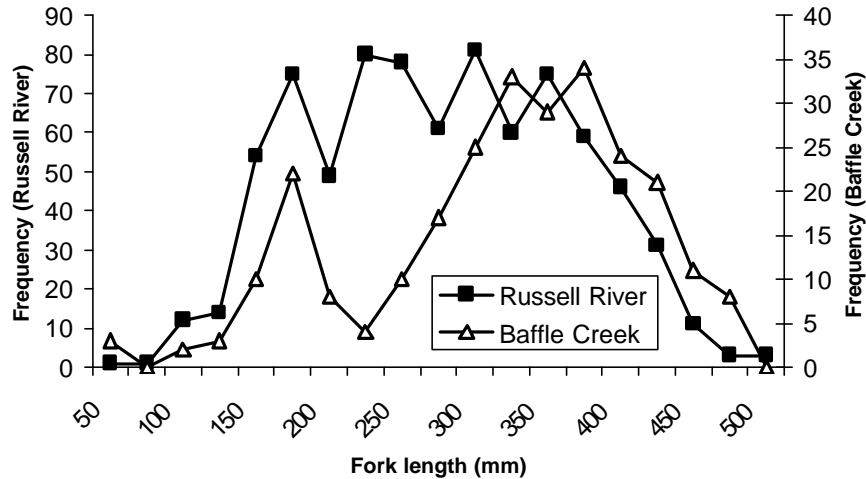


Figure 36. Length-frequency of mangrove jack from the Russell River and Baffle Creek.

Length-frequency plots from the Russell River and Baffle Creek (Figure 36) also support the proposition of recruitment variability. Fish appear to have fully recruited into the upper tidal areas where the sampling was undertaken at a length of about 175 mm LCF. In the Russell River, the number of fish in the size classes from 175mm to 375mm remains relatively constant and then declines probably because fish then begin to move to offshore habitats. Initially, the size-frequency plot for the Baffle Creek fish follows the same trend, rising to a high in the 175mm size class and then sharply dipping before rising again. This dip may indicate a recruitment failure in one or more year classes of mangrove jack in Baffle Creek.

Recruitment to offshore areas

Despite evidence of spawning activity in offshore areas (see Reproduction chapter) no juvenile fish were sampled in habitats other than in rivers, tidal creeks and gutters or in supra-littoral wetlands. The smallest fish that was sampled offshore from the commercial catches was 370 mm LCF.

Settlement

The 20-30 mm juvenile fish sampled in the Russell River estuary on the 26 February 2001 were caught in amongst coffee rock close to the river mouth. See Habitat section for details of habitat preferences of juvenile mangrove jack.

Discussion

Juvenile mangrove jack are recruited into rivers and inshore coastal areas at an early age. The smallest mangrove jack that were sampled in a river during this study were 20 mm long and about 32 days old. Fish of similar size were also sampled using rotenone in an earlier study in

supra-littoral tidal wetlands in Princess Charlotte Bay and in the Cairns region (DJ Russell, QDPI, unpublished data). Despite spawning occurring offshore, no evidence was found in this study of juvenile fish inhabiting offshore areas.

At most sites the relative abundance of juvenile mangrove jack was lower in 2000 than in 2001. The reasons for the relatively poor recruitment in 2000 is not clear but may be related to climatic conditions (Doi and Singhagraiwan, 1993) or hydrological factors in the rivers or on the offshore spawning grounds. Young-of the year juvenile fish were recruited into rivers from early in the year (from at least as early as February) and the timing corresponds roughly to midway through the northern monsoon season. Similar observations have been made in other studies. For example, Doi *et al.* (1998) noted that juvenile fish appeared to recruit into the estuary after the wet season and suggested that the upstream movement of *L. argentimaculatus* is governed by freshwater runoff resulting from high seasonal rainfall. In Thailand, as well as a spawning season peaking from late September to November, there appears to be spawning later in the year around March to April (Doi and Singhagraiwan, 1993).

Table 25. Total monthly and average rainfall for the first three months of 2000 and 2001 for Babinda (17.3431°S, 145.9219°E) and Innisfail (17.5250° S, 146.0344° E).

Both centres are in the Queensland wet tropics. (Source: QDPI Rainman software).

Location	Year	January (mm)	February (mm)	March (mm)	Average (mm)
Babinda	2000	309	1360	916	1146.25
	2001	511	1227	424	1040.75
Innisfail	2000	426	1020	672	1029.5
	2001	240	744	311	824

The magnitude of the wet season freshwater runoff may not necessarily be a significant contributing factor attracting post larval and juvenile fish into estuarine and coastal areas. At sites in wet tropics streams the relative abundance of mangrove jack juveniles was higher in 2001 than in 2000. However the total rainfall for the first three months of 2001 was 18% less for the Russell River (Babinda) and 39% less for Johnstone River (Innisfail) than for 2000 (Table 25) suggesting that other factors beside river flow may be impacting on recruitment. The lower average size of fish caught in 2001 compared with juvenile fish caught in 2000 also suggests that recruitment was greater in 2001.

In central Queensland, small numbers of juvenile mangrove jack have been found to move up through the vertical slot fish way on the Ben Anderson Barrage on the Burnett River into freshwater but there was no evidence of any large scale movements (A. Berghuis, QDPI, pers. comm.). Mangrove jack are among juveniles of a number of species including tarpon (*Megalops cyprinoides*) and barramundi (*Lates calcarifer*) that appear to move upstream in the Burnett River in early autumn regardless of the river flow (A. Berghuis, QDPI, pers. comm.). While mangrove jack are known to move considerable distances into freshwater (Merrick and Schmida, 1984), unlike barramundi, most fish remain in the estuary or the upper tidal areas. In this study the relative abundance of mangrove jack juveniles increased as distances from the sea decreased. At sites that were relatively large distances upstream (eg. Baffle Creek at > 30 km) the CPUEs for juvenile fish were low or zero but higher for larger fish suggesting that colonization upstream is incremental over an extended period, with larger and older fish more likely to be found further upstream.

There is evidence that some tidal weirs, even those with certain types of fishways, can have adverse impacts on upstream movements of catadromous fish (Kowarsky and Ross, 1981; Russell, 1991; Stuart and Berghuis, 2002). In this study juvenile mangrove jack were observed swimming in tidal waters below tidal barrages on the Haughton River, Alligator Creek and Ross River in the Townsville region. All these weirs did not have fishways and, despite intensive sampling, no mangrove jack were caught in their impounded waters. At least one type of fishway, the vertical slot design, does allow some upstream passage of mangrove jack. Other types, for example pool and weir designs, appear less effective in allowing passage of juvenile mangrove jack (Kowarsky and Ross, 1981; Russell, 1991). Temporary structures like sand dams, which are constructed to allow irrigation of adjacent agricultural land during dry periods, are less of a long term obstacle. Provided these structures block fish movement for only part of the year and are washed away by each wet season flow, mangrove jack may migrate to utilize upstream habitat. During this study a range of different sized mangrove jack were sampled upstream of sand dams. Juvenile mangrove jack utilize habitats in the upper tidal and lower freshwater reaches of rivers. It is critical for the maintenance of riverine populations of mangrove jack that permanent obstacles, such as tidal barrages, to fish moving into these habitats have suitable fishways incorporated into each structure.

Mortality and fishing yields

Introduction

The three parameters that are used in most classical fisheries models are recruitment growth and mortality. An estimation of mortality is an important parameter for management of a species as it relates to the loss of individuals and biomass from the population (Everhart *et al.*, 1975). Total mortality estimates (Z) are assumed to be constant over time and is composed of fishing mortality (F) and the instantaneous natural mortality (M). The relationship between these parameters is given by the equation:

$$\text{Equation 4 } Z = F + M$$

The effect of natural mortality is a logarithmic decrease in the number of fish present in each age class (Everhart *et al.*, 1975). Causes of natural mortality include disease, predation, and starvation while fishing mortality is explained by removal of fish from the fishery due to fishing. This could be the result of stress of capture, disease or injury caused by fishing or simply by physical removal.

There are three primary methods that are most commonly used for estimating mortality in fish species. These are estimates made using catch curves, virtual population analysis and tag-recapture data. As well, there are other equations available that are family specific or relate to other environmental parameters (Ralston, 1987).

The first primary method is to use a “catch curve”, which essentially is a plot of the natural log (\ln) of the frequency of individuals in each age group. The plot should result in a linear equation with the resulting slope equal to $-Z$ (Ricker, 1975). Fish that have not fully recruited to the population and older, less common fish are excluded from this analysis. This method generally results in the best estimates of mortality as it is an actual measure of the decline in abundance of age classes and is particularly sensitive where the sampling technique is non-selective (electrofishing). The second primary method of estimating mortality is to take a single sample of a population and then determine the number of fish in each age class. An assumption using this technique is that the age composition does not change from year to year. This is called virtual population analysis (VPA), and is used in instances where further samples cannot be reasonably obtained. The third primary method is to use tag and recapture data to estimate mortality. This uses the assumption that loss of individuals from a tagged population mirrors losses in the overall population (Ricker, 1975). This technique has not been used extensively in either groupers or snapper in the past because their lifecycles involve extensive migrations, and subsequent loss of tagged individuals (Ralston, 1987).

Assumptions include no mortality from tagging, equal probability of capture of tagged individuals and minimal tag loss, which are not necessarily all applicable to these species (Cappo *et al.*, 2000).

In the literature there are some general equations for estimating mortality in snappers and grouper. These equations estimate mortality using parameters including maximum length (Hoenig, 1983), the von Bertalanffy growth parameters (L_8 , K) and mean environmental temperature (T) (Pauly, 1980) and the von Bertalanffy rate of change of length or weight (K) (Ralston, 1987). Pauly (1983) also suggests the use of a correction factor of 0.8 to ameliorate the affect of schooling, a behaviour which is common in the red snappers of the GBR. Another general equation links length at maturity and mortality, however there is large range in the proportion of L_m/L_8 in the species used (Rikhter and Efanov (1976) in Sparre *et al.* (Sparre *et al.*, 1989)).

Different estimates of mortality for a single species can have large implications on other parameters calculated from these figures. For example, estimates of Z from catch curves derived using different ageing techniques (with associated errors) for three lutjanid species common to the GBR showed that using a less accurate ageing technique could overestimate Z by up to 100% (Newman *et al.*, 2000c). Resultant estimates of yield-per-recruit models suggested that maximum yield was obtained in fish at a younger age (whole otolith age) and the value of the yield was up to 50% less than sectioned otolith ages.

The availability of natural mortality (M) estimates and weight-at-age data allows the calculation of yield per recruit (YPR) (Ricker, 1975). The value where YPR is at maximum is also the value that allows maximum fishing mortality (F_{max}). Because there is only a need for minimal change in F to affect the YPR, a level of F is used where increase in YPR due to a small increase in F is 10% of the marginal YPR in a lightly exploited stock ($F_{0.1}$) (Deriso, 1987).

In this chapter we compare the different techniques to estimate mortality in mangrove jack populations in Queensland and use the best mortality estimates to develop a yield-per recruit model.

Methods

Catch Curves

Catch curves were produced according to Ricker (1975), where only fully recruited age groups were considered, and where older uncommon age classes were excluded from the analyses (Ricker, 1975). Full catch curves were produced using *Genstat*® and FAST software packages (Slipke and Maceina, 2001), and a linear regression fitted to the appropriate age classes. Both weighted and un-weighted regressions were used, with the regression with the best correlation coefficient eventually accepted. Catch curves were produced for all samples which had been aged and for both sexes as differential mortality is not uncommon in Lutjanidae (Ralston, 1987). Accurate catch information and samples for ageing was also obtained from a typical commercial fisher who fished remote offshore fishing grounds for reef species including mangrove jack where $Z \sim M$ (ie. the fishing mortality assumed to be negligible).

To determine if there were any differences between different fishing zones (inshore and offshore), separate catch curves were produced to attempt to elucidate fishing mortality in inshore and offshore areas (Ricker, 1975). The age classes included in these analyses were those that were only found in each area. For example, age classes that were not fully represented (through either initial recruitment or recruitment to adult fishery) and uncommon older fish were excluded where there were 3 continuous age classes where no fish were sampled. A comparison of weighted and unweighted datasets using the FAST software package determined the regression that best described the data (highest r^2 value).

Because only a small percentage (<5%) (see Figure 1 in Age and growth section) of fish sampled from electrofishing operations were destructively sampled to produce age/length keys, the available data set did not provide a truly accurate picture of length frequency that could be then used in catch curve analysis. To remedy this, the proportion of each age class in 50 mm increments were estimated from the aged samples, and these proportions were applied to all of the inshore fish such that they were assigned an estimated age. Total mortality was also estimated for populations in both Baffle Creek (southern Queensland) and in the Russell River (northern Queensland) to provide a basis to compare estimates with those calculated from tagging studies (see Map 1 in Materials and Methods section). The level of F was estimated from these plots by subtraction of the value of M (where $Z \sim M$) calculated from

data obtained from the remote offshore location from the Z value obtained from the inshore fishery (Equation 4).

Mortality estimates using other methods

A number of other methods are available to estimate mortality and these were used to compare with the values obtained with the three primary estimation methods described above. These general mortality equations were as follows:

Equation 5

Hoenig's (1983) method

$$\ln Z = 1.46 - 1.01 \ln t_{\max}$$

where: t_{\max} = maximum age in years

Equation 6

Pauly's (1980) method

$$\log_{10} M = -0.0066 - 0.279 \log_{10} L_8 + 0.6543 \log_{10} K + 0.4634 \log_{10} T$$

where: T = mean annual water temperature (25.7 °C for pooled data from (Newman *et al.*, 2000a))

Equation 7

Ralston's (1987) method

$$M = 0.666 + 2.52 K$$

Equation 8

Pauly's (1983) method

$$M = 0.8 * (\text{Pauly, 1980}) \quad (\text{to account for schooling behaviour})$$

Equation 9

The method of (Rikhter & Efanov (1976) in (Sparre *et al.*, 1989)

$$M = 1.521 / (T_m^{0.720}) - 0.155$$

where: T_m = age where 50% of fish are mature

The results of these equations are tabulated, along with estimates produced with trial values for other geographic locations or zones.

Tagging

As the duration of this project was less than three years, research tagging data was only available from two full years that could be used to estimate mortality and survival. Survival is related to total mortality through the expression:

Equation 10

$$S = e^{-Z}$$

Ricker (1975) outlines a method of determining all mortalities (Z , F and M) and S where marking is done throughout the year (section 5.2, pg.126). This method uses the equation:

Equation 11

$$R_{12} M_2 / R_{22} M_1 = A_2^2 / Z_2 - A_2$$

where: M_1 = number of fish tagged in first year

M_2 = number of fish tagged in second year

R_{12} = number of fish recaptured in second year (first year of tagging)

R_{22} = number of fish recaptured in second year (second year of tagging)

A_2 = Annual mortality rate (second year)

Z_2 = Total mortality rate (second year)

Trial values of mortalities were first produced using the above equation using trial values in the right-hand side of the equation (Ricker, 1975). By initially using these trial values, more accurate estimates of mortality and survival are produced.

Tag and recapture data obtained for the Russell River and Baffle Creek were used in this exercise because both of these systems had sufficient data and they would provide an insight into any differences between two geographically distinct locations in Queensland.

The tag data from the research sampling in these systems was also collated and run using the four standard models in program MARK (White, 2000). The input for this software package required the production of catch histories for each tagged fish over the life of the project, with

the data being in a binary form where '1' relates to a tagging or capture event and '0' relates to a sampling interval where the fish was not seen.

The complicated life history of mangrove jack where fish emigrate from inshore nursery areas to offshore breeding grounds prevented the use of recently developed open-population estimating software. Less than ten fish have been recaptured from offshore locations from this research project, which would not provide sufficient precision in estimation of parameters required to estimate movement. The AUSTAG dataset could be used in future to determine the rate of movement of mangrove jack in different fishing zones, but this is outside the scope of this report. The AIC (Akaike's Information Criteria) calculated in MARK was used to provide the model with the best precision. AIC is an index used which "penalises" the better fit of the more parameterised models for the reduced precision of the estimates themselves (White, 2000). A lower AIC value represents a better model precision.

Plots of the change in recapture rates over time periods were used to assess the applicability of using the tag data as recommended by (Ricker, 1975). Underlying sources of error include non-reporting of tags, type-I and II tag shedding, tagging mortality or behaviour effects, and other losses such as emigration or differential loss due to tagging. Tag shedding is further discussed in the Movement section of this report.

Changes in fishing mortality can be estimated through knowledge of the fishery and data from tagging exercises, however estimates on tag losses are also needed. The level of exploitation (u) is calculated by dividing the number of recaptured fish by the number of tagged fish (Ricker, 1975). Tag loss reduces the number of fish potentially able to be recaptured and therefore causes underestimate of exploitation. The number of fish tagged in the population is corrected by accounting for the estimate of tag loss. Irregularities due to non-reporting can be estimated by dividing the number of recaptures by the inverse of the estimated non-reporting rate.

Natural death (v) is the percentage of fish that die from other causes not related to fishing activity. Annual mortality incorporates the fish dying naturally while exploitation is occurring (Ricker, 1975).

Equation 12

$$AM = u + v$$

Annual survival is then:

Equation 13

$$S = 1 - AM$$

F and M can then be computed by:

Equation 14

$$F = u * Z / (1 - S)$$

and

Equation 15

$$M = v*Z/(1-S)$$

Conditional fishing mortality (cf) is the exploitation rate when no natural mortality occurs. In addition, conditional natural mortality (cm) is the death rate due to natural causes when no fishing mortality occurs simultaneously (Slipke and Maceina, 2001). These can then be calculated by:

Equation 16

$$cf = 1 - e^{-F}$$

and

Equation 17

$$cm = 1 - e^{-M}$$

The calculation of these equations allows the comparison of effects of changing this size at capture and therefore the flow-on effects of this mortality (Slipke and Maceina, 2001).

Yield per recruit (YPR)

Von Bertalanffy parameters and mortality estimates derived from catch curves were entered into program FAST (Slipke and Maceina, 2001) to determine YPR. FAST utilises the Jones modification to the Beverton-Holt method of determining yield (Ricker, 1975). The output of the yield contour plot was used to determine the impact of current exploitation and also the exploitation rate that maximises yield.

In addition, YPR was also computed using the method described by Ricker (1975) to model the impact of the size of recruitment to the fishery (minimum legal size (MLS)) and the associated F value. The parameters used in this model were a maximum age set at 11 years for the inshore fishery and a natural mortality set at 0.158 for the first 3 years and then 0.55 thereafter.

Results

Estimation of mortality rates from catch curves

Mangrove jack are a minor component of both the recreational and commercial fisheries in offshore areas. This species only contributes approximately 1% (with an annual variation between 0.87%-1.20%) of the total catch of a typical commercial line fisher that supplied accurate and detailed data on his catches in offshore areas on the Great Barrier Reef (Ray Walker, pers. comm.). In comparison, red emperor, large-mouth nannygai and small-mouth nannygai (*Lutjanus sebae*, *L. malabaricus* and *L. erythropterus*) contribute 13.9% (12.98%-14.45%), 14.55% (13.65%-16.19%) and 11.36% (10.53%-12.57%) of the total catch respectively. Overall mangrove jack only contribute <1% of the total commercial line-fishing

catch annually (CFISH data, 2002). The catch curve for this commercial fisher was used to estimate M on the basis that he is fishing what is essentially an almost unexploited stock. The fisher works along relatively remote reefs from Lizard Island north of Cooktown, down along the GBR through to Myrmidon Reef offshore of Townsville. The fishing effort for mangrove jack in much of this area is assumed to be minimal.

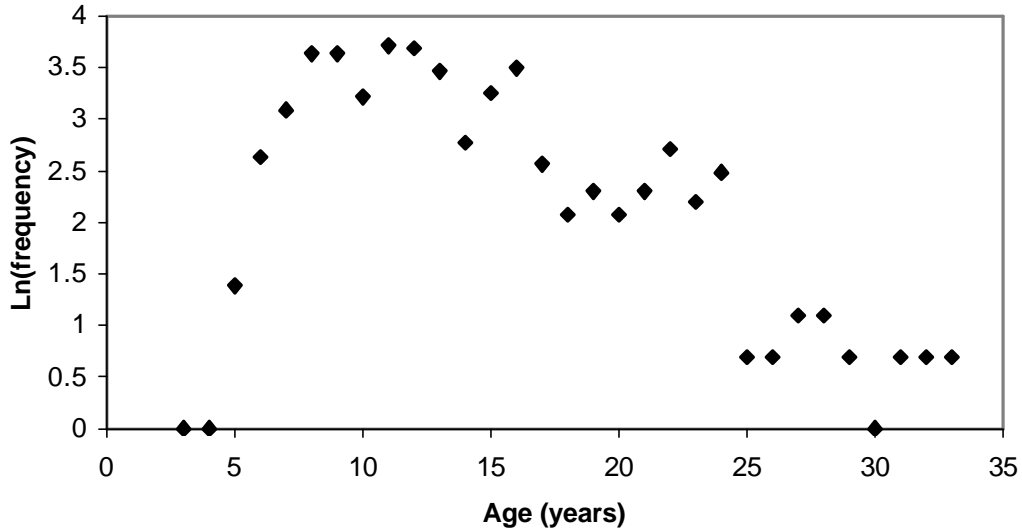


Figure 37. Total catch curve for mangrove jack from north Queensland from offshore locations.

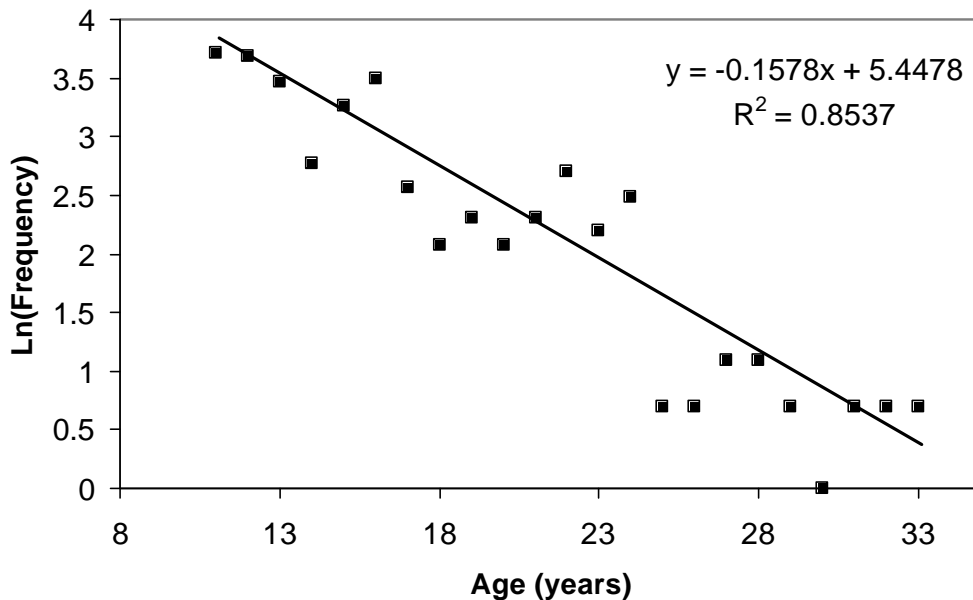


Figure 38. Catch curve for vulnerable age classes in offshore areas of North Queensland.

There were 435 fish between the ages of 3 and 33 years of age used in this analysis. Figure 37 shows that mangrove jack are not fully represented in the catch until the age of 8 years, where the curve is at a maximum for a further 5 years. The ascending portion of the curve

represents the proportion of fish migrating into the fishery (E) minus that lost to natural mortality (M). An estimate of this rate of migration is presented in the Movements section. A truncated catch curve between the age of 11 years and 33 years was used to determine M (Ricker, 1975)(Figure 38).

The resulting regression produced an estimate of $Z \sim M=0.158$ and a maximum age of approximately 35 years ($S=0.854$, $AM=0.146$). Long-lived species like mangrove jack must have low natural mortality rates to sustain their longevity in natural states (Ralston, 1987). Unfortunately few samples were available from other offshore locations around the state, with only 144 fish coming from recreational anglers fishing in offshore areas from throughout Australia. Because each age class was not adequately represented, and differential growth occurs between latitudes, an estimate of natural mortality or total mortality could not be made with a high level of precision. Furthermore, when the catch curve regressions for the separate sexes (north Queensland data only) were compared using ANCOVA, no significant difference ($p=0.138$, 1 d.f., $F=2.53$) was found, thus the data could be pooled to give an estimate of total mortality. Approximately equal number of fish (208 males and 219 females) over the same age range were used in this analysis, which would suggest that there is no selective mortality between sexes.

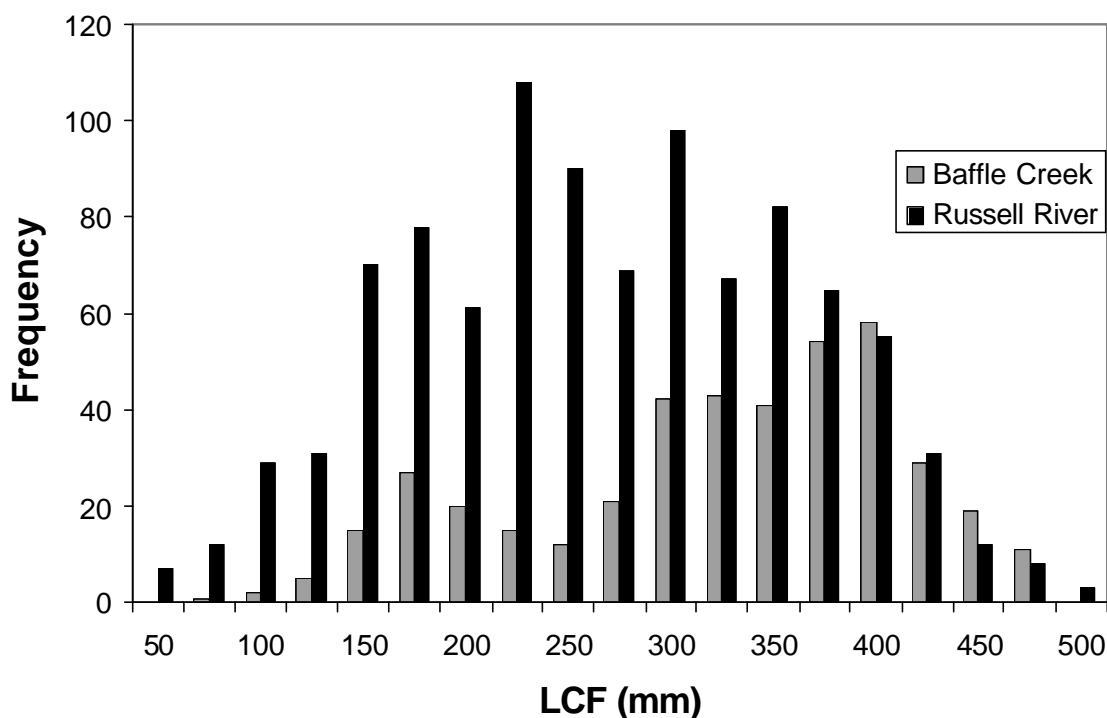


Figure 39. Length frequency of mangrove jack in 25 mm size classes (LCF) in Baffle Creek and the Russell River.

These fish were sampled by electrofishing.

Because only a small percentage of electrofished samples were kept for ageing (see Figure 15 in Age and growth section), percentages of fish in each 50 mm length class were assigned proportionally to age classes determined by ageing the limited samples that were taken (Ricker, 1975). This was instead of producing a catch curve based on length rather than age, which does not take into account differential growth rates. The assumption using this method

is that the variation in age estimates assigned to each age class were similar to the small sub-sample used.

Two separate trends are evident when comparing the length frequency plots of the Russell River (n=976) and Baffle Creek (n=415) (Figure 39); more fish were caught in the Russell River and different modes between the systems. In the Russell River, the number of fish caught increases until the 250 mm size class, with the numbers then decreasing. Cohorts appear to be visible at the 175, 225, 300 and 350 mm size classes. In Baffle Creek, there appears to be cohorts visible at the 175, 325 and 400 mm size classes, with the majority of fish in the larger size classes. Note that both frequency plots deplete at the same rate after about 400 mm, suggesting a similar pattern of emigration. Electrofishing is a relatively unbiased technique giving a representative sample of all sizes of fish (Wildman and Neumann, 2002).

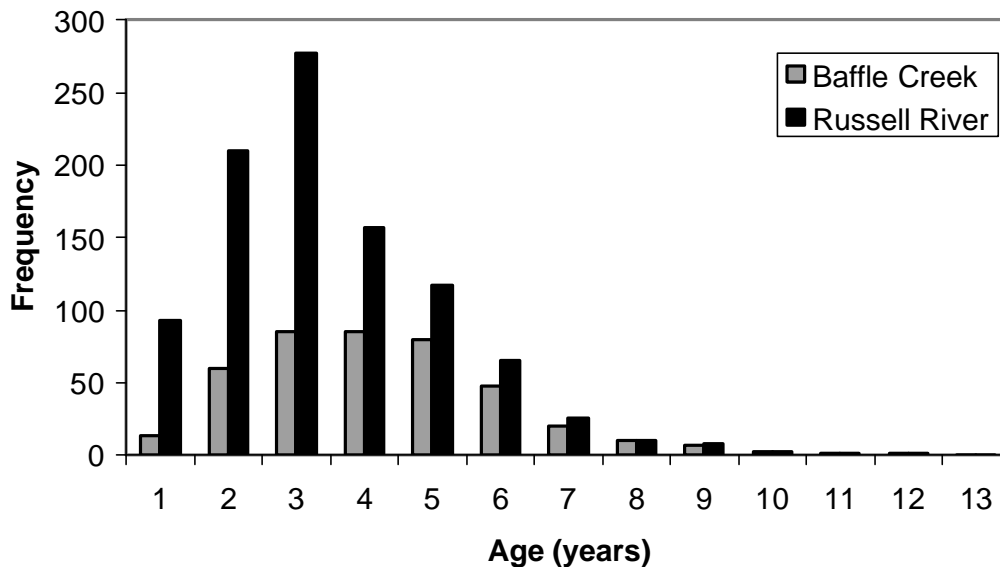


Figure 40. Age frequency of mangrove jack populations in Baffle Creek and the Russell River. Simulated from aged samples and applied to electrofishing data.

However, because only a small proportion of fish were kept for ageing analysis, the age structure of the fishery was simulated from the age-length data for each system (Figure 40). As there was extensive overlap in lengths between each age class, percentages of 50 mm length classes were assigned to age classes as determined by the proportions in the aged samples.

When the age-frequency data was plotted as catch curves (with only the fully recruited classes used) there was no significant difference in mortality (slopes) between the sites ($Z=0.661$)($p=0.266$, $F=1.35$, 1 d.f., $n=16$)(Figure 41).

Mortality estimates using other methods

The estimates of mortality of mangrove jack were estimated using the five other equations given in the methods section for the pooled data are given in Table 26.

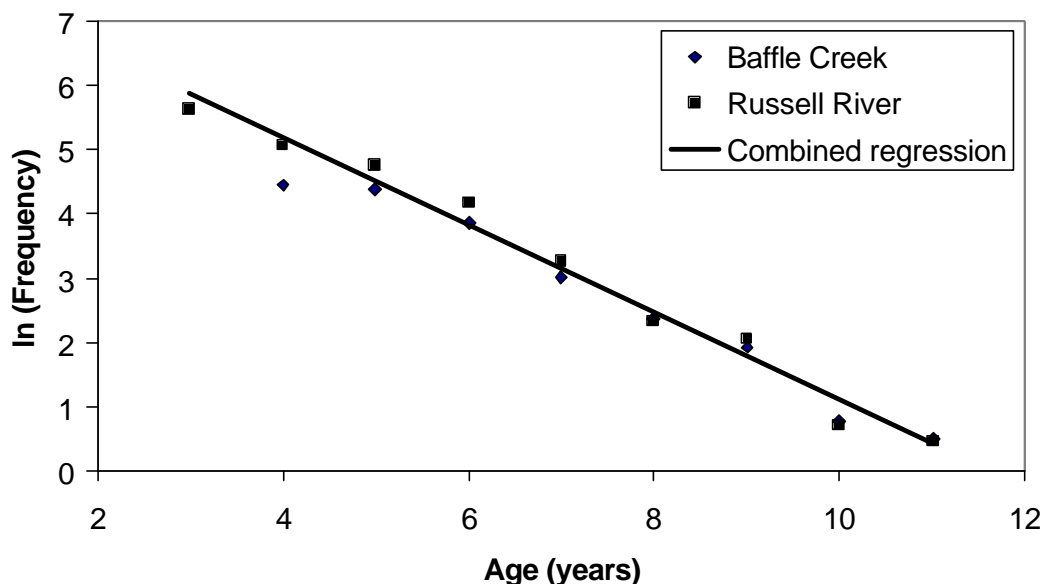


Figure 41. Catch curve for simulated age data for mangrove jack populations in Baffle Creek and the Russell River.

Table 26. Mortality estimates for mangrove jack in Australia from five standard equations.

* This is an estimate of Z not M.

	Hoening (1983)	Pauly (1980)	Ralston (1987)	Pauly (1983)	Rickter & Efanov (1976)
Pooled data	0.106*	0.232	0.503	0.185	0.157

The estimates do vary significantly with the largest value almost five times greater than the lowest. As mentioned previously such a wide range of estimates would result in significant differences in the estimates of *F* and excessive YPR (Newman *et al.*, 2000c).

Estimate of mortality and survival from tagging

Ricker's method

The number of mangrove jack tagged and recaptured over the two years of tagging for the Russell River and Baffle Creek are given in Table 27. The Russell River was sampled on 22 occasions, whereas Baffle Creek was only sampled six times. There was also a difference in sampling frequency; the sampling protocol was for the Russell River to be sampled monthly and Baffle Creek to be fished quarterly. Unfortunately unfavourable environmental conditions (eg. low flows) reduced the sampling frequency in Baffle Creek to three occasions in 2000, one in 2001 and one in 2002. Similarly, high salinities or high flows caused some of the scheduled monthly sampling sessions for the Russell River to be cancelled.

Table 27. Parameters used to compute estimates of mortality of mangrove jack from research tagging in years 2000 and 2001.

Explanation of parameters as outlined in methods section.

	Russell River	Baffle Creek
M ₁	568	268
M ₂	203	148
R ₁₂ research sampling	13	12
R ₂₂ research sampling	9	1
Z ¹ (S ¹)	2.81 (0.0602)	0.01 (0.99)
F ¹	0.7	-
M	2.11	-
Z ² (S ²)	2.28 (0.1023)	-
F ²	0.167	-

The values computed for Z for stocks in the Russell River was very high (2.81 and 2.28) and this resulted in a very low survival estimate (0.06 to 0.10). Due to the unequal sampling periods and reduced recaptures in the second year, only a preliminary value of total mortality could be computed (Z) as the estimates were actually less than those provided by Ricker (1975). Therefore, because of the lack of uniform sampling periods and the low recapture rate, it was concluded that this method was not a particularly useful means of estimating mortality.

Estimates from the software Program Mark:

Only recaptures from the research sampling were used in this method to estimate mortality and survival. However, it became apparent after running the program that, because of their complex life history, accurate estimates of survival probabilities could only be obtained where information on movement was incorporated into the analyses as there may be some behavioural impacts of tagging.

Table 28. Output of the four models in MARK for Baffle Creek.

Selected model is highlighted.

Model	AIC*	Number of parameters
Phi()p()	204.23	2
Phi()p(t)	210.77	6
Phi(t)p()	206.47	5
Phi(t)p(t)	206.72	7

Table 29. Output of the four models in MARK for the Russell River. Selected model is highlighted.

AIC (Akaike's Information Criteria) is used to determine the best model. Phi=survival probability, p=probability of recapture, ()=changing rates, (t)=time dependant rates.

Model	AIC	Number of parameters
Phi()p()	929.71	2
Phi()p(t)	852.97	22
Phi(t)p()	929.44	22
Phi(t)p(t)	880.33	41

The most parsimonious model for the Baffle Creek data was one with constant probability of survival and capture between events (Table 28 and Table 30). The results suggest that

the mangrove jack have a very low annual mortality (AM=0.107) and a probability of capture of 3.9%. In contrast, the most parsimonious model for the Russell River data is one with constant probability of survival but with varying probability of capture (Table 29 and Table 31). This tag data suggests that the annual mortality in the Russell River is similar (AM=0.144), however the probability of capture varied between ~0 to approximately 15%. There were three occasions (highlighted in Table 31) where the probability of capture was quite high (April – June 2000)(p=11.6-14.6%) which was at a time where the number of animals being captured was quite high. Other reasons for unequal and decreasing probability of capture may be due to seasonal offshore migration, environmental conditions or behavioural response to electrofishing intensity (monthly).

Table 30. Probability of survival and capture between events for tagging data from Baffle Creek.

Model is $\Phi(p)$.

Parameter	Estimate	SE	Upper CI	Lower CI
Probability of survival $\Phi()$	0.968	0.030	0.995	0.813
Probability of capture $p()$	0.039	0.014	0.078	0.019

Table 31. Probability of survival and capture between events for tagging data from the Russell River.

Model is $\Phi(p(t))$.

Parameter	Estimate	SE	Upper CI	Lower CI
Probability of survival $\Phi()$	0.841	0.029	0.890	0.776
Probability of capture $p_{(1)}$	$0.10 \cdot 10^{-15}$	$0.32 \cdot 10^{-8}$	$0.63 \cdot 10^{-8}$	$-0.63 \cdot 10^{-8}$
$P_{(2)}$	0.116	0.039	0.218	0.058
$P_{(3)}$	0.147	0.036	0.231	0.090
$P_{(4)}$	0.126	0.030	0.198	0.077
$P_{(5)}$	$0.34 \cdot 10^{-16}$	$0.52 \cdot 10^{-9}$	$0.10 \cdot 10^{-8}$	$-0.10 \cdot 10^{-8}$
$P_{(6)}$	$0.14 \cdot 10^{-18}$	$0.36 \cdot 10^{-10}$	$0.70 \cdot 10^{-10}$	$-.70 \cdot 10^{-10}$
$P_{(7)}$	0.048	0.015	0.087	0.027
$P_{(8)}$	0.036	0.013	0.073	0.017
$P_{(9)}$	$0.45 \cdot 10^{-16}$	$0.67 \cdot 10^{-9}$	$0.13 \cdot 10^{-8}$	$-0.13 \cdot 10^{-8}$
$P_{(10)}$	0.040	0.016	0.088	0.017
$P_{(11)}$	0.031	0.015	0.078	0.012
$P_{(12)}$	0.025	0.016	0.083	0.007
$P_{(13)}$	0.009	0.009	0.063	0.001
$P_{(14)}$	$0.38 \cdot 10^{-16}$	$0.82 \cdot 10^{-9}$	$0.16 \cdot 10^{-8}$	$-0.16 \cdot 10^{-8}$
$P_{(15)}$	0.012	0.012	0.087	0.002
$P_{(16)}$	0.074	0.037	0.189	0.027
$P_{(17)}$	0.031	0.020	0.103	0.009
$P_{(18)}$	0.023	0.017	0.096	0.005
$P_{(19)}$	0.025	0.019	0.103	0.005
$P_{(20)}$	$0.14 \cdot 10^{-16}$	$0.61 \cdot 10^{-9}$	$0.12 \cdot 10^{-8}$	$-0.12 \cdot 10^{-8}$
$P_{(21)}$	0.017	0.018	0.122	0.002

Estimate of fishing mortality, natural mortality and movement of inshore fish

One of the main techniques for evaluating the impacts of fishing is to estimate the level of fishing mortality. Using the total mortality calculated from the catch curves for the inshore fish ($Z=0.661$) and the level of natural mortality obtained from the “unexploited” offshore stocks ($M=0.158$), the predicted level of current fishing mortality is 0.503. However, because there are fish constantly leaving the inshore population through emigration (E), Equation 4 should be re-written

Equation 18

$$Z = F + (M + E)$$

Hence, without an estimate of the impact of emigration, the value calculated for F is an overestimate. Note that the fish emigrate from the inshore areas from the age of 3 years and are fully recruited to the offshore fishery by the 11+ age class (see Age and growth section and Figure 37). A better estimate of F may be calculated by inputting the exploitation estimates obtained from tagging into the FAST software.

Inputs for FAST

By combining the tagging from this research and Suntag data, it was found that there were 2 120 fish tagged in the Russell River with 63 recaptures ($\mu=2.97\%$) and 469 fish tagged in Baffle Creek with 11 recaptures by anglers ($\mu=2.35\%$). Using recreational tagging only, there were 554 mangrove jack tagged in the Russell River with 27 recaptures ($\mu=4.87\%$) and 92 mangrove jack tagged in Baffle Creek with 6 recaptures ($\mu=6.52\%$). There was a relatively large discrepancy in the total numbers of fish tagged in these systems and because the exploitation rate was relatively constant, a generic exploitation rate of 5.83% (Queensland State average from Suntag database) was used for further analysis. Overall, there were 31 offshore recaptures out of the 20 036 tagged mangrove jack (0.15% recapture rate) in the Suntag database.

Tag loss as determined by tag-shedding study experiments in Oonoonba and Walkamin, in addition to double tagging suggests that tag loss is approximated to be about 20% (see Movement section). Non-reporting was approximated for this species by the limited number of anglers that reported to Suntag that they either did not keep the tag number when reporting, or released the fish without recording the number. The estimated non-reporting rate was 5% (Bill Sawynok, pers. comm.). Non-reporting of tagged fish has been substantiated for other species and programs including a rate of 28% for saltwater fish in Texas (Matlock, 1981), 7% for Black Crappie (*Pomoxis nigromaculatus*) (Eder, 1990) and significantly increased reporting due to monetary rewards for two other species in impoundments in the USA (Haas, 1990). Exploitation rate was therefore calculated as:

$$\mu=(5.83)/(0.95)/(100*0.8)=7.67\%$$

$$\mu_{\text{offshore}} = (0.15)/(0.95)/(100*0.8) = \mathbf{0.2\%}$$

and

$$F \text{ (from Equation 14)} = 0.0767 * 0.661 / (1 - 0.52) = 0.106$$

$$F_{\text{offshore}} \text{ (from Equation 14)} = 0.002 * 0.661 / (1 - 0.52) = 0.003$$

and therefore:

$$M + E \text{ (from Equation 18)} = 0.661 - 0.106 = 0.555$$

Using the value of M established for offshore populations, emigration is calculated as:

$$E = 0.555 - 0.158 = 0.397$$

Conditional fishing and natural mortalities for the inshore population calculated from Equation 16 and Equation 17 is 0.101 and 0.146 respectively.

Yield per recruit

Using the FAST software and the pooled growth equation (see Age and growth section), the YPR model provided a yield contour plot for a cumulative mortality of 0.45, which approximates $M+E$ (Figure 42). With the current rate of exploitation (0.0767), the length at entry to the fishery producing maximal yield would be approximately 250 mm TL using the 21.0 kg yield contour. Maximum yield would be obtained at a size of approximately 175 mm (44.1 kg yield contour).

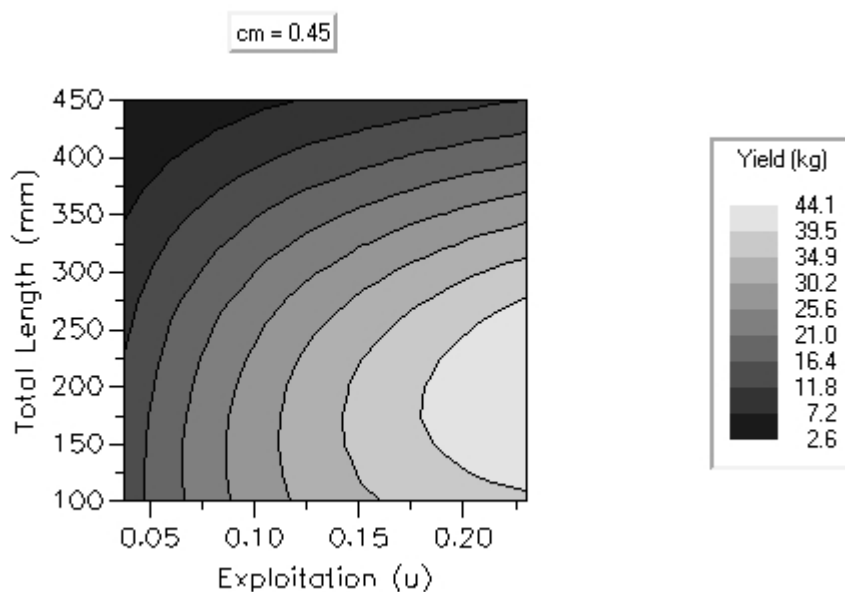


Figure 42. Yield contour plot of inshore fishery derived using a cumulative mortality of 0.45 ($M+E$).

Output from FAST using the Beverton-Holt method.

Table 32. Optimal $F_{0.1}$ and nominal yield-per-recruit (g).

This is for a range of ages-at-first capture (minimum legal size) for both Wet tropics and South-eastern Queensland zone growth parameters.

Zone	Wet tropics		South-eastern Queensland	
	$F_{0.1}$	YPR	$F_{0.1}$	YPR
1	0.20	307.55	0.22	410.45
2	0.28	314.71	0.32	411.48
3	0.47	299.08	0.47	383.79
4	0.67	252.08	0.68	320.40
5	0.83	188.02	0.80	239.53

To model the possible impacts of use of a minimum legal size (MLS) based on the age-at-capture, YPR was computed for both the wet tropics and southeastern Queensland zones (Table 32). Optimal values of fishing mortality were simultaneously established to match the yield (Deriso, 1987) using the method given by (Ricker, 1975).

The age-at-first-capture for optimal YPR for both the wet tropics and southeastern Queensland were 2 years (FL from von Bertalanffy equation of 183.14 mm and 236.3 mm respectively). This equated to a maximum fishing mortality of 0.28 and 0.32 for the wet tropics and southeastern Queensland, which is approximately 3 times the current estimated level of fishing mortality (0.101). If emigration is not taken into account, ie only $M=0.158$ is used, the resultant optimal age does not change, however, the yields and levels of fishing mortalities differ ($F_{0.1}=0.22$ and YPR=841g for wet tropics and $F_{0.1}=0.18$ and YPR=1112 g for southeastern Queensland). Both of these sizes of entry to the fishery are considerably

smaller than the current MLS in Queensland (350 mm TL) and the L_m established in this project (512.3 mm for females and 449.4 mm for males).

Discussion

Catch curves

The use of catch curves for mangrove jack resident offshore produced a low mortality rate of 0.158, where $M \sim Z$. These data were obtained from a commercial fisher who operated in remote areas where fishery exploitation by recreational and commercial fishers is thought to be relatively insignificant (Ray Walker, commercial fisher, pers. comm.). The fact that mangrove jack only contribute about 1% of his total catch combined with the low exploitation rate (0.2%) obtained from the tagging data suggests that the exploitation of offshore stocks is minimal and the mortality derived from the catch curve is $M \approx Z$.

Similarly low mortality rates have been found using catch curves for other lutjanids from the GBR: *L. carponotatus* ($M=0.199$) and *L. vitta* ($M=0.342$) (Newman *et al.*, 2000a); *L. erythropterus* ($M=0.1495$), *L. malabaricus* ($M=0.126$) and *L. sebae* ($M=0.147$) (Newman *et al.*, 2000c); and *L. adetti* ($M=0.235$) and *L. quinquelineatus* ($M=0.154$) (Newman *et al.*, 1996a).

Mangrove jack recruit to the offshore fishery beginning at an age of 3 years, but were not fully recruited until age 11 years (Figure 37). Due to the low exploitation rate for offshore areas and low catch rate for the commercial fishers, only 427 specimens were used for analysis of the offshore fishery in north Queensland. No comparison could be made of estimates of mortality in different geographical regions, however a comparison of the slopes of the regressions of catch curves for males and females from north Queensland found no significant difference ($p=0.138$). The sex ratio of sampled fish was relatively even (0.95:1 male:female) and concurs with the similar mortality estimates. This is in contrast to the prediction from the growth parameters (see Age and growth section), which were significantly different, suggesting mortality differences between sexes (Grimes, 1987).

Other studies of lutjanids have also found no significant differences between the mortalities of males and females (Newman *et al.*, 1996a; Newman *et al.*, 2000b), even though there were significant differences in growth parameters. Rocha-Olivares (Rocha-Olivares, 1997) suggested that the skewed sex ratio of *L. peru* may be explained by the significantly different total mortality rates he obtained in his study (also significantly different growth parameters). He found that the male *L. peru* grew to a larger size, were more dominant in the catches and had a higher Z (possibly due differential behaviour causing increased F).

Comparison of the total mortality estimates for two geographically separated streams (Russell River and Baffle Creek) suggests that was no significant difference ($p=0.266$) ($Z=0.661$)(Figure 41). Examination of the length-frequency and the simulated age-frequency plots also suggests that, although mortality estimates are similar, differential recruitment is present (Figure 39)(Figure 40). It is worth noting that the age that this fishery is fully recruited at approximately age 3 and then decreases with no fish present over 11 years of age. These ages correspond exactly to the age of the fish that are recruited into the offshore fishery.

Much of the age data for the inshore populations was inferred from a sub-sample of fish of known ages and therefore some care needs to be taken when drawing conclusions. However, there does appear to be some interesting trends even considering the possibility of ageing error and incorrect assignment of fish to year classes (Figure 40). For example, there appears to be less 1+ fish present in the Baffle Creek samples, which may be attributed to the site location. As the site at Baffle Creek is approximately 30 km from the mouth, there is poor

recruitment of mangrove jack until at least 150 mm when compared to sites closer to the river mouth (see Recruitment section). Conversely, the Russell River site is dominated by younger age classes, with abundance dropping steadily in older fish.

Mortality estimates using other methods

Using these other methods, estimates of mortality varied between 0.106 – 0.503, compared with the estimate derived from catch curves of $M=0.158$ (Table 26). The estimate using Hoenig's method (Hoenig, 1983) probably underestimated the value of M by using a maximum age of 39 years. These data were obtained for one specimen aged from vertebrae from southeastern Queensland (FL 1035 mm). Using a maximum age of 33 years (the biggest fish caught in north Queensland), the estimate of 0.126 is closer to that derived from catch curves. In contrast, the estimates obtained using the equation of Pauly (1980) and Ralston (1987) also produced probable overestimates (0.232 and 0.503) respectively. Newman *et al.* (1996a) also found these equations overestimated the mortality of the long-lived *L. quinquelineatus*. He suggested that the Pauly equation (Pauly, 1980) was derived using only few long-lived species and hence not suitable. Both of these equations use the growth coefficient (K) that is significantly different in slow-growing species that have a relatively rapid early growth (K) coupled with a rapid change to reduced growth.

Pauly (1983) sought to account for schooling activity in some species by introducing a compensating factor of 0.8; for mangrove jack, the estimate obtained using this compensating factor would be revised to 0.185. Evidence from fishing operations suggest that mangrove jack, while they do occasionally aggregate on snags, infrequently school, although there have been a few instances where there have been substantial hauls made by trawlers (W. Sawynok, Suttgart, pers comm.). The estimate (0.157) using techniques developed by Rikhter & Efanov (1976) on the other hand, is very similar to that derived from the catch curves and is produced by using an average age at maturity.

Use of the estimates from Pauly (1980) and Ralston (1987) would have the probable effect of underestimating F and consequently producing erroneous YPR estimates and subsequent over-utilisation of the stock (Newman *et al.*, 2000c, b).

Estimates from tagging

The use of tagging data to directly derive mortality estimates produced mixed and most probably erroneous results. As detailed in the project proposal, the method by Ricker (1975) which used tag-recapture data obtained in two consecutive years was used to estimate mortality. Using this method, total mortality (Z) for the Russell River was estimated to be 2.81 for year 1 and 2.28 for year 2. Total mortality was estimated to be less than 0.01 for Baffle Creek for both years (Table 27). These estimates are very different to those produced by catch curves for the same areas (Figure 41).

The population-modelling program, MARK, was also used to estimate probabilities of survival and capture. The results suggest a constant probability of survival (96.8%, $Z\sim 0.04$) and capture (3.9%) for Baffle Creek and constant probability of survival (84.1%, $Z\sim 0.11$) and time-dependant capture probability (~ 0 to 14.7%) for the Russell River.

Baffle Creek sampling was supposed to be conducted quarterly, however due to lack of flow, it was only sampled on 6 occasions with uneven sampling intervals. Seasonal variability in CPUE in the Russell River, with the resultant capture probabilities being very variable.

Both methods are flawed for a number of reasons. These techniques don't take into account factors such as tag loss, and loss of individuals due to emigration (closed population models) and uneven sampling periods as occurred at the Baffle Creek sites. Behavioural impacts of tagging could also play an important role in affecting results, with the capture probability

being highly variable but reducing in the last half of the experiment for the intensely sampled Russell River (Table 29).

Estimation of emigration and fishing mortality

The exploitation rates for Baffle Creek and the Russell River were very similar and within the range of the Queensland state average (5.83%) for fish tagged and released by recreational fishers (Suntag database 2002). Differences in rates could be explained purely by large differences in number of fish tagged, rather than any fishing differences. The exploitation rates determined including the tagging done by this project were underestimated primarily due to the fact that most mangrove jack are recaptured within a year and differences in fishing zones between researchers and anglers caused under representation due to limited movement of fish (see Movement section). The exploitation rate in inshore areas was increased by allowing for non-reporting and tag loss to produce a value of 7.67%. The corrected value of exploitation for the offshore fishery is estimated from recapture data at 0.2% and demonstrates possible differences in fishing effort between inshore and offshore zones. These differences are reflected in the catches of recreational and commercial fishers in 2001 (55 tonne: approx. 2 tonne) being inshore and offshore respectively (Queensland Fisheries Service CFISH and RFISH databases).

When exploitation rate is converted to F , this equates to values of 0.106 and 0.003 for the inshore and offshore fisheries respectively. Assuming natural mortality is similar in both fisheries, the level of emigration was calculated to be 0.397, which accounts for the vast majority of the loss of individuals from the inshore fishery. Interestingly, James Aumend (James Cook University) used changes in otolith microchemistry to determine the age of emigration from a number of the fish used in this study and found the modal age to be 6 years. By looking at Figure 5 in Appendix 3 and Figure 40 from this chapter, it can be seen that the large change in later year classes after 3 years is primarily due to the emigration seen in Figure 5 of Appendix 3.

Natural mortality has been assumed to be similar between the inshore and offshore habitats, however the level of predation on this species may be higher than originally thought. Sheaves (2001) suggests that the level of piscivory of shallow estuarine habitats is poorly understood, with many small piscivorous species able to impact substantially on larval and juvenile fish. Mangrove jack juveniles have to survive numerous days migrating in from offshore spawning grounds and then take up residence in suitable habitat in estuarine areas. This species has a high affinity to structure and may be able to utilise the small rock habitats to protect against substantial piscivory pressure both from larger individuals of the same and other species.

As emigration and fishing and natural mortality are occurring continuously, the emigration causes a disproportionate reduction of the number of fish in larger size classes and therefore tends to regulate F to some degree. Faunce *et al.* (2002) suggested that the protection of the gray snapper (*L. griseus*), a species with similar life history to mangrove jack, from fishing in the Crocodile Sanctuary of the Everglades National Park caused an increase in the number of fish above the MLS of 254 mm. However, the 3 sites located in this National Park were riverine, whereas the sites in the open areas were from estuarine or coastal sites. This species does appear to aggregate to a greater degree and hence may be less resilient to exploitation, however differences in length structure could equally be due to recruitment variability. The National Park site contained the larger fish, but there were no individuals less than 150 mm in length, compared to the open sites where these size classes averaged between 10-30% of the total number of individuals. Further protection of *L. griseus* is suggested through the increase in the MLS to 350 mm, which is the current MLS in Queensland for mangrove jack. Exploitation of the offshore breeding population and inshore fishery for *L. griseus* appears to be many times more than that impacting mangrove jack so although *L. griseus* has a similar

biology, it is exposed to greater exploitation both inshore and offshore and may require the increased MLS and protection from fishing in both these regions. Closure of fishing access to inshore areas inhabited by immature mangrove jack would no doubt cause a minor shift in length frequency to larger sizes, better long-term protection may be afforded by protection and rehabilitation of the habitat required by this species (see Habitat section).

Yield per recruit

Using the yield contour plot obtained from the Beverton-Holt method of computing YPR, suggests that at an exploitation rate of about 18%, the optimal size at entry to the fishery was very small (approximately 175 mm, yield contour 44.1 kg)(Figure 42). At the current level of exploitation, the optimal yield size-at-entry is approximately 250 mm (yield contour 21.0 kg). The maximum exploitation rate is approximately three times the current exploitation, with the current yield at the MLS of about a third of optimal yield.

Using the Ricker (1975) method, the optimal age/size at capture was optimised at age 2 (183.14 mm and 236.3 mm respectively for the wet tropics and southeastern Queensland)(Table 32). These lengths of capture are similar to the optimal size of capture by the previous method, but are both very much smaller than the current Queensland MLS of 350 mm TL. These lengths also are considerably smaller than the length at maturity (512.3 mm for females and 449.4 mm for males), with most individuals of 236 mm or less being still immature.

By using the value for natural mortality only and discounting emigration, values for optimal yield were many times larger and comparable with studies on other offshore lutjanid species (Newman *et al.*, 2000c). However, the optimal age at entry to the fishery for the three red snappers in their study (*L. erythropterus*, *L. malabaricus* and *L. sebae*) were significantly older (4, 6 and 7 years respectively). In addition, these ages corresponded closely to the length at maturity for all three species and were all above the current MLS for each species (Newman *et al.*, 2000c). The M (0.158) obtained for mangrove jack is very similar to that for these three species ($M=0.1495$, 0.126 and 0.147 respectively); however these values were calculated for the inshore fishery only where there is a continuous emigration of individuals older than three years to offshore areas.

This emigration to the offshore areas by mangrove jack accounts for the majority of loss of individuals for the inshore fishery. Because there is this large proportion of fish that are not exposed to fishing mortality in the inshore fishery (fish < 350 mm TL) and minimal offshore fishing mortality, the adult fishery is relatively healthy. In addition, mangrove jack are highly fecund (see Reproduction section), many of the traditional commercial fishing gears are ineffective at catching the species and the current level of exploitation is less than a third of the optimal fishing pressure. These factors, coupled with the present conservative Queensland MSL suggest an appropriate management regime is in place for the fishery in most parts of Queensland. In areas where recruitment may not be reliable such as in southern Queensland and northern New South Wales, the MLS acts as a buffer to reduce the level of fishing mortality on successful age cohorts.

Conclusion

The level of natural mortality is very low ($M=0.158$) and is comparable with values obtained in studies on other lutjanids from the Great Barrier Reef. Total mortality is higher in inshore areas ($Z=0.661$) than offshore areas ($Z-M=0.158$). Four of the five standard equations for estimating mortality produced values that were similar to those obtained from the catch curves.

Estimates of total mortality obtained from the tagging data were highly variable (Z ranging from 0.01 and 2.81) and proved unreliable due to at least one assumption not being met.

There was no significant difference in mortality rates between sexes even though there were significantly different estimates of growth parameters (see Age and growth section). Estimates of F are difficult to calculate without a reasonable estimate of emigration. Using tagging exploitation rates, the level of fishing mortality was estimated to be 0.101 for inshore areas and 0.002 for offshore locations.

Preliminary modelling suggests that the current level of fishing mortality is a third of the optimal level, with the corresponding yield also less than optimal. Optimal size at entry to the fishery is about 200 mm, whereas the current MLS is 350 mm TL in Queensland. Based on continual recruitment, the use of the current MLS, although conservative, is appropriate for management of the species in northern areas. In southern areas of Queensland and northern New South Wales, where recruitment appears to be less continuous, the conservative MLS may be more beneficial.

There is a substantial emigration of individuals from the inshore nursery areas, where most fishing occurs, to the offshore locations where exploitation is low. This element of their life history ensures that most of the fishing effort effects only a few age cohorts and that the very young fish and the breeding populations are substantially protected. Further, with the level of fishing mortality estimated to be quite low and less than natural mortality, the fishery does not appear to be fully exploited.

Genetic Population Structure

Introduction

The beneficial role that population genetics analyses of fisheries species can play in their sustainable management is widely known (Shaklee, 1983; Ovenden, 1990; Ward and Grewe, 1994). The key benefits include

- Direct statistical testing of the null hypothesis of a single panmictic population,
- Wide applicability across species,
- Utilisation of naturally occurring genetic variation eliminating the costs and assumptions of artificial tagging, and
- Unaffected by environmental conditions unlike meristic and morphometric characters (Shaklee and Bentzen, 1998).

For commercial marine species such as finfish, crustaceans and molluscs, genetic information on the degree of population subdivision in space and time is important to the design of management regimes. For example, the application of uniform catch quotas across an assemblage of genetically separate populations may result in local extinction, especially in hyperstable species where catch per unit effort does not decline at the same rate as biomass. For commercial estuarine and freshwater species, where the presence of population subdivision is likely to be more common and pronounced, there is an additional management consideration. Genetically distinct populations may be brought into contact and mixed, intentionally or accidentally, when a species is translocated for captive breeding, fisheries enhancement or other purposes (Skibinski, 1997). Although it has been argued that intraspecific hybridisation may not be harmful (Keenan, 1998), loss of local adaptations may occur when interbreeding disrupts genetically linked gene complexes and consequently reduces fitness. These complexes may confer advantages during periods of stress associated with environmental change or onset of disease (Allendorf *et al.*, 2001). Government fisheries protection agencies often have policies to protect endemic aquatic species against these, and other detrimental affects, of translocation (Anonymous, 1999).

Lutjanus argentimaculatus (Forsskål, 1775) or mangrove jack is a member of the most common genus in the snapper family (Lutjanidae). In a predominantly tropical marine Indo-Pacific group, it is the only Australian species of this group to enter freshwater where juveniles and sub-adults are common. Adults are found in marine habitats such as reefs in 100m of water where they may reach 16kg. Mangrove jack is distributed in northern and eastern Australia from Ningaloo Reef (WA) to Sydney (NSW) and is widespread in the Indo-Pacific from East Africa to Samoa and northwards to Japan (Allen *et al.*, 2002). A small commercial fishery operates in Queensland and north-west Western Australia where reef-dwelling individuals are taken by line and trap. The species has been domesticated and captive breeding occurs in south-east Asia and Australia (Doi and Singhagraiwan, 1993; Garrett, 1994). It is prized by anglers for its aggressiveness and has good table characteristics. In Queensland, selected inland freshwater impoundments may be stocked with captive-bred mangrove jack to promote recreational fishing opportunities.

There are a comprehensive set of precedents for genetic population subdivision in marine and estuarine fish and Shaklee (1998) has identified categories representing varying gradations of genetic subdivision. Some marine species are highly subdivided over small distances, for example the catfish (*Cnidoglanis macrocephalus*, (Ayvazian *et al.*, 1994) and the damselfish (*Acanthochromis polyacanthus*, (Doherty and Fowler, 1994). They both lack a pelagic larval

stage and have limited gene flow between adjacent populations. Mangrove jack has a pelagic larval stage, but its vagility and duration are unknown. In contrast, other marine species are subdivided over larger distances. Australian inshore marine species such as barramundi (*Lates calcarifer*, (Keenan, 1994) and school mackerel (*Scomberomorus queenslandicus*, (Begg *et al.*, 1998) that spawn in estuaries (barramundi) or coastal embayments (mackerel) along the Queensland coastline conform to this pattern. Mangrove jack spawn 40-50 km offshore (McDougall, pers. comm.), but the scale of their dispersal away from nursery freshwater and estuarine habitats is uncertain. Shaklee (1998) also distinguishes species that appear genetically homogeneous over a large range but are subdivided in one or more areas. An excellent example is Western Australian pink snapper, *Pagrus (Chrysophrys) auratus* that was genetically homogeneous over 4,500 km of its Australian distribution, yet subdivided within Shark Bay, Western Australia (Johnson *et al.*, 1986). Shared biogeographic features can be an important factor also. In Australia, the genetic division of populations of numerous species to the east and west of the shallow waters of Torres Strait; green turtles (*Chelonia mydas*, (Fitzsimmons *et al.*, 1997), barramundi (*Lates calcarifer*, (Chenoweth *et al.*, 1998b) and Spanish mackerel (*Scomberomorus commerson*, (Ovenden and Street, in prep) may be explained by pre-Pleistocene sea-level fluctuations that periodically interrupted the marine pathway between the Indian and Pacific Oceans. The distribution of mangrove jack in Australia straddles the Torres Strait region and if their vagility was similar to barramundi or Spanish mackerel, then genetic subdivision would be expected to the east and west of Cape York.

The purpose of this study was to assess the degree of population subdivision in mangrove jack throughout its Australian distribution. Its bi-phasic life history, where juveniles and sub-adults live in estuaries and freshwater and adults live in the sea, suggests that the scale of dispersal may be limited and the species may be genetically subdivided. Alternatively, dispersal may be extensive during the adult or larval phases leading to genetic homogeneity. Distinction between these hypotheses is important for the licensing of the species for aquaculture and impoundment stocking in Australia under regulations that limit translocations between areas occupied by genetically distinct populations. Two classes of genetic markers were used; nuclear (microsatellite) and mitochondrial loci. Both confer the advantage of being able to sample fish non-lethally and provide a powerful test of genetic homogeneity (Buonaccorsi *et al.*, 2001). Additionally, mitochondrial sequence data can provide insights into the evolutionary history of populations under certain circumstances (Ovenden and Street, in prep).

Objectives

The objective of this study was to

“determine the genetic stock structure of mangrove jack in Queensland and other parts of their range.”

Methods

Sampling

Microsatellites

Mangrove jacks were genotyped from four locations in Australia,

South-east Queensland (SQ, Gladstone to Gold Coast, approximately 151-153 °E, 24-28 °),

North Queensland (NQ, between Cape Melville and Hinchinbrook Island, approximately 145-146°E, 14-18°S),

Gulf of Carpentaria (GOC, north and south of Weipa approximately 141-142°E, 11-14° 30'S and southern Gulf approximately 138°E, 16°30'S), and

Western Australia (WA, west Sahul Banks approximately 123-124°E, 13-14°S and Port Headland to Dampier approximately 116-119°E 20°S).

Fish were sampled from estuaries and freshwater rivers (Gulf of Carpentaria, 109 samples) and offshore reefs (WA, 11 from reefs and 92 from rivers; NQ, 35 reef and 66 rivers and SQ, 3 reef and 61 rivers). They were taken from research, recreational and commercial catches during 1999, 2000 and 2001.

Approximately 10-20 mm² of fin tissue was removed non-lethally, were applicable, and stored in DMSO (20% dimethyl sulphoxide in 5M sodium chloride) at room temperature in the field and – 80°C in the laboratory. Many of the fish caught by recreational anglers and biologists were released.

A target of 100 fish were genotyped from each location. Binomial theory describes the probability of collecting an allele of frequency 'p' as:

$$N = \left\lceil \frac{\ln(1-\alpha)}{\ln(1-p)} \right\rceil / 2$$

where *N* is the number of individuals required and 'α' is the confidence level desired (Bartley *et al.*, 1995). For example, a minimum of 74 fish is needed to capture a rare allele of frequency 0.02 at a 95% confidence level.

MtDNA

Mangrove jack analysed for mtDNA were a subset of those genotyped with microsatellites, with the addition of samples from four sampling locations outside Australian waters in the Indo-Pacific (Table 33). The Indo-Pacific sampling locations were in Indonesia from the islands of Bali (138°E, 16°30'S), Java (138°E, 16°30'S) and Sumatra (138°E, 16°30'S) and in Samoa (138°E, 16°30'S).

Table 33. Numbers of mangrove jack sampled for mtDNA nucleotide sequence data.

These fish were from Australian [southern Queensland (SQ), northern Queensland (NQ), Gulf of Carpentaria (GOC) and Western Australia (WA)] and Indo-pacific [Bali (BALI), Java (JAVA), Sumatra (SUM) and Samoa (SA)] sampling locations. MtDNA sequence data was collected from the control region (CR) and the ATPase genes.

Sampling Location	MtDNA gene	
	CR	ATPase
SQ	5	10
NQ	6	11
GOC	5	9
WA	5	10
BALI	5	5
JAVA	2	4
SUM	3	4
SA	3	3
Total	34	56

Laboratory Protocol

DNA isolation

Genomic DNA was isolated from 10-25 mg of preserved fin tissue using commercial kits (Qiagen P/L, P.O. Box 25 Clifton Hill, Victoria 3068 DNeasy™ tissue kit or Biorad P/L, P.O. Box 210 Regents Park, New South Wales 2143 Aquapure™ genomic tissue kit).

Microsatellites

Microsatellite loci (44) developed by Van Herwerden (2000a) for coral trout (*Plectropomus laevis*) and red throat emperor (*Lethrinus miniatus*) during FRDC project 98/131 were evaluated as population genetic markers for mangrove jack. Four suitable loci were found and these were used in this study (Table 34).

Table 34. Characteristics of microsatellite markers in *Plectropomus laevis* and *Lethrinus miniatus*.

These were developed by Van Herwerden *et al.* (2000), including repeat motif, forward and reverse primers and Genbank accession numbers. These loci were used for mangrove jack in this study.

Locus	Species	Repeat motif	Primers (5' to 3')	Genbank Acc. No.
BST2.33	<i>Plectropomus laevis</i>	(TG) ₃₅ (CG) ₁₂	F TAATGCCACAAACCTGCTGG R ATGTTCCACAACGCCTGACAAACC	AF249850
BST6.39TG	<i>Plectropomus laevis</i>	(TG) ₁₇	F GCAGCATTAAGTGAGAGAGGC R GGATAATGTAGGGCCAGAGCG	AF249855
BST6.56	<i>Plectropomus laevis</i>	(TG) ₈	F ACGTGAGCATTGAGGGTAA R ATCTCCATCATCTGCTGCCCTGG	AF249857
90RTE	<i>Lethrinus miniatus</i>	(TG) ₁₇ TATGA G(TG) ₄	F ATGCTGTCCACTTCCTCCAGC R TTTCTCAAACCTCCTGCCCTCC	AF261002

Microsatellite amplification, gel separation and scoring were performed by the AGRF (Australian Genome Research Facility, Melbourne Division, Walter and Eliza Hall Institute, Post Office Royal Melbourne Hospital Victoria 3050) from aliquots of extracted DNA using primers designed by Van Herwerden (2000a) using standard PCR conditions for microsatellites. Genotype information was returned by email and collated into local databases.

MtDNA

Approximately 375bp of the 5' end of the control region (D-loop) was amplified as described (Ovenden *et al.*, in press), but using primers Pro889U20 (CCW CTA ACT CCC AAA GCT AG) and TDKD1291L21 (CCT GAA ATA GGA ACC AAA TGC).

Primers COIII.2 [(GTT AGT GGT CA(GT) GGG CTT GG(AG) Bermingham , <http://nmg.si.edu/bermlab.html>] and MJB1567U22 (CCT TAA CAT GCT CGC ACT ACT C) were used to amplify approximately 400 base pairs of the protein-coding ATPase 6 gene. This region was chosen as it spanned a cluster of single nucleotide polymorphisms revealed by a comparison of the sequence of several mangrove jack from the entire ATPase 6 and 8 genes.

Forward and reverse PCR primers for the control region and the ATPase 6 region were used to sequence purified PCR products. Sequences were obtained with an ABI automated

sequencer using the chain-termination method with big-dye terminators. Sequence data was aligned with Sequencher v 3.12 (Anon, 2000).

Analysis Methods

Microsatellites

Allele frequency estimation, linkage disequilibrium, heterozygosity and Hardy-Weinberg tests were calculated from raw data using Genepop-on-the-web (Morgan, 2000) based on Genepop v. 3.3 (Raymond and Rousset, 1995) (dememorisation 5000, batches 500, iterations per batch 1000). Linkage disequilibrium was tested using goodness of fit procedures and the null hypothesis was that genotypes at one locus were independent from genotypes at the other locus. Observed heterozygosity, the proportion of individuals in the sample that were observed to be heterozygous by direct counting, was calculated for each locus and for each population. Exact tests for conformance to Hardy-Weinberg proportions were performed using the complete enumeration method for each microsatellite locus and each population sample.

Several low frequency alleles for locus BST6.39TG were pooled so that their pooled frequencies summed across population samples was greater than 0.025. At this frequency, there would have been a greater than 95% chance of detecting them in 62 or more fish; the SQ population sample consisted of 62 fish. Pooled allele frequencies were used in subsequent tests for population subdivision.

The significance of difference in allele frequencies between populations was tested using an analog of Fisher's exact test, the log-likelihood G-test, using Genepop-on-the-web (dememorisation 5000, batches 500, iterations per batch 1000). Analysis of molecular variance (AMOVA) (Excoffier *et al.*, 1992) was used to assess the significance of differing geographical groupings by quantifying the inter- and intragroup component of total variance using analogs of F-statistics in Arlequin v 2.0 (Schneider *et al.*, 2000). Statistical significance was inferred from a null distribution constructed from a random allocation of genotypes to simulated populations that have the same sample sizes as the original populations.

MtDNA

The filter taxa utility of Macclade (Maddison and Maddison, 1992) was used to determine the number of haplotypes from sequence data from the control region and ATPase 6 regions. The molecular distance among sequences was calculated according to the Kimura 2-parameter method (Kimura, 1980) with mutation rates for variable sites assumed to follow the gamma distribution with shape 0.5 (Yang, 1996).

Maximum parsimony trees were constructed from sequence data using the heuristic search option of PAUP 4.0 b10 (Swofford, 1999). Indels were regarded as 'fifth' bases. Characters were not weighted. The strict consensus method was used to summarise MP trees where appropriate.

Results

Microsatellites

The number of fish genotyped from each population sample ranged from 61 to 63 across loci for SQ, 88-102 for NQ, 87-93 for GOC and 90-102 for WA. Locus BST 6.39TG had the most missing data where 5-10% of samples were not able to be reliably genotyped. The number of alleles per locus was ten for 90RTE and BST6.56, nine for BST2.33 and 24 for locus BST6.39TG after pooling selected low frequency alleles (Table 36). Locus BST6.39TG had the highest level of heterozygosity (proportion of total number of expected heterozygotes;

0.930). Loci 90RTE, BST6.56 and BST2.33 had lower levels of heterozygosity (0.786, 0.602 and 0.636).

Linkage disequilibrium may have been possible for one pair of loci (90RTE, BST2.33) in one population (WA). This comparison returned a p-value that was significant after Bon-ferroni adjustment (Rice, 1989, $p=0.00010$). There was no observable pattern of genotype association for these loci in the WA sample, and this pair of loci was not in linkage disequilibrium in the remaining three population samples. This suggested that it was unlikely that the two loci were physically linked in the genome. It was concluded that there was no significant association among genotypes at the four microsatellite loci.

Observed and expected genotypic proportions for microsatellite loci were in general agreement with the Hardy-Weinberg principle. For locus BST2.33, two population samples had significantly different proportions of observed compared to expected heterozygotes and homozygotes (NQ, $p\text{-value} = 0.0076$; GOC, 0.0219). In these populations the number of observed homozygotes (48, 48) was above that expected (41, 34) and the number of observed heterozygotes (53, 43) was less than expected (59, 57). Locus BST6.39TG showed a similar pattern in populations SQ and WA. However, none of the p-values across all four loci were significant for the population samples (SQ, $p\text{-value}=0.0613$; NQ, 0.1633; GOC, 0.1022 and WA, 0.1277) and it was concluded that the genotypes conformed to Hardy-Weinberg proportions. Excess homozygosity (heterozygote deficit) is often observed when microsatellite markers are used on wild populations, and a convincing explanation for the phenomenon has yet to be proposed. For interested readers, Lessios (1992) summarises the principles involved, while Bagley (1999) discusses its relevance to microsatellite loci.

Table 35. Microsatellite loci (above diagonal) that show significant ($p<0.05$) allele frequency variation between population pairs.

The populations are southern Queensland (SQ), northern Queensland (NQ), Gulf of Carpentaria (GOC) and Western Australia (WA). p-values given as superscripts.

	NQ	GOC	WA
SQ	None	BST6.39TG ^{0.00669}	90RTE ^{0.03110}
NQ	-	None	BST2.33 ^{0.00358}
GOC		-	BST2.33 ^{0.00174} , BST6.39TG ^{0.04513}
WA			-

Locus-by-locus exact tests of allelic frequencies revealed some significant differences between populations against a background of general allele frequency homogeneity. Allele frequencies for locus BST2.33 for the WA sample were significantly different from GOC and NQ, but not from the SQ sample. For locus BST6.39TG, the GOC sample was significantly different from the WA and SQ, but not from the NQ sample (Table 35). From the AMOVA of the microsatellite data, a small (0.17%) but significant ($p \sim 0$) proportion of overall variance among the four populations was due to variation between populations ($F_{ST} = 0.00172$). A likely biogeographic barrier (Torres Strait) was used to group the populations into Queensland east coast samples (SQ, NQ) and northern Australian samples (GOC, WA). The amount of overall variation that was attributed to the difference between these two regional groups following AMOVA was small, but significant ($F_{CT} = 0.00080$, $p \sim 0$).

Table 36. Allelic frequencies for four microsatellite loci for mangrove jack populations.

The populations are from southern Queensland (SQ), northern Queensland (NQ), Gulf of Carpentaria (GOC) and Western Australia (WA). Alleles are designated numerically and by their size in base pairs. For locus *BST6.39TG*, four alleles (666, 777, 888 and 999) represent low frequency alleles that were pooled.

Locus : *9ORTE*

Population	Allele designation and size in base pairs									2N
	1	2	3	4	5	6	7	8	9	
	175	183	185	189	191	193	195	197	199	
SQ	0	0	0	0.048	0.246	0.230	0.206	0.183	0.087	126
NQ	0	0.005	0	0.044	0.255	0.304	0.157	0.181	0.054	204
GOC	0.005	0.005	0	0.032	0.204	0.242	0.210	0.247	0.054	186
WA	0	0	0.005	0.025	0.211	0.265	0.176	0.294	0.025	204

Locus : *BST6.56*

Population	Allele designation and size in base pairs										2N
	1	2	3	4	5	6	7	8	9	10	
	129	131	133	135	137	139	145	147	149	151	
SQ	0.008	0.484	0.008	0	0.379	0.073	0.032	0.008	0	0.008	124
NQ	0	0.469	0.010	0	0.428	0.067	0.021	0	0.005	0	194
GOC	0	0.378	0.006	0.011	0.511	0.056	0.028	0.006	0.006	0	180
WA	0	0.393	0.015	0	0.505	0.051	0.010	0	0.020	0.005	196

Locus : *BST2.33*

Population	Allele designation and size in base pairs									2N
	1	2	3	4	5	6	7	8	9	
	179	181	183	191	193	195	197	199	201	
SQ	0	0.049	0	0	0	0.303	0.500	0.139	0.008	122
NQ	0	0.030	0.005	0	0	0.262	0.569	0.134	0	202
GOC	0	0.016	0	0	0	0.291	0.505	0.181	0.005	182
WA	0.006	0.044	0.006	0.039	0.033	0.261	0.472	0.139	0	180

Locus : *BST6.39TG*

Population	Alleles 1 to 12 and size in base pairs											
	1	2	3	4	5	6	7	8	9	10	11	12
	143	145	147	149	151	153	155	157	159	161	163	165
SQ	0.128	0.144	0.032	0.088	0.080	0.072	0.024	0.024	0.008	0.032	0.032	0.008
NQ	0.136	0.119	0.045	0.102	0.085	0.085	0.068	0.040	0.040	0.023	0.040	0.028
GOC	0.080	0.080	0.017	0.109	0.132	0.080	0.040	0.040	0.052	0.040	0.023	0.023
WA	0.139	0.117	0.028	0.111	0.117	0.072	0.061	0.022	0.006	0.039	0.033	0.028

Locus : *BST6.39TG cont.*

Population	Alleles 13 to 24 and size in base pairs												2N
	13	14	15	16	17	18	19	20	21	22	23	24	
	167	169	175	177	179	189	191	193	666	777	888	999	
SQ	0.016	0.008	0.040	0.008	0.040	0.016	0.008	0.008	0.016	0.040	0.040	0.088	126
NQ	0.011	0.028	0.017	0.023	0.040	0.023	0.006	0	0.011	0.006	0.011	0.011	176
GOC	0.040	0.029	0.006	0.023	0.040	0	0	0	0.057	0.023	0.057	0.006	174
WA	0.022	0.017	0.033	0.022	0.022	0.017	0.011	0	0.022	0.022	0.017	0.022	180

MtDNA

In 375 bp at the 5' end of the control region (GenBank accession number AY166828), 108 variable sites were observed, including seven single base pair indels and one five base pair indel across the sequences from 34 mangrove jacks. There were 33 haplotypes, one of which was represented by two fish. Intrapopulation control region sequence divergence was approximately 4 to 7% for the Australian sampling locations (Table 37). Non-Australian sampling locations were similar. Note that these diversity values are biased downwards as PAUP excludes indels during 'distance' analyses.

Table 37. Intrapopulation nucleotide diversity (Kimura's two-parameter) for 5' end of the control region.

The populations were southern Queensland (SQ), northern Queensland (NQ), Gulf of Carpentaria (GOC), Western Australia (WA), Bali (BALI), Java (JAVA), Sumatra (SUM) and Samoa (SA).

Population	Mean	Std Dev
SQ	0.061568	0.028410
NQ	0.047709	0.026498
GOC	0.075441	0.029484
WA	0.060961	0.010808
BALI	0.071664	0.018083
JAVA	0.057340	-
SUM	0.090327	0.010059
SA	0.059053	0.051142

Of the 108 polymorphic nucleotide positions in from the control region, 67 were parsimony informative (present in two of more fish). There were 255 MP trees that were 218 character state changes in length. The topology of the strict consensus tree (Figure 43) did not support the separation of Australian mangrove jack populations into a Queensland east coast versus a northern and western Australian stock. There was also no support for a genetic distinction between Australian and Indo-Pacific mangrove jack; for example, there were few clades (closely related groups) whose membership consisted only of Indonesian fish, or of Australian fish. Furthermore, the three Samoan fish appeared to be as related to fish from Australia as they were to fish from Indonesia and were not members of a unique or distantly-related clade.

Table 38. Numbers of identical ATPase sequences among six mangrove jack haplotypes.

The populations were collected from southern Queensland (SQ), northern Queensland (NQ), Gulf of Carpentaria (GOC), Western Australia (WA), Bali (BALI), Java (JAVA), Sumatra (SUM) and Samoa (SA).

Haplotype	SQ	NQ	GOC	WA	BALI	JAVA	SUM	SA
1	5	9	7	6	2	1	2	
2			1	1	1			
3					1		1	
4	4	1		1		1		1
5	1							1

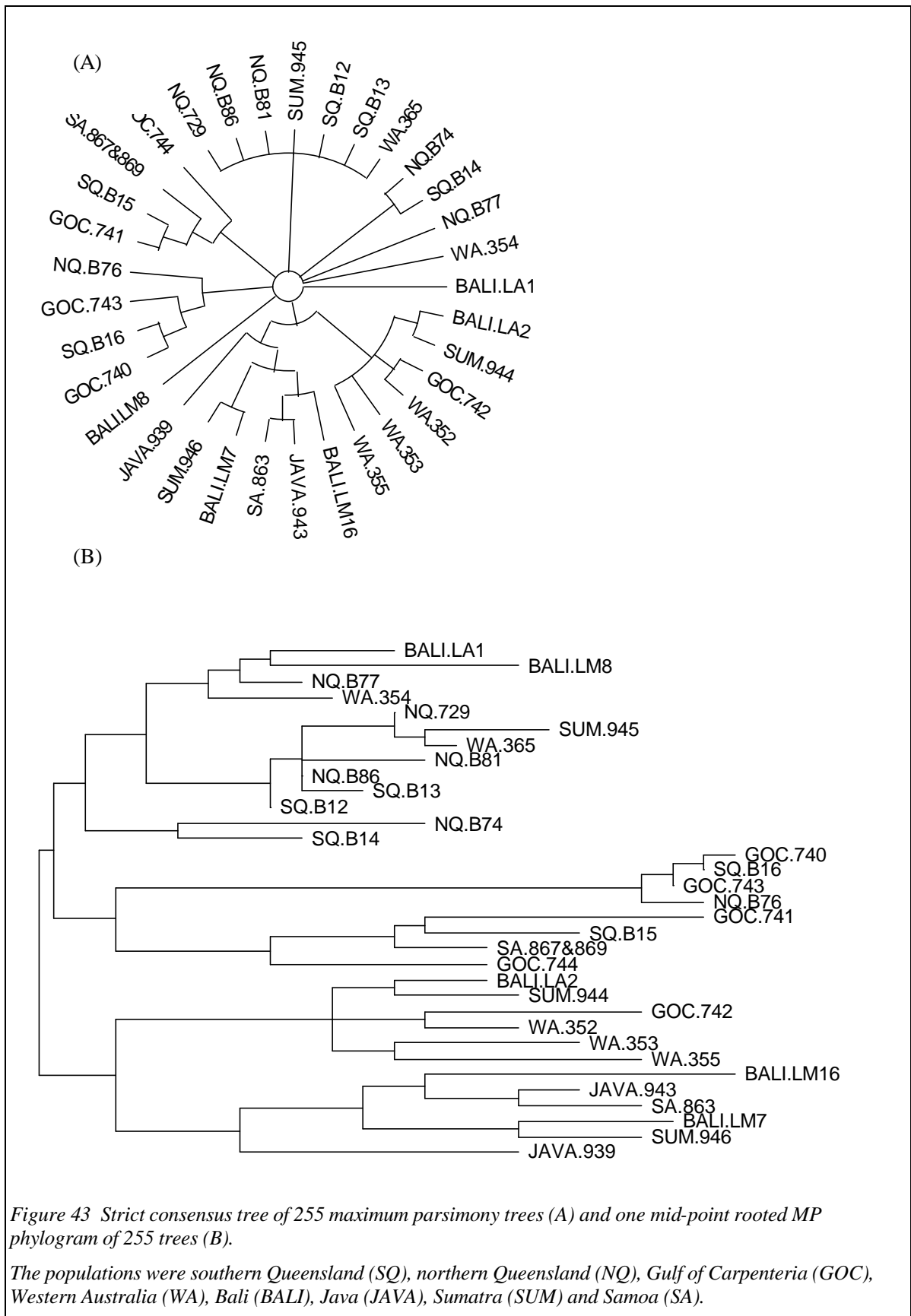


Figure 43 Strict consensus tree of 255 maximum parsimony trees (A) and one mid-point rooted MP phylogram of 255 trees (B).

The populations were southern Queensland (SQ), northern Queensland (NQ), Gulf of Carpentaria (GOC), Western Australia (WA), Bali (BALI), Java (JAVA), Sumatra (SUM) and Samoa (SA).

As expected, sequence variation in the ATPase 6 region was less extensive than the control region sequences. In 415 base pairs, (GenBank accession number AY166827) only 17 variable characters were observed among 56 fish; there were no indels. There were five haplotypes that were represented by more than one fish (Table 38) and eight fish had unique sequences. Haplotypes 1 to 5 represented fish from one or more population samples. Among the 17 polymorphic characters, only two were parsimony informative, leading to a single MP tree with no homoplasy (Figure 44). A pattern of genetic distinctiveness was not apparent among the population samples from the ATPase phylogenetic tree

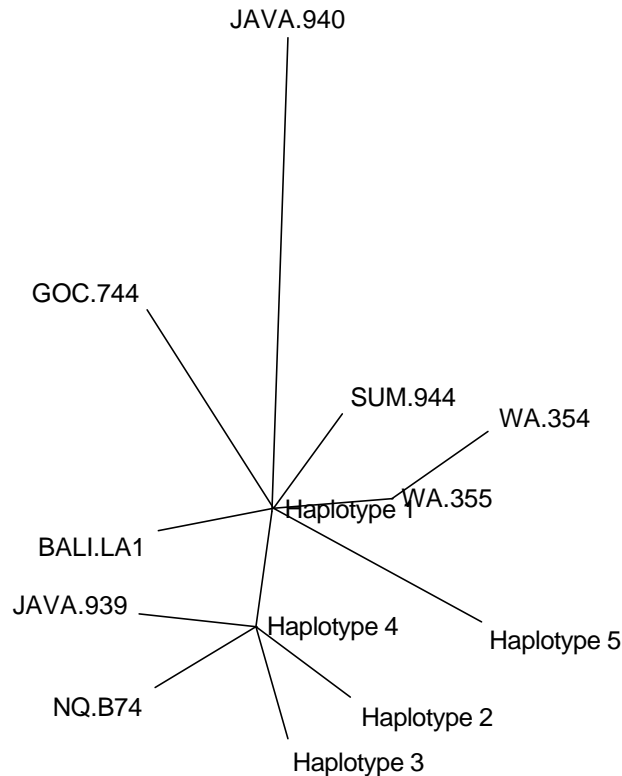


Figure 44. Unrooted maximum parsimony tree constructed from ATPase sequence data.

Each branch represents a fish or group of identical fish grouped into haplotypes (Table 6) [southern Queensland (SQ), northern Queensland (NQ), Gulf of Carpentaria (GOC), Western Australia (WA), Bali (BALI), Java (JAVA), Sumatra (SUM) and Samoa (SA)].

Discussion

Population Genetic Structure

From a genetic perspective, mangrove jack from different geographic areas in Queensland and Western Australia appear to be highly interconnected. Estimates of F_{ST} (microsatellites) were small (0.002) and the topological structure of MP trees from mtDNA sequence data showed no congruence with relationships between geographic areas. A high degree of interconnectedness on such a large scale is not without precedent. Ovenden (1992) surveyed mtDNA sequence variation in red rock lobster populations (*Jasus edwardsii*) over several

thousand kilometres of southern Australian coastline, including Tasmania, and reported an apparent lack of subdivision that was accounted for by extensive dispersal during their long larval phase. The tailor (bluefish, *Pomatomus saltatrix*) has a life-history more similar to mangrove jack. It spawns offshore but juveniles and sub-adults are commonly found in estuaries. Graves (1992) analysed mtDNA in over 400 North American tailor from populations along about 1000 km of mid-Atlantic coastline (North Carolina, Virginia and New Jersey) but reported no significant genetic differentiation. In Australia, Nurthen (1992) found no evidence for stock structure among east-coast tailor samples using nine allozyme loci. They did, however, find significant differences between samples from the western and eastern extremes of their distribution around the southern coastline of Australia. Jackass morwong (*Nemadactylus macropterus*) are distributed in the south from Brisbane in the east to Perth in the west, including Tasmania. The adults are demersal living at 40 to 400m while the juveniles are found on shallow reefs in Bass Strait and around Tasmania (Kailola *et al.*, 1993). Both allozyme (Elliott and Ward, 1994) and mtDNA (Grewe *et al.*, 1994) analyses concluded there was a lack of genetic differentiation among populations sampled throughout the entire range of the species in Australia, but there was significant, albeit minor, evidence of a genetically distinct population in New Zealand. As with other species with little or no genetic subdivision, the implication for mangrove jack populations is that they are connected via gene flow mediated by dispersal.

The high level of gene flow experienced by mangrove jack populations is emphasized by lack of (mtDNA) and minor (microsatellite) differences reported here between tropical eastern and western Australia. The Torres Strait region, in particular, is a major biogeographic boundary between the Pacific and Indian Oceans over which many marine species are genetically subdivided. During the Pleistocene period the wide continental shelf to Australia's north, including the Torres Strait, was exposed joining Papua New Guinea and Australia into a single land mass (Van Oosterzee, 1997). This vicariant event is reflected in the population genetics of at least three tropical Australian fish (*Lutjanus malabaricus*, (Elliott, 1996); *Lates calcarifer*, (Chenoweth *et al.*, 1998a) and *Scomberomorus commerson*, (Ovenden and Street, in prep) and green turtles (*Chelonia mydas*, (Norman *et al.*, 1994). The frequencies of the three most common *L. malabaricus* mtDNA RFLP haplotypes were 61%, 15% and 12% in the east coast of Queensland sample (near Townsville, Qld), but were 27%, 26% and 29% in samples taken from Gulf of Carpentaria and north-west Western Australia. Chenoweth (1998b) reported a net mtDNA sequence divergence of 4% (control region) and 0.47% (ATPase region) between *L. calcarifer* populations sampled to the east and west of Torres Strait. For *S. commerson*, Ovenden (Ovenden and Street, in prep) reported an mtDNA RFLP marker that was present in 20-25% of the fish sampled from the east Queensland coast (near Townsville, Qld), but only present in 2% of the fish sampled from the Gulf of Carpentaria. Similarly, Norman (1994) reported mtDNA clades that were endemic to either Pacific Ocean or Gulf of Carpentaria and Indian Ocean green turtle rookeries. The absence of mtDNA sequence variation between mangrove jacks that were sampled in south-east and northern Queensland compared to the Gulf of Carpentaria and Western Australia emphasizes the high vagility of the species despite the presence of a major biogeographic boundary. Any impact made by the successive formation of a land-bridge between northern Australia and New Guinea on the pattern of population subdivision in this species has presumably been erased by large amounts of larval or adult long-shore movement. Mark recapture data for adult mangrove jack revealed that some individuals are capable of moving several hundred kilometers from coastal habitats where they were tagged to offshore reefs where they were subsequently recaptured (see Movements section).

Evidence from mtDNA sequence data reported here suggests that the vagility of mangrove jack is also high enough to homogenise populations in Australia and near south-east Asian countries. This is in contrast to two recent studies conducted in this laboratory where genetic breaks have been demonstrated between northern Australian and Indonesian populations of snapper and mackerel. Ovenden (in press) reported that 14% of the total molecular variance

among restriction site mtDNA haplotypes for the goldband snapper (*Pristopomoides multidens*) was attributable to genetic distinction between south-east Asian and Australian samples. Indonesian (Kupang, West Timor) and Australian (Kimberley) sampling locales had significantly different haplotype frequencies (haplotype AACAA; Kupang 0.018, Kimberley 0.180; BACBA; Kupang 0.018, Kimberley 0.120 and BBCBA; Kupang 0.333, Kimberley 0.075) despite geographic separation over only 300-400 km. Over the same stretch of the Timor Sea, Spanish mackerel (*S. commerson*) mtDNA control region sequence data revealed that 19% of the observed sequence variation was due to genetic differences between the populations. Further, three of ten polymorphic allozyme loci were significantly different between Kupang and Kimberley (ADH, GPI-A and PGDH) while seven loci differed between Kupang and Queensland east coast samples (ACP, ADH, CK-A, EST-1, GPI-A, sIDHP and PGDH; (Ovenden and Street, in prep). While mtDNA has proven an excellent marker in other species to detect genetic subdivision between northern Australian and south-east Asian populations, no similar pattern is evident for mangrove jack. It would be worthwhile to validate the apparent high level of gene flow across national boundaries in mangrove jack with another genetic marker, such as microsatellites.

In population genetics there are two models to account for the generation of new, mutant alleles; the infinite allele (IAM) and stepwise mutation models (SMM). Assuming equilibrium has been achieved in a population between mutation and genetic drift, the models can be used to estimate expected heterozygosity, gene flow (Nm) and to partition genetic variance within and between populations (Nei and Kumar, 2000). The F_{ST} values presented in this report to partition microsatellite genetic variance assume the IAM; the SMM equivalent (R_{ST}) has not been presented. It is unclear which model is the most appropriate for microsatellite data, and several authors regard the solution to lie between the extremes of IAM and SMM (Bagley *et al.*, 1999; Nei and Kumar, 2000). Several recent applications of microsatellites to spatial differentiation in fisheries species have reported similar results using both F_{ST} and R_{ST} (Ruzzante *et al.*, 1996), (Smith and McVeagh, 2000) and (Waters *et al.*, 2000). Given the concordance between mtDNA and microsatellite results reported here for mangrove jack, it is unlikely that the implementation of R_{ST} would uncover hidden population subdivision.

Implementation of microsatellite loci in non-target species

Instead of developing microsatellite loci in this study that were unique to mangrove jack, we took advantage of the ability of some loci to cross species boundaries. Di-, tri- and tetra-nucleotide perfect and complex repeat loci developed by Van Herwerden (Van Herwerden *et al.*, 2000b; 2000a) for coral trout (*Plectropomus laevis*) and red-throat emperor (*Lethrinus miniatus*) were discarded if (1) PCR product was absent or smeared, (2) locus was monomorphic, (3) allele frequencies were out of Hardy-Weinberg or (4) more than two bands were observed per individual. Although only 11% (4/44) of loci met these criteria, the screening process was judged to be more cost and time efficient than developing, sequencing and optimising a new and specific set of loci for mangrove jack. Our allele size ranges largely agreed with that reported by Van Herwerden (2000b; 2000a); 90RTE, 190bp in *L. miniatus* and 175-199bp in mangrove jack; BST6.56, 121bp and 129-151bp; BST2.33, 230bp and 179-201bp; BST6.39TG, 133bp and 143-193). We did not confirm the nature of the repeat in mangrove jack by direct sequencing. Interestingly, three of the four loci selected for this study were independently found to amplify in a range of Serranid, Lethrinid, Lutjanid and Labrid species (BST2.33 and 90RTE (Van Herwerden *et al.*, 2000b) and in *Scomberomorus commerson* (90RTE and BST6.39TG, Ovenden, Broderick and Street, unpublished results). We endorse the approach of some journals to encourage authors of primer notes to report cross-species amplification, and anticipate the future where implementation of loci on novel species will be simplified by accumulation of such information.

Adaptive population divergence

Adaptive population divergence is the evolutionary response of a population to its environment. Its magnitude depends on the heritability of the adaptive trait and the strength of the selective pressure exerted on the trait by the environment. Adaptive traits, unlike molecular markers (traits) are rarely controlled by a single genetic locus, rarely exhibit discrete phenotypes and are subject to complex interactions such as dominance, epistasis and pleiotropy. Yet these traits make up the genetic architecture that confers on populations the unique characteristics that are perceived to be worthy of conservation. A recent meta-analysis (Reed and Frankham, 2001) has contributed to the controversy surrounding the extent to which variation in neutral molecular markers is positively correlated with variation in adaptive traits. Among 71 datasets, they found the correlation to be weak and concluded that the molecular measures of genetic diversity had only very limited ability to predict diversity in adaptive traits. Molecular markers are used extensively in conservation biology to rapidly, cheaply and non-invasively measure genetic divergence, but the study of Reed (2001) raises concerns over whether they should be used to define evolutionarily significant units (ESUs (Moritz, 1994) that may be worthy of special conservation status.

ESUs and their evolutionary potential are preserved in an effort to halt, and possibly, recreate lost biodiversity assuming evolutionary processes continue to operate. Despite their lack of correlation with adaptive traits, molecular markers have been successful in the identification of ESUs, provided as much ecological information as possible is included in the definition of the ESU (McKay and Latta, 2002). It is the use of molecular markers for more short-term actions, such as intentional translocations for economic or social benefit where the addition of ecological data is not only desirable, but essential. At this scale, ecological and climatic gradients become as relevant for assessing differentiation in population adaptive divergence as molecular markers (McKay and Latta, 2002). Precise matching between donor and recipient populations for presumed adaptive characteristics is an important precursor for success. This study was conducted to define the molecular boundaries of ESUs for mangrove jack in Queensland. Unless distinct ecological subunits are discovered within the range of mangrove jack in Queensland, the definition of an ESU for this species includes the entire Queensland population. However, no latitudinal gradients in abundance were reported for mangrove jack in seven streams along 320 kilometres of northern Queensland coastline (see Movements section). In circumstances where intentional translocations were intended to establish or supplement populations more precise ecological matching between donor and recipient populations is recommended.

Conclusions and recommendations

- Using molecular markers, and in the absence of conflicting ecological data or further genetic data, we regard Queensland populations of mangrove jack to belong to a single ESU.
- The addition of ecological information to the definition the Queensland mangrove jack ESU is desirable, particularly in light of the large ecological gradient experienced by the species in Queensland.
- Translocations for establishing *de novo* populations in land-locked freshwater impoundments in Queensland are unlikely to adversely affect population genetic structure of the species. We recommend the use of locally derived broodstock, where possible, to achieve maximum stocking success and to minimize potential disruption to locally adapted sub-populations.
- The probable existence of a single genetic ESU in eastern Queensland and the Gulf of Carpentaria and biological data suggesting both extensive dispersal of adults and eggs

and larvae and a substantial mixing of spawning fish from different river systems suggests that limited riverine translocations for stock enhancement purposes may be acceptable under some conditions. These conditions would be that donor and recipient populations are from the same region (eg. Gulf of Carpentaria, north, central or south-east Queensland). However, should circumstances arise where stocking is required between, rather than within regions, then caution needs to be applied and further genetic and ecological data should be collected to further refine the definition of the ESU.

Benefits

Benefits and beneficiaries

The outcomes from this project will benefit the management of the mangrove jack fishery in a number of ways. The biological information on mangrove jack, including population parameters and yield estimates, provides pertinent information on the present status of the stocks and the relevance of current management arrangements. The information in the report will be valuable for future stock assessments and long term monitoring of the fishery for this species. The methodologies developed for sampling mangrove jack in upper tidal and freshwater areas will be particularly useful for future monitoring of inshore mangrove jack stocks. The beneficiaries of this information are primarily the recreational and reef-line commercial fishing industries and fisheries and natural resources managers in Queensland, the Northern Territory and Western Australia.

In addition, the work on the genetic stock structure of mangrove jack in Queensland will provide the basis for the development of a translocation and stocking policy. This policy will facilitate the establishment of more 'put and take' mangrove jack fisheries in impoundments through the State. Quantifying the actual benefits of a mangrove jack 'put and take' recreational fishery is difficult however there has been a previous economic assessment of a barramundi impoundment fisheries (Rutledge *et al.*, 1990). This study estimated that for every dollar spent on enhancement generates \$31 of economic benefit for the Queensland economy. The primary beneficiaries are recreational fishers but past experience with impoundment barramundi fisheries has shown that there are flow-on benefits to many sectors of the community including the tourist industry, fishing guides, fish stocking groups and small businesses particularly sports and tackle retailers and other suppliers of fishing and boating gear.

The project outcomes and outputs may also benefit the emerging aquaculture industry for this species. For example, the development of a stocking and translocation policy should provide clear guidelines on origin of broodstock and determining the geographic areas where their progeny are permitted to be stocked.

As well as the recreational and reef-line commercial fisheries, another major beneficiary of this study is the environment. The project identified the importance of both complex structural habitat and coastal nursery swamps to mangrove jack. This information has provided direction to community ICM and Landcare groups on issues such as the importance of minimising coastal development and destruction of riparian forests. It also highlights the importance of the establishment of marine protected areas (eg. Fisheries Habitat Areas) to preserve critical mangrove jack juvenile habitat in the coastal zone.

Most of the benefits and beneficiaries listed above were identified in the original application.

Intellectual property and valuable information

No patentable inventions or processes have been developed during this project.

Dissemination of research results

Presentation of research results has been made to a wide variety of audiences including. During the project, preliminary results were presented to a wide range of audiences through:

- radio and television interviews including
 - WIN News, December 1999
 - ABC Radio Townsville, October, 2000
 - ABC Radio Far North *Fishtalk* program.
 - ABC Radio National, August 2002
 - ABC Radio Townsville, October 2002
- regular newsletters that were mailed or emailed to wide range of individuals and organisations. The newsletters were also published on the internet at www.dpi.qld.gov.au/mangrovejacknews/.
- Scientific papers presented to peers at national and international conferences, for example:
 - Australian Society of Fish Biology annual conference, 14-17 August, 2002.
 - 3rd World Recreational Fishing Conference, 21-24 May 2002.
- Presentations to industry bodies including
 - Australian National Sportsfishing Association Annual Conference, Yeppoon, 2000, 2001 and 2002.
 - Cooktown Tag and Release Tournament, October 1999, 2000, 2002.
 - Hinchinbrook Tag and Release Tournament, May 1999 and 2000.
 - Cairns Sportfishing Club
- Publication of results and background information on mangrove jack on the internet at www.dpi.qld.gov.au/fishweb/7096.html.
- Articles in popular fishing magazines and newspapers including:
 - *Cairns Post*, December 1999.
 - *The Courier Mail*, March 2000, November and December 2001, July 2002.
 - *Modern Fishing*, *Fish and Boat*, *Queensland Fishing Monthly*, *Fishing World* and *Fish'n'Boats'n'Bits*.
 - Regional Queensland newspapers, March 2000, November and December 2001, July 2002

The results chapters in this report will form the basis of scientific papers that will be submitted to peer-review journals.

Further Development

There are a wide range of activities that can be undertaken to further build on the outcomes and outputs of this project. These include:

- Examining the efficiencies of different types of fish way designs in facilitating the upstream passage of mangrove jack.
- Identify streams where fish barriers such as weirs and dams prevent the upstream passage of mangrove jack so that future remedial works can be undertaken.
- Obtaining more biological information from mangrove jack resident in other parts of Australia. While samples were collected from throughout tropical Australia during this project, from a management point of view it would be useful to collect more specific information about the growth rates and population parameters of mangrove jack in the Northern Territory and Western Australia. This is being partially addressed through the current FRDC project *Biological parameters for managing the fisheries of blue and king threadfin salmons, Malabar cod and mangrove jack in north-western Australia* (project number 2002/003).
- Obtain additional ecological information to assist in the definition the Queensland mangrove jack ESU. This is important, particularly in light of the large latitudinal gradient experienced by the species in Queensland.
- More work needs to be done to delineate the wider stock structure of mangrove jack. Genetic samples from other parts of the range of mangrove jack, particularly east Africa and the northern Indian Ocean would assist to determine if fish from these areas are the same genetic stock.
- Promote the importance of complex habitat such as snags and the sources of that habitat including mangrove and riparian forests to community organisations such as Integrated Catchment Management and Landcare groups, River Improvement Trusts and local government. The community needs to be educated that coastal development and destruction of riparian forests will adversely impact on mangrove jack populations. Giving presentations to community and industry groups and targeted media releases on the results of this project are assisting to reinforce these concepts. This process has already commenced.
- The development of a translocation and stocking policy for mangrove jack in Queensland. The information on the genetic stock structure of mangrove jack in Queensland presented in this report will assist in the development of this policy.
- Identification of specific mangrove jack nursery swamps with a view to protecting critical areas as Fish Habitat Areas. This is an important issue in areas where coastal development is occurring and
- Continued monitoring of mangrove jack stocks in Queensland as part of the long term monitoring program. Currently mangrove jack stocks do not appear to be heavily exploited but this may quickly change and a monitoring program to give early warning of pending problems is desirable. The Queensland Department of Primary Industries' Long

Term Monitoring Program is now using some of the sites and the techniques developed during this current program. While this monitoring will not be on as regular a basis as occurred during the research sampling, it will assist in providing a valuable insight into the health of mangrove jack stocks in Queensland. The impacts of fish trawls, which are now being tested in exploratory fisheries in the Gulf of Carpentaria, need to be closely monitored.

- A socio economic evaluation of the recreational, both stocked and wild, mangrove jack fisheries in Queensland would be desirable. Issues like the value of stocked mangrove jack to the community and attitudes to catch and release would be desirable outcomes of such a survey.
- Refinement of the commercial log book program to give more accurate statistics on offshore mangrove jack catches is desirable.
- Catch and release of fish, including mangrove jack, is widely practiced by recreational anglers. There are a number of relevant issues pertaining to catch and release including tagging mortality and tag shedding. The controlled tagging experiments conducted during this research project suggested that tag mortality was minimal in this species, however the proposed FRDC project on hooking mortality may provide more information under field conditions. The experiments also demonstrated that tag shedding can be high and it may be appropriate to train recreational anglers who are practicing tag and release of mangrove jack in the most appropriate tagging technique for this species to minimise tag loss.
- Recruitment of mangrove jack into inshore areas may be variable from year to year. Monitoring levels of juvenile recruitment into selected rivers may assist in predicting what fishing will be like in those estuaries in future years.

Planned Outcomes

The principal outcome that this project has delivered is a major contribution towards the ecologically sustainable management of mangrove jack in Australia and also to improved public awareness and knowledge of fishery management issues. Specific outputs that have contributed to this key outcome are:

- Enhanced knowledge of the complex, bi-phasic life cycle of mangrove jack populations including movements, reproduction, age and growth, habitat preferences and recruitment that will be the basis for developing future management strategies for the species.
- Estimation of population parameters, including fishing and natural mortality, which in turn, have led to the development yield-per-recruit models for the fishery. These models suggest that the current minimum legal size for the species, while conservative, is appropriate.
- Confirmation of the importance of structural habitat to coastal mangrove stocks and recognition that inappropriate anthropogenic activities including reclamation of coastal wetlands and clearing of riparian forest could have adverse impacts on the fishery.
- The probable existence of a single genetic ESU in eastern Queensland and the Gulf of Carpentaria has implications for the development of a translocation policy for this species. Impoundment stockings of mangrove jack are unlikely to adversely impact on genetic population structure and limited riverine translocations, provided donor and recipient populations are from the same region, should be acceptable.
- Collaboration and close contact between researchers and industry during the project and dissemination of information through quarterly newsletters, newspaper articles, a web site and numerous presentations to industry bodies provided commercial and recreational fishers with and increased understanding of the lifecycle of mangrove jack and the impact of fishing on this species.

Conclusions

Objective 1. Investigate the biology of mangrove jack in coastal rivers including habitat preferences, mortality, reproduction, recruitment, and movements;

This study has contributed significantly towards the ecological sustainable management of the mangrove jack fishery in Queensland by providing detailed information on the biology and life history of the species, its fishery and other anthropogenic influences affecting populations. Specifically, major outputs include:

Biology and life history

L. argentimaculatus has a complex life history involving both inshore and offshore phases. Juvenile mangrove jack were found in inshore habitats while mostly mature fish were sampled offshore. As well as moving from inshore areas to adjacent offshore reefs, mangrove jack are capable of making coastal movements in excess of 300 km and can move into water of depths of 120 m or more.

Fish begin to move offshore at an age of 2 years, with fish fully recruited to offshore habitats by age 11+. Mangrove jack is a long-lived species, with the maximum age within the population possibly in excess of 40 years.

The growth rate of mangrove jack appears to vary between geographic zones. Fish resident in southern Queensland appear to reach the current legal size (350mm TL) at a faster rate than fish from the northern zones. Growth rates slow substantially after the fish mature. Mangrove jack older than two years appear to grow at approximately 25 – 50 mm/ year. In their first two years (0+ and 1+), the growth rate of impoundment fish tends to be at least twice the growth of wild fish.

In Queensland, the spawning season of mangrove jack is over an extended period from about October to March with peak spawning between November and January inclusive.

Males tend to mature before females. The smallest mature male fish was 3 years, however most male mangrove jack did not mature before they were about 7+ years old. Female fish matured from 5+ years, with most fish specimens not maturing until they were 8+ years.

Mangrove jack are a relatively fecund species capable of releasing up to four million eggs in a single spawning.

Post larvae and juvenile fish are recruited into inshore areas during the first half of the year from about a size of about 20 mm LCF and an age of 32 days. They can remain in rivers to a maximum age of about 11 years but begin to recruit to offshore areas at about 2+ years old.

In rivers, mangrove jack are likely to be associated with complex structural habitat. Juvenile fish of less than 100 mm LCF are more likely to be caught on or near rocky habitat while larger fish are caught more often in snags.

In rivers, the size of mangrove jack generally increases with increasing distance from the river mouth.

Fishery structure and exploitation

Most of the fish harvested from the inshore fishery and conversely, are immature while most of the fish caught offshore are mature.

The preliminary data suggests that offshore stocks of mangrove jack in Queensland are only lightly exploited.

- Yield per recruit modelling of mangrove jack suggests that the current minimum legal size, while conservative, is not inappropriate from a yield perspective.

Anthropogenic impacts on habitat

- The results of this study also emphasised the importance of quality habitat to the continued viability of this fishery.
- Inappropriate clearing of coastal wetlands and riparian vegetation could pose a serious threat to mangrove jack stocks.
- The ongoing declaration of critical coastal wetlands as marine protected areas will assist to protect mangrove jack habitat. In particular, identifying important mangrove jack nursery areas and then targeting them for protection (eg. as Fish Habitat Areas) will assist to ensure future health of stocks.
- Physical barriers like tidal barrages are capable of blocking riverine movements of mangrove jack. It is important that tidal barrages in rivers where mangrove jack are found have appropriate fishways incorporated into their structure.

As well as management, industry and community need to be engaged to ensure the continued viability of the mangrove jack stocks. During the life of this project, we developed a close partnership with industry, particularly recreational fishers, and this has greatly assisted in promoting awareness of management issues related to the mangrove jack resource. In promoting importance of riparian habitat and coastal wetlands as fish habitat for mangrove jack, we have targeted Integrated Catchment Management and Landcare groups and communication with such organisations needs to continue. There is a need for ongoing monitoring of mangrove jack stocks, particularly in coastal river systems. This will be undertaken as part of the Queensland Department of Primary Industries' Long Term Monitoring Program using the some of the sites and the techniques developed during this current program. While this monitoring will not be on as regular a basis as occurred during the research sampling, it will provide a valuable and continuing insight into the health of mangrove jack stocks in Queensland.

Objective 2. Determine the genetic stock structure of mangrove jack in Queensland and other parts of their range

An outcome from the genetic stock structure work will be to provide information relevant to the development of a mangrove jack stocking and translocation policy. The following recommendations should be considered in the development of such a policy:

- Using molecular markers, and in the absence of conflicting ecological data or further genetic data, we regard Queensland populations of mangrove jack to belong to a single ESU.

- The addition of ecological information to the definition the Queensland mangrove jack ESU is desirable, particularly in light of the large ecological gradient experienced by the species in Queensland.
- Translocations for establishing *de novo* populations in land-locked freshwater impoundments in Queensland are unlikely to adversely affect population genetic structure of the species. We recommend the use of locally derived broodstock, where possible, to achieve maximum stocking success and to minimize potential disruption to locally adapted sub-populations.
- The probable existence of a single genetic ESU in eastern Queensland and the Gulf of Carpentaria and biological data suggesting both extensive dispersal of adults and eggs and larvae and a substantial mixing of spawning fish from different river systems suggests that limited riverine translocations for stock enhancement purposes may be acceptable under some conditions. These conditions would be that donor and recipient populations are from the same region (eg. Gulf of Carpentaria, north, central or south-east Queensland). However, should circumstances arise where stocking is required between, rather than within regions, then caution needs to be applied and further genetic and ecological data should be collected to further refine the definition of the ESU.

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Appendix 1

Project Staff

<i>Name</i>	<i>Position</i>
John Russell	Principal Fisheries Biologist & Principal Investigator
Andrew McDougall	Fisheries Biologist
Adam Fletcher	Fisheries Technician
Jenny Ovenden	Senior Fisheries Biologist
Raewyn Street	Fisheries Technician

Appendix 2

Habitat assessment datasheet

SITE NO	DATE	AMG COORDS	ZONE	pH
STREAM NAME		STREAM ORDER	SITE LENGTH	
AIR TEMP.	WATER TEMP.	% OXYGEN SAT.	TURBID(NTU)	
DISTURBANCE RATING				
AQUATIC VEG. SPECIES				
<u>STREAM STRUCTURE</u>				
<u>LEFT BANK</u>		<u>RIGHT BANK</u>	<u>POOLS/DEEP</u>	<u>RIFFLES / SHALLOW</u>
WIDTH	CONTINUITY	% TREES / SHRUBS	TOTAL LENGTH	MAX. DEPTH
% GRASSES	% OTHER	% NO VEG	AV. DEPTH	BANK FULL DEPTH
% GRASSES	% OTHER	% NO VEG	BANK FULL WIDTH	MAX. WIDTH
% NO VEG	% NO VEG	% NO VEG	MAX. WIDTH	AV. WIDTH
<u>COVER</u>		<u>SEDIMENTATION</u>	<u>BOULDER/COBBLE</u>	<u>COBBLE/GRAVEL</u>
BANK COVER (m)	OVERHANGING VEGETATION (m)	INVASIVE GRASSES	SAND	FINE MATERIAL
AQUATIC MACROPHYTES (m ²)	TOTAL NUMBER OF SNAGS	SPECIES		
		TOTAL LENGTH		
		MAX. WIDTH		
		P	R	
		R		
COMMENTS				

Index of disturbance ratings

1. Extreme disturbance

Valley flat: Crops or pasture on both sides, sparse or no riparian tree buffer on either side.

Banks/stream: Channellised; or weeds or grasses choking watercourse; or extensive trampling by cattle; or evidence of discharges; or stagnant water with significant decaying organics; and no canopy cover in small streams.

2. High disturbance

Valley flat: Crops or pasture on both sides but a limited riparian buffer of grasses, shrubs and some trees present on one or both sides. Trees less than 25% of total riparian vegetation.

Banks/stream: Exotic grasses or weeds extend into the channel; tree canopy shading only part of smaller creeks. Where there is adjacent pasture, cattle have unlimited access to the riparian zone.

3. Moderate disturbance

Valley flat: Agricultural land on one or both sides but limited riparian buffer of grasses, shrubs and some trees present on one side and treed riparian buffer on other side; or trees at least 25% - 50% of riparian vegetation.

Banks/stream: Banks well treed on at least one side providing adequate canopy; in smaller streams this may influence the other bank; exotic grasses and weeds may intrude into the stream; feral and domestic animals may damage stream bed and banks.

4. Low disturbance

Valley flat: Agricultural land present on one or both sides but functional treed riparian buffer of less than 30 m on both sides. Where pastures are present, they are fenced, preventing stock access to the stream.

Banks/stream: Exotic grasses limited to stream edge; banks well treed providing a substantial canopy for smaller streams; only occasional evidence of disturbances in the riparian zone by feral animals or cattle.

5. Undisturbed

Valley flat: Riparian vegetation undisturbed for at least 30 m on either side of bank; no evidence of disturbance by feral animals.

Habitat assessment explanatory notes

Riparian vegetation continuity

- a. Without breaks in the native riparian vegetation
- b. Breaks few, narrow, and less than 25% of total length
- c. Many breaks, narrow and less than 50% of total bank length
- d. Length of breaks exceed that of native riparian vegetation

Riparian composition

Tree/shrubs- All tree and shrub species

Grasses –All grasses including invasive and exotic species

Bare- Mimimum vegetation cover (<10%)

Other- Rocks, organic debris etc.

Stream structure

Sedimentation

- a. No apparent unstabilised material in channel.
- b. Traces of unstabilised silt, sand, or gravel in quiet areas.
- c. Quiet areas covered by unstabilised materials, deep pools restricted to areas of greatest scour.
- d. Pools shallow, filled with silt, sand or gravel; riffles contain noticeable silt deposits.
- e. Streambed covered with varying degrees of transported material; substrates relatively uniform along stream length.
- f. Stream channel nearly or completely filled with unconsolidated, transported material.

Substrate type

Boulder / cobble	> 25 mm in size
Cobble / gravel	2 - 25 mm
Sand	0.0625 - 2 mm
Fine material (Silt)	< 0.0625 mm

Flow types

Pools / deep areas	slow flowing, laminar flow, > 1 metre deep
Riffles / shallow areas	faster flow, rippled surface, < 1 metre deep

Instream cover

Bank cover	total length (left and right bank) of steep or undercut banks or root systems or rocks.
Overhanging vegetation	total length (left and right bank) of branches hanging in or just above the water surface.
Aquatic macrophytes	total area (m ²) of floating and submerged plants, filamentous algae and reeds.
Snags	total number of woody debris such as large branches and trees in the streambed.

Appendix 3

Otolith microchemical analysis

An assessment of the use of otolith microchemical analysis for identifying ontogenetic migrations of mangrove jack *Lutjanus argentimaculatus* from estuarine nursery habitats to offshore waters of the Great Barrier Reef.

James F Aumend

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Introduction

In recent years, many studies have demonstrated the utility of the microchemical analysis of otoliths for identifying the movement of fishes among locations or habitats. This field of research is based on the permanency of incorporation of trace elements from the surrounding water into otoliths as they accrete (Campana and Nielsen 1985), an attribute of otolith growth that provides the mechanism by which otoliths can record elemental and isotopic 'fingerprints' or 'signatures' distinctive to the locations or habitats in which fishes have resided (Campana *et al.* 1995, Thorrold *et al.* 1998a,b, Patterson *et al.* 2001). The majority of such studies undertaken to date have reconstructed the movements of anadromous fishes between fresh and marine waters on the basis of changes in otolith Sr concentration which is lower in freshwater owing to its lower water concentration (eg. Secor 1992, Rieman *et al.* 1994). However the movements of wholly marine fishes have also been identified using other elements or otolith fingerprints comprising several elements (Campana *et al.* 1999, Thorrold *et al.* 1997).

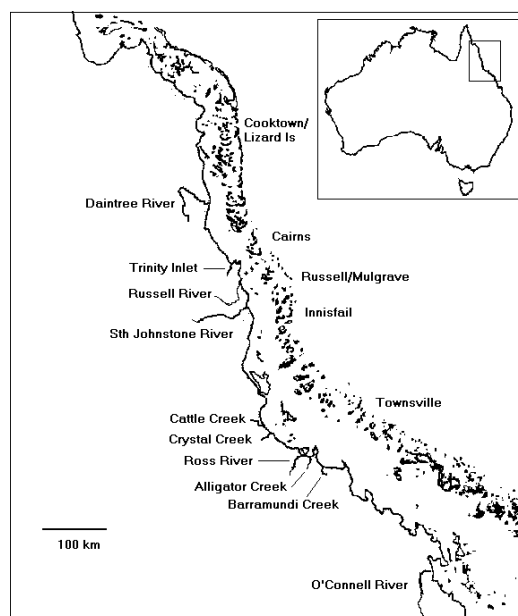
The assessment of differences in trace-element composition between the inner juvenile 'core' and the outer 'adult' regions of otoliths has also shown considerable promise as a technique for retrospectively identifying the nursery grounds of adult fishes (Gillanders and Kingsford 1996, Milton *et al.* 1997, Thorrold *et al.* 2001). A number of other studies have identified strong differences in the otolith composition of juvenile fish residing in different coastal nursery grounds (Thorrold *et al.* 1998a,b, Gillanders and Kingsford 2000, Patterson *et al.* 2001, Gillanders 2002). These authors have thus suggested that it may subsequently be possible to retrospectively identify the nursery of origin of adult fish captured further offshore. In a recent study which demonstrated

the potential of otolith microchemical analysis for nursery identification, Thorrold *et al.* (2001) quantified for the first time in a wholly marine fish, rates of homing from offshore waters to natal estuaries for spawning by 2 year old weakfish *Cynoscion regalis* in the eastern USA. Homing rates were determined on the basis of the otolith composition of juveniles of the same cohort collected 2 years previously. Clearly, the analysis of otolith microchemistry shows considerable promise as a technique for retrospective nursery identification, and for potentially answering other questions relating to movement and migration in fisheries biology that have, as yet, proven difficult to address by more conventional means.

This report presents the results of microchemical analysis of otoliths of mangrove jack *Lutjanus argentimaculatus* from waters of the Great Barrier Reef (GBR) and the adjacent coast. The study was undertaken as a preliminary assessment of the viability of more widely applying this technique to identify ontogenetic migrations of this and other *Lutjanus* spp. from coastal nursery habitats to adult grounds in offshore GBR waters. The specific aspects of study undertaken in this research included: (1) determining whether juvenile mangrove jack have otolith trace element compositions that are distinctive to particular estuaries or coastal areas in tropical north Queensland and which might thus facilitate retrospective nursery identification; (2) examination of patterns of microchemical change along otolith growth axes of offshore fish that might reflect a juvenile life spent in coastal waters prior to a migration offshore; and (3) comparison of the microchemical composition of the juvenile 'core' of otoliths of offshore fish and otoliths of the coastal juveniles to assess patterns of similarity that might suggest migration pathways and the relative contribution of particular coastal areas to adult populations in different offshore areas.

Methods

The assessment of spatial variability in coastal juvenile otolith composition was conducted using sagittal otoliths of juvenile mangrove jack from each of 10 estuaries along the North Queensland coast (Fig 1). Otoliths of 10 fish were analysed for most estuaries, however otoliths of only 8 fish from Trinity Inlet and 7 from Barramundi Creek were available. For 5 estuaries (Daintree R, Russell R, Sth Johnstone R, Crystal Ck, O'Connell R), fish were primarily 200-250mm fork length, and were collected in Feb/Mar 2001 by QDPI staff using electro-fishing equipment. A few smaller and larger fish were included in these 5 samples but were only marginally outside this size range. The only statistically significant difference between these estuaries in fork length and otolith weight was between Crystal Ck and the O'Connell River with fish from the later estuary being slightly smaller and having lighter otoliths than those from Crystal Ck. For the five estuaries not sampled for 200-250mm fish in Feb/Mar 2001, fish varied in size, but were mostly ~350-400mm fork length and were collected at different times over the preceding 3 years by line-fishing, and from donations of filleted fish frames by recreational fishers.



*Figure 1 Map of sampling region showing the ten estuaries and five offshore areas of the Great Barrier Reef from which otoliths of mangrove jack *L. argentimaculatus* were collected for microchemical analysis.*

Otoliths of estuarine fish were cleaned, dried and stored following rigorous decontamination and contamination avoidance procedures. One otolith of each pair was digested in analytical grade HNO₃, and analysed by solution-based inductively-coupled plasma mass spectrometry (SB-ICPMS) at the Advanced Analytical Centre, James Cook University. As preliminary analysis showed that, of a suite of divalent metals, only magnesium (Mg), manganese (Mn), strontium (Sr) and barium (Ba) were present in concentrations reliably above the detection limits of the ICPMS instrument, these four elements and calcium Ca were assayed in the analysis sequence. A 1:100 dilution of otolith acid digests (1% acid) was used in the assay of Mg, Mn and Ba and matrix-matched standard additions used for instrument calibration. External calibration and 1:1000 dilutions were used in the assay of Ca and Sr owing to their high concentrations in otoliths, and the rapid accumulation of Ca on sampling and skimmer cones and consequent decline in sensitivity of the ICPMS instrument when using 1:100 dilutions. Other elements that are known to occur in otoliths in comparably high concentrations such as Na, P, K, Cl and S were not considered owing to their high degree of physiological regulation, a factor that limits their likely use as environmental indicators (Campana 1999). Sample sequence was block randomized by estuary such that one random sample from each estuary was analysed in turn. Ca concentration was quantified as a reference to which concentrations of other elements were standardised, and elemental concentrations were converted to molar ratios to Ca. Standardising to Ca facilitated comparison with the results for offshore fish. Instrument drift was accounted for by including a 100ppb rhodium Rh internal standard in otolith sample solutions, and adjusting elemental concentrations for the relative deviation of the measured of Rh concentration in each sample from its known 100ppb concentration.

To examine patterns of change in otolith microchemistry that might reflect the ontogenetic coast to reef migration of *L. argentimaculatus*, 50 sectioned sagittal otoliths comprising 9-11 fish from each of 5 offshore reef areas were analysed by laser-ablation-ICPMS (Fig 1). In this study, otolith material along growth axes was ablated by an ArF eximer laser and the vapourised material entrained in an argon/helium gas stream for assay by ICPMS. As in the analysis of estuarine fish, LA-ICPMS analysis assayed Mg, Ca, Mn, Sr and Ba concentrations, and data were standardized to Ca concentration. This is necessary in LA-ICPMS as the amount of otolith material ablated may vary among laser pulses, precluding the standardising of elemental concentrations to otolith weight as is possible in SB-ICPMS. In preparation for analysis, otoliths of offshore fish were sectioned transversely with a low speed diamond wafering saw, polished and thoroughly cleaned to remove possible contamination introduced during the sectioning process. Prior to the analysis transect in which a 65 μ m laser beam width was used, a 364 μ m beam width pre-ablation transect was made along the axis to be analysed to further remove surface contaminants and expose a clean surface for analysis. Calibration of the ICPMS instrument was achieved by ablating a NBS 612 glass standard, and drift accounted for reanalyzing the glass standard prior to and after each analysis run of 6 otolith sections and interpolating a linear mass-response drift (6 sections were mounted on each slide with a transect distance for each section of ~10mm). Continuous LA-ICPMS transects in which pulses of the laser beam overlapped were made from the dorsal to ventral edges of each otolith section, and passed through the nucleus such that a continuous record of microchemical change during the life of the individual was obtained. As this approach meant that two growth axes were analysed either side of the nucleus, it was thus possible to assess the reliability of assay data by comparing results from both growth axes. LA-ICPMS analysis was conducted at the Research School of Earth Sciences, Australian National University.

To correlate elemental concentrations measured by LA-ICPMS with the age of fish at the time of incorporation in the otolith, sections were digitally photographed and annulus distances measured manually using image analysis software. Annulus distances could be matched with otolith composition at different distances along laser transects as the sample stage in the laser ablation cell was mounted on a track attached to a DC motor and moved at a constant rate beneath the laser beam. Mean Mn/Ca Sr/Ca and Ba/Ca molar ratios over the first one, two and three years of life, and the last two years of life were calculated from LA-ICPMS transect data to broadly compare otolith composition during putative coastal and offshore life, and with estuarine juveniles. The age at which changes in otolith microchemistry indicated first movement offshore was also identified for each fish.

Statistical methods employed to examine otolith composition data included univariate analyses of variance (ANOVA) of individual element/Ca ratios, and multivariate classification techniques that examined otolith composition as an integrated multi-elemental 'fingerprint'. As both SB-ICPMS and LA-ICPMS analysis of otoliths of several *Lutjanus* spp. in the wider research aspects of this project showed that otolith Mg concentration appears to be strongly related to physiological processes, it was excluded from statistical analysis as it is unlikely to reflect environmental differences. Hence multi-elemental fingerprints included Mn/Ca, Sr/Ca and Ba/Ca ratios. Quadratic discriminant function analysis (QDFA) using a jack-knife cross-validation technique was used to assess the ability to predict the estuary of capture of juvenile *L. argentimaculatus*, and the coastal or offshore origin of different species on the basis of their otolith fingerprints. QDFA was also used to classify offshore *L. argentimaculatus* to estuaries using discriminant functions derived from estuarine fish. Otolith element/Ca ratios were log transformed to meet the assumptions of ANOVA. Although variance of Mn/Ca data remained heterogeneous despite transformation, ANOVA was still employed as it is robust to moderate heterogeneity if sample sizes are not very small or unbalanced.

Although temporal variability and ontogenetic effects are likely to influence spatial patterns in otolith composition and confound comparisons of fish of different age, larger fish from the 5 estuaries other than those sampled in Feb/Mar 2001 for 200-250mm fish were still included in this study to provide a broader geographic assessment of estuarine juvenile otolith composition. However, as the primary assessment of spatial variability in estuarine otolith composition, analyses were performed separately including only the 5 estuaries sampled in Feb/Mar 2001 for fish of 200-250mm. In statistical analyses including fish from all 10 estuaries, the effect of otolith weight (as a proxy for age) was removed from Ba/Ca and Mn/Ca data as these ratios showed significant inverse relationships with otolith weight, and as LA-ICPMS transects generally showed a trend of decline in these ratios during juvenile life. This was done by pooling all estuarine samples and subtracting the slope of the relationship multiplied by otolith weight from Ba/Ca and Mn/Ca data. Although variability in otolith composition between years may also have an influence in spatial comparisons including fish from all 10 estuaries, it was considered prudent to remove the effect of otolith weight to at least broadly account for apparent ontogenetic effects and permit a more valid comparison. As such, the results of analyses including all 10 estuaries should be interpreted with caution and with consideration of possible temporal variability and the detrending of Ba/Ca in Mn/Ca data to account for the inclusion of fish of different ages from different estuaries. As Ba/Ca and Mn/Ca vs otolith weight regressions were non-significant for fish from the 5 estuaries in which fish were around 200-250mm and collected Feb/Mar 2001, the effect of otolith weight was not removed from Ba/Ca and Mn/Ca data in analyses including only those estuaries.

Results

Strong differences were found in otolith Mn/Ca, Sr/Ca and Ba/Ca ratios of juvenile mangrove jack between the five estuaries sampled in Feb/Mar 2001 (Fig 2). Differences were highly significant for all element/Ca ratios in univariate analyses of variance ($df = 4,45$, $F > 25.00$, $p < 0.0001$), and post hoc Tukeys HSD tests showed they primarily reflected variability due to: the much higher otolith Mn/Ca of Johnstone River fish than those from the other four estuaries, high Sr/Ca of Daintree River fish and low otolith Sr/Ca of O'Connell River fish, and the highly variable otolith Ba/Ca ratios of fish from the different estuaries. Mean otolith Ba/Ca varied between estuaries over a range of ~5-40 $\mu\text{mol/mol}$, suggesting that this element may be of particular use as a marker of fish from different estuaries. The high otolith Mn/Ca of Johnstone River fish also suggests that Mn may be a strong marker of residency in particular estuaries.

Spatial variability between the additional five estuaries were not as great however, and otolith Mn/Ca and Ba/Ca in these estuaries in which samples included older fish exhibited generally lower ratios than 200-250mm fish collected in Feb/Mar 2001 (Fig 2). Fish from these estuaries did not exhibit any differences in Mn/Ca ratios when corrected for otolith weight, however corrected otolith Ba/Ca ratios were significantly different among estuaries ($df = 4,40$, $F = 14.470$, $p < 0.0001$), reflecting lower levels in Cattle Creek fish than in the other estuaries. Otolith Sr/Ca was also significantly different among estuaries ($df = 4,40$, $F = 8.530$, $p < 0.0001$), as a result of lower ratios in fish from Ross River than the other four estuaries. Differences over all ten estuaries for Sr/Ca and corrected Mn/Ca and Ba/Ca ratios were also highly significant ($df = 9,85$, $F > 16.00$, $p < 0.0001$), largely reflecting the patterns of otolith composition described above for each of the two groups of five estuaries, and the consequent additional instances of variability between individual estuaries when both groups were combined in analysis.

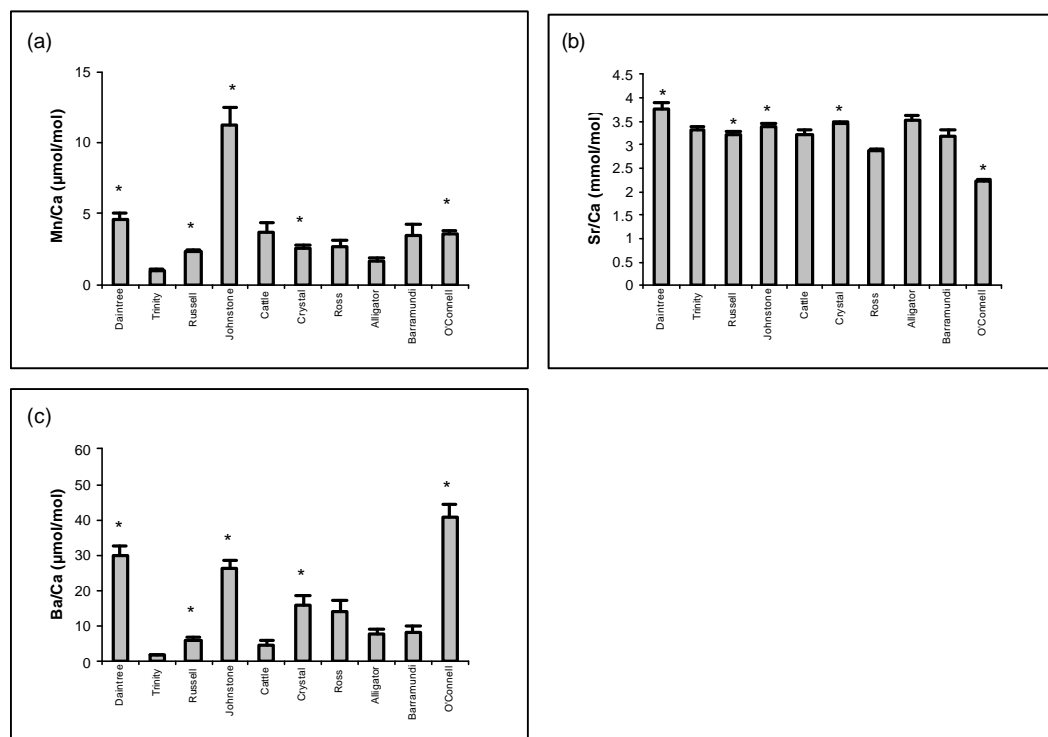


Figure 2 a-c: Mean otolith Mn/Ca, Sr/Ca and Ba/Ca ratios (\pm SE) of juvenile mangrove jack collected from 10 North Queensland estuaries. Asterisks denote samples including only fish around 200-250mm FL collected in Feb/Mar 2001. Other samples include juveniles of varying age collected over the preceding 3 years.

Classification of fish to their estuary of capture using QDFA of multi-elemental otolith fingerprints produced high levels of accuracy when only 200-250mm Feb/Mar 2001 fish were included in analysis (Table 1a). Overall accuracy was 90%, and for the Johnstone and O'Connell Rivers, classification rates were 100%. Overall classification accuracy when all 10 estuaries were included in QDFA fell to 67.3%, as classification rates to the five estuaries including larger fish were mostly only 30-60% (Table 1b). However Ross River fish were classified with an accuracy rate of 90%. Classification accuracies for the five Feb/Mar 2001 estuaries declined on average by only 10-20% (ie. 1 or 2 samples) when all ten estuaries were included (Table 1b), suggesting that otolith microchemical composition does represent a distinctive marker of residency in particular estuaries. However, low classification rates for some estuaries (eg. Alligator Creek and Barramundi Creek) indicates that this is not the case for all estuaries. As misclassifications were not always to the adjacent estuary or to estuaries in the same region (Tables 1a,b), otolith composition does not appear to be a clear marker of juvenile life spent in particular regions of the coast adjacent to the Great Barrier Reef.

(a)	DR	RuR	JR	CrC	OCR					
Daintree R (10)	90	0	0	10	0					
Russell R (10)	0	80	0	20	0					
Johnstone R (10)	0	0	100	0	0					
Crystal Ck (10)	20	0	0	80	0					
O'Connell R (10)	0	0	0	0	100					

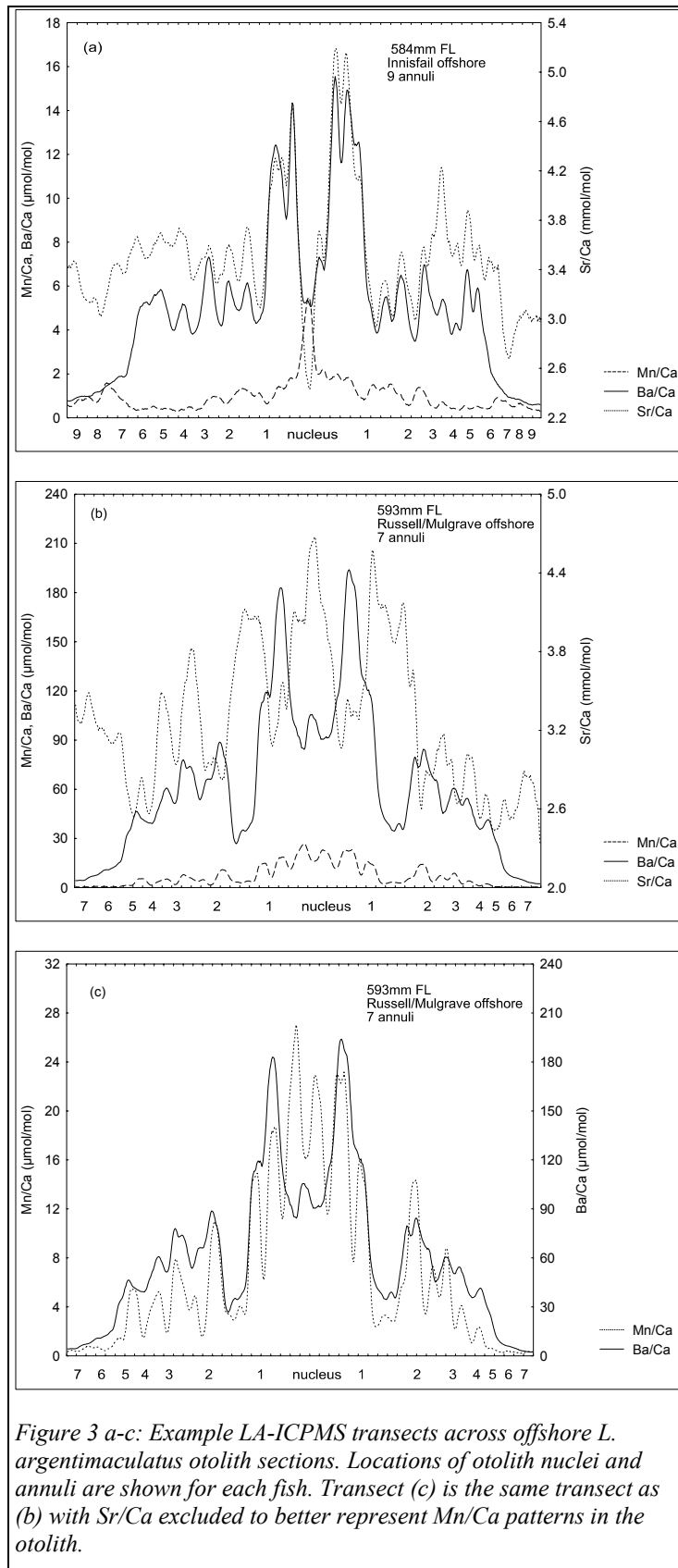
(b)	DR	TI	RuR	JR	CaC	CrC	RoR	AC	BC	OCR
Daintree R (10)	70	10	0	0	0	10	0	10	0	0
Trinity Inlet (8)	0	62.5	0	0	0	12.5	12.5	12.5	0	0
Russell R (10)	0	0	80	0	0	20	0	0	0	0
Johnstone R (10)	0	0	0	80	0	0	0	0	20	0
Cattle Ck (10)	0	0	10	0	60	10	0	0	20	0
Crystal Ck (10)	20	10	0	0	10	60	0	0	0	0
Ross R (10)	0	0	0	0	0	0	90	10	0	0
Alligator Ck (10)	20	20	0	10	0	0	10	40	0	0
Barramundi Ck (7)	0	0	0	0	14.3	0	42.9	14.3	28.6	0
O'Connell R (10)	0	0	0	0	0	0	0	0	10	90

Table 1 a,b: % Results of cross-validated classification of juvenile mangrove jack to their estuary of capture using quadratic discriminant functions. Table 1a includes only the five estuaries in which fish were collected in Feb/Mar 2001 (overall 90% of cases were correctly classified). Table 1b includes fish from all 10 estuaries, in which Mn/Ca and Ba/Ca data were adjusted to remove the influence of age (overall 67.4% of cases were correctly classified). Numbers in parentheses are numbers of fish analysed from each estuary

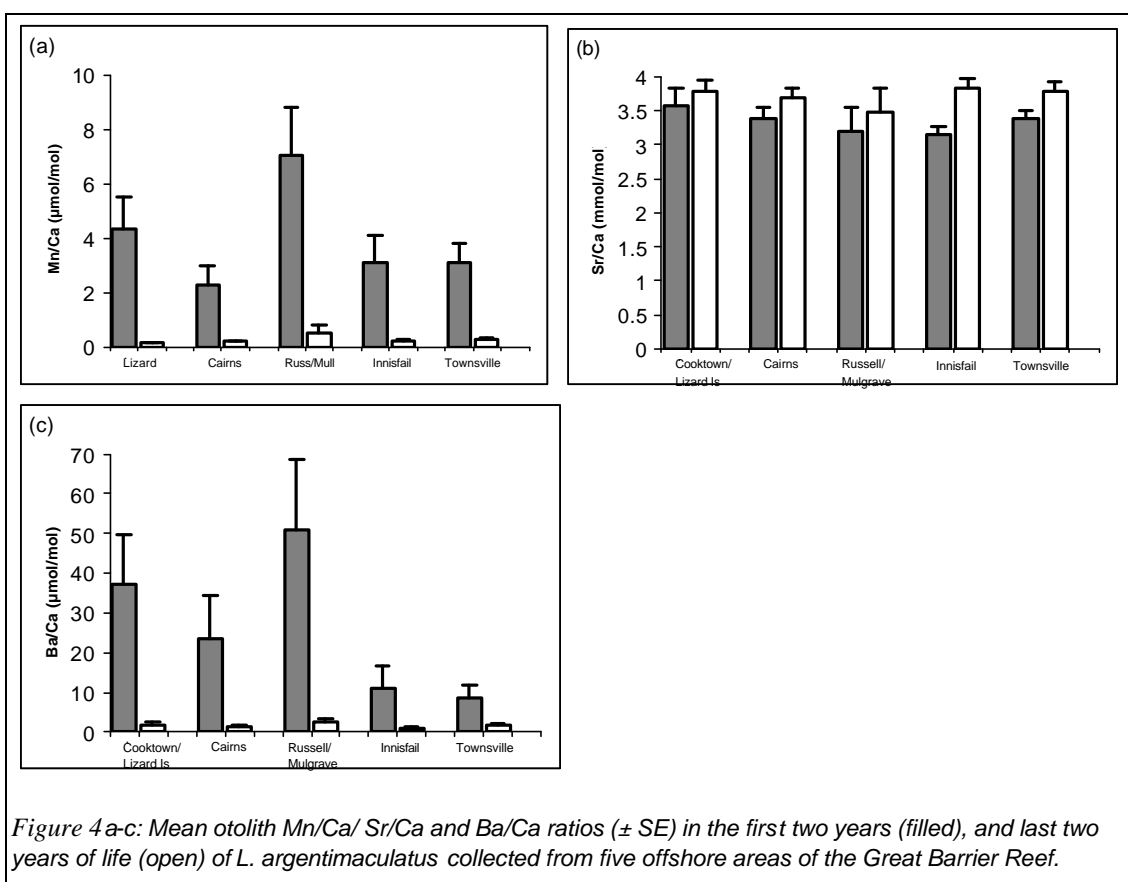
Laser-ablation ICPMS transects across sections of offshore *L. argentimaculatus* otoliths showed strong patterns of change in otolith microchemistry that appear to correspond with what would be

expected in a fish with a juvenile life spent in coastal waters prior to a migration offshore. Figure 3 shows two example LA-ICPMS transects across sections of offshore *L. argentimaculatus* otoliths, with distance along the transect replaced by the position of otolith annuli. Marked declines in otolith Ba/Ca at ages of around 5 and 6 respectively would appear to reflect movement downstream or out of estuaries and away from terrestrial sources of Ba. This apparent movement is relatively rapid in many fish, yet in others it is a more gradual process generally concluding with a steeper decline presumably reflecting ultimate departure from the estuary. Such gradual decline (Figs 3b,c), may reflect a gradual movement downstream after an early life spent in upper estuarine reaches. Mn/Ca invariably showed highest levels in very early life and generally declining levels thereafter (Figs 3a,c). In many fish, Mn/Ca exhibited similar patterns to Ba/Ca and showed declines at around the same age at which Ba/Ca indicated movement offshore (Fig 3c). However in others, Mn/Ca fell to very low levels while Ba/Ca still indicated residency in estuaries (Fig 3a). Peaks in otolith Ba and Mn during juvenile life are likely, in part, to reflect high river flow and fluvial trace-element input in the summer wet season, though seasonal changes in temperature, physiology and otolith precipitation rate may also be causal factors. As declines in Mn/Ca and Ba/Ca often corresponded with otolith annuli (Figs 3a,c), it would also appear that these effects are important factors in the incorporation of these elements. Sr/Ca also showed peaks and declines throughout its life that may reflect high wet season flow events where declines correspond with peaks in Mn/Ca and Ba/Ca (Fig 3b), and physiological effects where Sr/Ca peaks correspond with declines in Mn/Ca and Ba/Ca in annuli (Fig 3a). Clearly, incorporation of Mn, Ba and Sr into otoliths appears a complex interaction of both physiology and environment.

The example transects also show the degree of variability in juvenile otolith composition observed among offshore mangrove jack. Figure 3a shows a fish with relatively low otolith Mn/Ca and Ba/Ca during juvenile life. Although variable, around 70% of fish had relatively low juvenile Mn/Ca and Ba/Ca ratios. In contrast, around 30% of fish had much higher juvenile ratios (Fig 3b,c). Differences in Mn/Ca and Ba/Ca between these groups are clearly great, and in comparison of the two fish shown in Figure 3, are about an order of magnitude different. While not all high juvenile Mn/Ca and Ba/Ca fish had levels as great as those in Fig 3b, several did and others were even higher. Individuals of both types were observed among offshore fish from all of the offshore areas, however high juvenile Mn/Ca and Ba/Ca was most commonly observed in fish from the Russell/Mulgrave and Cooktown/Lizard Island offshore areas. It is possible that high juvenile Mn/Ca and Ba/Ca fish represent those that spent their juvenile life in upper estuarine or freshwater reaches of rivers and creeks, and that declines in Mn/Ca and Ba/Ca reflect both movement to downstream reaches as well as offshore. However, otolith Sr/Ca would be expected to be a strong marker of life in upper estuarine or freshwater reaches, with lower values expected at lower salinities. Although a number of fish with high otolith Mn/Ca and Ba/Ca ratios also had low Sr/Ca during juvenile life suggesting possible juvenile residency in brackish or freshwater, several did not, and some had otolith Sr/Ca considerably higher than fish with low Mn/Ca and Ba/Ca. In addition, a number of fish with low juvenile Mn/Ca and Ba/Ca had lower Sr/Ca ratios than some of the juveniles with high otolith Mn/Ca and Ba/Ca.



Comparison of the otolith composition of juvenile and adult parts of otoliths of offshore *L. argentimaculatus*, using mean Mn/Ca, Ba/Ca and Sr/Ca ratios over the first 2 years and the most recent 2 years of life for each individual showed marked differences (Fig 4). Mn/Ca and Ba/Ca in the last 2 years of life were much lower than in the first 2 years, and presumably in part reflect movement to offshore waters beyond the strong influence of the coast and its fluvial sources of these elements. In contrast, Sr/Ca was higher in the last 2 years of life, and may reflect residency during offshore life in waters of more consistently high salinity. While these patterns of change in Mn/Ca, Ba/Ca and Sr/Ca would appear to reflect movement away from the coast, and likely do, it is also possible that physiological or ontogenetic effects may be involved in changes in otolith composition over the life of fish.



The age of fish determined from counts of the number of otolith annuli laid down prior to the time at which declines in Mn/Ca and/or Ba/Ca indicate movement out of estuaries varied between 3 and 8, with most appearing to begin to move offshore at ages of 5-7 years (Fig 5). Only 2 of the 50 offshore *L. argentimaculatus* analysed had low Mn/Ca and Ba/Ca throughout life and did not exhibit the declines in these elements that are presumably indicative of migration offshore. These were an 18 year old fish from Cooktown/Lizard offshore, and an 8 year old from the Cairns offshore region.

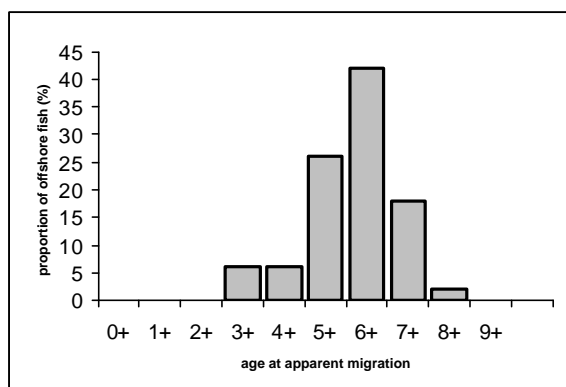


Figure 5 Age of offshore fish determined from annulus counts at which declines in otolith Mn/Ca and/or Ba/Ca indicated movement out of estuaries.

Classification of offshore mangrove jack to estuaries using quadratic discriminant functions derived from the analysis of estuarine fish to classify them using mean otolith compositions of the first two years of life showed that fish from each offshore region were largely classified to only two estuaries, and there were no clear patterns of classification to the estuary or estuaries in the adjacent coastal region (Table 2). In classifications using functions derived from discriminant analysis including only the five estuaries sampled in Feb/Mar 2001, the majority of fish with lower levels of otolith Mn/Ca and Ba/Ca were assigned to Crystal Creek (Table 2a), reflecting the moderate levels of Mn/Ca and Ba/Ca in otoliths of many offshore fish and those of juveniles from that estuary. Offshore fish with high Ba/Ca and low Sr/Ca were classified to the O'Connell River as juveniles from that estuary had this combination of Sr/Ca and Ba/Ca ratios. Other patterns included the classification of fish with particularly low Mn/Ca and Ba/Ca to the Russell River, those with high Mn/Ca to the Johnstone River, and those with high Ba/Ca and Sr/Ca to the Daintree River (Table 2a).

When functions derived from discriminant analysis including juveniles from all ten estuaries were used to classify offshore fish, patterns of classification to estuaries changed substantially and most fish were assigned to Cattle Creek (Table 2b). This largely comprised a shift to classification to Cattle Creek of many of those fish that were classified to Crystal Creek when functions from only the five Feb/Mar 2001 estuaries were used. However, some fish were still classified to Crystal Creek. The number of fish assigned to the O'Connell River also fell in classifications to ten estuaries, as those offshore fish with low otolith Sr/Ca were more commonly classified to the Ross River, as juvenile fish from this estuary also had lower Sr/Ca than other estuaries but otolith Ba/Ca was not as high as in the O'Connell River. Fish with high levels of otolith Mn/Ca and Ba/Ca were variably classified to different estuaries depending on their particular relative levels of these ratios and Sr/Ca. Classifying offshore fish to the ten estuaries using the mean otolith composition of the first year and first three years of life produced generally similar patterns and proportions of fish assigned to each estuary as when the means of the first 2 years of life were used.

(a)	DR	RuR	JR	CrC	OCR
Cooktown/Lizard Is (10)	1	1	0	6	2
Cairns (9)	1	2	1	3	2
Russell/Mulgrave (11)	3	0	2	2	4
Innisfail (9)	1	1	1	4	2
Townsville (11)	0	1	2	6	2

(b)

	DR	TI	RuR	JR	CaC	CrC	RoR	AC	BC	OCR
Cooktown/Lizard Is (10)	0	0	0	0	4	2	2	2	0	0
Cairns (9)	1	0	0	0	3	3	1	0	0	1
Russell/Mulgrave (11)	1	1	0	1	1	1	4	1	0	1
Innisfail (9)	0	0	0	0	5	2	2	0	0	0
Townsville (11)	0	0	1	0	7	2	0	0	1	0

Table 2a,b: Results of classification of offshore mangrove jack to estuaries using quadratic discriminant functions derived from analysis of juveniles from those estuaries. Mean otolith composition of the first two years of life of each offshore fish were used in classifications. Table 2a shows results when functions from analysis of only the five estuaries in which fish were collected in Feb/Mar 2001 were used. Table 2b shows results using functions derived from analysis when juveniles from all 10 estuaries were used, and in which Mn/Ca and Ba/Ca data were adjusted to remove the effect of otolith weight/age. Numbers in parentheses are numbers of fish analysed from each offshore region.

Discussion

The results of this study show that the pre-requisite factors necessary for retrospective nursery identification do appear to be present in the otoliths of *L. argentimaculatus* from waters of the Great Barrier Reef and the adjacent coast. There are strong differences in the otolith composition of juveniles from different estuaries and high classification accuracy rates of fish of similar size collected at the same time, thereby providing the baseline variability in coastal juvenile composition required if one is to suggest an offshore fish originated from a particular coastal area or estuary. There is also higher Mn/Ca and Ba/Ca in the juvenile parts than the recently deposited

parts of otoliths of offshore fish that would appear to indicate coastal juvenile life. Marked declines in concentrations of these terrestrially associated elements also occur in otoliths at ages of 3-8 years, and presumably reflect migration offshore. Lower Sr/Ca ratios in the juvenile than recently deposited parts of otoliths of offshore *L. argentimaculatus* may also indicate a movement to waters of more consistently high salinity. On the basis of these patterns, it would appear that the microchemical analysis of otoliths does hold potential as a method of retrospectively identifying the estuary of origin of offshore *L. argentimaculatus*.

However, the assessment of retrospective nursery identification undertaken in this study produced uncertain results, in that the majority of offshore fish were assigned to particular estuaries. This appears largely to be a consequence of the small number of estuaries from which juveniles were collected, and the fact that otolith compositions of juveniles in some of the estuaries were considerably different to those of most offshore fish, suggesting they may not be widely representative of patterns of spatial variability in estuarine juvenile otolith composition. Sampling more estuaries would be likely to overcome this pattern of classification, as additional estuaries would likely include those in which juvenile otolith composition were more like those observed in juvenile parts of otoliths of offshore fish. Consequently, classifications using discriminant functions derived from the analysis of juveniles including such estuaries would result in offshore fish being more variably classified to those estuaries in which juveniles had otolith composition most like the juveniles parts of their otoliths. Yet this would not necessarily represent a more accurate classification of offshore fish to their estuary of origin as similarities between offshore fish and juveniles from particular estuaries may not mean they spent their juvenile life in that estuary. As they are older individuals, and even if the estuary in which they spent their juvenile life was represented in estuarine samples used to derive discriminant functions, temporal variability in otolith composition in estuaries may still result in their being classified to a different estuary.

Temporal variability in otolith composition of juvenile *L. argentimaculatus* was not assessed in this study, however it has been reported in juvenile fish in temperate Australian estuaries (Gillanders and Kingsford 2000, Gillanders 2002). These authors have suggested that for nursery identification to be possible it may be necessary to compile a 'library' comprising juvenile otolith fingerprints for each estuary in each year, and to use discriminant functions derived for a particular year-class of juveniles to later classify offshore fish that spent their juvenile life in an estuary in that year. In tropical estuaries, temporal variability in otolith composition of juveniles from the same estuary is also likely as rainfall and river flow can vary greatly between years (Furnas and Mitchell 2001). Furthermore, life-history transects across otoliths do show variability in otolith composition during juvenile life. Hence such temporal variability is likely to complicate, and perhaps preclude the retrospective identification of the estuaries of origin of offshore fish. While it may be possible to compile a library of otolith composition for all estuaries in each year in regions where there are only a relatively small number of estuaries along the coast (Gillanders and Kingsford 2000, Gillanders 2002), the number of estuaries in tropical North Queensland from which juvenile *L. argentimaculatus* may have migrated offshore is so large that making collections from all estuaries in all years to compile a library would probably not be viable owing to the effort and expense involved in collection, sample preparation and analysis. In addition, if analysis included samples from a large number of estuaries it is likely that similarities in otolith composition of juveniles from different estuaries would be more common, resulting in the misclassification of offshore fish to putative natal nurseries.

Although a number of studies have reported strong differences in otolith composition of juvenile fish from different estuarine nursery grounds and have suggested that retrospective nursery identification of offshore adults should be possible (Thorrold *et al.* 1998a,b, Gillanders and Kingsford 2000, Patterson 2001, Gillanders 2002), only two have reported a successful application to date. Thorrold *et al.* (2001) estimated rates of homing of 60-81% for spawning by 2 year old weakfish, *Cynoscion regalis* (Sciaenidae), to five estuaries in the eastern United States on the basis of the trace element and stable carbon and oxygen ratios of otolith cores and otolith compositions of juveniles collected in those estuaries two years previously. However, this study compared only a small number of estuaries, and minimized potential error in classification due to temporal variability by analyzing otoliths of juvenile and adult fish of the same cohort. Milton *et al.* (1997) collected adult terubok, *Tenualosa toli* (Clupeidae), from a number of coastal sites and the two main spawning estuaries of the species in Sarawak and found that although temporal variability was apparent between years, the core composition of coastal adults was similar to fish from the spawning estuaries, suggesting that most had originated from those estuaries. Attempted retrospective identification of the particular coastal nursery of origin of offshore fish that may have originated from any of a large number of estuaries, or of species that remain offshore after emigrating from nursery estuaries has not yet been reported.

In a study of blue groper *Achoerodus viridis* (Labridae) in waters near Sydney, Gillanders and Kingsford (1996) found differences in the otolith composition of juveniles between estuarine seagrass habitats and nearby coastal rocky reefs where adults also occur, and used these to estimate the relative contribution of recruits from each juvenile habitat to adult populations by analysing the core of otoliths of the older adults. While this study essentially does represent retrospective nursery identification of adult fish of different ages to juveniles, its results reflect only juvenile habitat differences as no assessment of spatial variability in juvenile otolith composition was conducted. Nevertheless, an ability to assess the relative contribution of recruitment from different nursery habitats to adult populations is obviously of considerable value in attaining an understanding of the wider ecology of species in which juveniles occur in nursery habitats different to those in which adults occur.

Although the attempted retrospective identification of nursery estuaries in the present study produced uncertain results, and its wider application also appears unlikely to produce definitive results owing to the number of estuaries in which juvenile *L. argentimaculatus* occur, the analysis of otoliths of offshore *L. argentimaculatus* does appear to provide retrospective nursery habitat information similar to that of Gillanders and Kingsford (1996). The higher levels of otolith Mn/Ca and Ba/Ca in juvenile than in recently deposited parts of otoliths of offshore *L. argentimaculatus*, and the marked declines in these ratios at ages of 3-8 in life-history transects strongly suggest coastal juvenile life prior to an offshore migration. As the great majority of offshore *L. argentimaculatus* analysed in this study appear on the basis of these patterns to have spent their juvenile life in coastal habitats (48/50 or 96%), impacts on coastal juvenile populations of this species will thus directly impact on offshore populations, as migration from coastal habitats appears the primary source of recruits. While low juvenile Mn/Ca and Ba/Ca and a lack of a distinct decline in these ratios in two fish may reflect the residency of these fish in non-estuarine waters during juvenile life, it does not necessarily indicate a life spent entirely in offshore waters. Habitats such as headlands, islands, or estuaries on islands with minimal catchments may also be habitats in which such fish may have resided (Sheaves 1995), and where water Mn and Ba concentrations are likely to be lower than in coastal estuaries. However it is also possible that these fish have spent their entire life in the offshore adult habitat.

As dissolved concentrations of Mn and Ba typically exhibit downstream declines in estuaries (Coffey *et al.* 1997, Klinkhammer and McManus 2001), the particularly high levels of Mn/Ca and Ba/Ca in juvenile parts of otoliths of some offshore fish may reflect residency in upstream reaches of estuaries. However Sr/Ca ratios in these fish did not always indicate a juvenile life spent in waters of lower salinity than those with lower otolith Mn/Ca and Ba/Ca. It has been suggested that analysis of otolith Sr/Ca to reconstruct salinity history may be more applicable where fish have resided in waters of very low salinity, and that there may be a threshold effect, above which otolith Sr/Ca does not reflect salinity (Gillanders and Kingsford 2000). Such a threshold effect may in part explain the high Mn/Ca and Ba/Ca of some fish in the absence of low Sr/Ca if they resided in waters far enough upstream in estuaries to be exposed to higher concentrations of Mn and Ba than downstream fish, but of salinities still above the threshold salinity. By corollary, those fish with high Mn/Ca and Ba/Ca and low Sr/Ca may represent those that did reside in waters of lower salinity than the threshold level. Those fish with low Mn/Ca and Ba/Ca and low Sr/Ca may represent those that resided in upstream reaches of estuaries with low inputs and upstream concentrations of Mn and Ba. Sampling fish from different sites along the estuarine gradient of several estuaries, including those in upstream brackish and freshwater reaches would be required to test this hypothesis. If such differences in otolith composition do exist along estuarine gradients, it would further suggest that the retrospective identification of the estuary of origin of offshore fish is not possible, as similarities in otolith composition amongst fish from different estuaries will presumably be common as a result of variability within estuaries.

On the basis of the results of this study, and the large number of estuaries along the coast of tropical Queensland, retrospective nursery identification of adult *L. argentimaculatus* from offshore waters of the GBR appears limited to determining whether an individual spent its juvenile life in coastal waters, and perhaps whether it resided in upstream reaches of estuaries. Identification of the region in which a fish spent its juvenile life also appears unlikely as juveniles from estuaries relatively close to one another (eg the Johnstone and Russell Rivers) had very different otolith composition. However, it is possible that retrospective identification of particular nursery estuaries or regions may become more viable in the future if analytical techniques continue to improve in sensitivity. If additional elements such as copper, lead and zinc which are known to occur in otoliths in very low concentrations were able to be reliably quantified and included in otolith fingerprints, it would obviously increase the likelihood of identifying fingerprints distinctive to particular estuaries in the juvenile parts of otoliths of offshore adults.

Despite the uncertainties of retrospective identification of nursery estuaries, it strongly appears that the migration of *L. argentimaculatus* from coastal nurseries to the GBR is recorded in their otolith microchemistry, and the age at which fish moved offshore can be identified. Similar clear patterns have been observed in life-history transects across otolith sections of other GBR lutjanids that use coastal waters as nursery grounds including *L. russelli*, *L. malabaricus* and *L. erythropterus*. In these species the migration apparent in otolith microchemistry also corresponds with ages at which these species appear to begin to move offshore. Hence it would appear that it can be reliably determined whether an offshore adult population is sustained entirely by the recruitment of migrants from coastal nursery grounds. While clearly not as useful as specific nursery identification in terms of attaining a knowledge of recruitment patterns and movement and mixing of offshore populations, such information nevertheless provides insights into the ecology of such species that would be impossible to assess by other means.

Tag-recapture is obviously the most reliable method of nursery identification as it provides incontrovertible evidence of residency in the estuary in which it was tagged, and movement to the offshore location of capture. However tag-recapture programs can be time-consuming and expensive, and if recapture rates are low, or for species in which tagging of juveniles in coastal habitats has not been undertaken or has been only small-scale, otolith microchemistry does appear a useful identifier of a juvenile life spent in coastal waters prior to a migration offshore. Where it can be shown that offshore populations are sustained largely or entirely by recruitment of juveniles from coastal nurseries, such information lends strong support to the conservation of these juvenile populations and their nursery habitats.

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Appendix 4

Number of fish caught, tagged and recaptured at each site on each sampling occasion.

Site types are p -primary, s secondary and e extra.

Code	Location/ (Lat./Long.)	Site type	Conductivity (ms/cm)	Date	No. caught	No. tagged	No. recaptured
BA1	Baffle Creek 151.8596, -24.5964	s	0.576	08-Dec-99	13	9	
			0.369	21-Mar-00	27	25	
BA2	Baffle Creek 151.8471,-24.5983	s	0.367	30-May-00	16	15	1
			0.234	28-Nov-00	26	20	4
			1.04	06-Mar-01	19	10	6
			0.461	06-Mar-02	34	34	3
BA3	Baffle Creek 151.8792, -24.584	s		22-Mar-00	71	64	1
				31-May-00	81	80	5
				08-Aug-00	1	0	
				29-Nov-00	29	28	1
			1.78	07-Mar-01	8	4	2
				06-Mar-02	43	40	2
BA4	Baffle Creek 151.8997,-24.5239	s	2.19	07-Mar-02	44	42	1
BO1	Boyne River 151.3181, -24.0367	e	1.37	10-Dec-99	2	0	
BR1	Barron River 145.6789, -16.8757	e		20-May-99	8	0	
			0.108	17-Jan-00	10	2	
				22-Mar-01	1	0	
				17-Jul-01	4	0	
BR2	Barron River 145.6812,-16.8748	e	0.068	19-Jan-01	19	19	
BR3	Barron River 145.6588,-16.8754	e	0.104	14-Jan-02	38	34	
CA1	Calliope River 151.1215, -23.9912	e	0.663	10-Dec-99	2	1	
			0.796	19-Mar-00	0	0	
CA2	Calliope River 151.1433,-23.9833	e	1.04	30-Nov-00	6	0	
			0.11	07-03-02	0	0	
CK1	Endeavour River 145.2020,-15.4277	e		01-Oct-99	4	2	
CR1	Crystal Creek 146.3057, -18.9359	e	0.079	03-Dec-00	186	163	
			1.94	18-Jan-01	102	92	14
				20-Feb-01	7	7	
			0.096	03-Mar-01	98	75	13
				03-Dec-01	3	3	

Code	Location/ (Lat./Long.)	Site type	Conductivity (ms/cm)	Date	No. caught	No. tagged	No. recaptured
				02-Feb-02	3	0	
			0.09	20-Feb-02	55	45	
			0.081	11-Mar-02	97	66	1
DA1	Daintree River 145.3172, -16.2524	p	0.05	24-Jan-00	25	24	
			0.051	30-Mar-00	10	10	
			0.053	19-Apr-00	15	15	
			0.045	23-May-00	25	25	4
			0.051	26-Jun-00	41	40	2
			0.055	20-Jul-00	29	29	5
				22-Aug-00	0	0	1
			0.063	19-Sep-00	14	12	8
			0.234	12-Oct-00	1	1	
				20-Oct-00	10	10	5
			0.181	09-Nov-00	9	8	3
				15-Dec-00	8	6	2
			0.058	11-Jan-01	5	5	2
			0.049	19-Mar-01	7	7	1
			0.05	18-Apr-01	6	6	2
			0.052	15-May-01	18	14	4
			0.059	16-Jul-01	15	11	3
			0.057	15-Aug-01	6	5	2
			1.49	18-Sep-01	4	4	1
			0.062	10-Oct-01	16	13	1
			0.732	15-Nov-01	3	2	
			0.509	11-Dec-01	13	10	2
			0.388	28-Feb-02	3	2	
DA2	Daintree River 145.3421, -16.2563	p		24-Jan-00	3	2	
				30-Mar-00	19	19	
				19-Apr-00	32	32	1
				23-May-00	32	31	
			0.053	26-Jun-00	17	14	1
			0.056	20-Jul-00	7	6	2
			0.26	19-Sep-00	12	12	
			0.87	20-Oct-00	11	10	2
			0.214	09-Nov-00	9	3	
			0.052	15-Dec-00	20	19	4
			0.073	11-Jan-01	8	7	4
			0.049	19-Mar-01	11	11	1
			0.05	18-Apr-01	10	8	1
			0.052	15-May-01	8	3	

Code	Location/ (Lat./Long.)	Site type	Conductivity (ms/cm)	Date	No. caught	No. tagged	No. recaptured
			0.068	16-Jul-01	10	3	
			5.46	18-Sep-01	10	3	1
			0.09	10-Oct-01	14	7	2
				11-Dec-01	2	2	
			0.795	28-Feb-02	3	1	
DA3	Daintree River 145.4047,-16.2720	e		23-May-00	18	18	
				20-Jul-00	4	4	
				19-Mar-01	21	10	
				18-Apr-01	20	16	
EU1	Euleilah Creek 151.9072,-24.4987	e		29-Nov-00	1	1	
HE1	Herbert River 146.2044, -18.591	s	0.069	13-Dec-99	27	22	
			0.06	28-May-00	8	8	1
			0.078	01-Sep-00	17	13	3
				06-Sep-00	8	0	
				08-Sep-00	1	0	
			0.066	17-Jan-01	6	6	1
			0.074	05-Jun-01	11	10	1
			0.088	04-Sep-01	15	13	3
			0.053	03-Dec-01	6	6	
			0.07	12-Mar-02	8	4	
HE2	Herbert River 146.29, -18.5416	s	0.063	26-May-00	25	15	
			0.145	01-Sep-00	60	44	2
			0.207	04-Sep-00	42	39	
			0.074	17-Jan-01	57	49	3
			0.083	04-Jun-01	110	91	5
			0.218	04-Sep-01	71	61	4
			0.386	12-Mar-02	29	19	1
HE3	Herbert River 146.2895,-18.5408	e	0.02	26-May-00	24	20	1
JR1	Johnstone River 146.0010,-17.4894	e		01-Mar-01	7	5	
MU1	Mulgrave River 145.9099, -17.1904	e		09-Nov-99	12	12	
MU2	Mulgrave River 145.9132, -17.1799	e		09-Nov-99	2	2	
MU3	Mulgrave River 145.9064, -17.1753	e		09-Nov-99	16	16	
MU4	Mulgrave River 145.8839, -17.1502	p	0.041	10-Nov-99	35	35	
			0.04	27-Jan-00	22	21	2
			0.045	09-Mar-00	6	6	
			0.039	17-Apr-00	3	3	

Code	Location/ (Lat./Long.)	Site type	Conductivity (ms/cm)	Date	No. caught	No. tagged	No. recaptured
			0.03	22-May-00	7	7	1
			0.035	22-Jun-00	14	14	2
			0.049	24-Jul-00	13	13	3
			0.056	10-Oct-00	9	8	3
				07-Nov-00	10	8	1
			0.039	11-Dec-00	10	10	2
			0.042	09-Jan-01	2	2	3
				14-Feb-01	6	6	1
			0.041	20-Mar-01	6	6	2
			0.036	20-Apr-01	1	1	
			0.047	24-May-01	6	6	
			0.045	11-Jul-01	6	5	
			0.053	14-Aug-01	2	2	1
			0.062	17-Sep-01	5	5	1
			0.06	11-Oct-01	11	9	2
			0.062	12-Nov-01	9	7	1
			0.075	12-Dec-01	8	5	1
			0.067	09-Jan-02	11	9	
			0.061	25-Feb-02	5	3	
MU5	Mulgrave River 145.8334, -17.1023	e	0.046	12-Nov-99	6	6	
MU6	Mulgrave River 145.9833, -17.2091	p	0.056	27-Jan-00	78	63	
			0.049	09-Mar-00	52	38	2
			0.041	17-Apr-00	31	23	4
			0.033	22-May-00	75	64	11
			0.038	22-Jun-00	57	47	9
			0.057	25-Jul-00	41	31	2
			0.63	20-Sep-00	68	56	1
			1.06	10-Oct-00	43	24	5
			1.68	07-Nov-00	4	2	2
			0.165	11-Dec-00	22	16	5
			0.098	09-Jan-01	17	9	
			0.049	14-Feb-01	17	11	1
			0.045	20-Mar-01	24	8	4
			0.037	20-Apr-01	37	9	3
			1.06	16-May-01	31	7	
			0.064	11-Jul-01	27	11	
				18-Jul-01	40	2	
			0.084	31-Jul-01	1	1	
			1.45	14-Aug-01	21	9	
			0.433	31-Aug-01	12	12	

Code	Location/ (Lat./Long.)	Site type	Conductivity (ms/cm)	Date	No. caught	No. tagged	No. recaptured
				11-Sep-01	6	0	
			0.97	11-Oct-01	14	6	
MU7	Mulgrave River 145.933, -17.2166	e	0.085	10-May-00	99	54	
NC1	Ninds Creek, Johnstone River 146.0584,-17.5277	e	0.202	01-Mar-01	28	16	
NJ1	North Johnstone River 146.0343, -17.4940	p	0.062	28-Jan-00	20	19	
				15-Mar-00	15	15	
			0.034	18-Apr-00	29	25	1
			0.038	25-May-00	12	7	3
				27-Jun-00	14	10	3
			0.076	26-Jul-00	20	19	2
			1.31	18-Sep-00	9	9	2
			2.6	09-Oct-00	7	86	2
			5.09	08-Nov-00	7	6	
			0.101	12-Dec-00	4	4	
			0.05	19-Jan-01	7	4	
			0.038	01-Mar-01	8	3	1
			0.04	23-Mar-01	23	4	
			0.036	19-Apr-01	24	2	
			0.043	17-May-01	10	0	
				15-Jun-01	10	0	
	11-Jul-01	6	1				
	1.43	25-Oct-01	11	3	1		
	0.416	01-Mar-02	19	13			
NJ3	North Johnstone River 146.0012, -17.4902	e		01-Jun-99	10	10	
				28-Sep-99	3	3	
				22-May-01	7	6	
			0.076	01-Mar-02	1	1	
NJ4	North Johnstone River 146.0012, -17.4997	e		01-Jun-99	1	1	
				28-Sep-99	3	3	
				28-Jan-00	6	6	
				15-Mar-00	4	4	1
				26-Jul-00	1	1	
				22-May-01	1	1	
				01-Mar-02	1	1	
NJ5	North Johnstone River 145.9949, -17.5088	e		28-Sep-99	4	4	
				22-May-01	3	3	
NJ6		e		01-Jun-99	5	5	

Code	Location/ (Lat./Long.)	Site type	Conductivity (ms/cm)	Date	No. caught	No. tagged	No. recaptured
	North Johnstone River 145.9871, -17.5062			28-Sep-99	3	3	
			0.039	22-May-01	1	1	
OC1	O'Connell River 148.6128, -20.5661	s	0.578	18-Jan-00	5	2	1
			0.274	23-Mar-00	29	26	
			0.42	01-Jun-00	44	33	8
			0.461	02-Dec-00	41	37	5
				20-Feb-01	1	0	
			0.266	04-Mar-01	23	13	3
				09-Mar-02	9	8	
OC2	O'Connell River	s	0.493	02-Sep-00	33	31	
			0.5	06-Jun-01	46	42	2
			0.558	06-Sep-01	47	43	10
			0.706	05-Dec-01	21	21	9
				09-Mar-02	21	8	1
PN1	Pine Creek, Trinity Inlet 145.8201, -16.9936	e	0.05	06-Mar-00	11	11	2
			0.052	06-Apr-00	13	11	
			0.05	05-May-00	7	7	3
			0.613	13-Dec-00	4	4	1
RA1	Raglan Creek 150.5190, -23.4470	e	0.36	11-Dec-99	5	2	1
			0.573	30-Nov-00	3	2	
RE1	Repulse Creek 148.7716, -20.4102	e		19-Jan-00	13	8	
				02-Jun-00	9	0	
RE2	Repulse Creek 148.7631, -20.4161	e		19-Jan-00	1	1	
RU1	Russell River 145.9487, -17.2727	p	0.092	25-Jan-00	35	33	
			0.026	27-Mar-00	71	62	
			0.031	20-Apr-00	65	60	8
			0.024	24-May-00	56	51	18
			0.039	23-Jun-00	169	160	18
			0.031	25-Jul-00	6	6	
			0.037	06-Aug-00	85	80	22
			4.28	21-Sep-00	22	20	12
			4.77	06-Oct-00	17	15	8
			0.389	03-Nov-00	12	9	
			0.042	14-Dec-00	18	16	8
			0.035	10-Jan-01	12	7	5
			0.03	21-Mar-01	21	17	2
			0.029	17-Apr-01	20	17	2
			0.081	14-May-01	18	9	2
2.34	13-Jul-01	16	12	1			

Code	Location/ (Lat./Long.)	Site type	Conductivity (ms/cm)	Date	No. caught	No. tagged	No. recaptured
			0.206	20-Aug-01	47	33	6
			3.71	13-Sep-01	24	8	4
			2	17-Oct-01	11	9	2
			1.36	13-Nov-01	17	11	2
			1.51	12-Dec-01	9	6	
			0.343	25-Feb-02	20	14	1
RU2	Russell River 145.9526, -17.3275	e	0.033	16-Nov-99	5	4	
			0.04	24-Nov-99	8	8	
				25-Jan-00	4	4	
			0.023	20-Apr-00	3	3	
			0.023	23-Jun-00	2	2	
RU3	Russell River 145.9447,-17.2454	e		20-Apr-00	12	12	
				09-May-00	114	58	
				26-Feb-01	25	21	
RU4	Russell River 145.9512, -17.3292	e		22-Mar-01	24	24	
RU5	Russell River 145.9535,-17.3081	e		12-Jan-00	2	0	
				25-Jan-00	4	4	
RU7	Russell River 145.9634,-17.2288	e	0.685	26-Feb-01	26	13	
SJ1	South Johnstone River 146.0275,-17.5496	p		22-Oct-98	1	1	
				02-Jun-99	43	43	1
				29-Sep-99	5	5	
				11-Oct-99	9	9	2
				22-Oct-99	25	22	2
			0.041	18-May-00	73	67	2
			0.042	27-Jun-00	45	43	5
				26-Jul-00	17	16	12
			0.517	18-Sep-00	11	10	4
			2.82	09-Oct-00	16	15	4
			0.177	08-Nov-00	14	13	4
			0.368	12-Dec-00	6	6	1
			0.092	19-Jan-01	11	8	1
			0.041	01-Mar-01	19	15	4
			0.044	23-Mar-01	20	15	10
			0.04	19-Apr-01	7	6	
			0.046	17-May-01	13	5	2
			0.043	21-May-01	24	19	
			0.052	12-Jul-01	30	20	7
			0.092	03-Oct-01	37	26	2
			0.21	25-Oct-01	15	7	1

Code	Location/ (Lat./Long.)	Site type	Conductivity (ms/cm)	Date	No. caught	No. tagged	No. recaptured
			0.378	26-Feb-02	20	16	
TI1	Hills Creek, Trinity Inlet 145.775, -16.9734	e		15-Apr-00	1	0	
				12-Oct-01	12	12	
			34.6	29-Oct-01	3	2	
				21-Jan-02	1	0	
TV1	Ross River 146.8059,-19.3054	e	0.23	19-Feb-02	2	1	
WR1	Wrights Creek 145.7641, -17.0121	e	0.097	12-Jan-00	16	9	
			0.071	06-Mar-00	1	0	
			0.071	06-Apr-00	34	28	2
			0.074	05-May-00	19	16	4
			2.38	14-Dec-00	7	6	
			0.073	22-Feb-01	5	5	1
WR2	Wrights Creek 145.7600,-17.0100	e		12-Jan-00	1	0	