LARVAL FISH ASSEMBLAGES IN THE LOWER RIVER MURRAY, AUSTRALIA: EXAMINING THE INFLUENCE OF HYDROLOGY, HABITAT AND FOOD.

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Frontispiece: The Lower River Murray, Plush's bend boat ramp at sunset (photo courtesy of Matt Pellizzari)

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KATHERINE JANE-MAREE CHESHIRE

9th June 2010

The Lower River Murray, South Australia (photo courtesy of Kelly Marsland).

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~

This thesis is dedicated to my Mum and Dad…

For choosing a life dedicated to science, for inspiring and encouraging my love of science and the environment from a very early age and for the unconditional love and support that is given every day…

~

Chowilla (photo courtesy of Lesley Alton).

ABSTRACT

The major assumption of currently accepted fish recruitment hypotheses (e.g. *flood pulse concept* and *flood recruitment model*) is that in the absence of overbank flows the main river channel does not provide adequate food and habitat for larvae and juveniles. However, periods of low flows are common throughout floodplain rivers, and there are a wide diversity of life history strategies exhibited by riverine fish. Therefore, the broad applicability of these assumptions to the management of all fish species and floodplains rivers has been questioned. The *low flow recruitment hypothesis* pioneered the concept that some fishes can successfully spawn and recruit during low flows by utilising main channel habitats. Characteristics of the river channel, flow regime and level of regulation are often distinctly different both within and between rivers, and many of the recruitment models and indeed the life history strategies of fishes, remain untested in alternative floodplain river systems.

River regulation has resulted in altered flow regimes in river systems throughout the world, and in turn, has a range of negative impacts on the fish populations. The Murray-Darling Basin is Australia's largest river catchment and has been severely affected by river regulation. To test some of the assumptions of the previously described recruitment models larval fish and zooplankton sampling was conducted in the main channel environments of the Lower River Murray, South Australia. In comparison to the rest of the Murray-Darling Basin, the Lower River Murray is unique due to the combination of four distinct geomorphologic regions, the absence of significant tributaries, and the high degree of regulation. Extensive river regulation has drastically reduced the natural flow variability of the Lower River Murray. Furthermore, there has been little work on the spawning and larval assemblages within this region.

Larval fish sampling is often used for studying the early life history of fishes, but sampling gear and diel timing of sampling can bias results. Pelagic plankton tows were the single most effective method for collection of most species. Diel variation was identified for many species; with most exhibiting higher abundances during the night, although one species occurred in higher abundances during the day. Given these results the sampling regime for this project utilised both day and night pelagic plankton tows.

Annual differences in the larval assemblages in relation to variations in hydrology and environmental variables were investigated across four years, including a year of increased flow and a water level raising, and three years of low regulated flow with stable water levels.

The main channel environment of the Lower Murray supported larvae from all life history strategies. The larval assemblage differed between years; the flow pulse year was consistently different from the subsequent three low flow years. Three responses to varying hydrology were identified in the larval assemblage: larvae that were 1) positively correlated to increased flow, 2) negatively correlated to the increased flow and 3) correlated to temperature. The low flow recruitment hypothesis was supported, with a number of small-medium bodied native species spawning under low flow conditions in the river channel. However, golden perch and silver perch (flow cued spawners), were only present during the flow pulse year. Environmental flows are therefore vitally important for the management and restoration of some native fish species.

Strong within year variability was inherent in the data due to the seasonal variation in spawning time of fishes. The timing of peak spawning in the Lower River Murray was compared to other studies throughout the Basin. The broad spawning patterns identified for individual species were similar to seasonal spawning guilds identified for Australian species in previous studies. These spawning guilds were spring/summer and summer spawners. Understanding the timing of spawning of key species within a region will ensure that management actions can be targeted at providing benefits for species of interest.

The key assumption of many recruitment models is that the main river channel is an area of low productivity, and therefore it does not provide adequate food for developing larvae, which is particularly pronounced in years of low flow. Zooplankton sampling was conducted during the spring/summer of 2006 in the pelagic zone of the main river channel in a typical low flow year. Although temporally and spatially restricted, results indicated that during a low flow year an abundant prey source does exist in the main river channel in the Lower River Murray. Furthermore the prey was abundant in the pelagic zone of the open water, where traditionally pelagic zooplankton abundances have been documented to be relatively low. This suggests that in the absence of floodplain inundation developing larvae have adequate access to food in this lowland temperate system.

The inundated floodplain is generally recognised as important habitat for developing, larvae, consequently the importance of the main channel environment is frequently overlooked despite many studies highlighting the importance of shallow, still littoral zones. Larval fish were sampled in three main channel habitats: backwaters, open water and still littoral zones. Larvae of key species successfully spawned and utilised these main channel habitats during a low flow year. Specifically, still littoral zones and backwaters were important main channel habitats for developing fish larvae, providing support for the applicability of the low flow recruitment hypothesis to the Lower River Murray.

Some species (namely the small – medium bodied natives were able to spawn and recruit in the Lower River Murray under low flow conditions, but these were also able to spawn under the higher flow conditions. However, during the low flow years there were no larvae golden perch or silver perch collected, suggesting that these species were not spawning under the low flow conditions. This study has highlighted that a number of species will spawn and develop as larvae in the heavily regulated weir pool environment. In addition, adequate food and habitat were available for developing fish larvae in the absence of floodplain inundation in the Lower River Murray. However, for species with specific flow requirements (such as golden perch and silver perch, and potentially Murray cod and freshwater catfish) continued low flow conditions may pose a significant threat. In heavily regulated systems, environmental water allocations should be considered to manage and potentially restore declining fish populations, and the benefit of within channel flow pulses should not be underestimated.

Sunset in Chowilla (photo courtesy of Lesley Alton).

CHAPTER 1: GENERAL INTRODUCTION AND STUDY AREA

The most diverse vertebrate group worldwide are fishes, with more than 30,000 species documented, they constitute over half the total described vertebrates (Nelson 2006). An estimated 10,000 are freshwater species (Matthews 1998). Fishes vary in their morphology, habitat associations and basic biology including behaviour, diet, lifespan and spawning attributes (Nelson 2006). For humans, fishes provide an essential food source, and therefore, have economic, recreational and cultural value within society. Furthermore, fish play a crucial role in ecosystems, particularly nutrient cycles, food webs and productivity. Riverine fishes are dependent on the morphology, hydrology and water quality of the river, and thus, are widely used as indicators of broader ecological health within a system (Karr 1981, 1991; Gatz and Harig 1993; Chaves and Alipaz 2007). Unfortunately, many are under threat because of human activity. To assist in the management and rehabilitation of fish populations we require a greater understanding of the factors controlling population dynamics of each species.

1.1 EARLY LIFE HISTORY OF FISH

The early life history of fish is the period from fertilisation through the embryonic and larval periods, up to the early portion of the juvenile stage (Trippel and Chambers 1997). Changes in the abundance and mortality of early life history stages have long been recognised as critical to understanding fluctuations in fish populations (Hjort 1914; May 1974). Numerous studies have been conducted on the distributions of eggs, and the distribution and behaviour of larvae, given the importance of the early life stage to population and fisheries studies (Kelso and Rutherford 1996). Data on the early life history stages are essential to determine critical spawning cues, timing of spawning, spawning and nursery areas, the influences of environmental change on larval survival, and ontogenetic shifts in habitat and diet.

The early life history stage is a time of exceptionally high mortality, often in the order of 90-99% (Kelso and Rutherford 1996; Trippel and Chambers 1997). Mortality is attributed to a range of features including, inherited defects, egg quality, starvation, predation, and environmental variability such as changing physiochemical conditions. These events can result in variations in mortality during the larval period, which subsequently results in changes in the adult fish populations (Kelso and Rutherford 1996; Houde 1997; Trippel and Chambers 1997). The importance of understanding the variability in the abundances of early life stages has long been recognised in marine systems particularly for commercially important species. In contrast, freshwater studies have predominately focussed on understanding adult fish biology and ecology, with comparatively few investigating the early life history.

The relationship between food availability, larval survival and subsequent recruitment was first examined by Hjort (1914), who identified that larvae need to encounter high densities of appropriately sized prey to survive. Hjort (1914) found that high mortality rates occurred during the shift from endogenous to exogenous feeding, and termed this the *critical period* (May 1974). Differences in mortality rates during this time can have substantial implications for future year class strengths. The *match/mismatch hypothesis* (Cushing 1990) is a widely accepted model developed to explain the relationship between the timing of occurrence of fish larvae and their prey items (zooplankton). It was developed for temperate marine systems, where fish spawn during a fixed period, and zooplankton occurrence and abundance varies with environmental conditions (Cushing 1990). The match/mismatch hypothesis proposes that larval growth and survivorship relies upon the degree of temporal overlap between the spawning season and zooplankton occurrence (Figure 1.1) (Cushing 1990). In this framework, a match occurs when the peak abundances of both larvae and zooplankton overlap, thus resulting in a strong recruitment year (Figure 1.1a). In a mismatch the larval spawning season does not overlap with the peak abundance of zooplankton, resulting in a poor recruitment year (Figure 1.1b). The assumption is that fish spawning is consistently timed, but that the variation occurs in zooplankton abundances due to changing environmental variables such as temperature (Cushing 1990). While some marine studies have supported this hypothesis (e.g. Cushing 1990; Fortier *et al.* 1995; Gotceitas *et al.* 1996), others have rejected this hypothesis based on variation in the timing of fish spawning (e.g. Johnson 2000; Wright and Trippel 2009).

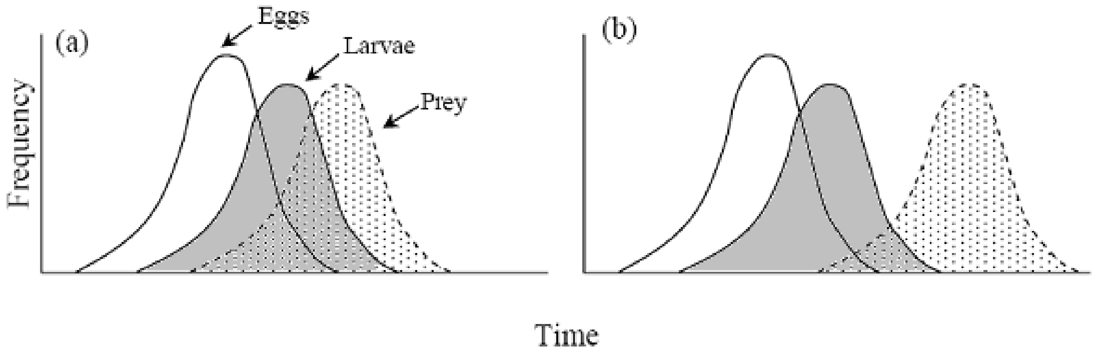

Figure 1.1. The match/mismatch hypothesis (adapted from Cushing 1990), where (a) is the match scenario and (b) is the mismatch scenario. The production of eggs, larvae and prey (zooplanktons) are shown as distributions in time.

For most species the highest mortality occurs during the larval phase; therefore, the progression of fish into juveniles is the most widely accepted definition of recruitment in fish ecology (Trippel and Chambers 1997). Food availability was initially accepted as the biggest driver for mortality (Houde 2002), but more recent studies have suggested that predation may have a more significant effect (Houde 1997), however it is still largely unknown and likely to vary through time and across species. Given the wide acceptance of the critical period and match/mismatch hypothesis, many fish recruitment models assume the most limiting factor during early life history is prey resources and other aspects such as growth and predator avoidance are subsequently linked to prey (Cushing 1990; Jobling 1995; Houde 1997). Consequently, the rates of larval growth, survival and recruitment are frequently linked to prey availability; and this has formed the basis for many recruitment models currently in use.

1.2 RECRUITMENT MODELS IN FLOODPLAIN RIVERS

The *flood pulse concept* is a riverine production model designed for large unaltered river systems with productive floodplains in the temperate, subtropical, or tropical regions (Junk *et al.* 1989). This model suggests that the majority of the primary production within rivers originates directly or indirectly from interactions with the floodplain and not from downstream transport of organic matter produced elsewhere in the basin (Junk *et al.* 1989). Thus, in the flood pulse concept the flux of nutrients occurs horizontally, rather than longitudinally as proposed in one of the other widely accepted river productivity models the *river continuum concept* (Vannote *et al.* 1980). The flood pulse concept proposes that the strength of fish recruitment is linked to prey availability as a function of floodplain inundation. In temperate river systems high temperatures and high flows must coincide for successful utilisation of the floodplain by fishes for spawning and recruitment. Thus emphasising the importance of the lateral connectivity between the river channel and the floodplain for primary production, and providing spawning cues, food and habitat for fish (Junk *et al.* 1989).

Observations from temperate Australian floodplain rivers have resulted in two models: the *flood recruitment model* (Harris and Gehrke 1994) and the *low flow recruitment hypothesis* (Humphries *et al.* 1999). The flood recruitment model was developed based on the principles of the flood pulse concept for Australian temperate rivers. The flood recruitment model proposes two methods by which increased flows may enhance recruitment in river fish; flooding as a direct stimulus for spawning of some species, or flooding indirectly enhances the survival of larvae and juveniles by providing suitable food and habitat on the inundated floodplain (Harris and Gehrke 1994). The main assumption is that the main river channel does not support sufficient densities of prey during low flow conditions (Lake 1967a; Rowland 1992; Harris and Gehrke 1994).

Of particular importance to the application of models such as the flood pulse concept and the flood recruitment model is the annual predictability of large overbank floods. However, periods of low flows are common throughout floodplain rivers. Consequently, a wide diversity of life history styles are exhibited by fish in these systems including those that utilise main channel habitats rather than the floodplain (Junk *et al.* 1989).

The low flow recruitment hypothesis (Humphries *et al.* 1999) pioneered the concept that main channel habitats were potentially important during the early life history of some fish species in Australian rivers. This model proposes that some fishes can successfully recruit in the main channel environment during low flows, by residing in food rich habitats, such as still, shallow littoral zones and backwaters. Furthermore, it emphasises that some fishes do not require high flows to initiate spawning. Particular species will use these main channel habitats under low flows over summer as these areas provide warm, low velocity habitats with suitable concentrations of appropriately sized prey (Humphries *et al.* 1999).

The key concepts behind the models are similar; flow regime is important for fish spawning and recruitment, and the mid river channel does not provide suitable habitat or abundant food, therefore specific habitats are required to ensure sufficient growth and survival of larvae (Junk *et al.* 1989; Harris and Gehrke 1994; Humphries *et al.* 1999). These models differ in that the flood pulse concept and the flood recruitment model highlight the importance of high flows to initiate spawning and provide floodplain habitats. Whilst the low flow recruitment hypothesis emphasises that some fish can spawn under low flows and that specific main channel habitats can provide necessary conditions for developing larvae. Importantly, it should be highlighted that these models do not contradict one another, but rather deal with the different features of the flow regime.

Whilst floodplain inundation has been recognised as important to the life cycles of many fish species (Welcomme 1985; Winemiller 2005) many others do not require inundated floodplain habitats for reproduction and are able to successfully spawn and recruit in main channel habitats (Haines and Tyus 1990; Turner *et al.* 1994; Watkins *et al.* 1997; King 2004b). Recent studies have demonstrated successful spawning and recruitment of many species in the main channel during low flow conditions (Humphries *et al.* 2002; King 2004b; Zeug and Winemiller 2008). Main channel habitats, such as littoral areas, backwaters and embayments, commonly enhance larval survivorship and subsequent recruitment (Haines and Tyus 1990; Tyus 1991; Sempeski and Gaudin 1995; Watkins *et al.* 1997; King 2004b).

1.3 THE MURRAY-DARLING BASIN

The Murray-Darling Basin is Australia's largest river catchment occupying a large region of south-eastern Australia (between 24 and 37 ° S and 138 and 153 ° E). The Basin covers an area of just over 1 million km^2 or 14 % of Australia's land area (Walker and Thoms 1993; Newman 2000) (Figure 1.2). The headwaters originate in the Great Dividing Range and flow westward as a complex system of streams and rivers ending at the Murray Mouth in South Australia. Most of the Basin is in semi-arid to arid climatic regions; consequently rainfall is low and evaporation rates are generally high. The Basins climate is characterised by highly variable annual rainfall, which can lead to a high variability in river flows, including severe flooding and drought conditions (Young 2001). The climatic conditions are complex, with four dominant climatic characteristics being proposed by Walker (2006b): the north is subtropical, the east is cool and humid, the south is temperate, with the intervening area being dry and hot. Annual discharge of the Basin is low with an annual discharge of 8,489 GL from 1894-1993 (Walker *et al.* 1995) which is equivalent to the daily discharge of the Amazon River (Young *et al.* 2001). However, discharge in the Basin has decreased in recent years, with current modelling suggesting that median annual discharge has decreased to approximately 2,900 GL (Eaton, J pers comm.).

Since 1857 flow regulation structures and storages (dams and weirs) have been constructed throughout the Murray-Darling Basin to mitigate large floods and retain water primarily for irrigation (Arthington and Pusey 2003). The rivers throughout the Basin are now primarily used as irrigation conduits, with most of the large water storages occurring in the headwaters. Approximately two-thirds of the water that would have originally reached the River Murray estuary (commonly called the Murray Mouth) is now diverted for irrigation each year (Crabb 1997).

The type of river regulation varies throughout the Basin, the Darling River is comparatively unregulated (although many of the Darling tributaries also have water storages), and the River Murray is quite heavily regulated. Lake Dartmouth on the Mitta Mitta River is the largest water storage in the catchment; whilst the Hume dam (upstream of Albury) is the largest storage on the River Murray, with another 13 weirs downstream. Ten of these weirs occur in the Lower River Murray (downstream of the Murray-Darling junction) making it the most heavily regulated region. The weirs form the river into deep, low velocity pool environments (Walker and Thoms 1993).

Flow regulation has impacted hydrology within the Murray-Darling Basin on three temporal scales: the flood pulse (days to weeks), the flow history (weeks to years), and the flow regime (decades or longer) (Walker *et al.* 1995). River regulation has impacted natural flow variability by reducing the frequency and duration of major flooding events and the magnitude of smaller within channel pulses, and by maintaining a relatively stable water level within the weir pools in the Lower River Murray (Maheshwari *et al.* 1995). Consequently longitudinal linkages within the channel, and lateral linkages with the floodplain have deteriorated (Walker 2006b).

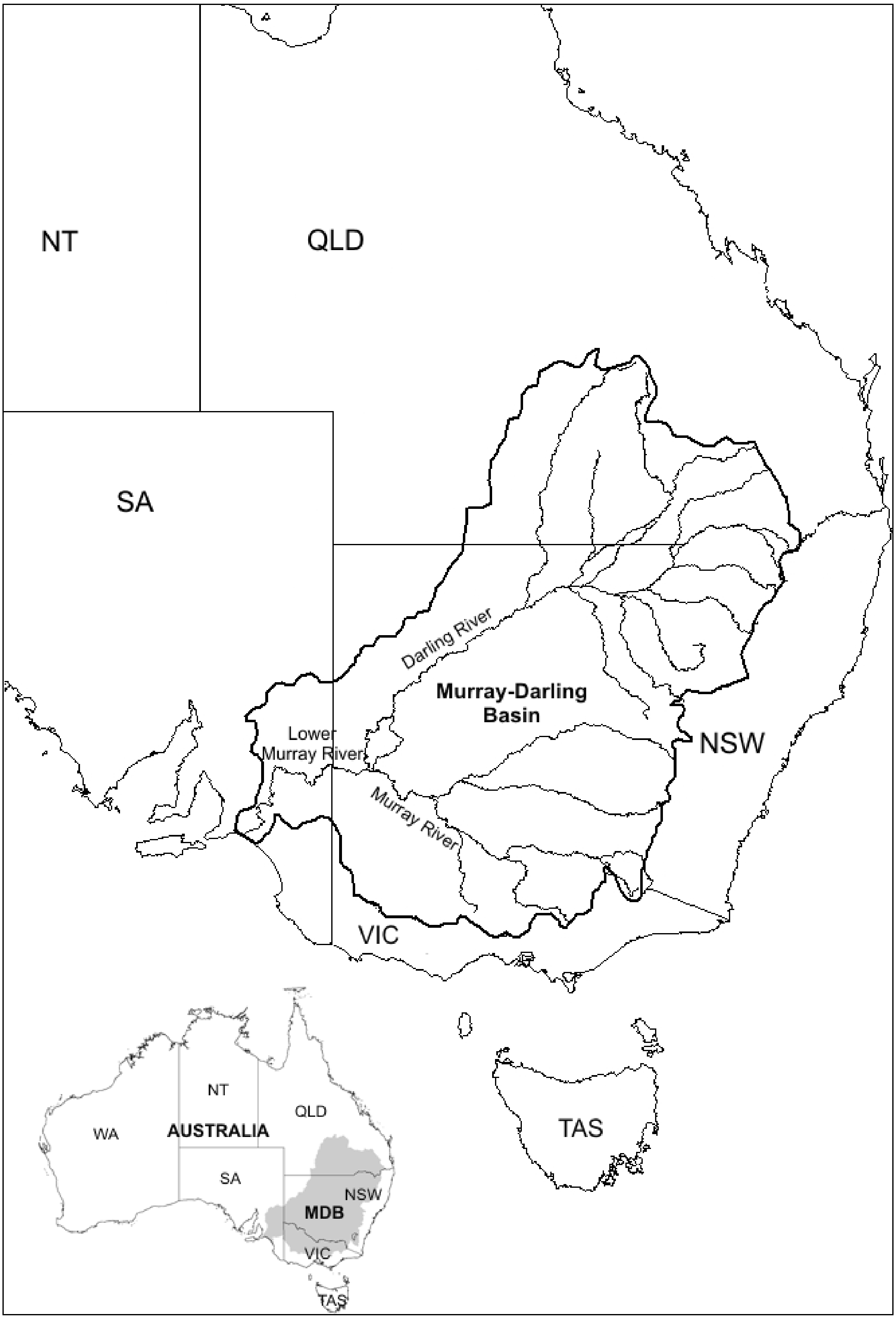

Figure 1.2. A map of the Murray-Darling Basin in Australia.

1.3.1 Murray-Darling Basin fish fauna

The freshwater fish fauna of Australia is commonly described as depauperate, with only about 300 species of native freshwater fish, of which 46 are recorded in the Murray-Darling Basin (McDowall 1988; Lintermans 2007). Comparatively, the Colorado River, shares a similarly impoverished fish fauna with roughly 32 species, while the Mississippi-Missouri system has more than 370 species, and the Amazon more than 1,300 species (Cadwallader and Lawrence 1990; Minkley 1991; Lintermans 2007). Geographical isolation, historically low rainfall, highly variable nature of historical flow and a high percentage of aridity are often implicated as reasons for Australia's low diversity freshwater fish community (Allen 1989; Cadwallader and Lawrence 1990; Lintermans 2007). The Murray-Darling Basin has a high percentage of introduced species, 12 have been recorded, some of which have been translocated from within Australia, while others are exotic to the continent (Allen 1989; Cadwallader and Lawrence 1990; Lintermans 2007).

Native fish populations in the Murray-Darling Basin have suffered severe declines in distribution and abundance following European settlement, although to-date no extinctions have occurred (MDBC 2004). However, some localised extinctions of fishes have occurred, and many species are considered threatened listed under state or national conservation listings (Lloyd and Walker 1986; Schiller and Harris 2001; MDBC 2004). Declines in the range and abundance of native fish in the Basin have been attributed to changes to the flow regime, construction of regulatory structures, removal and decrease of in channel habitat, over fishing, and the introduction of exotic species (Cadwallader 1977; Koehn and O'Connor 1990; MDBC 2004). Of all these factors, river regulation is thought to have had the most profound impact due to the substantial alteration of the flow regime (Cadwallader 1978; Gehrke *et al.* 1995; Humphries *et al.* 2002). The physical presence of dams, weirs and levees can limit access to suitable habitats, affect water quality, change the bank structure, act as fish passage barriers by obstructing movement and affect conditions for spawning, survival and dispersal of eggs and larvae (Cadwallader 1978; Gehrke *et al.* 1995; Humphries *et al.* 2002).

In March 2003, the Ministerial Council endorsed a basin-wide Native Fish Strategy (2002-2012) with the overall goal to "rehabilitate native fish communities in the Murray-Darling Basin back to 60% of their estimated pre-European settlement levels after 50 years of implementation" (MDBC 2004). A driving action of the Strategy is "fish habitat rehabilitation", which includes rehabilitation of degraded fish habitats and restoration of more natural flow regimes where possible. However, a greater understanding of the role of flow regimes in the life history of native fish is needed before management strategies can be effectively implemented.

Various attempts have been made to broadly group/classify the life history strategies of the Murray-Darling Basin fishes (e.g. Harris and Gehrke 1994; Humphries *et al.* 1999; Schiller and Harris 2001; King 2002; Growns 2004). Life history/reproductive strategies for Murray-Darling Basin fish have been developed based on the *opportunistic*, *equilibrium* and *periodic* strategies (*sensu* Winemiller 1989b; Winemiller and Rose 1992). Life cycle characteristics such as spawning cues, timing and duration of spawning period, egg development, larval feeding and parental care are used in the categorisation.

Humphries *et al.* (1999) classified the key species of the Murray-Darling Basin into four life cycle styles: Mode 1 (equilibrium), Mode 2 (periodic) and Mode 3a & b (opportunistic) (Table 1.1). Growns (2004) classified fish into five reproductive guilds using numerical analysis (Table 1.2). Schiller and Harris (2001) classified the species of the Murray Darling Basin to guilds based on the associations between floods and spawning (Figure 1.3). King (2002) presented a generalised conceptual model under both high and low flow conditions, which was later expanded in conjunction with the CRCFE (2003) (Table 1.3). The classifications differ in the grouping of species, primarily due to the aspects considered when developing the models, although inherent similarities are present. Humphries *et al.* (1999) developed a detailed classification considering aspects of the reproductive cycle including spawning cues, and egg and larval development as well as the influence of flow on breeding strategies. Growns (2004) employed numerical analysis of 13 life history traits including all aspects of the reproductive cycle, age at maturity, spawning migration and maximum size. These results were very similar to Humphries *et al.* (1999), but differed slightly because equal weighting was applied to all life history traits. In contrast, Schiller and Harris (2001) only used the associations between floods and spawning events, or influence of habitat to group the species, producing a much more generalised classification of the life cycles. King (2002) included the influence of flows and habitat type on spawning and recruitment success; this model differed to the others models as it emphasised the importance of larval survivorship and recruitment.

Table 1.1. Life cycle styles for key Murray-Darling Basin fishes (from Humphries *et al.* 1999).

Variable	Mode 1	Mode 2	Mode 3a	Mode 3b
Duration of spawning	Short	Variable	Long	Short
Spawning style	Single spawning, approx same time each year	Single spawning, timing, delay	Protracted, serial or repeat	Single spawning
Spawning time	October - December	October-March	September-March	Late winter or summer
Cues for spawning	Circannual rhythm and min. temp.	Rising water level (?) and min. temp.	Uncertain	Uncertain
Number of eggs	1,000's-10 000's	100 000's	100's-1, 000's	100's-1, 000's
Type of eggs	Demersal	Semi-buoyant or planktonic	Planktonic or demersal	Planktonic or demersal
Parental care of embryo/larvae	Yes	No	No	No
Incubation period	10+days	Hours	<10 days	<10 days
Size of embryo at hatching	6-9 mm	3-6 mm	3-4 mm	2-7 mm
Time to first feeding	ca. 20 days	ca. 5 days	ca. 3 days	ca. 3 days
Development of embryo/larva at first feeding	Advanced, large gape, well-formed fins, highly mobile	Undeveloped, small gape, limited mobility	Undeveloped, small gape, limited mobility	Undeveloped, small gape, limited mobility
Examples of species [1]	Murray cod, trout cod, freshwater catfish, river blackfish (*Gadopsis marmoratus*)	Golden perch, Silver perch	Australian smelt, flathead gudgeon	Carp gudgeon, Murray rainbowfish, Southern pygmy perch (*Nannoperca australis*)

Table 1.2. Guild definitions based upon numerical analysis (sourced from Growns 2004).

Guild	Definition
A	Adhesive, demersal eggs with no parental care
B	Low fecundity, small non-adhesive demersal eggs with short incubation times
C	Show parental care, including nest building and protection of young
C1	As in guild C but species display a spawning migration and have very large eggs and young
C2	As in guild C but species do not generally undergo a spawning migration and have large eggs
D	Single spawning species with high fecundity, non-adhesive eggs with no parental care
D1	As in guild D but undergo a spawning migration
D2	As in guild D but display no spawning migration
E	Low fecundity with long incubation of young associated with live bearing

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[1] Note: scientific names can be found in table 1.4 for key species.

Table 1.3. Spawning and recruitment strategies of Murray-Darling Basin fish species (from CRCFE 2003; King 2002).

Strategy	Species	Hydrology	Habitats
Flood spawners	Golden perch and silver perch	Spawn and recruit following flow rises. Major spawning events occur during periods of floodplain inundation	Main channel and anabranches
Wetland specialists	Australian smelt, bony herring, carp gudgeon, Southern pygmy perch, hardyhead, *Galaxias rostratus*	Spawn and recruit during in-channel flows	Floodplain wetlands and lakes, anabranches and billabongs
Main channel generalists	Australian smelt, bony herring, flathead gudgeons, redfin perch	Spawn and recruit in high or low flow	Main channel
Main channel specialists	Murray cod, trout cod, river blackfish, two-spined blackfish	Spawn and recruit under high or low flow	Main channel. Woody debris important habitat attribute
Low-flow specialists	Crimson-spotted rainbow fish, Carp gudgeon, gambusia	Only spawn and recruit during low flow	Main channel or floodplain habitats
Freshwater catfish	*Tandanus tandanus*	Spawn any flow conditions, recruitment needs further investigation	Builds nests in coarse sediment beds (usually sand or gravel)
Flood opportunists	Carp	Spawning under any conditions recruitment may be enhanced under higher flows	Main channel/ wetlands/ floodplain

NOTE:
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Figure 1.3. Reproductive guilds of native MDB fish species based on associations between floods and spawning (from Schiller and Harris 2001).

The influence of the flow regime, habitat requirements, and food availability for spawning and the early life stages of fish play a major role in all of the recruitment models and classifications of life history styles; although for many Murray-Darling Basin fish species further research is required (Table 1.4). Most of our understanding has been developed in the mid and upper Murray and some of the Murray tributaries (e.g. Lake 1967a, b; Humphries *et al.* 1999; Humphries and Lake 2000; Meredith *et al.* 2002; King 2004b, 2005). Furthermore, Humphries *et al.* (1999) highlighted that the relationships between fish recruitment and flows may potentially vary between different climatic regions. In addition, available habitats will depend on the geomorphology of the surrounding landscape. Many of these recruitment models and life history strategies remain un-tested in different regions of the Basin, where characteristics of the river channel, flow regime and level of regulation are often distinctly different.

Table 1.4. Status of available information on life history strategies, habitat associations and dietary preference for early life stages for Murray-Darling Basin fish species investigated in this study. CU, = comprehensive understanding, FR= further research required. References are noted numerically.

Common name (*Scientific name*)	**Life history strategies**	**Habitat associations**	**Dietary composition**
Australian smelt (*Retropinna semoni*)	CU [25, 35, 36, 37, 41]	CU [3, 4, 5, 8, 9, 17, 23]	CU [25, 26, 27]
Bony herring (*Nematalosa erebi*)	FR [8, 9, 14, 35, 41]	FR [3, 8, 9, 14]	FR
Carp gudgeon (*Hypseleotris* spp.)	CU [25, 35, 36, 37, 41]	CU [1, 3, 4, 5, 8, 9, 10, 11, 17]	FR [1, 10, 26, 33]
Flathead gudgeon (*Philypnodon grandiceps*)	FR [25, 35, 36, 37, 41]	CU [1, 3, 4, 5, 8, 9, 13, 17]	FR [25]
Hardyhead (*Craterocephalus* spp.)	FR [25, 35, 36, 41]	FR [3, 34]	FR [12]
Murray rainbowfish (*Melanotaenia fluviatilis*)	FR [25, 35, 36, 41]	CU [3, 4, 15]	FR [25, 26]
Murray cod (*Maccullochella peelii peelii*)	CU [25, 35, 36, 37, 38, 40, 41]	CU [3, 4, 5, 8, 9, 16, 17, 18, 19]	CU [25, 26, 29, 33, 39]
Golden perch (*Macquaria ambigua*)	FR [7, 25, 35, 36, 37, 41]	FR [2, 6, 8, 9, 11, 20]	FR [2, 28, 3, 392]
Freshwater catfish (*Tandanus tandanus*)	FR [25, 35, 36, 41]	FR [8, 9, 11, 21]	FR [35, 18]
Silver perch (*Bidyanus bidyanus*)	FR [7, 25, 35, 36, 37, 41]	FR [8, 11, 22, 24]	FR [28, 30, 31, 33]

1 (Gehrke 1992), 2 (Gehrke 1991), 3 (Meredith *et al.* 2002), 4 (Humphries *et al.* 2002), 5 (Humphries and King 2003), 6 (Gehrke 1990), 7 (Mallen-Cooper and Stuart 2003) 8 (Cheshire and Ye 2008), 9 (Leigh *et al.* 2008), 10 (Meredith *et al.* 2003), 11 (Lake 1967a, b) 12 (Wedderburn *et al.* 2007), 13 (Tredwell and Hardwick 2003), 14 (Puckridge and Walker 1990), 15 (Lake 1971), 16 (Humphries and Lake 2000), 17 (King *et al.* 2003), 18 (Koehn and Harrington 2005), 19 (Gilligan and Schiller 2004), 20 (Koehn and Nicol 1998), 21 (Clunie and Koehn 2001a), 22 (Clunie and Koehn 2001b) 23 (Cadwallader and Backhouse 1983), 24 (Geddes and Puckridge 1989), 25 (Humphries *et al.* 1999), 26 (King 2005), 27 (Lieschke and Closs 1999) 28 (Arumugam and Geddes 1987), 29 (Rowland 1992), 30 (Warburton *et al.* 1998) 31 (Merrick and Schmida 1984), 32 (Rowland 1996), 33 (McDowall 1996). 34 (Llewellyn 1971) 35 (Schiller and Harris 2001), 36 (King 2002; CRCFE 2003), 37 (King *et al.* 2007), 38 (Koehn and Harrington 2006), 39 (Tonkin *et al.* 2006), 40 (Humphries 2005), 41 (Growns 2004).

1.4 AIMS OF THE THESIS

Native fish populations are recognised as an indicator of ecological health within a riverine ecosystem (Karr 1981, 1991; Gatz and Harig 1993; Chaves and Alipaz 2007). To manage fish populations, a better understanding of the factors that govern population dynamics is required. The role of the flow regime, habitat use and dietary associations in the life cycle of fish in temperate floodplain rivers is continually being developed. Specifically, there is a need to understand the interactions between various flow components (e.g. low flow, within channel flows, flow pulse and overbank floods) and the interactions with fish spawning and larval survivorship. Although some work has been conducted, much more is needed, particularly in heavily regulated systems. A greater understanding of spawning and larval survivorship in response to low flows and flow pulses will benefit recovery of fish populations. The assumptions of the flood recruitment model and the low flow recruitment hypothesis are similar, suggesting that the mid river channel does not provide adequate habitat or prey densities for larval survivorship (Harris and Gehrke 1994; Humphries *et al.* 1999). The flood recruitment model suggests that some species require high flows as spawning stimulus and to provide adequate food and habitat on the inundated floodplain (Harris and Gehrke 1994). In contrast, the low flow recruitment hypothesis highlights that some species can successfully spawn and recruit during low flow years and that appropriate densities of food do exist in specific main channel habitats (Humphries *et al.* 1999).

In heavily regulated rivers such as the Lower River Murray, the river no longer resembles the historical and natural characteristics of the system. Therefore, it is important to understand whether the currently accepted recruitment models are applicable for management and restoration of native fish in these systems. Principally, this thesis aims to:

1. Compare gear types and diel catch variations in larval fish assemblages to determine the most suitable sampling method for the main channel of the Lower River Murray (Chapter 2),
2. Compare and contrast annual variation in larval fish assemblages between years with varied hydrologies (Chapter 3),
3. Describe seasonal variation in the spawning of key species under low flow conditions in the Lower River Murray (Chapter 4),
4. Describe zooplankton densities and dietary composition of Australian smelt in the mid river channel under low flow conditions (Chapter 5),
5. Compare and contrast the use of main channel habitat types (backwaters, open water, and still littoral zones) by larvae of key species during low flow conditions (Chapter 6).

1.4.1 Notes on chapter style

Each data chapter (Chapters 2 to 6) has been written in a style that can be read as a separate study. Tables and figures appear within the text and all references cited in this thesis are compiled at the end of the thesis not at the end of each chapter.

1.5 STUDY REGION

This study was conducted in the main channel of the Lower River Murray in South Australia. This region is located downstream of the Murray-Darling junction (Figure 1.4). Walker (2006b) described the Lower River Murray as a ‘distinctive environmental unit’; due to the combination of four distinct geomorphologic regions and compared to the rest of the river, the absence of significant tributaries, and the high degree of regulation.

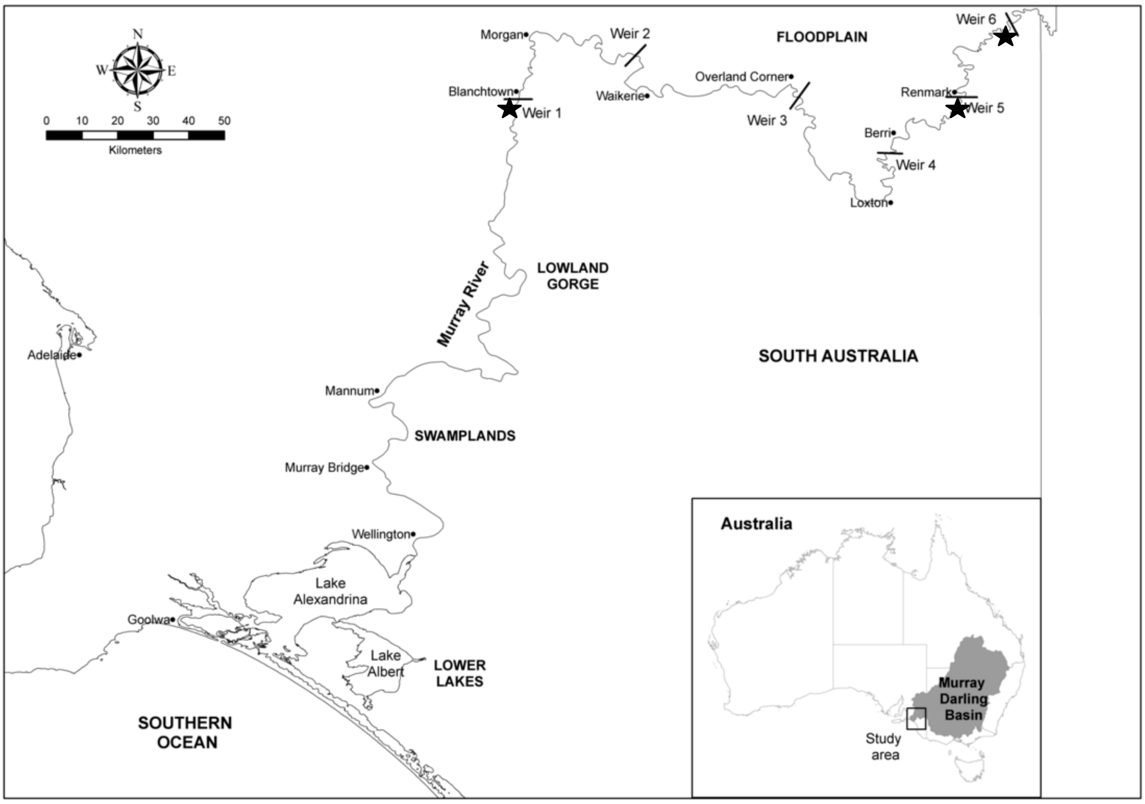

Figure 1.4. The Lower River Murray and geomorphic regions in South Australia, inset shows extent and position of the Murray-Darling Basin. Sampling sites are denoted by the (★).

The Lower River Murray consists of four distinct regions: *floodplain, lowland gorge, swamplands* and *lower lakes* (Figure 1.4). The floodplain region, occurs from the Murray-Darling junction to Overland Corner (near the town of Morgan), it comprises a wide floodplain (10-20 km) with a variety of aquatic habitats (Figure 1.5) including anabranches, extensive wetlands and woodlands (Walker and Thoms 1993; Young 2001). The lowland gorge, from Overland Corner to Mannum, is an incised section with limestone cliffs and a constrained floodplain (2-3 km wide) (Figure 1.6); wetlands are typically channel margin swales rather than oxbows (Pressey 1986). The swamplands region, from Mannum to Wellington, is bordered by reclaimed swamplands, now used for pasture and forage crops (Figure 1.7). The lower lakes region, so called due to the freshwater evaporation basins, Lake Alexandrina and Lake Albert, occurs from Wellington to the sea (Figure 1.8). The Murray Mouth (Figure 1.9) has been dredged since 2001 to avoid closure caused by depleted flow volumes and accumulated sediments.

Figure 1.5. The main channel typical of the floodplain region in the Lower River Murray, downstream of Weir 6, South Australia.

Figure 1.6. The main channel in the gorge region of the Lower River Murray, downstream of Weir 1, South Australia (photo courtesy of Jason Higham).

NOTE:
This figure is included on page 16
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Figure 1.7. The main channel in the swamplands region of the Lower River Murray, downstream of Mannum, South Australia (photo courtesy of the MDBA).

Figure 1.8. Aerial view of the Lower Lakes Region, South Australia in 2000 (photo courtesy of Paul Jennings).

NOTE:
This figure is included on page 17
of the print copy of the thesis held in
the University of Adelaide Library.

Figure 1.9. Aerial view of the Murray Mouth in 2004 (photo courtesy of the MDBA).

The Lower River Murray is heavily controlled by weirs, barrages, causeways, channelled diversions, off stream storages, and the influence of upstream dams. In South Australia, the river is regulated by the presence of six weirs (Figure 1.4). These structures have left almost no naturally free flowing river; with essentially the river upstream of Blanchetown now a series of isolated, slow flowing, deep weir pools (Walker 2006b). The impact of flow regulation structures escalates when many structures occur within close proximity, as occurs in the Lower River Murray region, potentially surpassing the effects of dams (Walker 2006b). River regulation has resulted in a significant reduction in total discharge, reduced seasonal variation in the discharge volume and water level, and the frequency, magnitude and duration of floods (Maheshwari *et al.* 1995; Walker 2006b). Furthermore, since 1996 there have been extreme dry conditions throughout most of the Basin, and inflows have dropped to record lows (Figure 1.10). Since 2002, the conditions have been defined as one of the most severe hydrological droughts recorded, with above average ambient temperatures also adding to the impacts (MDBC 2007; Murphy and Timbal 2008).

Water use throughout the Murray-Darling Basin is heavily regulated and minimum discharge entitlements for irrigation and human consumption are set for each state. Environmental water is allocated through a number of agencies within South Australia, however, these allocations are not used as discharge through the main channel but generally applied by pumping water to off channel habitats such as floodplains and wetlands (Gippel 2003). The discharge entitlements across the South Australian border are between 3,000 and 7,000 ML per day depending on the time of year, however, under current conditions these entitlements have not been met since 2006 (Figure 1.11). Consequently, longitudinal linkages within the channel and lateral linkages with the floodplain have been substantially reduced (Walker 2006b).

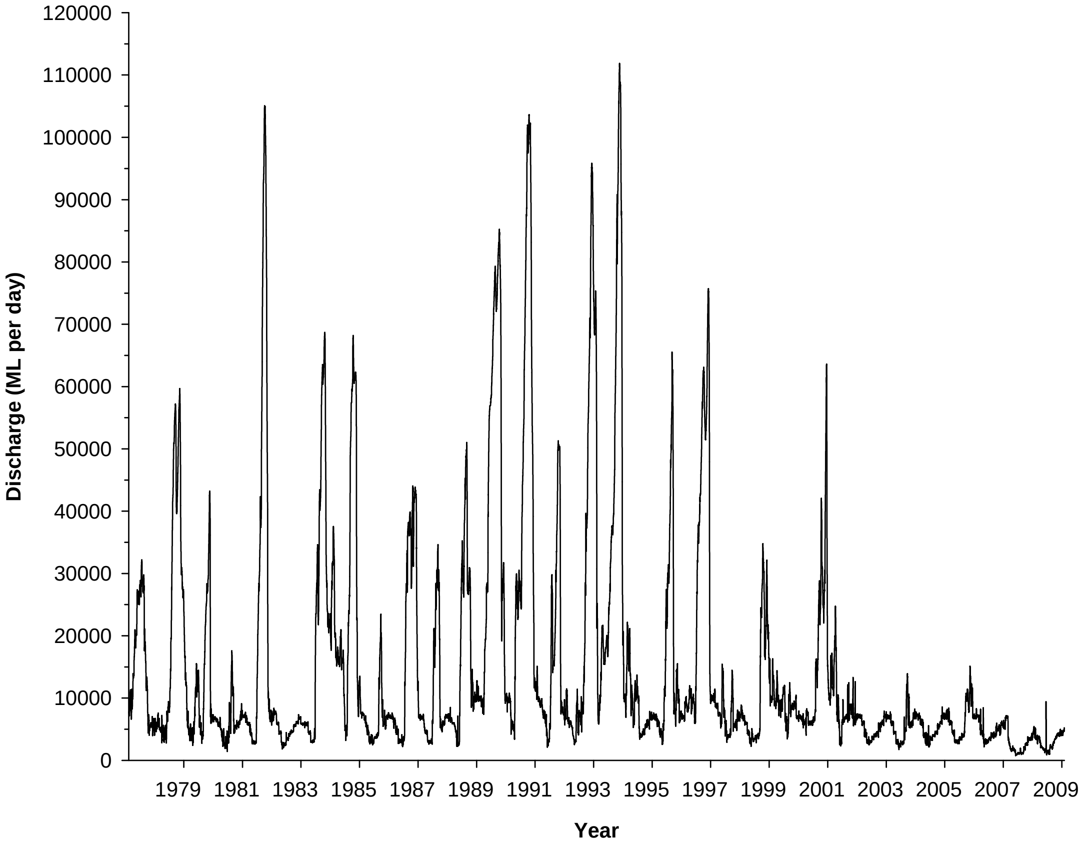

Figure 1.10. Discharge (ML per day) in the River Murray as measured at the South Australian border 1977 to 2009.

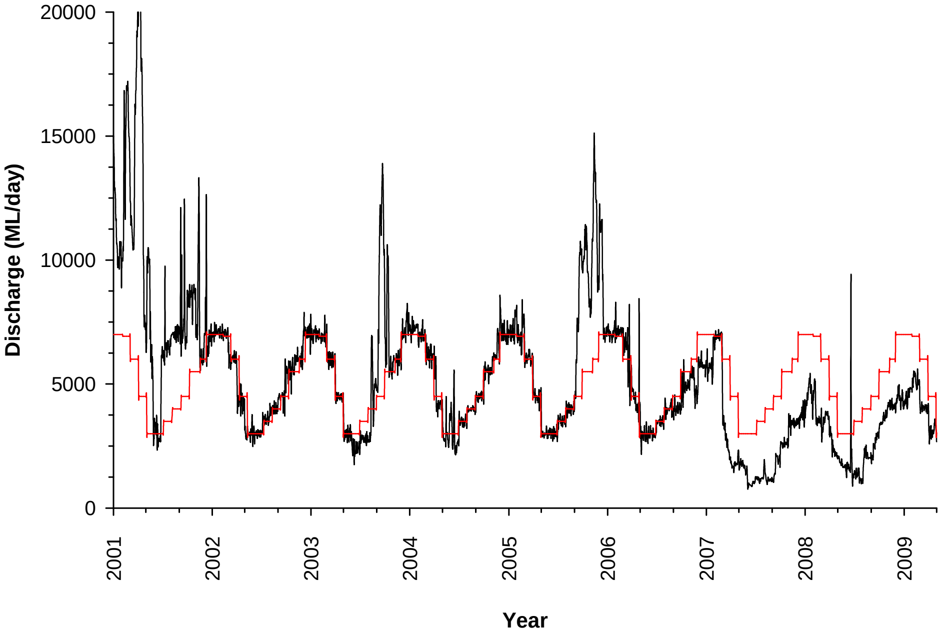

Figure 1.11. Discharge (ML per day) in the River Murray as measured at the South Australian border 2001 to 2009, entitlement allocations are presented in the red lines

CHAPTER 2: DETERMINATION OF DIEL VARIATION AND COMPARISON OF THREE GEAR TYPES FOR SAMPLING LARVAL FISH IN A REGULATED LOWLAND RIVER

ABSTRACT

Larval fish sampling is a useful technique for studying the early life history of fishes, however, there are biases associated with the sampling gear and an understanding of these will improve interpretation of results. The River Murray is a heavily regulated temperate river system and the main channel of the lower section is composed of a series of wide, deep, low velocity pools, with very little structural complexity. Larval fish assemblages were sampled using pelagic plankton tows (day and night), drift nets and light traps over two years, of differing hydrology in the main channel of Lower River Murray, South Australia. These sampling gears were compared to determine which gear type(s) most effectively sampled the fish community. Furthermore, day and night comparisons of the pelagic plankton tows were conducted to determine if species exhibited diel variation. Pelagic plankton tows were the single most effective method for collection of most species. Drift nets only collected larval fish in the higher discharge year. Diel variation was identified for many species; most exhibited higher abundances during the night although one species occurred in higher abundances during the day. A sampling regime which includes a diversity of gear types, and both day and night samples will sample the larval community in complex habitats more effectively, while a specific method and/or time may be most suitable when targeting a particular species or group of species.

2.1 INTRODUCTION

Sampling larval fish can help determine important biological information about fish assemblages, and ecologically or economically important species including the potential effects of environmental change on recruitment. Fish exhibit a wide range of characteristics including habitat associations and basic biology, particularly behaviour (Nelson 2006). Therefore, a diverse range of sampling gears has been developed. When targeting a fish assemblage rather than a particular species, it is unlikely that a single method will provide a full and absolute description of the fish assemblage present (e.g. Weaver *et al.* 1993; Brind'Amour and Boisclair 2004; Niles and Hartman 2007). Different habitats may also require different gear types to sample fish effectively, which inherently complicate quantitative comparisons between habitats (Kelso and Rutherford 1996; Rozas and Minello 1997; Niles and Hartman 2007). Gear types include: active gear types such as plankton tows, hand trawls, electrofishing and pump sampling; and passive gear types including drift nets, light traps and emergence traps (Kelso and Rutherford 1996), all with various advantages and disadvantages depending on the nature of the study (Table 2.1).

The susceptibility of a species to particular gear types will vary between species with differing early life histories as well as within species, depending on factors such as development and size. Gear selection requires consideration of the target species, sampling habitats and the overall objectives of the study; and is therefore one of the most important aspects of the experimental design in any study (Kelso and Rutherford 1996; Rozas and Minello 1997). Because of the vast potential for selectivity within each method a variety of gear types are often used in combination in an effort to achieve a relatively unbiased sampling protocol across the larval fish community (Kelso and Rutherford 1996).

When comparing community studies it is important for investigators to ensure results are not confounded by the ability for gears to collect particular species, as a function of differences in development at hatching, swimming ability, behaviour and microhabitat association. Furthermore, many researchers have demonstrated strong variations in diel abundance of the larval stage. Larval fish are frequently collected in greatest abundances during the night (Gale and Mohr 1978; Carter *et al.* 1986; Gehrke 1992; Gadomski and Barfoot 1998), although less common increased day time abundances have also been observed (Munk and Kiorboe 1985; Leis 1991; Haldorson *et al.* 1993; Brodeur and Rugen 1994). Diel patterns however, are often specific to individual species and can be complicated by gear avoidance and macrohabitat associations (Gallagher and Conner 1983; Holland and Sylvester 1983; Holland 1986; King 2004b). Very little work has been conducted in Australia on diel variation in larval stages of freshwater fish (although see Gehrke 1992; King 2004b). Studies into juvenile and adult populations have documented diel variation in assemblage composition (Baumgartner *et al.* 2008b). Diel variations in assemblage composition suggest that maintaining a consistent sampling time may be necessary to avoid confounding results, and that day and night sampling may be needed to represent the fish assemblage.

There are a wide range of gear types available for larval fish sampling within river systems (Gehrke 1992; Humphries *et al.* 2002; King and Crook 2002). Previous studies in Australian river systems have employed drift nets, fyke nets, plankton tows, light traps, sweep net electrofishing (SNE), hand trawl nets and pump sampling (Table 2.1). The Lower River Murray is a very distinct region. It is heavily regulated, resulting in the main channel being characterised by deep pools and low velocities. In addition very little work has been conducted on the larval assemblages in this area, therefore it is important to investigate a range of gear types and determine which are most appropriate for sampling assemblages of fish in a low flow river.

The main aim of this study was to compare and contrast the total abundance, species richness and assemblage structure collected by three different gear types; to identify the method, or combination of gear types, that effectively sampled the broadest range of species within the Lower River Murray region. Given the characteristics of the main channel and the larvae within the region pelagic plankton tows, drift nets and light traps were trialled. Second, diel variations for the assemblage and individual species were compared between day and night plankton tow samples, to determine if differences between day or night could influence the sampling regime.

Table 2.1. List of commonly used gear types for sampling larval fish in the River Murray, Australia, with a description of the method, advantages and disadvantages of each type and references.

Gear type	Description	Advantages	Disadvantages	References
Plankton tows (Active)	Often paired nets, constructed from nylon mesh (100-1000 µm), with removable collection jar at the end. Large mouth opening, between 0.4 - 1 m. Fitted with a flow meter in each mouth opening. Towed with a boat generally for a set period of time.	• Easy to use, day and night • Large sample volumes • Suitable for large deep areas (> 1m) • Not reliant on water flow • Quantitative (with flow meter)	• Cannot sample complex habitats • Nets clog easily with large amounts of debris • Increased processing time • Only used in open water, as difficult to navigate around structures • Potential for gear avoidance	**General:** Nesler (1988), Kelso and Rutherford (1996), Rozas and Minello (1997) **Australian use:** Humphries and Lake (2000), Humphries *et al.* (2002), Meredith *et al.* (2002), Koehn and Harrington (2005)
Hand trawls (Active)	Standard plankton net (100-500 µm mesh) with a removable collection jar, attached to a rope of set length. Smaller mouth opening than drift nets or plankton tows, generally <0.5 m. Can attach flow meters. Net thrown from shallow margins into deeper areas and quickly retrieved.	• Easy and quick to use, day and night • Suitable for shallow, more complex habitat areas • Not reliant on water flow • Numerous, small samples per site • Quantitative (with flow meter)	• Small sample area • Potential for gear avoidance • Some damage to fish • Labour and time intensive • Difficult to use around snags	**General:** Kelso and Rutherford (1996) **Australian use:** King *et al.* (2003), King (2004b, 2005)
Pump sampling (Active)	A centrifugal pump is used to sample a known volume of water with an intake hose positioned at a selected depth or depth integrated in the water column, into a mesh net or filtering system.	• Easy to use, day and night • Sample structurally complex areas • Not reliant on water flow • Quantitative	• Limited effective sampling area • Fish may be able to avoid intake hose • Damage to fish	**General:** Gale and Mohr (1978), Kelso and Rutherford (1996) **Australian use:** Gehrke (1992)
Electrofishing Sweep net (SNE) or point abundance (PA) (Active)	Electricity is pulsed into the water, immobilising fish that are then collected with sweep net. PA sampling uses small targeted samples with electrofisher, with net swept through stunned area. SNE has modification to anode pole or backpack, allowing sweeping action and electrofishing at the same time.	• Easy to use, day and night • Sample shallow, structurally complex habitats • Not reliant on water flow • Numerous, small samples per site • Not biased to species or development stage • Semi-quantitative (SNE timed, PA estimated volume)	• Restricted to wadeable depths • Small sample area per sample • Damage to fish	**General:** Copp (1989) King and Crook (2002) **Australian use:** King *et al.* (2003), King (2004b, 2005), Koehn and Harrington (2005)

Gear type	Description	Advantages	Disadvantages	References
Drift nets (Passive)	Stationary set of plankton nets (100-500 µm mesh), with removable collection jar at end. Set in flowing habitat to capture drifting eggs/larvae, mouth opening generally 0.5 m, fitted with flow meter. Nets are set for a standard amount of time (frequently over night) and retrieved.	• Easy to use, day and night • Horizontal and vertical sampling replication possible • Quantitative (with flow meters) • Sample large volumes of water	• Nets clog easily with large amounts of debris • Increased processing time • Reliant on significant water flow • Potential for significant damage to fish • Relies on drifting behaviour of fish	**General:** Franzen and Harbight (1992), Kelso and Rutherford (1996), Gilligan and Schiller (2004) **Australian use:** Humphries and Lake (2000), Humphries *et al.* (2002), Meredith *et al.* (2002), Humphries and King (2003), King *et al.* (2003), Gilligan and Schiller (2004), King (2004b, 2005), Humphries (2005), King *et al.* (2005), Koehn and Harrington (2005), Baumgartner *et al.* (2006), King *et al.* (2008a), Leigh *et al.* (2008)
Light traps (Passive)	Modified quatrefoil light trap, four plexiglass cylinders with a central light source, and collection sieve. Traps are set overnight and retrieved the next morning.	• Easy to use • Able to sample a wide range of habitat types (deep, shallow, complex, simple) • Collects species/ stages that exhibit positive phototaxis, and may not be collected by other methods	• Only effective at night • Mostly qualitative method, although can be quantified to time • Effective only in still or slow flowing habitats • Effectiveness decreases in highly turbid waters • Unknown amount of fish and invertebrate predation possible within the trap. Some authors suggest exclusion mesh but limits capture of larger fish • Species specific: (but known to capture wide range of Murray fish)	**General:** Floyd *et al.* (1984), Secor et al (1992), Gehrke (1994), Hernandez and Lindquist (1999), Niles and Hartman (2007), Porter *et al.* (2008) , Vilizzi *et al.* (2008) **Australian use:** Humphries and Lake (2000), Humphries *et al.* (2002), Meredith *et al.* (2002), Humphries and King (2003), Gilligan and Schiller (2004), Koehn and Harrington (2005), Leigh *et al.* (2008), Vilizzi *et al.* (2008)

2.2 MATERIALS AND METHODS

2.2.1 Study sites

The present study occurred in the main channel of the Lower River Murray in South Australia (Figure 1.4). The South Australian section of the River Murray is a heavily regulated lowland temperate river. The main channel is characterised by deep pools with low velocity. Sampling was conducted at three sites: in the tailwaters 5 km downstream of Weir 1 (Site 1, 34°21.138' S, 139°37.061' E), Weir 5 (Site 5, 34°13.246' S, 140°45.0909' E) and Weir 6 (Site 6, 33°59.725' S, 140°53.152' E) (Figure 1.4). Sampling occurred in the tail water reaches of each stretch of river. The area surrounding Site 1 is the gorge region, and the area surrounding Site 5 and Site 6 is the floodplain region (see Chapter 1 for a detailed discussion). Despite the surrounding characteristics of the floodplains being different, the main channel habitat is generally similar, being wide, deep, slow flowing pool habitats with very little connectivity between reaches.

2.2.2 Sampling regime

Larval fish were sampled during the austral spring/summers of 2005/06 and 2006/07. This sampling period was selected based on the peak spawning season and larval abundances suggested from other studies within the river system (Humphries *et al.* 2002; Meredith *et al.* 2002). Sampling was conducted fortnightly from September through December, and monthly in January and February (Table 2.2). Each site was sampled during the day and at night, of the same day, and all three sites were sampled within a four-day period each fortnight. During 2005/06 Site 1 was not sampled during the first trip in late September.

Table 2.2. Sampling trips and dates presented in the analyses and graphs[2].

Trip Number	Sampling week	Date presented in analyses/graphs
1	25-29 Sept	28 Sep
2	09-13 Oct	11 Oct
3	23-27 Oct	24 Oct
4	06-10 Nov	08 Nov
5	20-24 Nov	24 Nov
6	04-08 Dec	07 Dec
7	18-22 Dec	21 Dec
8	22-26 Jan	25 Jan
9	19-23 Feb	21 Feb

2.2.3 Collection and processing of fish larvae

Plankton tows were conducted using a set of paired square-framed (0.5 x 0.5 m) bongo nets with 500 μm mesh, and each net was 3 m long. Nets were equipped with 30 cm

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[2] Note all sampling days were used for analyses and but for aesthetics in graphs only one date from this period was presented.

pneumatic floats either side of the frame, which allowed the frame to be positioned 5 cm below the water surface. Nets were towed in circles behind a boat using a 20 m rope, in the centre of the main river channel. Three day and three night, 15-min tows were conducted at each site, with the aim of determining diel variation in catches with this method. This time period was chosen based on preliminary testing (Qifeng Ye, SARDI, unpublished data). The volume of water filtered through each net was estimated using a General Oceanics™ flow meter, fixed in each of the mouth openings. Catch was standardised to fish per 1000 m^3.

Modified quatrefoil light traps (Secor *et al.* 1992), (30 x 30 cm) constructed from perspex were used to target habitats that were not effectively sampled by plankton tows. A yellow 12 hour Cyalume® lightstick was used as the light source, and 5 mm stretched mesh was fitted to prevent predation by larger fish (Vilizzi *et al.* 2008). Three light traps were deployed at each site before sunset each afternoon, and retrieved in the same order before 24:00 hours of the same night. The length of sampling time (time in the water) for each trap was recorded to allow standardisation to fish per 6 hours.

Three drift nets were set just below the surface of the water in areas with suitable flow velocities. The 1 m long drift nets were constructed of 500 μm mesh, with a 0.5 m diameter opening fitted with a General Oceanics™ flow meter to determine the volume of water filtered. A small float was attached to the top of each net ensuring the mouth remained at the water surface. Nets were tied to immovable objects, deployed before sunset and retrieved the following morning before 08:00 hrs. Catch was standardised to fish per 1000 m^3.

Each sample was washed into separate buckets where fish were euthanased using high concentrations of clove oil. Samples were then preserved in 95% ethanol *in situ* and returned to the laboratory for sorting using magnification lamps and dissecting microscopes. All larvae were identified to species level where possible, using published descriptions (Lake 1967b; Puckridge and Walker 1990; Neira *et al.* 1998; Serafini and Humphries 2004) with the exception of carp gudgeon (*Hypseleotris* spp.) and hardyhead (*Craterocephalus* spp.). These two genera were treated as a species complex due to close phylogenetic relationships and very similar morphologies within the genus (Bertozzi *et al.* 2000; Serafini and Humphries 2004). It is most likely that the hardyhead collected in this study were the un-specked hardyhead (*Craterocephalus stercusmuscarum fulvus*) as the Murray hardyhead (*Craterocephalus fluviatilis*) are not found in the main channel environment (Wedderburn *et al.* 2007), however, as a precaution these species have been grouped. Each fish was categorised according to developmental stage, namely larvae (up to the development of caudal fin rays and pelvic fins forming) or juvenile/adult (rays in all fins fully developed) (Kelso and Rutherford 1996), but only larvae were included in the analyses.

2.2.4 Data analysis

The fish assemblage was characterised using four descriptors: total abundance, species richness (number of species), assemblage structure (standardised abundance of each species) and individual species abundances. Total abundance was calculated as the sum of the standardised abundance for all species collected per replicate. Standardised abundance of species for each method was calculated based on the catch, which was standardised to fish per 1000 m^3 of water (plankton tows and drift nets) or to fish per 6 hrs (light traps). For plankton tow data left and right nets were pooled for analysis following standardisation of fish per 1000 m^3.

Environmental variables and assemblage descriptors were tested for normality and homogeneity of variance. Given that very few variables met the assumptions, data were analysed using permutational analysis of variance (PERMANOVA) for univariate and multivariate data (Anderson 2001b). Prior to analysis the standardised larval assemblage data were fourth-root transformed. Transformation was not conducted on total abundance, species richness or individual species abundances. In all cases larval data were examined using Bray-Curtis similarity measures (Bray and Curtis 1957). Two three-way designs were developed for both univariate and multivariate analyses. The factors were *year, site* and *gear type,* or, *year, site* and *day/night* to analyse for differences between selected sampling gear types and diel variation, respectively. In all analyses *year* and *site* were treated as random factors while *gear type* or *day/night* were treated as fixed factors. Using both of the three-way designs univariate analyses were conducted for each of total abundance, species richness data and individual species abundances for those species that were collected in multiple gear types. Further multivariate analyses were performed using both three-way designs on the larval assemblage to determine if differences occurred as a function of *gear type* or *day/night.* Unrestricted permutations of data were performed for all analyses, with 999 permutations for the test to detect differences at $\alpha=0.05$ (Anderson 2001a). Where significant effects were detected pairwise analyses were performed to identify where the differences occurred.

2.3 RESULTS

2.3.1 Gear comparisons

Eleven species were collected throughout the study, comprising nine native and two exotic species. Overall, the small-medium bodied native species Australian smelt (*Retropinna semoni*), carp gudgeon (*Hypseleotris* spp.), flathead gudgeon (*Philypnodon grandiceps*), and bony herring (*Nematalosa erebi*), were the most abundant and dominated the larval catch (Table

2.3). Total species richness was higher in 2005/06 than 2006/07 due to the presence of golden perch (*Macquaria ambigua*) and silver perch (*Bidyanus bidyanus*) larvae (Table 2.3).

All species were recorded in plankton tows, whereas light traps and drift nets collected fewer species (Table 2.3). Six species were collected in light traps, comprising the small-medium bodied highly abundant species (Table 2.3). During 2005/06 drift nets caught larvae of five species; in contrast only one species was collected in 2006/07 and this was a single fish.

Table 2.3. Total raw number of fish larvae collected at all sites using drift nets, light traps, and day and night plankton tows (PT) during 2005/06 and 2006/07.

Species list	**2005/06**				**2006/07**			
Common name (*Scientific name*)	**Drift net**	**Light trap**	**Day PT**	**Night PT**	**Drift net**	**Light trap**	**Day PT**	**Night PT**
Australian smelt (*Retropinna semoni*)	348	285	902	1,047	0	4,949	6,700	7,762
Bony herring (*Nematalosa erebi*)	25	2	926	184	0	159	5,025	665
Carp gudgeon (*Hypseleotris* spp.)	113	333	1,619	860	0	930	2,972	2,149
Flathead gudgeon (*Philypnodon grandiceps*)	203	74	553	503	1	1,217	2,034	4,013
Hardyhead (*Craterocephalus* spp.)	0	25	3	4	0	158	0	20
Murray cod (*Maccullochella peelii peelii*)	1	0	1	8	0	0	0	8
Freshwater catfish (*Tandanus tandanus*)	0	0	1	20	0	0	0	8
Golden perch (*Macquaria ambigua*)	0	0	5	33	0	0	0	0
Silver perch (*Bidyanus bidyanus*)	0	0	7	3	0	0	0	0
Carp (*Cyprinus carpio*)	0	0	6	24	0	8	5	5
Redfin (*Perca fluviatilis*)	0	0	0	2	0	0	2	61
Total number of species	**5**	**5**	**10**	**11**	**1**	**6**	**6**	**9**
Total number of individuals	**695**	**724**	**4,033**	**2,699**	**1**	**7,427**	**16,744**	**14,700**

Total catch from plankton tows accounted for greater than 75% of the catch for individual species in both years with the exception of hardyhead (*Craterocephalus* spp.) which were captured in greater abundance in light traps (Table 2.4). Drifts nets were only effective during 2005/06 under increased flow but did not collect a large number of individual compared with tows (Table 2.4).

Table 2.4. Percent (%) occurrence of each species across key gear types: plankton tows (day and night combined), light traps and drift net samples for all sites combined during 2005/06 and 2006/07. NC indicates that no fish of this species were collected.

Species List	**2005/06**			**2006/07**		
Common name (Scientific *name*)	**Plankton tows (%)**	**Light trap (%)**	**Drift net (%)**	**Plankton tows (%)**	**Light trap (%)**	**Drift net (%)**
Australian smelt (*Retropinna semoni*)	75.5	11.0	13.5	74.5	25.5	0.0
Bony herring (*Nematalosa erebi*)	97.6	0.2	2.2	97.3	2.7	0.0
Carp gudgeon (*Hypseleotris* spp.)	84.8	11.4	3.9	84.6	15.4	0.0
Flathead gudgeon (*Philypnodon grandiceps*)	79.2	5.6	15.2	83.3	16.8	0.0
Hardyhead (*Craterocephalus* spp.)	21.9	78.1	0.0	11.2	88.8	0.0
Murray cod (*Maccullochella peelii peelii*)	90.0	0.0	10.0	100.0	0.0	0.0
Freshwater catfish (*Tandanus tandanus*)	100.0	0.0	0.0	100.0	0.0	0.0
Golden perch (*Macquaria ambigua*)	100.0	0.0	0.0	NC	NC	NC
Silver perch (*Bidyanus bidyanus*)	100.0	0.0	0.0	NC	NC	NC
Carp (*Cyprinus carpio*)	100.0	0.0	0.0	55.6	44.4	0.0
Redfin perch (*Perca fluviatilis*)	100.0	0.0	0.0	100.0	0.0	0.0

Total abundance and species richness differed between gear types, although the patterns were not consistent between years and sites (Figure 2.1; Table 2.5). There was a significant difference in total abundances between years (Figure 2.1a & b) due to variations in environmental parameters (see Chapter 3). Total abundance collected by light traps was consistently less than plankton tows in both years, and drift nets in 2005/06 (Figure 2.1a & b). During 2005/06 there was no significant difference between total abundance collected by plankton tows and drift nets. During 2006/07 there were significant differences in total abundance between all gear types at each site, with plankton tows collecting the greatest total abundance, followed by light traps (Figure 2.1b). Species richness in 2005/06 was significantly greater in plankton tows than all other gear types at all sites; in addition drift nets at Site 5 and Site 6 also collected greater number of species than light traps (Figure 2.1c).

The only difference detected in species richness during 2006/07 was between drift nets and both light traps and plankton tows as drift nets only collected one species (Figure 2.1d). The assemblage structure differed by both method and year although not independently (Table 2.5). *Post hoc* tests indicated that significant inter-annual variation was detected for each of the gear types although the magnitudes of differences were not consistent between years.

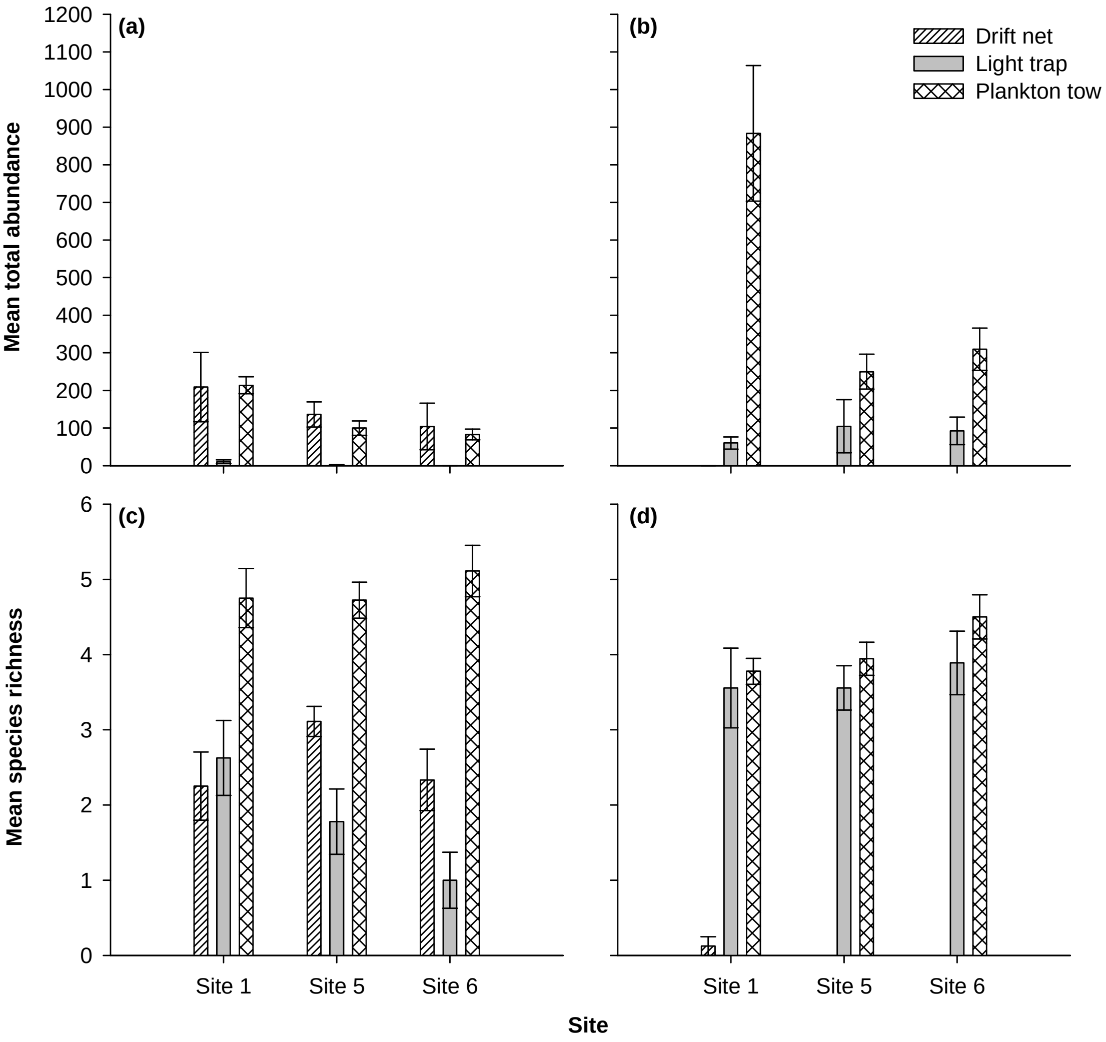


Figure 2.1. Comparison of mean total abundance ± standard error during (a) 2005/06 and (b) 2006/07 and mean species richness ± standard error during (c) 2005/06 and (d) 2006/07 at Sites 1, 5 and 6. Data are the standardised abundance of fish per 1000 m^3 of water filtered for drift nets and plankton tows and fish per 6 hrs light traps.

Table 2.5. Three-way univariate (total abundance and species richness) and multivariate (assemblage structure) PERMANOVA for differences in catch rates among year, site and gear type. Bold text indicates significant value.

		Total abundance		Species richness		Assemblage structure	
Source of variation	***df***	**MS**	***p***	**MS**	***p***	**MS**	***p***
Year	1	7952.1	0.155	5282.1	0.106	7642.9	0.061
Site	2	2385.8	0.520	348.6	0.697	1119.4	0.726
Gear Type	2	77154.0	0.416	32434.0	0.306	63659.0	0.186
Year x Site	2	3086.9	**0.029**	547.1	**0.046**	1956.5	**0.014**
Year x Gear Type	2	64181.0	**0.002**	20325.0	**0.001**	38449.0	**0.001**
Site x Gear Type	4	4952.8	0.432	442.7	0.525	1755.0	0.167
Year x Site x Gear Type	4	4278.5	**0.001**	484.0	**0.029**	985.9	0.138
Residual	193	1199.4		195.2		686.9	

Only Australian smelt, bony herring, carp gudgeon, and flathead gudgeon were collected using all three gear types (Table 2.3), therefore individual species analyses were restricted to these four species (Figure 2.2). Individual species abundances showed significant year-gear type interactions (Figure 2.2; Table 2.6). During 2005/06 the abundances of Australian smelt and carp gudgeon collected were similar between plankton tows and drift nets (Figure 2.2a & c). Flathead gudgeon were collected in greater abundances in drift nets (Figure 2.2d) and bony herring were collected in greater abundances in plankton tows (Figure 2.2b). In 2006/07 plankton tows collected significantly greater abundances of all species than light traps (Figure 2.2).

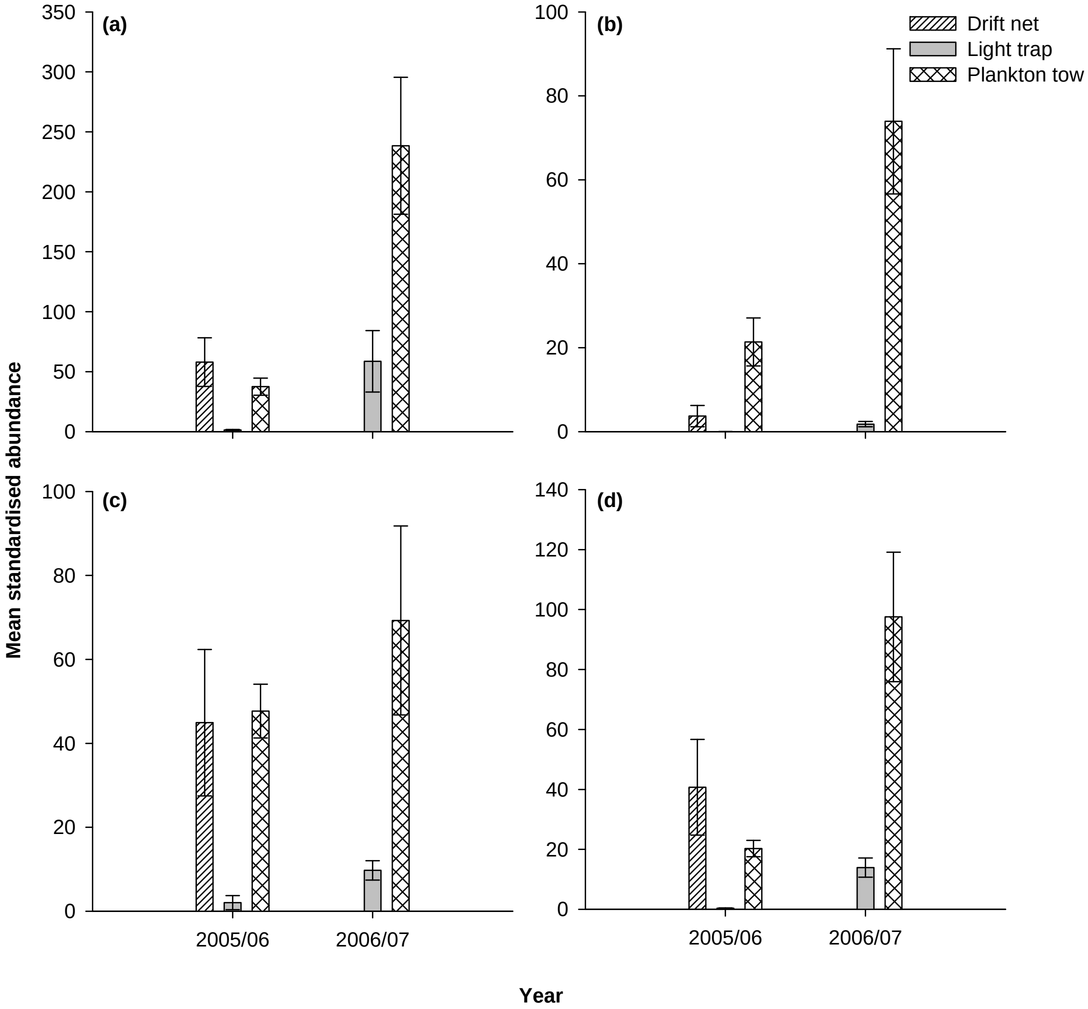


Figure 2.2. Comparison of mean abundance ± standard error of individual species between drift nets, light traps and plankton tow sampling during 2005/06 and 2006/07 for (a) Australian smelt, (b) bony herring, (c) carp gudgeon and (d) flathead gudgeon. Data are the standardised abundance of fish per 1000 m^3 of water filtered for drift nets and plankton tows and fish per 6 hrs light traps.

Table 2.6. Three-way univariate PERMANOVA for differences in collection of individual species among year, site and gear type. Bold text indicates significant value.

		Australian smelt		Bony herring		Carp gudgeon		Flathead gudgeon	
Source of variation	*df*	**MS**	*p*	**MS**	*p*	**MS**	*p*	**MS**	*p*
Year	1	3674	0.182	90	0.911	3609	0.231	480	0.610
Site	2	874	0.596	492	0.551	758	0.571	422	0.728
Gear Type	2	29117	0.179	34309	**0.018**	36966	0.166	35731	0.252
Year x Site	2	1172	0.146	506	0.458	1136	**0.027**	1015	**0.029**
Year x Gear Type	2	12783	**0.003**	3747	**0.036**	15959	**0.001**	19902	**0.004**
Site x Gear Type	4	843	0.301	1208	0.219	457	0.316	926	0.365
Year x Site x Gear Type	4	452	0.627	494	0.544	288	0.401	671	0.053
Residual	193	637		615		302		295	

2.3.2 Diel comparisons (plankton tows only)

Total abundance showed a significant year x site x day/night interaction (Table 2.7). Total abundance was significantly greater during the day in 2005/06 at Site 6 and significantly greater during the night in 2006/07 at Site 1. There was no significant effect of day/night sampling on species richness, although 2005/06 had higher species richness (Table 2.7). There was a significant diel difference in the assemblage structure of larval fish in 2005/06 but not during 2006/07 (Table 2.7).

Table 2.7. Three-way univariate (total abundance and species richness) and multivariate (assemblage structure) PERMANOVA for differences among year, site and day/night. Bold text indicates significant value.

		Total abundance		Species Richness		Assemblage structure	
Source of variation	***df***	**MS**	***p***	**MS**	***p***	**MS**	***p***
Year	1	24916.0	0.054	1682.4	**0.009**	4405.6	**0.009**
Site	2	8624.4	0.323	324.6	0.211	2102.3	0.244
Day/Night	1	4659.8	0.132	2272.1	0.065	3040.5	0.123
Year x Site	2	5823.0	**0.001**	35.2	0.828	1117.9	0.074
Year x Day/Night	1	650.3	0.852	75.0	0.565	1104.0	**0.025**
Site x Day/Night	2	2222.2	0.566	243.1	0.391	625.1	0.096
Year x Site x Day/Night	2	2577.9	**0.035**	170.7	0.300	279.8	0.867
Residual	94	997.2		140.6		584.6	

Abundances of several species individually differed significantly between day & night, although for the majority of species year and/or site were also a significant factor (Figure 2.3; Table 2.8). Bony herring was the only species that differed significantly only between day and night, occurring in greater abundances during the day (Figure 2.3b; Table 2.8). Australian smelt showed a significant difference between day and night but only at Site 6 in 2006/07 where abundances were greater in the day (Figure 2.3a) with the main differences occurring between sites and years (Figure 2.3a; Table 2.8). Hardyhead showed a significant difference between day and night, but this was only during 2006/07 at Site 6 where greater abundances occurred at night (Table 2.8; Figure 2.3c). Golden perch and carp (*Cyprinus carpio*) also showed significant differences between day and night samples, although only in 2005/06 (Figure 2.3c & d; Table 2.8). No significant differences in abundance between day and night tows were found for carp gudgeon, flathead gudgeon, Murray cod (*Maccullochella peelii peelii*), freshwater catfish (*Tandanus tandanus*), silver perch or redfin perch (*Perca fluviatilis*) (Table 2.8), although the total numbers of individuals caught for many of these species were very low (Table 2.3).

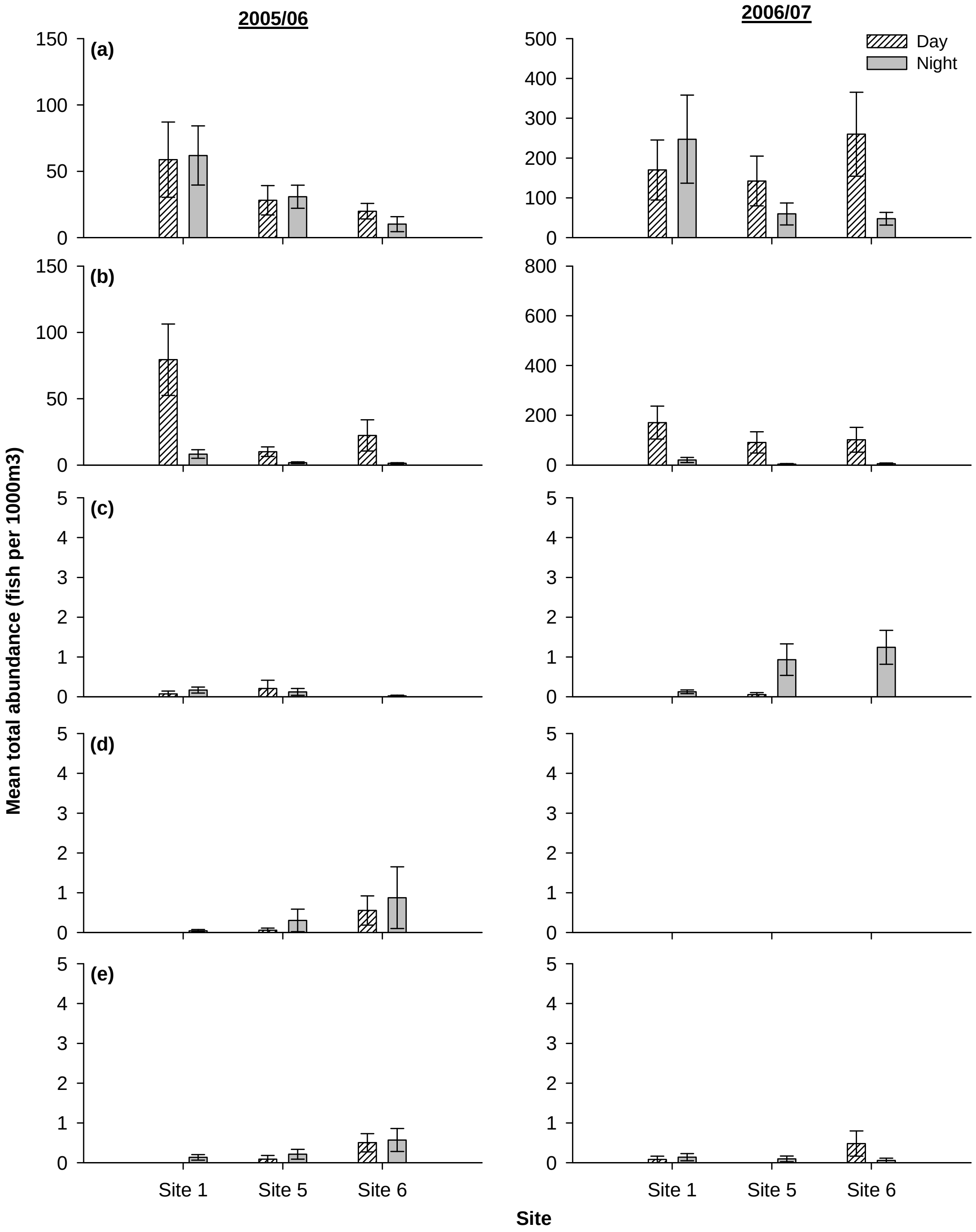


Figure 2.3. Comparison of mean abundance ± standard error of individual species between day and night plankton tows during 2005/06 and 2006/07. For (a) Australian smelt, (b) bony herring, (c) hardyhead, (d) golden perch and (e) carp at Sites 1, 5 and 6. Data are the standardised abundance of fish per 1000 m^3 of water filtered.

Table 2.8. Three-way univariate PERMANOVA testing for differences among year, site and day/night for individual species using standardised plankton tow data (fish per 1000 m^3). Bold text indicates significant value.

		Australian smelt		Bony herring		Carp gudgeon		Flathead gudgeon		Hardyhead		Murray cod		Freshwater catfish		Golden perch		Silver perch		Carp		Redfin perch	
Source of variation	*df*	**MS**	*p*	**MS**	*p*	**MS**	*p*	**MS**	*p*	**MS**	*p*	**MS**	*p*	**MS**	*p*	**MS**	*p*	**MS**	*p*	**MS**	*p*	**MS**	*p*
Year	1	2482	**0.025**	1484	0.392	1533	**0.024**	2356	0.337	151	0.890	329	0.418	484	0.149	2541	0.070	2119	0.143	2982	0.055	1717	0.198
Site	2	868	0.124	3443	0.102	745	0.053	550	0.786	70	0.953	76	0.823	139	0.343	312	0.490	656	0.587	1417	0.156	769	0.411
Day/Night	1	431	0.968	3621	**0.007**	234	0.864	1256	0.268	2596	0.154	1456	0.054	1867	0.076	531	0.514	28	0.535	1520	0.625	682	0.520
Year x Site	2	111	0.955	1241	0.266	97	0.786	1421	**0.005**	1339	**0.007**	339	0.185	111	0.604	312	0.183	656	**0.012**	394	0.330	506	0.075
Year x Day/Night	1	1627	**0.004**	79	0.453	630	0.095	570	0.488	525	0.517	83	0.411	198	0.214	531	**0.030**	28	0.371	2127	**0.014**	599	0.082
Site x Day/Night	2	809	**0.014**	248	0.190	455	0.135	641	0.438	500	0.641	82	0.544	80	0.406	14	0.546	21	0.527	368	0.095	501	0.104
Year x Site x Day/Night	2	75	0.972	90	0.968	117	0.748	588	0.046	804	**0.036**	90	0.605	60	0.774	14	0.954	21	0.849	38	0.894	68	0.743
Residual	94	720		926		250		237		236		188		211		193		152		343		214	

2.4 DISCUSSION

The Lower River Murray in South Australia has 24 species of native freshwater fish and 11 introduced (McDowall 1996; Allen *et al.* 2002; Hammer and Walker 2004). However, only 11 native and four introduced have been commonly recorded as adults in the main channel in the study area (Davies *et al.* 2008; Zampatti *et al.* 2008). Of these species only four species were not recorded as larvae in this study; these were native Murray rainbowfish (*Melanotaenia fluviatilis*) and dwarf flathead gudgeon (*Philypnodon* sp.) and introduced species, *Gambusia* (*Gambusia holbrooki*) and goldfish (*Carassius auratus*). It is unclear why these species were not collected, it may be due to misidentification, gear avoidance or potentially too low numbers to be effectively collected. However, the gear types used in the study generally represented the majority of the fish in the community, although there were clear variations in the assemblage composition collected by the different gears.

Plankton tows collected the highest diversity and abundance of larvae, providing the most effective method for collection in the main channel environment. Plankton tows collected the full suite of species sampled as larvae across all three sites, while light traps and drift nets collected a reduced number of species. Hardyhead was the only species to be collected in greater numbers in light traps and drift nets. Although drift nets only captured larval fish in the higher discharge year (2005/06), discharge volume and in-stream velocities during the regulated year (2006/07) were too low for this method to be effective. These results indicate that in years of average to below average flows drift nets are not a suitable method for sampling the main channel in South Australia. This study has demonstrated that in heavily regulated systems with wide, deep channels and low flow velocities, plankton tows are the most effective sampling gear and a combination of plankton tows and light traps will provide a comprehensive picture of the larval community. Furthermore, to ensure adequate representation of the larval assemblage samples should be taken both day and night, although night sampling will account for most species in reasonable abundances.

Gear variation is most likely attributable to differences in environmental factors and behavioural characteristics. Drifting species (after, Humphries and King 2003), such as, Murray cod, Australian smelt, flathead gudgeon and carp may be more readily collected using plankton tows and drift nets as they sample the pelagic environment. Non-drifting species often prefer structurally complex areas and are often associated with edge habitats, and therefore are likely to be more easily collected in the light traps. Light traps are also capable of sampling phototactic species, which may be hard to collect in high abundances with other gear types. These results are consistent with previous studies where behavioural characteristics of the larvae such as movement patterns, microhabitat associations, and swimming ability have influenced gear variation (Gehrke 1994; Rooker *et al.* 1996;

Hernandez and Lindquist 1999; Nabil *et al.* 1999; Humphries *et al.* 2002; Lindquist and Shaw 2005).

Bias associated with gear types is well documented in larval fish studies most often resulting in size and species selectivity. Both passive and active sampling gear types will inherently have bias associated with their use (e.g. Kelso and Rutherford 1996; Hernandez and Lindquist 1999; Lindquist and Shaw 2005), however an understanding of these biases can allow users to be more confident in their approach to sampling. Environmental factors including habitat complexity, water depth, water velocity, lunar phases, and turbidity can affect the physical ability of the gear to collect larvae. Light traps are able to sample more diverse habitats than plankton tows, and are more suitable for structurally complex areas. Drift nets however, rely on medium to high velocity water to work effectively. Plankton tows effectively sample open water areas where other gears cannot, however, there is also potential for gear avoidance by fish with enhanced swimming ability. Plankton tows appear to be particularly effective in low flow main channel regions, as there is no minimum requirement for velocity and they can effectively sample large deep areas (Kelso and Rutherford 1996; Rooker *et al.* 1996; Hernandez and Lindquist 1999; Nabil *et al.* 1999; Humphries *et al.* 2002; Lindquist and Shaw 2005).

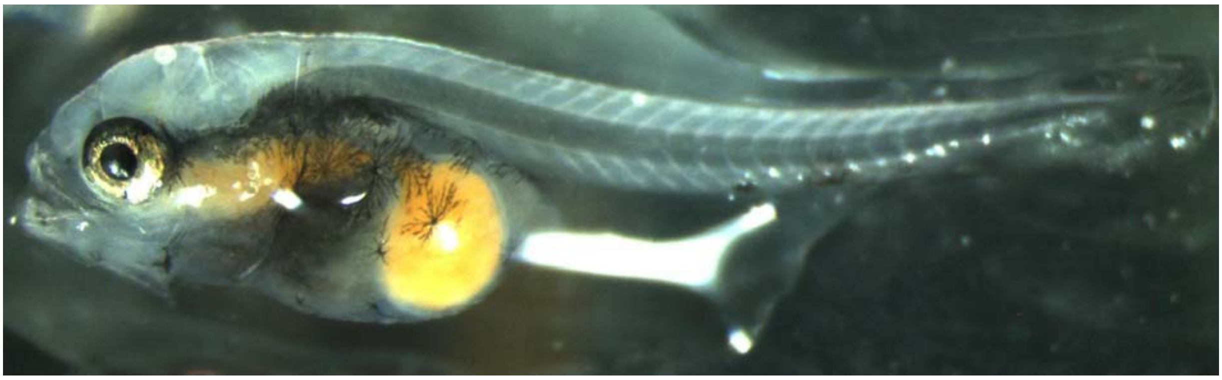
Diel variation in catch rates of larvae has been consistently documented in river systems around the world, with larvae generally being collected in higher abundances at night (Gale and Mohr 1978; Gallagher and Conner 1983; Gehrke 1992; Gadomski and Barfoot 1998). Total abundance, assemblage composition and abundance of individual species were all identified to show diel variation, although this was not consistent between years due to significant variation in the assemblage composition. Most species that showed significant diel variation were present in higher abundances during the night sampling, with the exception of bony herring. Interestingly, these diel patterns differ from those identified for the majority of species as adults within the same region (Baumgartner *et al.* 2008b). Diel variation in the larval stage is likely to differ from the adult stage as a result of feeding behaviour and physical development, and may be due to selective drift, or vertical migrations in the water column. Diel variation is also a function of species specific behavioural and environmental characteristics, particularly for species that can strongly regulate their movement as larvae such as Murray cod (Humphries *et al.* 2002; Humphries and King 2003; Humphries 2005). Increased abundance at night is often attributed to reduced avoidance of sampling gear, loss of visual orientation and rheotactic response, diel shifts in vertical migrations to avoid predators and ultraviolet radiation, and synchronicity with prey items (Muth and Schmulbach 1984; Carter *et al.* 1986; Corbett and Powles 1986; Gehrke 1992; Forrester 1994). In Australian billabongs avoidance of sampling gear and ontogenetic shifts in vertical migration have been attributed to diel patterns observed in carp gudgeon and flathead gudgeon larvae (Gehrke 1992). These patterns were suggested to be linked to the visual avoidance of gear during the day by more developed larvae, visual avoidance of predators by less developed larvae at night and a synchronicity between larvae and prey items (Gehrke 1992). However, most of the theories surrounding diel patterns require further research for specific species, particularly in Australia.

Some species do not exhibit significant diel variation (e.g. Muth and Schmulbach 1984), whilst some oceanic and coastal water species occur in higher abundance in surface waters during the day (e.g. Munk *et al.* 1989; Leis 1991; Sabates 2004). However, very few riverine species have been found to have greater larval abundances during the day, with the exception of the early developmental stages of carp gudgeon and flathead gudgeon larvae (Gehrke 1992). This study found that larval bony herring consistently occurred in greater abundances during the day. Bony herring have been collected in other studies in the Lower River Murray region, however, these studies did not conduct daytime sampling and therefore can not be used for comparison (Vilizzi *et al.* 2007; Leigh *et al.* 2008). Bony herring are from the Clupeidae family, the Atlantic herring (*Clupea harengus harengus*) a marine clupeid, has also been documented to occur in greater abundances as larvae in surface waters during the day. Atlantic herring larvae are visual feeders, having demonstrated a minimum light intensity for feeding (Batty 1987), thus, the day time vertical migration is thought to be a function of light intensity and food availability (Munk and Kiorboe 1985; Munk *et al.* 1989). Given similar lineage, this may explain why bony herring is the only species to occur in higher larval abundances during the day. In the Macintyre River, Australia adult bony herring showed an increase in feeding activity at midday (Medeiros and Arthington 2008), supporting the notion the greater larval abundances during the day may be driven by food. This potential niche specialisation may allow bony herring to reduce both competition and predation threats, and may have contributed to the successful nature of this species throughout the Basin, and should be investigated further.

This study has demonstrated that bias occur as a result of the sampling regime, specifically, gear type and timing of sampling. Most species were collected in higher abundances at night or showed no diel variation. One species, however, occurred in higher abundances during the day. A sampling regime which includes a diversity of gear types and day and night sampling will more effectively sample the larval community in complex habitats, while a specific method and/or time may be most suitable when targeting an individual species, or group of species. While it is not always appropriate for studies to conduct preliminary investigations into gear and diel variation, an understanding of how these factors may influence results will be beneficial.

Murray cod (*Maccullochella peelii peelii*) postflexion larvae.

Freshwater catfish (*Tandanus tandanus)* metalarvae (photo courtesy Sandra Leigh).



Golden perch (*Macquaria ambigua*) preflexion larvae (photo courtesy Simon Westergaard).

CHAPTER 3: VARIATION IN LARVAL FISH ASSEMBLAGES IN A HEAVILY REGULATED RIVER DURING DIFFERING HYDROLOGICAL CONDITIONS

ABSTRACT

Freshwater flows and flooding influence spawning and recruitment of many riverine fish, yet natural flows have been drastically altered through extensive river regulation. Consequently, many native fishes have declined in range and abundance in response to changes to the flow regime. The combined effects of drought and river regulation are likely to have drastic impacts on fish spawning and the survival of larvae. This study investigates the larval assemblages in a heavily regulated lowland temperate river system across varying annual hydrological and environmental variables. Larval fish were sampled in the Lower River Murray during the spring/summer period of 2005 under increased flow and a water level raising, and 2006, 2007 and 2008 under low regulated flow with stable water levels. The main river channel supported larvae from 11 species and a wide range of life history strategies. Three responses to varying hydrology were identified in the larval assemblage: larvae that were 1) positively correlated to increased flow, 2) negatively correlated to the increased flow and 3) correlated to temperature. The majority of small-medium bodied native species spawned under low flow conditions, while a flow pulse of even a small magnitude was required to induce spawning in two species previously defined as flood-cued spawners. This study suggests that in heavily regulated floodplain rivers flow pulses may provide a useful management and restoration strategy particularly during otherwise stable low flow situations.

3.1 INTRODUCTION

Many river systems around the world have been heavily modified to serve the needs of an ever-growing human population. Indeed, many of the worlds river systems are now so regulated that the river flows below dams and weirs are not in any way representative of the natural flow regime within the system (Richter *et al.* 2003). River regulation is so extensive across the world that nearly two-thirds of the planet's largest rivers are fragmented by dams and diversions(Revenga *et al.* 2000); and more than 800,000 dams block flow (Rosenberg *et al.* 2000). Whilst regulating rivers provide benefit for humans, it has caused considerable ecological damage (Baron *et al.* 2002).

River regulation impacts on natural flow regimes by reducing the frequency and duration of major flooding events, the magnitude and frequency of smaller within channel flows, and by maintaining a relatively stable water level (Maheshwari *et al.* 1995; Richter *et al.* 1996). This change to a system dominated by low flows, with unseasonal higher flow periods, and a lack of natural drying events has been termed the *anti-drought* (McMahon and Finlayson 2003). *Hydrological drought* in aquatic ecosystems, is most frequently a term applied to a '…low-flow period, which is unusual in its duration, severity or intensity…' (Humphries and Baldwin 2003; p. 1142). In most areas drought is a natural phenomenon, for which many plants and animals are adapted (McMahon and Finlayson 2003). In some systems these periods of hydrological droughts may result in the isolation of pools and/or the complete drying of edge, off-channel habitats or the entire riverbed (e.g. Boulton 2003; Humphries and Baldwin 2003; Lake 2003). In heavily regulated systems, however, the combination of the anti drought and hydrological drought result in flow that is permanently regulated to lower levels, but the water level remains stable and constant. The ecological impact of this combination of drought on rivers are rarely considered together (Humphries and Baldwin 2003; McMahon and Finlayson 2003; Walker 2006b).

Flow regimes are thought to be a major factor influencing fish spawning and recruitment in large rivers (Welcomme 1985; Sparks 1995; Agostinho *et al.* 2004; Winemiller 2005), although evidence to support this is limited (Humphries *et al.* 1999). Following the principles of the *flood pulse concept* (Junk *et al.* 1989), the *flood recruitment model* was developed for Australian temperate rivers by Harris and Gehrke (1994). The flood recruitment model proposes two methods by which increased flows may enhance recruitment in river fish: flooding directly triggers spawning, or flooding indirectly enhances the survival of larvae and juveniles by providing suitable food and habitat on the inundated floodplain (Harris and Gehrke 1994). However, despite widespread acceptance of the principles of the flood pulse concept and flood recruitment model, many fishes in temperate rivers have developed adaptations to spawn and recruit during periods of low flow (Humphries *et al.*

1999; King *et al.* 2003; Mallen-Cooper and Stuart 2003; Humphries *et al.* 2006; Zeug and Winemiller 2008). The *low flow recruitment hypothesis* (Humphries *et al.* 1999), highlights that some species can successfully spawn and recruit during within channel flows. Furthermore, the relationships between flows and the life history of fishes may vary between differing climatic regions (Humphries *et al.* 1999); emphasising the need to investigate these relationships within the system of interest.

Water management techniques, particularly the use of environmental water allocations, is a potentially viable option for the restoration of declining fish populations (Marchetti and Moyle 2001; Arthington *et al.* 2006). While the use of environmental water is becoming increasingly popular as a management option there are few applied examples, and there have been varied levels of success for native fish (e.g. Nesler *et al.* 1988; Travinchek *et al.* 1995; Freeman *et al.* 2001; King *et al.* 2009b; King *et al.* 2010). Further investigations into the application of environmental water allocations for management of fish populations is required in an effort to determine when and how available water could be best applied and optimised for fish. Hydrological manipulation, for example, changing the water levels in weir pools, is another method that has been recently employed in the Lower River Murray in an attempt to improve river health. By raising or lowering the level of water, managers aim to mimic natural historical variability and increase floodplain inundation (DWLBC 2000). However, simply raising and lowering water levels is not sufficient to reproduce the outcomes of flood pulses through the surrounding floodplain (Toth *et al.* 1998). The benefits to fish populations by manipulating water levels is yet to be determined, particularly in Australia.

River regulation has had a profound impact on the natural flow regime in the Lower River Murray, and for many years it has been subjected to anti-drought conditions (McMahon and Finlayson 2003; Walker 2006b). Furthermore, since 1996 there have been extreme dry conditions throughout most of the Basin, and inflows have dropped to record lows. Since 2002, the conditions have been defined as one of the most severe hydrological droughts recorded, with above average ambient temperatures also adding to the impacts (MDBC 2007; Murphy and Timbal 2008).

Changes to the flow regime as a result of river regulation are one of the major causes for declines in riverine fish communities (Minkley 1991; Gehrke *et al.* 1995; Walker *et al.* 1995; Lemly *et al.* 2000; de Groot 2002; de Leeuw *et al.* 2007; Agostinho *et al.* 2008; Winter *et al.* 2009). This is true for the native fish throughout the Murray-Darling Basin which have declined in abundance and distribution following extensive modification of the system (Cadwallader 1978; Gehrke *et al.* 1995; Humphries *et al.* 2002). Knowledge of the role of flows and flooding in the life history cycles of many of the Murray-Darling Basin fish have improved in recent years (Humphries *et al.* 1999; CRCFE 2003; King *et al.* 2003; Leigh *et al.* 2008), however, further research is required. The need to understand how hydrology affects spawning and recruitment has been highlighted as a crucial component of improving fish populations (Humphries and Lake 2000). The currently accepted recruitment models (flood recruitment model and the low flow recruitment hypothesis) have not yet been investigated in the Lower River Murray, where hydrological conditions are quite different to elsewhere in the Basin. Therefore, it is important to understand the life history strategies of key species, and changes in populations during critical conditions, in an effort to improve knowledge for native fish conservation and manage these species accordingly.

This study assesses how changes in hydrology influence spawning of native fish over four years in the Lower River Murray, South Australia. The first year of the study (2005) was higher in flow (volume), with a significant period of within channel flow pulse. The following three years (2006, 2007 and 2008) were hydrologically similar, with severe drought and low flow conditions occurring. The specific aims were to compare annual variability in larval assemblages between a within channel flow pulse year and low flow years; to determine whether the larval fish assemblages differ spatially between two differently managed sites; and if spatial or annual differences occur in larval fish assemblages, to assess if these can be linked to changes in physical and/or environmental variables. It was predicted that the diversity of species and life history strategies would be higher in the flow pulse year, and that in the low flow years there would be a dominance of low-flow recruitment specialists, as a function of hydrology. Furthermore, it was anticipated that larval assemblage descriptors would be broadly consistent between sites, but that differences may occur as a function of variations in physical parameters.

3.2 MATERIALS AND METHODS

3.2.1 Study sites

The present study occurred in the main channel of the Lower River Murray in South Australia (Figure 1.4). The Lower River Murray has no significant tributaries and its hydrology is generally determined by flows from the mid- and upper- Murray and the Darling rivers. The main channel in this region is heavily regulated by the presence of six weirs (Figure 1.4), which have altered the natural hydrology resulting in a series of lentic weir pool habitats, where historically there was a highly variable lotic system (Walker 2006b). The Lower River Murray encompasses four distinct geo-morphological zones including the floodplain and gorge regions, each with distinct ecological features (see Chapter 1 for a detailed discussion).

Sampling was conducted at two sites: in the tailwaters 5 km downstream of Weir 1 (Site 1, 34°21.138' S, 139°37.061' E), and Weir 6 (Site 6, 33°59.725' S, 140°53.152' E) (Figure 1.1). The area surrounding Site 1 is the gorge region, and the area surrounding Site 6 is the floodplain region. Despite the surrounding floodplains being different, the main channel habitat is generally similar with wide, deep, slow flowing pool habitats. The downstream regulation of the two sites, however, is substantially different. Site 6 occurs in the tail water regions of the weir pool of Weir 5 and in this area the water level is managed and held at a stable level in years of low flow. Site 1 occurs downstream of the first weir in the Lower River Murray, immediately prior to the Lower Lakes and Murray Mouth. While the study site is not within the lakes region or subjected to the physical effects of the lakes region, the river is not regulated by the presence of a downstream weir. Therefore, Site 1 is subjected to more impacts of the drought including evaporation, and a drawdown of the water level in years of low flow.

3.2.2 Hydrology

In this study flow will refer to the water discharge (ML per day) through the river, not the velocity of water. Flow in the Lower River Murray is extensively regulated. In recent years, the flow within the system has been little more than the irrigation allowance. However, 2005 was an above regulation flow year, when a significant period of within channel flow pulses occurred. This increase in flow was the result of a combination of increased tributary inflows and the release of the Barmah Millewa Forest environmental water allocation (BM-EWA) (King *et al.* 2008b). Although, the BM-EWA was not specifically aimed at increasing flows in the Lower River Murray this release resulted in a within channel flow pulse in South Australia. In conjunction with this flow pulse, the level of the weir pool at Weir 5 was raised by 50 cm, as a management intervention. The following three years (2006, 2007 and 2008) were hydrologically similar, being drought years of below regulation flow and weir heights were maintained at the regulated pool level.

3.2.3 Sampling regime

Larval fish were sampled during the austral spring/summers of 2005, 2006, 2007 and 2008. This sampling period was selected based on the peak spawning season and larval abundances determined upstream in the river (Humphries *et al.* 2002; Meredith *et al.* 2002). Day and night samples were taken to gain a representative picture of the whole larval assemblage (see Chapter 2). Sampling was conducted fortnightly from October through December, resulting in six trips per year (Table 3.1). Each site was sampled during the day and at night, of the same day, and both sites were sampled within a three-day period.

Table 3.1. Sampling trips and dates used in the analyses and graphs[3].

Trip Number	Sampling week	Date used in analyses/graphs
1	09-13 Oct	11 Oct
2	23-27 Oct	24 Oct
3	06-10 Nov	08 Nov
4	20-24 Nov	24 Nov
5	04-08 Dec	07 Dec
6	18-22 Dec	21 Dec

3.2.4 Collection and processing of fish larvae

An earlier study (Chapter 2) found that plankton tows were the best single method for collecting a representative sample of the fish species that occur in the main channel and were therefore utilised for this study. Three day and three night plankton tows were conducted at each site. Plankton tows were conducted using a set of paired square-framed bongo nets with 500 μm mesh; each net was 0.5 x 0.5 m and 3 m long. Nets were equipped with 30 cm pneumatic floats either side of the frame, which allowed the frame to sit 5 cm below the water surface. Nets were towed for 15 minutes in circles behind a boat using a 20 m rope, in the centre of the main river channel (mid river channel habitat). The volume of water filtered through each net was determined using a General Oceanics™ flow meter, placed in the centre of the mouth openings, which was then used to standardise larval abundance of fish per 1000 m^3. The sum of the standardised abundance for each species was taken from both left and right plankton tow nets for analysis.

Each sample was washed into separate buckets where fish were euthanased using high concentrations of clove oil. Samples were preserved in 95% ethanol *in situ* and returned to the laboratory for sorting using magnification lamps. All larvae were identified to species level, where possible with the aid of published descriptions (Lake 1967a; Puckridge and Walker 1990; Neira *et al.* 1998; Serafini and Humphries 2004), with the exception of carp gudgeon (*Hypseleotris* spp.) and hardyhead (*Craterocephalus* spp.). Each of these two genera were treated as a species complexes due to their close phylogenetic relationships and very similar morphologies making clear identifications difficult (Bertozzi *et al.* 2000; Serafini and Humphries 2004). It is most likely that the hardyhead collected in this study were the un-specked hardyhead (*Craterocephalus stercusmuscarum fulvus*) as the Murray hardyhead (*Craterocephalus fluviatilis*) has not been recently found in the main channel environment (Wedderburn *et al.* 2007).

3.2.5 Measurement of environmental variables

Data for flow (discharge in ML per day), water level (mAHD, level relative to Australian Height Datum), water temperature (°C), and conductivity (μS/cm @ 25 °C) were obtained

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[3] Note all sampling days were used for analyses and but for aesthetics in graphs only one date from this period was presented.

for both sites, throughout the entire season, from the Department of Water, Land and Biodiversity Conservation, Knowledge and Information Divisions, Surface Water Archive (http://e-nrims.dwlbc.sa.gov.au/swa/). The mean daily values from the week of sampling for were used to generate a set of six replicates for each site and trip, to correspond to the six larval replicates.

3.2.6 Data analysis

The fish assemblage was characterised using three descriptors: total abundance, species richness, and assemblage composition. Total abundance of larvae was calculated as the sum of the standardised abundance for all species occurring within a given replicate. Species richness was recorded as the total number of species occurring in each replicate. Assemblage composition was defined as the standardised abundance (fish per 1000 m^3) of each species recorded for each replicate.

Larval assemblage data were fourth root transformed, to prevent highly abundant species from influencing the similarity measure (Clarke and Ainsworth 1993). All biological analyses were examined using Bray-Curtis similarity measures (Bray and Curtis 1957). All environmental variables were examined using a normalised Euclidean distance similarity measure. The patterns in larval assemblages between years, sites and trips were examined using non-metric multidimensional scaling (NMS) ordination.

Prior to analysis the environmental variables and assemblage descriptors were tested for normality and homogeneity of variance. Given that very few variables met the assumptions, data were analysed using permutational analysis of variance (PERMANOVA) for univariate and multivariate data (Anderson 2001b). In all analyses, total abundance, species richness and environmental parameters were examined using univariate designs; while assemblage structure was examined using a multivariate design. Both the univariate and multivariate analyses were performed using a three-way design; with *year, site* and *trip* as random factors, to determine if differences could be detected. Unrestricted permutations of data were performed for all analyses, with 999 permutations for the test, to detect differences at $\alpha=0.05$ (Anderson 2001b). Where significant differences occurred pairwise analyses were performed.

Mantel's test (Mantel 1967) was used to determine if there was a significant correlation between the larval assemblage composition and the environmental variables. The correlations between environmental variables and species abundances were further analysed using NMS ordination. Environmental variables and species were added as vectors onto the NMS plots using a 0.1 R^2 cut-off. Correlations between the NMS axes, environmental variables and individual species were examined using Pearson (r) and Kendall (τ) correlation coefficients, where the Pearson (r) correlation explained the amount of variance attributable, and Kendall (τ) correlation provided the significance of the correlation (McCune and Mefford 1999).

3.3 RESULTS

3.3.1 Environmental variables

Flow (discharge in ML per day) was significantly greater in 2005 than the subsequent years, and also varied between sites and trips (Figure 3.1a; Table 3.2). Throughout 2005, flow exceeded the summer entitlement allocation of ~ 7, 000 ML per day, however, due to continuing drought conditions it remained below the entitlement allocation in 2006, 2007 and 2008 (Figure 3.1a). There were significant differences in the patterns of water level changes between years for each of the sites (Figure 3.1b; Table 3.2). Water level was consistently higher at Site 6 than at Site 1, as a result of management of the weir pools (Figure 3.1b). At Site 6 there was a step change in the water level from 2005 to 2006; but no further decrease occurred during 2007 and 2008 (Figure 3.1b). Water level at Site 1, however, showed a consistent decline in each subsequent year (Figure 3.1b). Variations in mean daily water temperature reflected seasonal variation (Figure 3.1c; Table 3.2). Within sites, the seasonal pattern was relatively consistent, with temperature increasing steadily from spring to summer. Temperatures in 2007 and 2008 were slightly lower when compared to 2005 and 2006, although this was not significant (Figure 3.1c). There were significant differences in the changes in conductivity between years, sites and within seasons (Figure 3.1d; Table 3.2). Conductivity was consistently highest at Site 1 (Figure 3.1d) and increased in each consecutive sampling year to 2007, but decreased in 2008. At Site 6 conductivity was consistent during 2005 and 2006, but was higher in 2007 and 2008 (Figure 3.1d). Changes in conductivity were primarily driven by flow and water level, and proximity to the Murray Mouth.

Table 3.2. Three-way univariate PERMANOVA for differences among years, sites and trips for flow, water level, temperature and conductivity. Bold text indicates significant value.

		Flow		Water level		Temperature		Conductivity	
Source of variation	***df***	**MS**	***p***	**MS**	***p***	**MS**	***p***	**MS**	***p***
Year	3	84.09	**0.001**	0.19	0.009	1.13	0.583	19.16	0.217
Site	1	0.68	0.773	286.26	**0.001**	0.21	0.742	164.52	**0.003**
Trip	5	1.32	0.285	0.00	0.126	15.95	**0.001**	1.33	0.594
Year x Site	3	1.41	**0.001**	0.04	**0.001**	2.06	0.347	10.95	**0.001**
Year x Trip	15	0.94	**0.001**	0.00	0.59	1.20	0.771	0.74	**0.003**
Site x Trip	5	0.19	0.304	0.00	0.103	1.51	0.537	0.96	**0.002**
Year x Site x Trip	15	0.14	**0.001**	0.00	**0.001**	1.73	**0.001**	0.17	**0.001**
Residual	240	0.02		0.00		0.61		0.03	

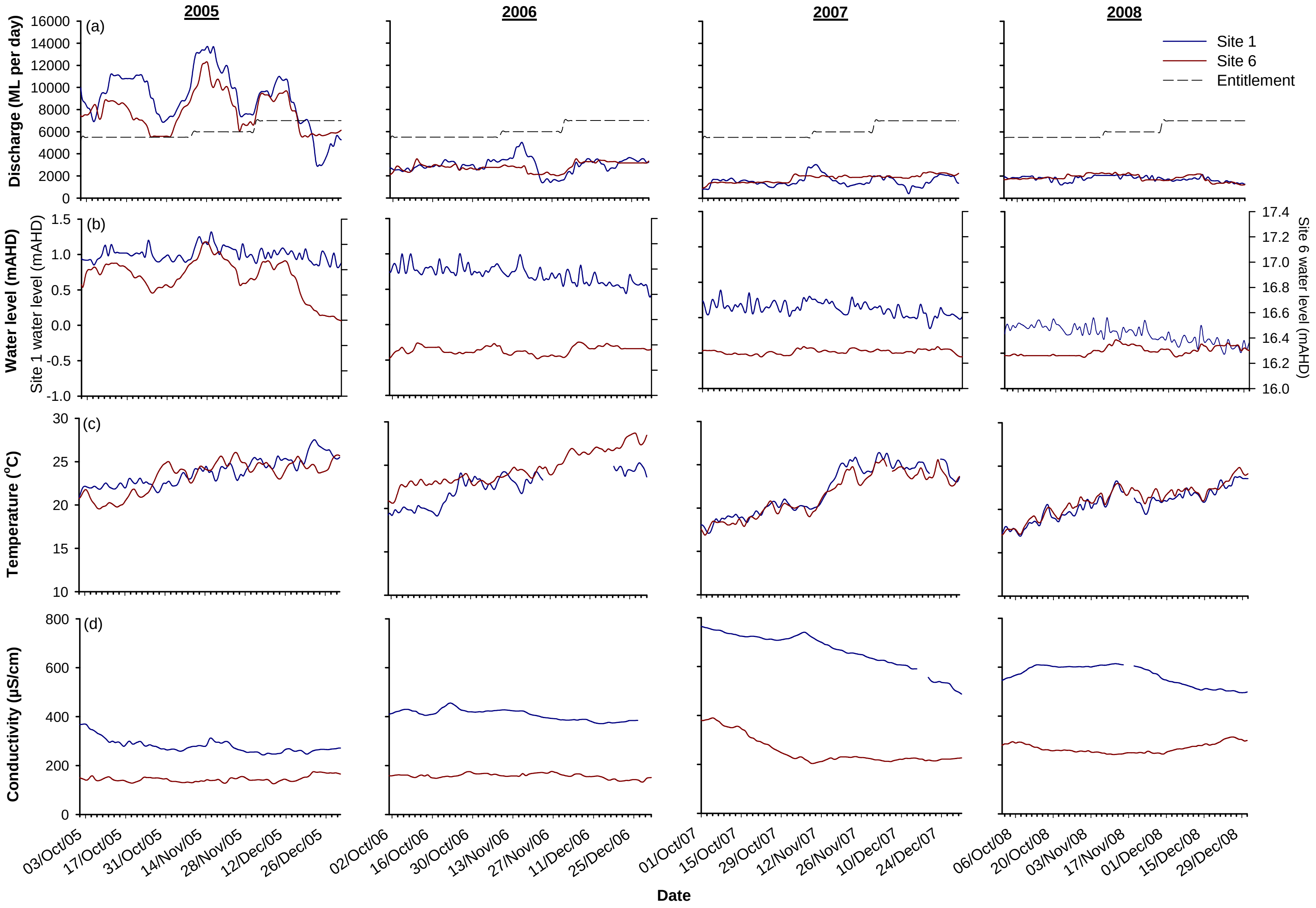

Figure 3.1. Comparison of the environmental variables between 2005, 2006, 2007 and 2008, at Sites 1 and 6, for a) flow (discharge ML per day), b) water level (mAHD, AHD = Level relative to Australian Height Datum), c) mean daily water temperature (°C) and d) mean daily electrical conductivity (µS cm^{-1} at 25 °C). The entitlement allocation is the irrigation allowance released into South Australia.

Water level raising

The increase in flow and the weir raising at Site 6 in 2005 is estimated to have inundated a total of 254 hectares of the floodplain, accounting for approximately 2% of the total floodplain in the area. The inundated area was generally concentrated around low lying areas. Without the raising the estimated area inundated at Site 6 would have been 7 hectares, thus, by raising the weir pool an additional 247 hectares was inundated (Table 3.3).

Table 3.3. The estimated total area inundated through raising Weir 5, at given flow rates, shaded row indicate level of weir pool raise (sourced from DWLBC).

NOTE:
This figure is included on page 49
of the print copy of the thesis held in
the University of Adelaide Library.

3.3.2 Annual and spatial variations in larval fish assemblages

Catch summary

Eleven species, nine native and two exotic, were collected throughout the study. The small-medium bodied native species, Australian smelt (*Retropinna semoni*), bony herring (*Nematalosa erebi*), carp gudgeon (*Hypseleotris* spp.) and flathead gudgeon (*Philypnodon grandiceps*) were the most abundant in all years (Table 3.4). During 2005, a total of 3,593 fish larvae were collected (both sites combined), which increased over the following two years (15,094 in 2006; 36,821 in 2007). Total larval numbers decreased in 2008 (14, 668 larvae). In all years, greater than half the catch was from Site 1 (72.9 % in 2005, 65.4 % in 2006, 56.1% in 2007, 62.7 % in 2008; Table 3.4).

Table 3.4. Species list and total raw numbers of fish larvae collected at Site 1 and Site 6 during 2005, 2006, 2007 and 2008.

	Site 1				Site 6				Total			
Common name (Scientific *name*)	**2005**	**2006**	**2007**	**2008**	**2005**	**2006**	**2007**	**2008**	**2005**	**2006**	**2007**	**2008**
Australian smelt (*Retropinna semoni*)	1,100	4,964	8,951	630	193	3,143	7,797	3,421	1,293	8,107	16,748	4,051
Bony herring (*Nematalosa erebi*)	633	1,435	117	348	83	618	3,530	357	717	2,053	3,647	705
Carp gudgeon (*Hypseleotris* spp.)	694	998	4,885	1,991	359	565	2,486	350	1,053	1,563	7,371	2,341
Flathead gudgeon (*Philypnodon grandiceps*)	186	2,475	6,674	6,931	282	821	2,336	489	468	3,296	9,010	7,420
Hardyhead (*Craterocephalus* spp.)	3	5	14	34	1	51	10	1	4	56	24	35
Murray cod (*Maccullochella peelii peelii*)	2	0	0	0	2	3	2	0	4	3	2	0
Freshwater catfish (*Tandanus tandanus*)	1	0	0	0	7	3	1	4	8	3	1	4
Golden perch (*Macquaria ambigua*)	0	0	0	0	26	0	0	0	26	0	0	0
Silver perch (*Bidyanus bidyanus*)	0	0	0	0	9	0	0	0	9	0	0	0
Carp (*Cyprinus carpio*)	2	0	9	109	10	1	4	3	12	1	13	112
Redfin (*Perca fluviatilis*)	0	0	5	0	1	12	0	0	1	12	5	0
Total number of individuals	**2,621**	**9,877**	**20,655**	**10,043**	**972**	**5,217**	**16,166**	**4,625**	**3,593**	**15,094**	**36,821**	**14,668**
Yearly percentage catch at each site	**72.9**	**65.4**	**56.1**	**62.7**	**27.1**	**34.6**	**43.9**	**37.3**				

Assemblage structure

The larval fish assemblages varied among years; 2005 differed significantly from the subsequent three years (2006, 2007 and 2008), although there were also site and trip effects (Figure 3.2; Figure 3.3; Figure 3.4; Table 3.5). The vertical distribution of samples along axis 2 indicates trip variation as a significant driver of assemblage structure (Figure 3.3; Table 3.5). Trips were separated between early season and late season, most likely due to seasonal spawning behaviour (see Chapter 4 for a detailed discussion). Spatial variation contributed to differences in the larval assemblage, although the variation between sites was subtle (Figure 3.4; Table 3.5).

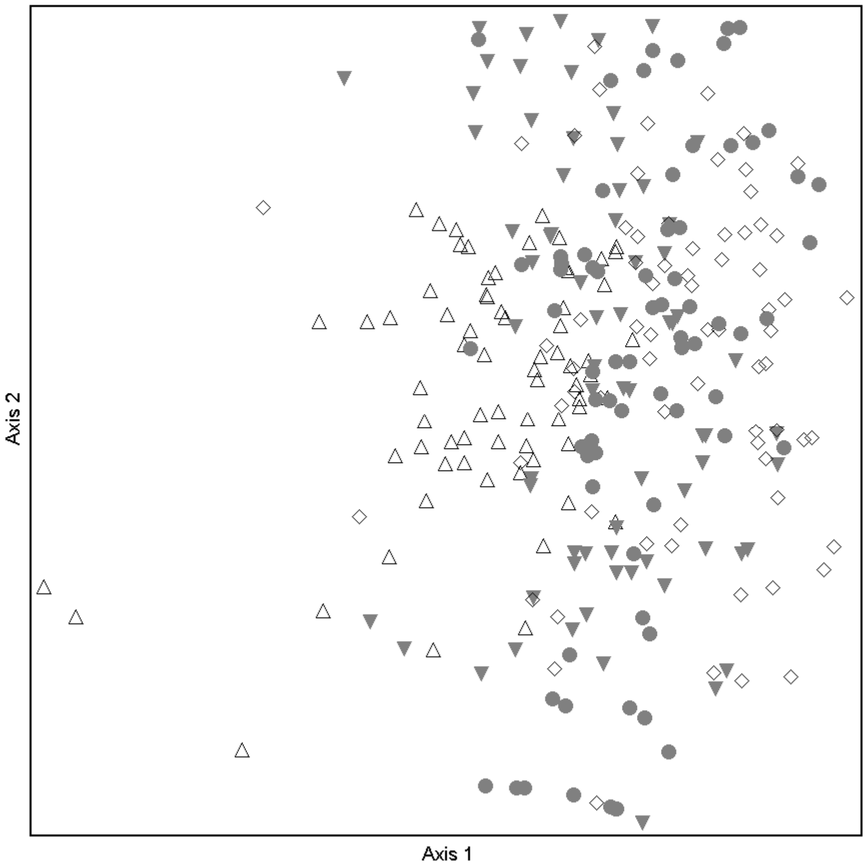

Figure 3.2. NMS ordination (stress 0.11) showing the annual separation of the larval assemblages, during 2005 (△), from 2006 (▼), 2007 (◇) and 2008 (●).Note sites and trips are not distinguished.

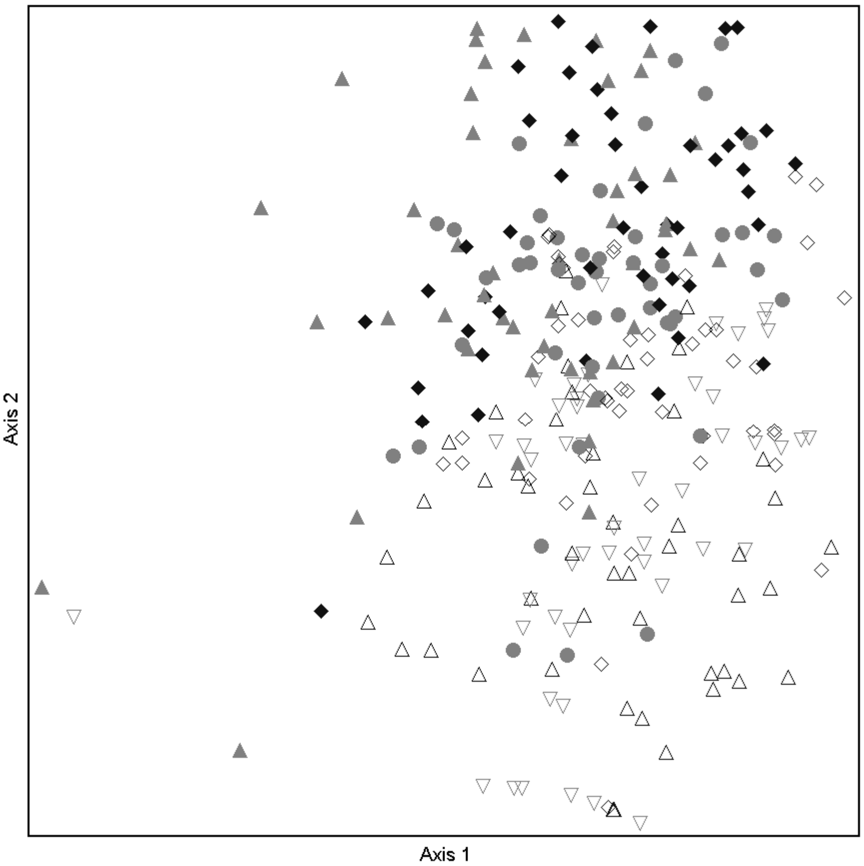

Figure 3.3. NMS ordination (stress 0.11) showing the seasonal separation of the larval assemblages from early season trips (1 △, 2 ▽, 3 ◇) and late season trips (4 ●, 5 ◆, 6 ▲) across the four years of sampling. Note years and sites are not distinguished.

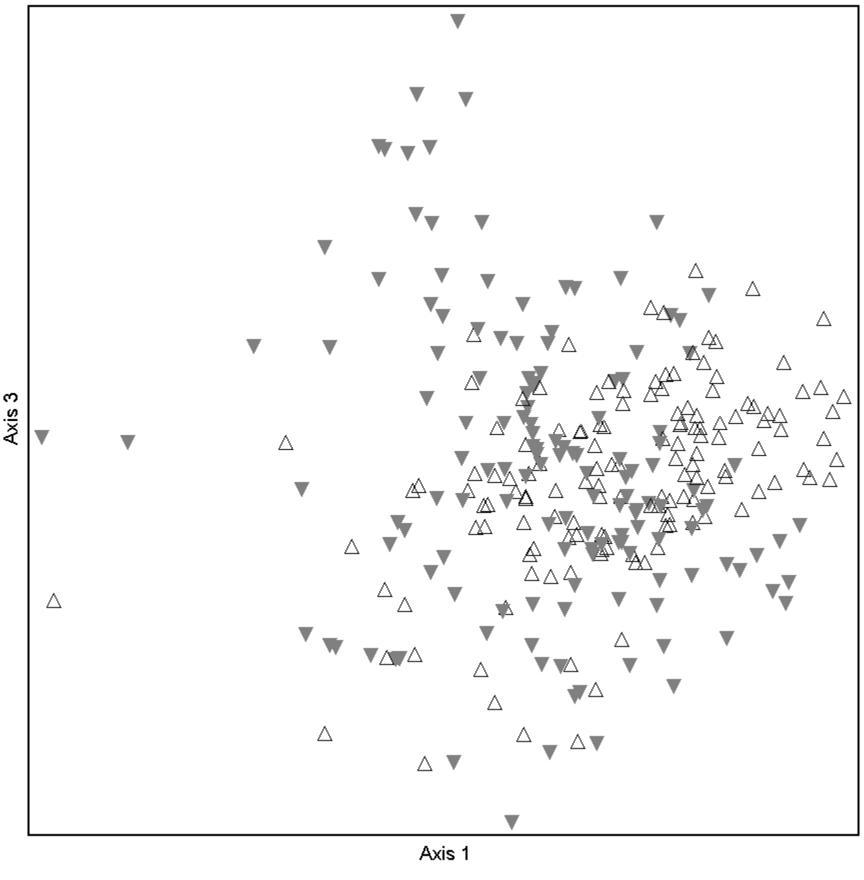

Figure 3.4. NMS ordination (stress 0.11) showing the spatial variation in dispersion of samples from Site 1 (△) and Site 6 (▼). Note years and sites are not distinguished.

Table 3.5. Three-way multivariate PERMANOVA for differences among years, sites and trips for assemblage composition. Bold text indicates significant value.

Source of variation	*df*	MS	*p*
Year	3	38516	0.076
Site	1	27906	0.343
Trip	5	19259	**0.001**
Year x Site	3	24651	**0.001**
Year x Trip	15	4992	0.243
Site x Trip	5	4108	0.537
Year x Site x Trip	15	4247	**0.001**
Residual	240	1664	

Total larval abundance and species richness

Total abundance and species richness were investigated to determine if these factors were contributing to the differences in the assemblage composition between 2005 and the three subsequent years. Patterns of annual and spatial variation were not consistent for either total larval abundance or species richness; neither year nor sites were individually responsible for the observed differences (Figure 3.5; Table 3.6). Trip differences were again identified as a strong factor (see Chapter 4 for a detailed discussion). Total abundance at Site 1 was significantly higher in all sampling trips than at Site 6, with the exception of the last three trips in 2007 (Figure 3.5). Total larval abundance during 2005, was significantly lower than all other years (Figure 3.5). Species richness did differ between the years, but there were only significant differences during October and early November (Figure 3.5). Spatial variation in species richness did not follow any consistent patterns (Figure 3.5; Table 3.6).

Table 3.6. Three-way univariate PERMANOVA for differences among years, sites and trips for total abundance and species richness. Bold text indicates significant value.

		Total abundance		Species richness	
Source of variation	*df*	MS	*p*	MS	*p*
Year	3	48943.0	**0.002**	772.8	0.906
Site	1	12447.0	0.504	1062.5	0.774
Trip	5	3605.7	0.322	1788..0	**0.004**
Year x Site	3	12376.0	**0.008**	2229.8	**0.010**
Year x Trip	15	1968.2	0.892	343.1	0.875
Site x Trip	5	3732.9	0.326	390.1	0.693
Year x Site x Trip	15	3115.6	**0.001**	564.4	**0.002**
Residual	240	1147.9		230.8	

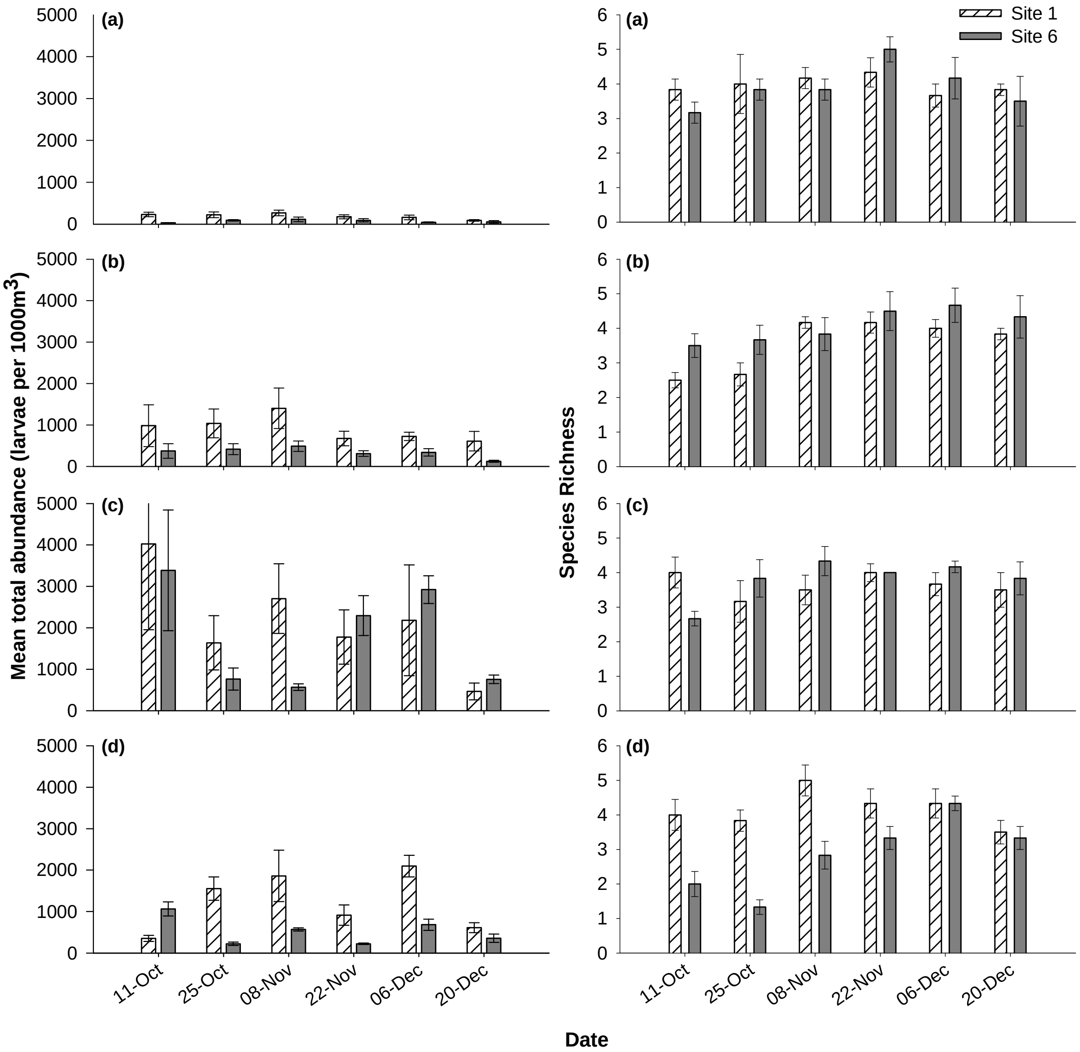

Figure 3.5. Annual and spatial comparisons of changes in mean total abundance (fish per 1000 m^3 ± standard error) and mean species richness (± standard error) for (a) 2005, (b) 2006, (c) 2007 and (d) 2008 at Sites 1 and 6.

Species abundance

The year 2005 was characterised by a different larval assemblage driven by the different abundance of individual species. 2005 was characterised by the lowest abundances of the small-medium bodied species and the exotic species (redfin perch, *Perca fluviatilis* and carp, *Cyprinus carpio*) (Figure 3.6; Figure 3.8). However, two of the large bodied species golden perch (*Maquaria ambigua*) and silver perch (*Bidyanus bidyanus*) were only collected in 2005 (Figure 3.7). Differences also occurred between sites, with higher abundances of the small-medium bodied natives consistently recorded at Site 1 (Figure 3.6). Murray cod (M*accullochella peelii peelii*) and freshwater catfish (*Tandanus tandanus*) were collected at both sites in 2005, but were not collected from Site 1 from 2006 to 2008 (Figure 3.7).

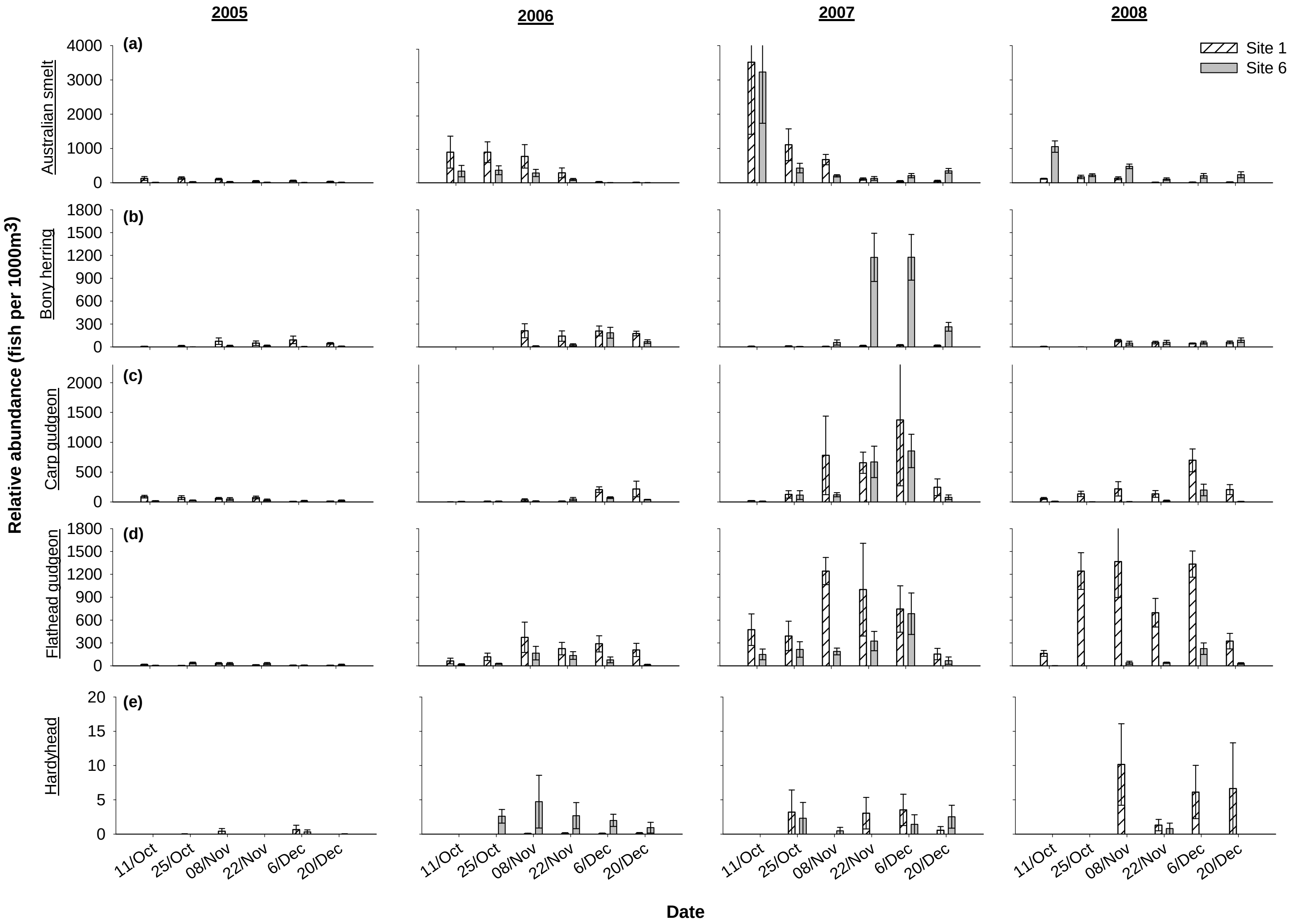

Figure 3.6. Mean relative abundance of larvae (fish per 1000 m³) ± standard error for the small-medium bodied native species at Sites 1 and 6 in the River Murray, South Australia, during 2005, 2006, 2007 and 2008. Note that y-axis values vary between species, but not years.

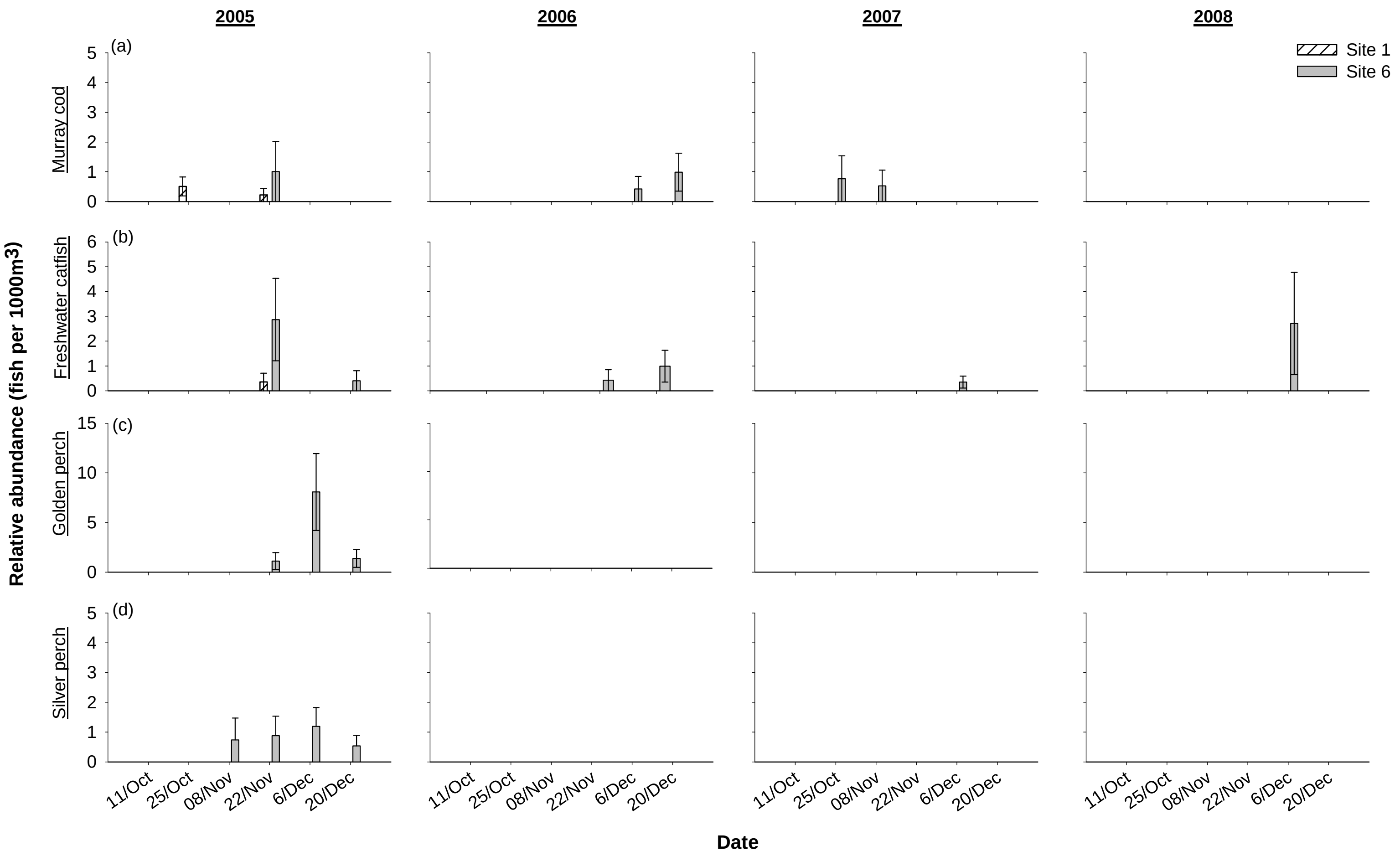

Figure 3.7. Mean relative abundance of larvae (fish per 1000 m³) ± standard error for the large bodied native species at Sites 1 and 6 in the River Murray, South Australia, during 2005, 2006, 2007 and 2008. Note that y-axis values vary between species, but not years.

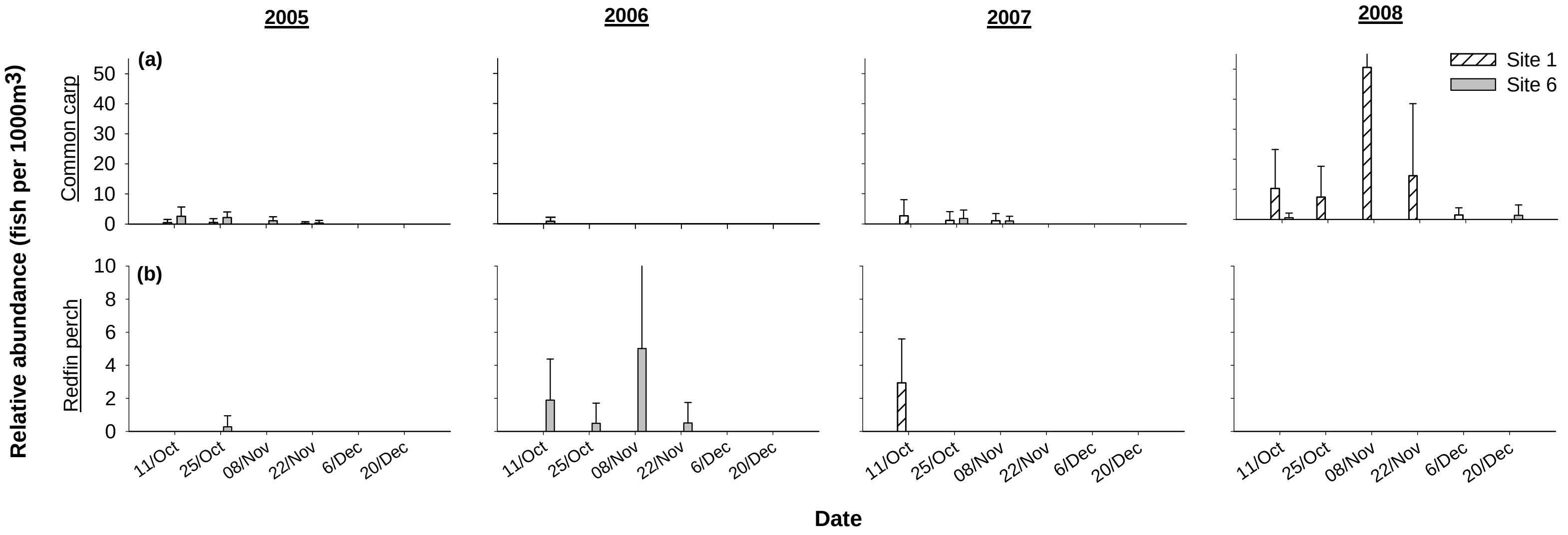

Figure 3.8. Mean relative abundance of larvae (fish per 1000 m^3) ± standard error for the exotic species at Sites 1 and 6 in the River Murray, South Australia, during 2005, 2006, 2007 and 2008. Note that y-axis values vary between species, but not years.

Linking environmental variables to larval fish assemblage

Hydrology and environmental variables were significantly correlated to changes in the larval assemblage (Mantel test: $r = 0.18, p < 0.0001$). The spread of the years across axis 1 was linked to a strong negative correlation with flow and water level and a positive correlation with conductivity (Table 3.7; Figure 3.9). Significant positive correlations with axis 1 were identified for Australian smelt, bony herring carp gudgeon, flathead gudgeon, hardyhead and carp (Table 3.7; Figure 3.9). This indicates that these species are negatively correlated to flow and water levels, but positively correlated to conductivity. Species with significant negative correlations to axis 1 were golden perch and silver perch, indicating a correlation with higher flows and lower conductivities (Table 3.7; Figure 3.9).

Axis 2 was significantly positively correlated to temperature and negatively correlated to water level (Table 3.7; Figure 3.9). Australian smelt and redfin perch had significant negative correlations with axis 2, indicating higher abundances during lower temperatures (Table 3.7; Figure 3.9). Bony herring, carp gudgeon, flathead gudgeon and hardyhead had strong positive correlations, indicating that increases in abundance correlated with higher temperatures (Table 3.7; Figure 3.9).

Axis 3 was not significantly correlated to any of the environmental variables (Table 3.7). However, there were significant positive correlations for carp gudgeon, flathead gudgeon, golden perch, silver perch, carp and redfin perch. Negative correlations were also present for Australian smelt and bony herring (Table 3.7). This indicates that there are further driving forces of larval assemblage that have not been included in these analyses.

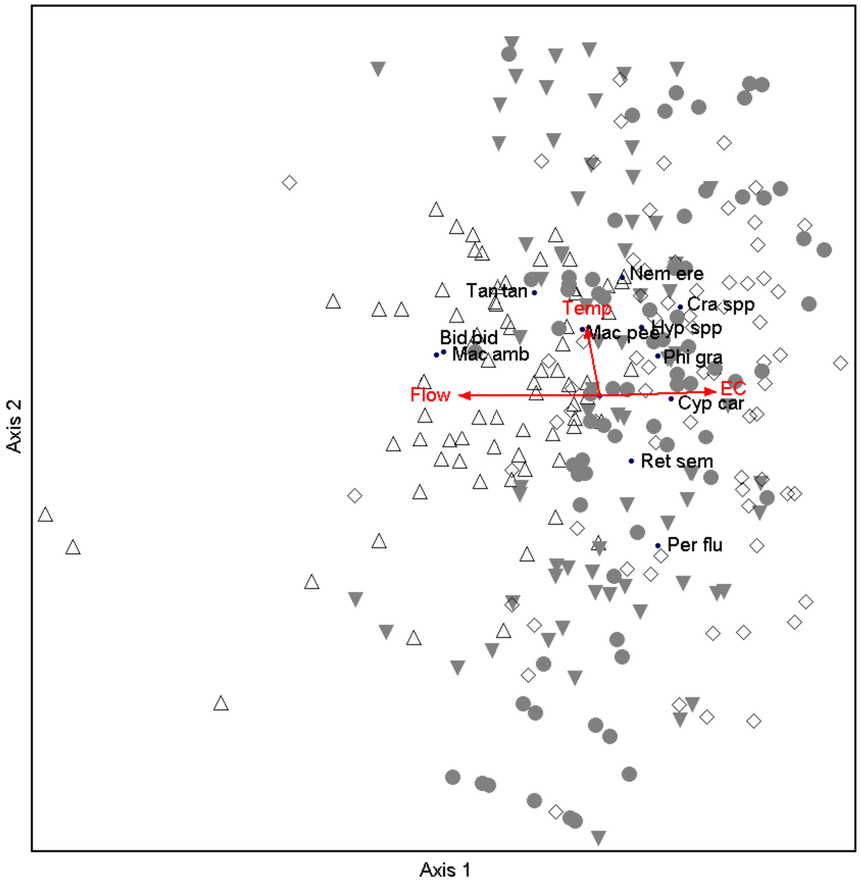

Figure 3.9. NMS ordination (stress 0.11) showing the annual separation of the larval assemblages, particularly 2005 (△), from 2006 (▼), 2007 (◇) and 2008 (●). Note sites and trips are not distinguished. Correlations for species (•) and environmental variables (→) are overlaid; vectors indicate the strength of the correlation with axes. See Table 3.7 for species and environmental variable codes.

Table 3.7. Pearson (r) and Kendall (τ) correlation coefficients between each NMS axis and the environmental variables and species. Where Pearson (r) correlation explained the amount of variance attributable, and Kendall (τ) correlation provides the significance of the correlation (Critical τ value for significance at 0.05 = 0.124). Bold text indicates significant correlations.

		Axis 1		Axis 2		Axis 3	
Variable	NMS codes	*r*	τ	*r*	τ	*r*	τ
Flow	Flow	**-0.49**	**-0.375**	0.04	0.014	0.23	0.075
Conductivity	EC	**0.44**	**0.315**	0.09	0.053	0.04	-0.040
Temperature	Temp	-0.15	-0.086	**0.34**	**0.377**	-0.03	-0.080
Water level	WL	**-0.25**	**-0.329**	**-0.22**	**-0.130**	0.04	0.009
Australian smelt	Ret sem	**0.47**	**0.336**	**-0.65**	**-0.467**	**-0.30**	**-0.242**
Bony herring	Nem ere	**0.21**	**0.126**	**0.71**	**0.584**	**-0.45**	**-0.345**
Carp gudgeon	Hyp spp	**0.55**	**0.391**	**0.60**	**0.454**	**0.23**	**0.184**
Flathead gudgeon	Phi gra	**0.81**	**0.680**	**0.37**	**0.236**	**0.23**	**0.169**
Hardyhead	Cra spp	**0.30**	**0.218**	**0.21**	**0.175**	0.12	0.077
Murray cod	Mac pee	-0.03	-0.053	0.07	0.044	0.09	0.035
Freshwater catfish	Tan tan	-0.11	-0.111	0.11	0.097	0.17	0.094
Golden perch	Mac amb	**-0.22**	**-0.181**	0.04	0.047	**0.29**	**0.170**
Silver perch	Bid bid	**-0.24**	**-0.163**	0.04	0.029	**0.19**	**0.139**
Carp	Cyp car	**0.24**	**0.139**	-0.01	-0.056	**0.30**	**0.265**
Redfin perch	Per flu	0.10	0.066	**-0.18**	**-0.158**	**0.19**	**0.159**

3.4 DISCUSSION

Composition of fish fauna in the main channel environment

Native fish throughout the Murray-Darling Basin have undergone significant declines in range and abundance (Gehrke *et al.* 1995), this is particularly advanced in the Lower River Murray (Lloyd and Walker 1986). The results from this study indicate that the main channel of the Lower River Murray provides suitable spawning and larval habitat for many native fish species. Eleven species were collected as larvae in this study, comprising nine native and two exotic species. Adults of all of these species have been recently recorded within the main river channel (Baumgartner *et al.* 2008b; Davies *et al.* 2008), and as larvae in adjacent off channel habitats in South Australia (Leigh *et al.* 2008). The most abundant species were the small-medium bodied native species (Australian smelt, bony herring, carp gudgeon, flathead gudgeon and hardyhead), which were collected in all years. Large bodied native species (Murray cod, freshwater catfish, golden perch and silver perch) were also recorded but in very low abundances and largely in only one year. Murray cod and freshwater catfish were collected in three of the four years, but golden perch and silver perch were only collected in the higher flow year.

Life history/reproductive strategies for Murray-Darling Basin fish have been developed based on the opportunistic, equilibrium and periodic life history strategies (Winemiller 1989b; Winemiller and Rose 1992; Humphries *et al.* 1999; see Chapter 1, Table 1.1). The results from this study support the categories previously defined by Humphries *et al.* (1999). The main channel environment in the Lower River Murray supported larvae from the three strategies. The small-medium bodied species group encompass a wide range of spawning strategies, akin to the opportunist strategy (Winemiller and Rose 1992) including: main channel generalists, wetland specialist and low flow specialists (King 2002). The large bodied species exhibit characteristics of the periodic (golden perch and silver perch) and equilibrium (Murray cod and freshwater catfish) spawning strategies (Winemiller and Rose 1992; Humphries *et al.* 1999), but due to low numbers of individuals collected in this study it is difficult to confidently determine these groupings.

Spatial variation in larval assemblages: regional differences

Spatial variation in fish assemblages has been documented throughout the worlds river systems, with variations related to a variety of factors, including environmental conditions, food availability and habitat morphology (e.g. Schlosser 1982; Oberdorff *et al.* 1993; Ostrand and Wilde 2002; Habit *et al.* 2007). The larval assemblages varied considerably between the two sites surveyed in this study, perhaps indicating broader regional differences along the river. These differences in community composition were mostly driven by the total abundance of larvae and individual species abundances. The larvae of small-medium bodied natives were more abundant in Site 1, while larvae of the large bodied species, although present in low abundances throughout the study, were consistently more abundant in Site 6.

Spatial differences in abundance and assemblage composition between sites were most likely driven by differences in habitat characteristics and potentially hydraulic conditions. In the gorge region surrounding Site 1, the increased number of wetlands may result in a higher abundance of adult fish for the small-medium bodied species, as these are able to utilise both the wetlands and main channel environments for spawning and recruitment (CRCFE 2003; Humphries *et al.* 1999; King *et al.* 2003). Site 6 is in the floodplain region, which is potentially a more favourable habitat for spawning and recruitment of large bodied species. This region has a higher degree of habitat diversity and structural complexity (pool habitats, woody debris, undercut banks, ledges and vegetation), and the presence of anabranches that provide areas with varied flow velocities, abundant woody debris and diverse vegetation (Young 2001; Walker 2006b). Although further research is needed on the habitat requirements of larval and juvenile Murray cod, golden perch and freshwater catfish, these habitat characteristics are important for adult fish (Koehn and Nicol 1998; Clunie and Koehn 2001a).

The contrasts in the degree of downstream regulation at the two sites may explain the variation between the larval assemblages. The presence of a weir downstream of Site 6 not only maintains the water level at a height significantly above that of Site 1, but in low flow years holds the water at a stable level. There were significant differences in the patterns of water level changes between years for each of the sites. At Site 6 there was a step change in the water level from 2005 to 2006; and no further decrease given management of the weir pool, whilst water level at Site 1 consistently declined in each subsequent year. The decrease in water level at Site 1 may have increased the concentration of larvae, making them easier to catch and producing an apparent increase in the abundance of small-medium bodied larvae. However, this is likely to have been a minor influence, and should have the same implications for large bodied species, which did not occur.

This study did not determine if the higher abundances of the small-medium bodied species at Site 1 is reflected in the adult populations, and this would be an interesting area of future research. This study also only examined one site in the gorge and floodplain regions, and hence it is difficult to comment confidently on regional differences without sampling multiple sites in each region.

Annual variation in larval assemblages: influence of hydrology

The larval assemblage differed between years; with 2005 (the flow pulse year) being significantly different from the subsequent three low flow years. There were some differences between the low flow years but no consistent pattern was identified. Three responses to varying hydrology were identified in the larval assemblage: larvae that were 1) positively correlated to increased flow, 2) negatively correlated to the increased flow and 3) correlated to temperature. The correlation between temperature and changes in larval fish assemblage most likely reflects seasonal differences in spawning of individual species (also see Chapter 4 for more details; King *et al.* 2003). Conductivity was negatively correlated to water level and flow. None of the values for conductivity measured across the years are outside the range for growth and survival of larvae (>12,500 µS/cm for short term exposures, McNeil, unpublished data). Therefore, conductivity is unlikely to be shaping the changes in the assemblage and the response of individual species in each year.

The role of hydrology (flows and flooding) in the life cycles of freshwater fish have been studied throughout Australia (e.g. Lake 1967a; Harris and Gehrke 1994; Humphries *et al.* 1999; King *et al.* 2003) and the world (e.g. Junk *et al.* 1989; Winemiller and Rose 1992; Dudley and Platania 2007; Zeug and Winemiller 2008). Whilst floodplain inundation is important for life cycles of some fish species (Welcomme 1985; Winemiller 2005), others do not require regular floodplain inundation and are able to successfully spawn and recruit in main channel habitats (Haines and Tyus 1990; Turner *et al.* 1994; Watkins *et al.* 1997; Humphries *et al.* 2002; King *et al.* 2003; King 2004b). In this study, the annual patterns of abundance for the small-medium bodied natives conform with, and support, the predictions of the low flow recruitment hypothesis (Humphries *et al.* 1999). The small-medium bodied native species all exhibited an opportunistic life history strategy, significantly increasing in abundance during the low flow years. Recent studies in Australia have demonstrated successful spawning and recruitment of many species in the main channel during low flow conditions (Humphries *et al.* 2002; King 2004b) and without floodplain inundation (Humphries *et al.* 2002; Balcombe *et al.* 2007; King *et al.* 2009b). The low flow recruitment hypothesis has also recently demonstrated application in temperate floodplain rivers outside of Australia, for example, the Brazos River, Texas; where during periods of low flow, recruitment of most species and abundances of their prey items were highest (Zeug and Winemiller 2008).

Humphries *et al.* (1999) propose in the low flow recruitment hypothesis that larvae utilise embayments, backwaters and shallow littoral zones under low flow conditions, as these areas are warm and there is a higher concentration of available prey items. However, King

(2004a) identified that prey concentration in these habitats was not greater than in the main river channel in a tributary of the mid Murray. Periods of low flow are often associated with greater zooplankton densities, thus in the main channel the combination of lower velocities and increased food availability may provide suitable conditions for the growth and survival of larvae (Gehrke 1992; King *et al.* 2003; King 2005; Zeug and Winemiller 2008). However, prolonged low flow and drought conditions can have disadvantages for species that are not low flow opportunists.

The impact of prolonged low flows and drought are not easily separated, for example, both can impact on water quality by decreasing dissolved oxygen, and increasing temperature and salinity (McNeil 2004). Under drought and low flow conditions many fishes will make use of refuge habitats; that provide suitable conditions for growth and survival (Lake 2003; Magoulick and Kobza 2003). However, high concentrations of organisms within these refuges may increase both predation and competition for resources, potentially resulting in lower recruitment into the juvenile and adult populations (Copp 1992; Magoulick and Kobza 2003). In heavily regulated main river channels there is little potential for areas to dry into isolated pools and therefore, less threat of decreased water quality. Thus, in these environments refugia (e.g. backwaters, embayments and still littoral zones) may be more important for providing areas that enhance the growth of larvae (as suggested by the low flow recruitment hypothesis), rather than areas that avoid unsuitable conditions. However, the current study focused on the weir pools of the main river channel, not specific refuge habitats. It should be highlighted that during low flows the velocity in the main channel of the Lower River Murray is much lower than less regulated rivers. The use of the main channel environment for spawning and by larvae is not unusual as many fish complete their life cycle in the main channel of rivers, particularly under drought conditions and in altered rivers that have been disconnected from off-channel habitats (Junk *et al.* 1989; Galat and Zweimuller 2001). The low flow pattern throughout the River Murray is predicted to continue for a number of years, as a result of the combination of drought and current water resource patterns (Lintermans and Cottingham 2007; MDBC 2008), which emphasises the need for environmental water to be considered as a management option.

River regulation

Heavily regulated rivers are harsh environments for all fish, but particularly for those that are flow dependent for some aspect of their life cycle (Walker 2006b). Those species that require specific conditions for spawning, recruitment and dispersal of their eggs and larvae are likely to be the most affected by changes to the natural flow regime (Mackay 1973; Welcomme 1985; Jobling 1995; Humphries *et al.* 2002). While not all species rely on flows to stimulate migrations or spawning; the larval survival and subsequent recruitment of many species may benefit from enhanced river flows (Gehrke *et al.* 1995; Humphries *et al.* 1999; King *et al.* 2008b). Large bodied, longer lived species are likely to be able to withstand unfavourable spawning and recruitment conditions for some period of time. However, fish with long life spans, slow growth rates, low fecundity and late age to maturity are also potentially more at risk and slower to recover from population decreases (Denney *et al.* 2002; Baker *et al.* 2009). Under prolonged drought conditions excessive periods of low flow and drying of off-channel habitats may compound, restricting the reproduction and recruitment in some of the large bodied species.

The hydrology during the four years of the study was varied. The increased flow in 2005 was still well below that required to inundate the floodplain (~50, 000 to 60,000 mega litres per day is required for an over bank flood), thus, the flow event can be categorised as a within channel flow pulse. This increase occurred as a result of increased inflows into upstream tributaries and the release of 513GL as a part of the Barmah-Millewa environmental water allocation (King *et al.* 2009b; King *et al.* 2010). This environmental water allocation resulted in significant increases in the spawning activity of golden perch and silver perch; and enhanced recruitment success of golden perch, silver perch, Murray cod and trout cod throughout the Barmah-Millewa Forest (King *et al.* 2009b; King *et al.* 2010). Golden perch recruitment in the Lower River Murray was also enhanced during 2005 (Ye *et al.* 2008), however, further studies are needed on the recruitment success of the other large bodied species in the Lower River Murray. The spawning of the large bodied species (particularly golden perch and silver perch) in the Lower River Murray were similar, although to a much lesser magnitude, to that recorded in the Barmah-Millewa Forest where the environmental water allocation was targeted (King *et al.* 2009b; King *et al.* 2010). The differences lie in the spawning effort. In the Barmah-Millewa study, golden perch and silver perch spawned in all years in low numbers, but had massive increases in abundance of eggs and larvae during the flow event (King *et al.* 2009b). Whereas, no eggs or larvae of golden perch or silver perch were collected in the Lower Murray during the low flow years, and only a small number were recorded during the flow pulse. However, it should also be highlighted that there were significant variations in the volume of the flow events between the two regions. In the current study the within channel flow pulse would be equivalent to the non flood events in the Barmah-Millewa (King *et al.* 2009b).

Although the flow pulse was moving down the Lower River Murray, an increase in amount of floodplain inundation occurred as a direct result of raising the water levels in the weir pool, but this occurred across a minimal amount of the floodplain. The abundance and distribution of Murray cod and freshwater catfish larvae was greater, and golden perch and silver perch were only present during the flow pulse and water level raising. It is, however, unlikely that raising the water level alone initiated spawning in any the large bodied native species. Murray cod and freshwater catfish are not believed to be initiated to spawn by changes in water levels or flow (Davis 1977a, b; Humphries *et al.* 2002; King *et al.* 2003; Gilligan and Schiller 2004; Humphries 2005; Koehn and Harrington 2006; King *et al.* 2008b). Golden perch and silver perch have previously been defined as flood-cued spawners as they were believed to be stimulated to spawn by flooding (Lake 1967a; Mackay 1973; Reynolds 1983; Rowland 1983) as prescribed by the flood recruitment model (Harris and Gehrke 1994). However, this has been questioned in more recent studies (Humphries *et al.* 1999; King *et al.* 2003) as spawning activity has been recorded for both golden perch and silver perch associated with moderately variable within channel flows (Mallen-Cooper and Stuart 2003; King *et al.* 2008b).

Weir pool manipulation is unlikely to be a replacement for increasing flow rates, as a simple manipulation of water level is not sufficient to reproduce the outcomes of flood pulses (both hydrological and biological) through the river and floodplain (Toth *et al.* 1998). During a static rise, without flow the inundated areas may become stagnant water bodies, low in dissolved oxygen and high dissolving organic matter, tannins, polyphenols and ligins which are potential hazardous to larvae (Gehrke 1991). Unfortunately, it was not possible to isolate the effect of weir pool raising in the current study given it was undertaken in conjunction with an increase in flow. Further investigation is needed to identify and quantify the ecological outcomes of weir pool manipulation and different regimes of raising and drawing down of weir pools (timing, duration, rate etc) to maximise the ecological benefits. Nevertheless, when used in conjunction with elevated flow or environmental flow allocations, manipulating weir pool height may have the potential to increase the environmental benefit.

The challenges in restoring and managing native fish populations in heavily regulated systems are vast. Environmental water is potentially the primary management mechanism. This study has highlighted that a number of species will spawn and develop as larvae during low flow conditions in the heavily regulated weir pool environment. However, for species with specific flow requirements continued low flow conditions may pose a significant threat. In heavily regulated systems, environmental water allocations should be considered to manage and potentially restore declining fish populations, however, further research is need to establish the required volume, pattern and intervals at which flow events are needed to maintain populations. Furthermore, this study suggests that in heavily regulated floodplain rivers, flow pulses may provide a useful management and restoration strategy particularly during drought and otherwise stable low flow conditions.

CHAPTER 4: SEASONAL LARVAL FISH DYNAMICS DURING A LOW FLOW YEAR IN A HEAVILY REGULATED, LOWLAND, TEMPERATE RIVER SYSTEM, AUSTRALIA

ABSTRACT

Temperate riverine fishes generally have defined spawning seasons and can be grouped into spring, spring/summer and summer seasonal spawning guilds. Seasonal variation has previously been identified in larval assemblages in the Lower River Murray, however few studies have investigated the spawning seasons for fishes in this region. The main objective of this study was to explore whether temporal shifts in larval assemblages during a typical low flow year in the Lower River Murray are consistent with seasonal spawning guilds previously identified for Murray-Darling Basin species. Larval fish were sampled using a combination of pelagic plankton tows and light traps, fortnightly during the spring/summer of 2006/07, a typical low flow year. Timing, abundance and duration of preflexion larvae were staggered for each species, and showed minor differences between sites for individual species. Seasonal variation contributed more strongly to observed differences in the preflexion larval assemblage than spatial variation. Although temporally restricted, the results indicate that the seasonal spawning guilds were consistent with those previously identified, suggesting that these can be extrapolated to management of fish in the Lower River Murray. However, there were differences in the protracted or discrete nature of spawning for some species between the Lower River Murray and previous studies. Given that riverine fish populations throughout the world are under threat given changes to the flow regime, further understanding of the specific seasonal dynamics within a system are important for structuring management and restoration programs to mitigate the threats.

4.1 INTRODUCTION

Rehabilitation of native fish populations requires a thorough understanding of the population dynamics and ecological requirements for key species. Population dynamics are defined as the variations in time and space in the sizes and densities of populations (Begon *et al.* 1996). Changes in populations are regulated by the rates of births and deaths, emigration and immigration (Begon *et al.* 1996). Most fishes exhibit an annual cycle of reproduction, while some spawn at two to three year intervals and others reproduce only once in their lifetime (Bye 1984). In temperate systems, most species have a defined spawning season to ensure that the early life stages are provided with optimal conditions for survival and development (Bye 1984; Jobling 1995). A combination of warmer temperatures and greater production of food is thought to benefit most species spawning during spring/summer in temperate rivers (Jobling 1995). Additionally there are frequently separations between the spawning seasons of species into spring, spring/summer and summer (e.g. Turner *et al.* 1994; King *et al.* 2003).

Changes to flow regime and the construction of regulatory structures in the Murray-Darling Basin have contributed significantly to the decline of native fish populations (Cadwallader 1978; Gehrke *et al.* 1995; Humphries *et al.* 2002). Consequently, the return of a more natural flow regime, for example through the provision of environmental water has been highlighted as a promising restoration strategy to improve native fish populations (Poff *et al.* 1997; MDBC 2002, 2004; Arthington *et al.* 2006; Walker 2006a; Chapter 3). Environmental water allocations are aimed at replicating components of the natural flow regime, including the magnitude, timing, pattern, frequency, duration and rate of change of key events (King *et al.* 2003; Arthington *et al.* 2006; Walker 2006a). Therefore, a detailed understanding of the spawning and larval assemblage dynamics is necessary to ensure water management strategies are targeted to create suitable conditions.

Numerous studies have been conducted on spawning and larval dynamics, focussing on determining spawning and nursery areas, temporal and spatial differences in spawning characteristics and ontogenetic shifts in habitat and diets (e.g. Holland 1986; Humphries *et al.* 2002; King 2004b, 2005; Tonkin *et al.* 2006). Using a review of key studies throughout the mid and upper River Murray and surrounding tributaries seasonal spawning activity has been investigated, resulting in the suggestion of three seasonal spawning guilds (Figure 4.1) (King *et al.* 2003). These spawning guilds were spring (August/September to October/November), spring/summer (September/October to December) and summer (November to March) (Figure 4.1) (King *et al.* 2003). Whilst strong inter-annual pattern of change in larval assemblages among different flow years have been documented in the Lower River Murray, there was also evidence of strong seasonal structuring in the larval assemblages (Chapter 3). However, the spawning seasons of fishes within the Lower River Murray have not been investigated in detail (but see Chapter 3 and Leigh *et al.* 2008) and it remains to be seen whether the seasonal spawning guilds previously defined for Murray-Darling Basin fishes are also applicable to this region.

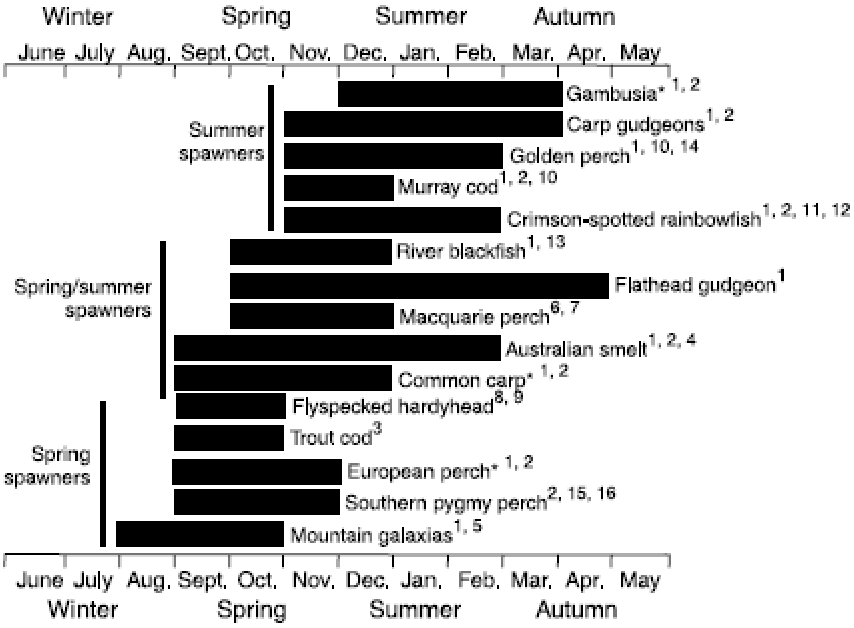

Figure 4.1. The estimated spawning calendar of most fish species known from the Ovens River. Note that the carp gudgeon estimated spawning calendar is for the species complex. An asterisk (*) indicates introduced species. The spawning calendar is based on the following estimates: (1) Humphries *et al.* (2002); (2) King (2002); (3) Ingram and Douglas (1995); (4) Milton and Arthington (1985); (5) O'Connor and Koehn (1990); (6) Cadwallader and Rogan (1977); (7) Appleford *et al.* (1998); (8) Milton and Arthington (1983); (9) Llewellyn (1979); (10) Cadwallader (1977); (11) Backhouse and Frusher (1980); (12) Milton and Arthington (1984); (13) Jackson (1978); (14) Mackay (1973); (15) Llewellyn (1974); and (16) Humphries (1995). (Reproduced from King *et al.* (2003)).

The main objective of this study was to explore whether within season temporal shifts in larval assemblages during a typical low flow year in the Lower River Murray are consistent with the seasonal spawning guilds identified by King *et al.* (2003). Specifically it aimed to determine if there were differences in the peak timing of preflexion larval occurrence and total larval abundance in three sites within the Lower River Murray; then to determine if timing, patterns and abundance of the preflexion larval assemblage (as an indicator of spawning activity) and individual species demonstrated temporal and spatial variation; and finally to identify if the spawning of individual species correlates to spring, spring/summer and summer seasonal guilds. It is hypothesised that the assemblage composition and individual species abundances would change within a spawning season due to individual species biology and that the seasonal spawning of species would reflect the guilds previously defined.

4.2 MATERIALS AND METHODS

4.2.1 Site description

The present study occurred in the main river channel of the Lower River Murray in South Australia, in both the gorge and floodplain regions (Figure 1.4). Sampling was conducted at three sites: in the tailwaters 5 km downstream of each of Weir 1 (Site 1, 34°21.138' S, 139°37.061' E), Weir 5 (Site 5, 34°13.246' S, 140°45.0909' E) and Weir 6 (Site 6, 33°59.725' S, 140°53.152' E) (Figure 1.4). The area surrounding Site 1 is the gorge region and the area surrounding Sites 5 and 6 is the floodplain region (see Chapter 1 for detailed site information). Despite the surrounding characteristics of the floodplains being different the main channel habitat is generally similar, being wide, deep, slow flowing pool habitat.

4.2.2 Sampling regime

Larval fish were sampled during the spring/summer of 2006/07; this year was selected as it is most representative of the low flow conditions that the Lower River Murray is currently experiencing. The sampling period was selected based on estimates of spawning times from previous studies within the river system (Humphries *et al.* 2002; Meredith *et al.* 2002). Sampling was conducted fortnightly from September through December, and monthly in January and February, resulting in nine trips (Table 4.1). Each site was sampled during the day and at night of the same day, and all three sites were sampled within a four-day period.

Table 4.1. Sampling trips and dates used in the analyses and graphs[4].

Trip Number	Sampling week	Date used in analyses/graphs
1	25-29 Sept	28 Sep
2	09-13 Oct	11 Oct
3	23-27 Oct	24 Oct
4	06-10 Nov	08 Nov
5	20-24 Nov	24 Nov
6	04-08 Dec	07 Dec
7	18-22 Dec	21 Dec
8	22-26 Jan	25 Jan
9	19-23 Feb	21 Feb

4.2.3 Collection and processing of larvae

Plankton tows were conducted using a pair of square-framed, 0.5 x 0.5 m, 3 m long bongo nets with 500 μm mesh. Nets were equipped with 30 cm pneumatic floats either side of the frame, so the frame sat 5 cm below the water surface. Nets were towed behind a boat in circles using a 20 m rope, in the centre of the main river channel. Three day and three night 15-min tows were conducted at each site. The volume of water filtered through each net was determined using a General Oceanics™ flow meter fixed in the centre of the mouth openings. Plankton tow data were standardised to number of larvae per 1000 m^3 of water filtered, and left and right plankton tow nets were grouped for analysis.

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[4] Note all sampling days were used for analyses and but for aesthetics in graphs only one date from this period was presented.

Modified quatrefoil light traps (Secor *et al.* 1992), (30 x 30 cm) constructed from perspex were used to target areas that may not be effectively sampled by plankton tows. A yellow 12 hour Cyalume® glow stick was used as the light source, and 5 mm stretched mesh was fitted to prevent predation on captured larvae by larger fish (Vilizzi *et al.* 2008). Three light traps were deployed at each site before sunset each afternoon and retrieved before 24:00 hours of the same night. Light trap data were standardised to number of larvae per 6 hours.

Each sample was washed into separate buckets where fish were euthanased using high concentrations of clove oil. Samples were preserved in 95% ethanol *in situ* and returned to the laboratory for sorting using magnification lamps. All larvae were identified to species level, where possible, using published descriptions (Lake 1967a; Puckridge and Walker 1990; Neira *et al.* 1998; Serafini and Humphries 2004), with the exception of carp gudgeon (*Hypseleotris* spp.) and hardyhead (*Craterocephalus* spp.). Each of these two genera were treated as species complexes due to their close phylogenetic relationships and very similar morphologies making clear identification difficult (Bertozzi *et al.* 2000; Serafini and Humphries 2004). It is most likely that the hardyhead collected in this study were the un-specked hardyhead (*Craterocephalus stercusmuscarum fulvus*) as the Murray hardyhead (*Craterocephalus fluviatilis*) is not found in the main channel environment (Wedderburn *et al.* 2007), however, these species have been grouped as a precaution. Each fish was categorised according to developmental stage, as preflexion larvae (no curvature at tip of notochord), postflexion (upward flexion of notochord, caudal fin rays developing), metalarvae (caudal fins rays developed and pelvic fins forming) or juvenile/adult (rays in all fins fully developed) (Kelso and Rutherford 1996). Juvenile and adult fish were not included in the analysis.

4.2.4 Data analysis

Due to inherent differences in collection efficiency for each method (see Chapter 2) larval abundance was presented as an *integrated catch index* (ICI). ICI combines the standardised catch for a set of gear types to provide a replicate sample. Thus, the ICI was comprised of the standardised catch from one of each of the gear types to provide a replicate sample as follows:

$$\text{ICI} = \text{LT} + \text{DPT} + \text{NPT}$$

Where:
LT= 1 x light trap sample standardised to fish/6 hrs
DPT = 1x day-time tow sample standardised to larvae/1000 m^3
NPT = 1 x night-time tow sample standardised to larvae/1000 m^3

This provided three replicate ICI's for each site during each trip; these replicates were used for all statistical analysis.

The patterns in preflexion larval assemblages between trips were examined using non-metric multidimensional scaling (NMS) ordination (McCune and Mefford 1999). The preflexion larval assemblage data were fourth-root transformed to prevent highly abundant species from influencing the similarity measure (Clarke 1993). All biological analyses were examined using Bray-Curtis (Sorenson) similarity measures (Bray and Curtis 1957).

Assemblage data were analysed using permutational multivariate analysis of variance (PERMANOVA) (Anderson 2001b) using a two-way design, with *site* and *trip* as random factors. Unrestricted permutations of data were performed for all analyses, with 999 permutations for the test, to detect differences at $\alpha=0.05$ (Anderson 2001b). Where significant differences occurred in the PERMANOVA pairwise analyses were performed to determine where the differences occurred.

NMS ordination in conjunction with the pairwise analyses was used to define seasonal groupings for preflexion larval abundance. To identify which seasonal grouping was most representative for individual species *Indicator Species Analysis* (Dufrene and Legendre 1997) was performed on the untransformed larval assemblage data. Indicator species analysis combines information on the concentration of species abundances within a defined group and the faithfulness of occurrence of a species in a particular group. For example, a perfect indicator of a particular group would be exclusively present within the defined group (Dufrene and Legendre 1997). This test provides indicator values for each species in each group on the basis of the standards of the perfect indicator (McCune *et al.* 2002). Statistical significance of each defined indicator is tested by the Monte-Carlo randomisation technique, where data are computed against 4999 runs of the randomised data. A species that is not deemed to be a significant indicator of any particular group is either uncommon (found in low abundances) or widespread (occurs in more than one group in similar abundances) (Dufrene and Legendre 1997).

4.3 RESULTS

4.3.1 Catch summary for all developmental stages

A total of 43,230 fish were collected as larvae throughout the sampling; this included all the developmental stages. Abundance at Site 1 was highest, whereas Sites 5 and 6 were similar (Table 3.4). Nine species, seven native and two exotic, were collected. Small-medium bodied native species, Australian smelt (*Retropinna semoni*), carp gudgeon (*Hypseleotris* spp.), flathead gudgeon (*Philypnodon grandiceps*), and bony herring (*Nematalosa erebi*), dominated the larval catch (Table 3.4). Species richness was higher at Sites 5 and 6, due to the presence of freshwater catfish (*Tandanus tandanus*) and Murray cod (*Maccullochella peelii peelii*) larvae (Table 3.4).

Table 4.2. Combined total number of all developmental stages for fish larvae collected at Sites 1, 5, and 6 using both light traps and plankton tows.

Common name (Scientific *name*)	**Site 1**	**Site 5**	**Site 6**
Australian smelt (*Retropinna semoni*)	12,124	4,575	6,508
Bony herring (*Nematalosa erebi*)	3,031	1,087	1,508
Carp gudgeon (*Hypseleotris* spp.)	3,927	702	1,185
Flathead gudgeon (*Philypnodon grandiceps*)	5,381	1,450	1,470
Hardyhead (*Craterocephalus* spp.)	9	93	81
Murray cod (*Maccullochella peelii peelii*)	0	1	5
Freshwater catfish (*Tandanus tandanus*)	0	1	5
Carp (*Cyprinus carpio*)	7	7	8
Redfin (*Perca fluviatilis*)	3	2	60
Total number of individuals	**24,482**	**7,918**	**10,830**
Percentage of total catch	**56.6**	**18.3**	**25.1**

To determine if total larval abundance or only preflexion larval abundances can be used to elucidate spawning activity, patterns in peak timing of abundances were examined. The developmental stages for the small-medium bodied natives indicated that timing of peaks in larval abundances was not always consistent with the timing of peaks in preflexion larval abundance (Figure 4.2). Peaks in total abundance of Australian smelt and carp gudgeon were not consistent with preflexion abundances at Sites 1 and 5 (Figure 4.2a & c). Peaks in total abundance were consistent with peaks in preflexion abundance for bony herring and flathead gudgeon at all sites, and for carp gudgeon but only at Site 5 (Figure 4.2b, c & d). The spawning period is best represented by the occurrence of preflexion larvae, therefore, the following analyses focus only on preflexion larvae.

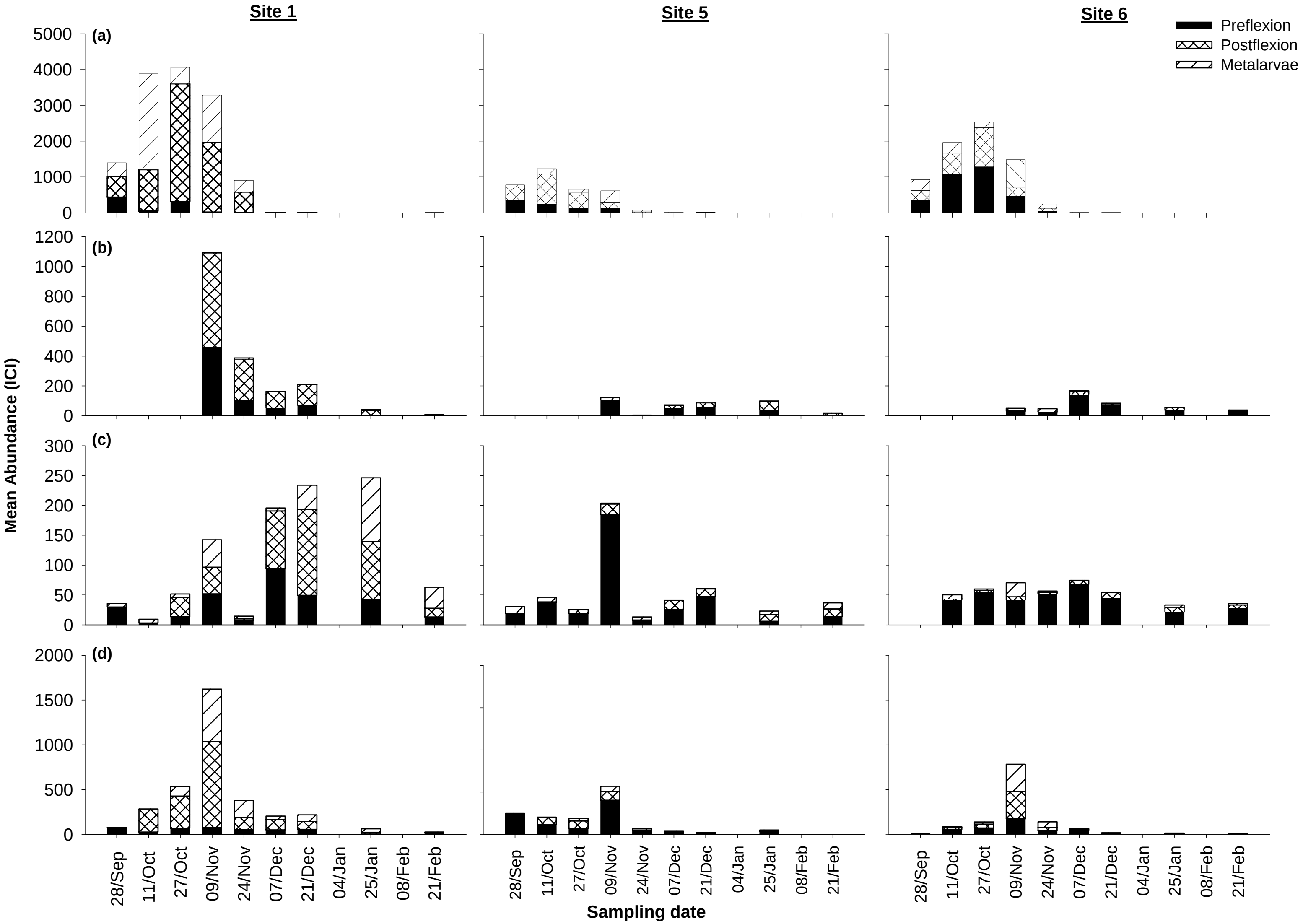

Figure 4.2. Abundance (ICI) of (a) Australian smelt, (b) bony herring, (c) carp gudgeon and (d) flathead gudgeon for preflexion, postflexion and metalarval developmental stages at Site 1, Site 5 and Site 6.

4.3.2 Spatial and seasonal variation in preflexion larval assemblage

A total of 11,794 preflexion larvae were collected at all sites throughout the sampling. Abundance of preflexion larvae was highest at Site 6 (46.3 % of total catch), whilst abundance at Sites 1 and 5 were similar (27.5 % and 26.2 %, respectively) (Table 4.3). Murray cod, freshwater catfish and redfin perch were not collected as preflexion larvae.

Table 4.3. Combined total number of preflexion fish larvae collected at Sites 1, 5, and 6 using both light traps and plankton tows.

Common name (Scientific *name*)	**Site 1**	**Site 5**	**Site 6**
Australian smelt (*Retropinna semoni*)	555	903	2,571
Bony herring (*Nematalosa erebi*)	962	611	1,181
Carp gudgeon (*Hypseleotris* spp.)	815	558	958
Flathead gudgeon (*Philypnodon grandiceps*)	895	902	582
Hardyhead (*Craterocephalus* spp.)	5	95	97
Murray cod (*Maccullochella peelii peelii*)	0	0	0
Freshwater catfish (*Tandanus tandanus*)	0	0	0
Carp (*Cyprinus carpio*)	3	18	59
Redfin (*Perca fluviatilis*)	0	0	0
Total catch at each site	**3,241**	**3,088**	**5,465**
Percentage of total catch	**27.5**	**26.2**	**46.3**

Variations in larval assemblage were driven by changes in the magnitude of differences between sites and trips, as indicated by the site x trip interaction (Figure 4.3 and Table 4.4). Seasonal variation (as represented by trip), was the most consistent driver for variations in larval assemblage (Figure 4.3a). The NMS ordination and pairwise results indicated that the larval assemblages were significantly different between early (trips 1 – 3) and late (trips 4 – 9) seasons (Figure 4.3a). Broadly, the NMS and pairwise results indicated there were very few differences in assemblage structure between sites (Figure 4.3b). These differences can be accounted for by minor variations in the duration and specific timing of peak abundances between sites for individual species, however, the broad scale patterns were consistent (Figure 4.4, Figure 4.6 and Figure 4.5).

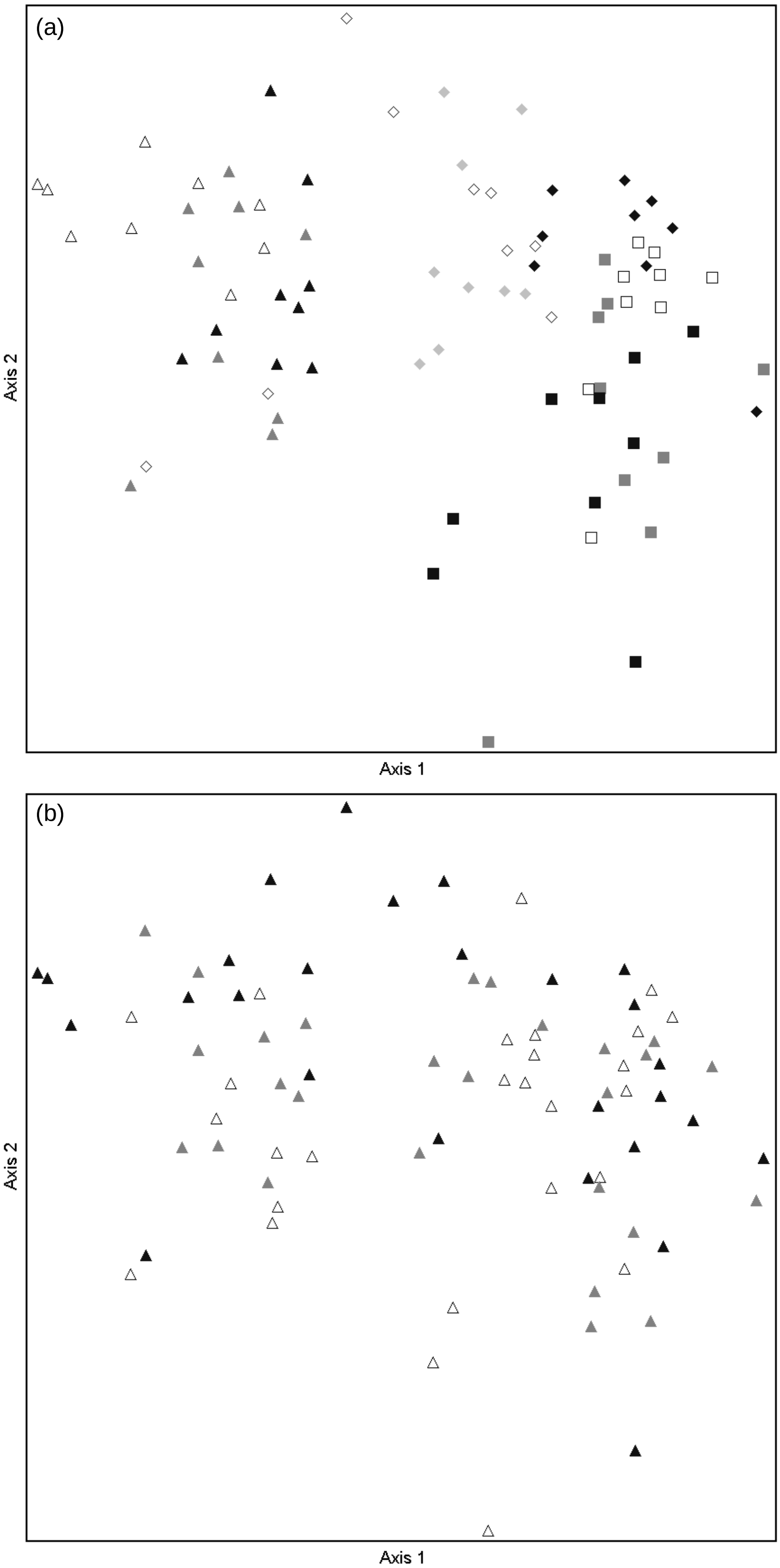

Figure 4.3. NMS ordination (stress 0.12) showing a) the seasonal separation of the preflexion larval assemblages from early season (trips △1, ▲ 2, ▲ 3) and late season (trips ◇ 4, ◆ 5, ◆ 6, □ 7, ■ 8, ■ 9) (note sites are not distinguished) and b) the spatial dispersion of the larval assemblages between Site 1 △, Site 5 ▲ and Site 6 ▲ (note trips are not distinguished).

Table 4.4. Two-way multivariate PERMANOVA for differences among sites and trips for assemblage composition. Bold text indicates significant value.

Source of variation	df	MS	*p*
Site	2	1968	**0.039**
Trip	8	6193	**0.001**
Site x Trip	16	620	**0.004**
Residual	54	265	

4.3.3 Seasonal spawning patterns

Using the early and late trip groupings identified in the NMS and pairwise results two categories were defined: spring/summer (trips 1 – 3) and summer spawners (trips 4 - 9). Indicator species analysis identified that some species were significantly correlated with one of these seasonal groupings, whilst others showed no significant association. Spring/summer spawners were Australian smelt, carp and redfin perch (Table 4.5); preflexion larvae were present from late September with highest abundances occurring during September and October (Figure 4.4). Summer spawners included bony herring and carp gudgeon (Table 4.5). Although both were present throughout most of the season, peak abundance occurred in December (Figure 4.5). Flathead gudgeon, hardyhead, freshwater catfish and Murray cod were not significantly associated with either spring/summer or summer (Table 4.5). Flathead gudgeon and hardyhead preflexion larvae had a protracted spawning and were present throughout the sampling period (Table 4.5; Figure 4.6). Murray cod occurred in very low numbers that overlapped the seasonal groupings (Table 4.5; Figure 4.6). Freshwater catfish was identified to be a summer spawner, with 100% of larvae being collected in summer although this was not significant due to low abundances (Table 4.5; Figure 4.6). Abundance of each species varied between sites and slight variations occurred for the patterns, however the overall seasonal groupings were not affected (Figure 4.4; Figure 4.6; Figure 4.5).

Table 4.5. Indicator species analysis. Relative abundance (%) of each species in each seasonal category (defined from NMS ordination and pair wise results). Indicator group identifies the seasonal grouping each species most likely corresponds to. Monte Carlo test of significance (p (MC)) indicates the probability of each species as an indicator of the corresponding indicator group. Bold text indicates significant indicator group.

Species	Total abundance (ICI)	Relative abundance (%) of species in seasonal groupings		Indicator group	*p* (MC)
		Spring/ Summer	Summer		
Australian smelt	15,241	80	20	Spring/ summer	**0.0002**
Bony herring	12,231	0	100	Summer	**0.0002**
Carp gudgeon	8,084	37	63	Summer	**0.0026**
Flathead gudgeon	9,469	49	51	Spring/ summer	0.7055
Hardyhead	284	35	65	Summer	0.0596
Murray cod	21	53	47	Spring/ summer	0.8470
Freshwater catfish	23	0	100	Summer	0.5595
Carp	486	86	14	Spring/ summer	**0.0002**
Redfin perch	56	100	0	Spring/ summer	**0.0002**

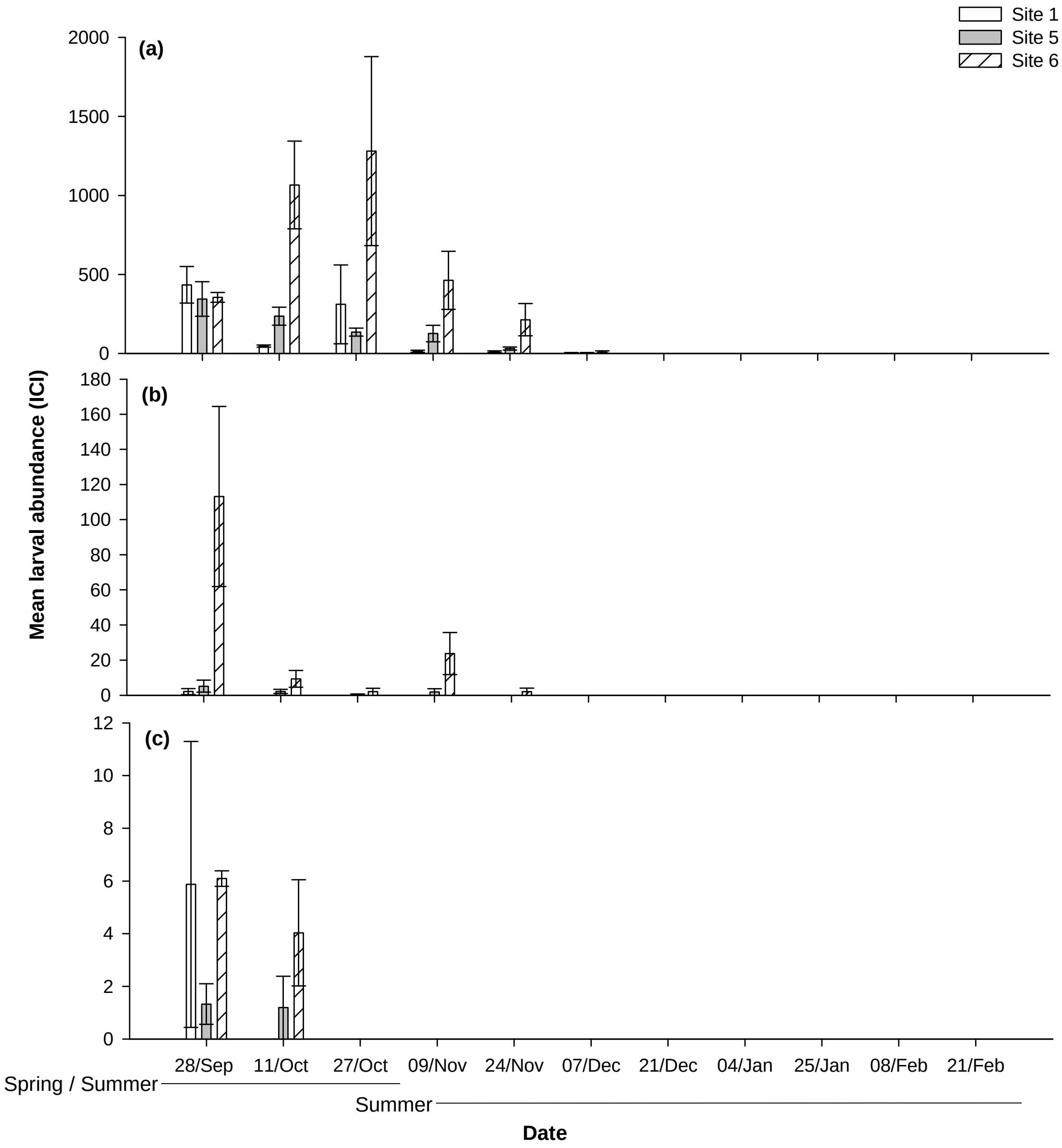

Figure 4.4. Mean preflexion larval abundance (ICI ± standard error) throughout the sampling for spring/summer spawning species, (a) Australian smelt, (b) carp* and (c) redfin perch*#. Note * indicates postflexion larvae that were corrected for approx age, # indicates very low numbers of individuals.

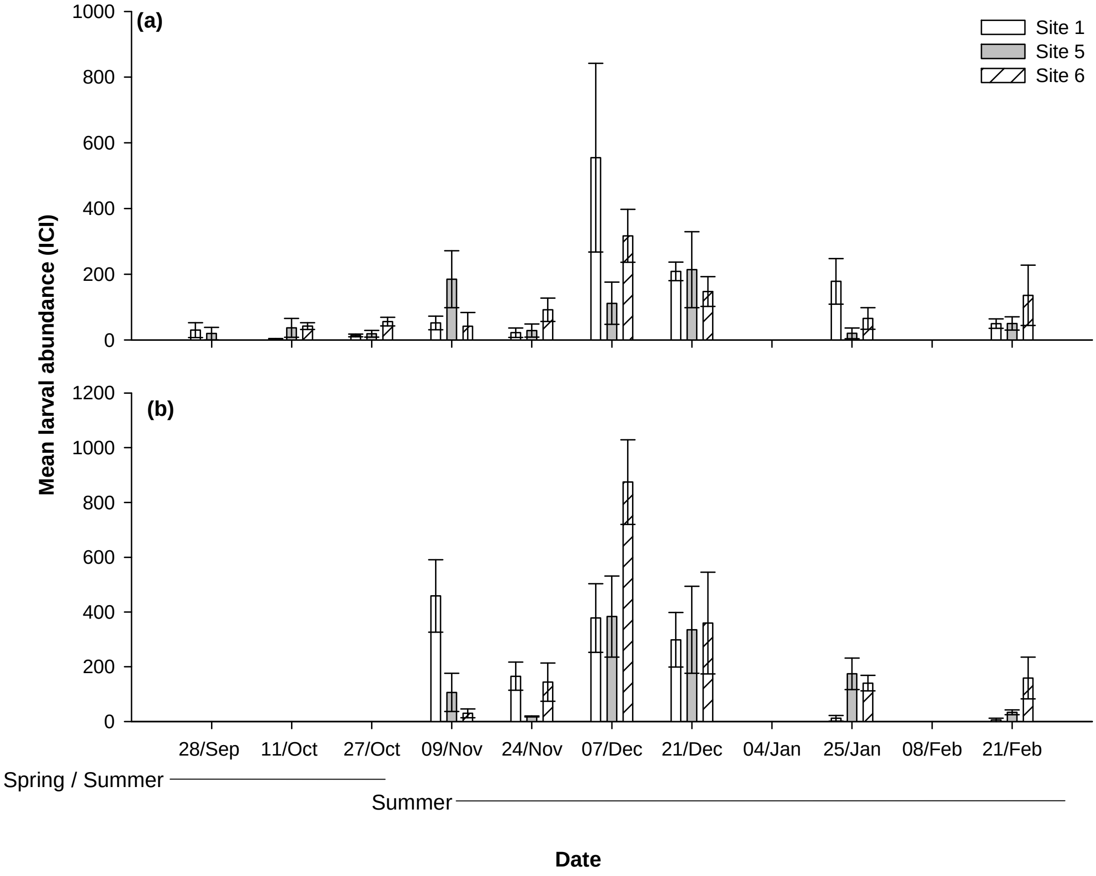

Figure 4.5. Mean preflexion larval abundance (ICI ± standard error) throughout the sampling for summer spawning species (a) carp gudgeon and (b) bony herring.

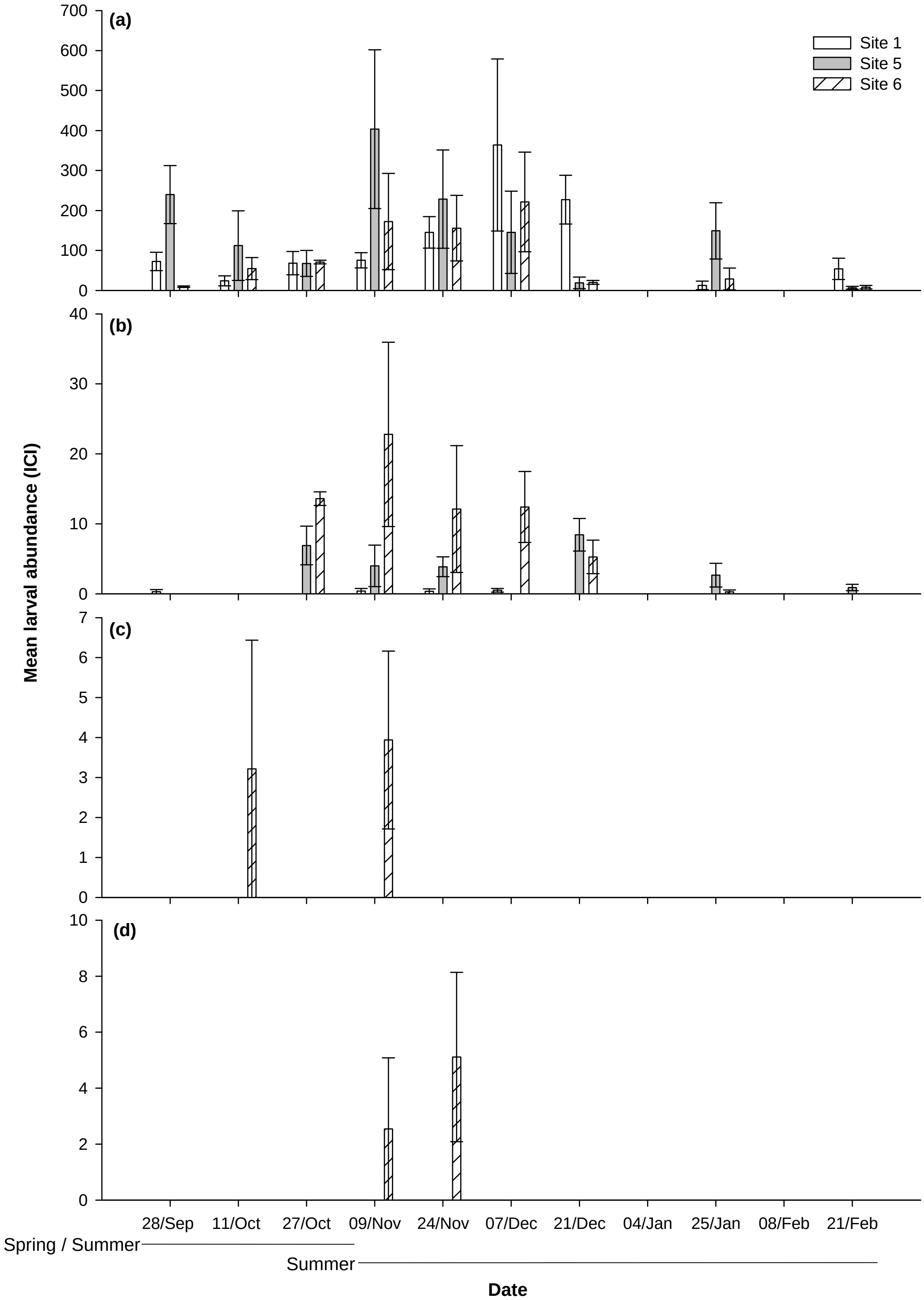

Figure 4.6. Mean preflexion larval abundance (ICI ± standard error) throughout the sampling for non grouped species, (a) flathead gudgeon (b) hardyhead, (c) Murray cod*#, and (d) freshwater catfish*#. Note * indicates postflexion larvae that were corrected for approx age, # indicates very low numbers of individuals.

4.4 DISCUSSION

The most abundant species were the small-medium bodied natives, whilst large bodied and exotic species were collected in lower abundances. The fish species collected represented opportunistic (Australian smelt, bony herring, carp gudgeon, flathead gudgeon and hardyhead) and equilibrium (Murray cod and freshwater catfish) spawners, whilst no periodic (golden perch and silver perch) spawners were collected under the low flow conditions (Chapter 3; Winemiller 1989b; Winemiller and Rose 1992; Humphries *et al.* 1999). Furthermore, the equilibrium spawners were only present at two of the three sites, both in the floodplain section. Peaks in total larval abundance did not consistently represent the timing of peaks of preflexion larvae for the highly abundant species, in the Lower Murray River. Therefore, abundance of preflexion larvae was used to estimate spawning activity rather than the later developmental stages. Timing, abundance and duration of preflexion larvae were staggered for each species, and differed between sites for individual species. Seasonal variation contributed more strongly to observed differences in the preflexion larval assemblage than spatial variation across the three sites samples.

Strong within season patterns have been identified in the larval assemblages in both temperate (e.g. Holland 1986; Turner *et al.* 1994) and tropical (e.g. Bialetzki *et al.* 2005; Agostinho *et al.* 2008; Sousa and Freitas 2008) river systems throughout the world. The timing of the spawning period and the timing of the main spawning event is an important aspect of fish ecology, as it aims to place larvae into the system under the optimal conditions for growth and survival (Bye 1984; Jobling 1995; Yaron and Sivan 2006). Although a number of years of investigation under a range of environmental conditions are required to confidently elucidate spawning time and required conditions, this study aimed to compare the timing of spawning across a number of species in a typical low flow year with those of a previous study which grouped similar species into three categories (King *et al.* 2003).

Larvae showed strong seasonal patterns in presence and abundance, which is consistent with other studies throughout the River Murray system (e.g. Humphries *et al.* 2002; King *et al.* 2007; Vilizzi *et al.* 2007; Leigh *et al.* 2008). Seasonal spawning guilds have previously been suggested for Murray-Darling Basin fishes by King *et al.* (2003) using a review of larval occurrence from other studies through out the Murray region (Figure 4.1). Using timing and abundance of preflexion larvae for individual species, the spawning patterns identified in this study were consistent with the spring/summer and summer seasonal spawning guilds for all species. The spring only period (defined by King *et al.* 2003) was not sampled during this study.

To maximise resources and suitable conditions fishes can also have discrete (small targeted period aimed at suitable conditions) or protracted (longer period aimed at increasing the chance of encountering suitable conditions) spawning periods. The protracted spawning of carp gudgeon identified in the Lower River Murray contrasts with the discrete period found in the tributaries of the mid Murray (Humphries *et al.* 2002). In contrast, carp gudgeon in the Ovens River and the Barmah Millewa Forest were protracted spawners, although spawning occurred later in the season starting in October (King *et al.* 2003; King *et al.* 2007). However, the spawning period in the Lower River Murray appeared to be approximately one month longer, starting in September, which is consistent with results from the Chowilla Anabranch in South Australia (Leigh *et al.* 2008) and the Lindsay Island system in Victoria (Meredith *et al.* 2002). It is likely that temperature differences between the regions may explain the variations in seasonal activity; Eleotrid spawning has been documented to be highly temperature dependent. (McDowall 1996). Additionally, the differences may be a result of the carp gudgeon being treated as a complex (Bertozzi *et al.* 2000), as individual species may spawn at slightly different times resulting in an overlap and the appearance of a protracted season. Genetic studies in the future may allow for isolating the spawning times of individual *Hypseleotris* species and to determine if the nature of spawning varies throughout the Basin. The spawning period identified for Australian smelt was shorter in the Lower River Murray than in the tributaries of the mid Murray, where they have been documented to occur for up to nine months of the year (Humphries *et al.* 2002). The shorter time frame for Australian smelt identified in this study is also consistent with results from the Chowilla Anabranch and Lindsay Island systems (Meredith *et al.* 2002; Leigh *et al.* 2008). Carp have previously been identified to spawn in the Lower River Murray for up to seven months, starting in November (Smith and Walker 2004). In the Barmah-Millewa region carp larvae have been recorded from October to December, and have also demonstrated a highly flexible spawning strategy, spawning for a second discrete period in February during a flood event (King *et al.* 2007). In contrast, preflexion larval carp were collected earlier in the current study, being present from September to November, suggesting a prolonged, although early (potentially spring) spawning season. Similarly, in both the Chowilla Anabranch and Lindsay Island systems, carp larvae had early protracted spawning seasons and were present from September to December, but were suggested to have peaked in abundances prior to the sampling commencement (Meredith *et al.* 2002; Leigh *et al.* 2008).

Many international studies have documented significant longitudinal variations in larval, juvenile and adult fish assemblages. A variety of factors including environmental conditions, food availability and habitat morphology have been identified as key drivers for spatial variation (e.g. Schlosser 1982; Oberdorff *et al.* 1993; Ostrand and Wilde 2002; Habit *et al.* 2007; Baumgartner *et al.* 2008a). In this study, there were few consistent patterns identified in the spatial variations of larval assemblages within one year. There were differences in the abundance of each species between sites, which were most likely due to the surrounding habitat characteristics (see Chapter 3 for a detailed discussion). However, the overall spawning time did not differ between sites within the Lower River Murray.

In temperate systems, spawning time is often correlated with day length, temperature and food availability. Seasonal timing of spawning within a year is likely to be driven by these factors, however, long term (preferably greater than 5 years) studies are required before environmental correlates for spawning activity can be accurately identified. The temporal patterns identified in this study may relate to a variety of influences, which will have evolved as specific strategies through time, including water quality tolerances and influence of resource competition. King *et al.* (2003) highlights that seasonal pattern in spawning may also influence the ability of individual species to utilise increases in flow.

This study was temporally restricted with only one sampling year, however, the results indicate that the seasonal spawning guilds were consistent with those previously identified. This suggests that these spawning guilds can be extrapolated to management of fish in the Lower River Murray. An understanding of seasonal spawning variability and environmental correlates is particularly important in systems where management strategies are being defined for rehabilitation. River regulation is believed to be the driving factor of the decline of many Murray-Darling Basin fishes (Cadwallader 1978; Gehrke *et al.* 1995; Humphries *et al.* 2002; Richter *et al.* 2003), and fishes throughout the world (Toth *et al.* 1998; Dudley and Platania 2007; Zeug and Winemiller 2008; Winter *et al.* 2009), hence using environmental water allocations to provide a more natural flow regime is highlighted as a key restoration action for native fish (Marchetti and Moyle 2001; Arthington *et al.* 2006). For fish that are reliant on increased flows for spawning or larval survival, managed flows should consider attributes of the flow regime, such as timing and duration (Poff *et al.* 1997), in relation to life history strategies and timing of spawning for the key species within the system (Nesler *et al.* 1988; Travinchek *et al.* 1995; King *et al.* 1998; Freeman *et al.* 2001). Given that riverine fish populations throughout the world are under threat, further understanding of the specific seasonal dynamics within a system are important for management and rehabilitation.

CHAPTER 5: ZOOPLANKTON AND LARVAE: LINKAGES BETWEEN SPATIAL AND SEASONAL DIFFERENCES IN THE MAIN CHANNEL OF A LOWLAND TEMPERATE RIVER, AUSTRALIA

ABSTRACT

Food is often thought to be a major limiting factor in the survival of the early life stages of fish. The assumptions of key floodplain river recruitment models suggest that in the absence of overbank flows the main river channel does not provide adequate densities of pelagic zooplankton for larval survivorship, however recent studies have disproved this theory. The Lower River Murray, South Australia is a heavily regulated floodplain river, which is currently experiencing extreme low flow conditions. This study examined the zooplankton composition in the main channel environment in an attempt to relate these to larval fish populations. The aims were first to describe the riverine zooplankton composition and densities in the main channel during a low flow year; second, to describe the dietary composition of Australian smelt; and finally, to compare the dietary composition of Australian smelt with the riverine zooplankton composition. Zooplankton sampling was conducted during spring/summer of 2006/07 during a typical low flow year in the main channel of the Lower River Murray, South Australia. The riverine zooplankton composition was dominated by rotifers, which are a known food source for larvae. Densities of zooplankton were generally greater than the acceptable range for survival of larvae in the wild. The pelagic zone of the main channel of the Lower River Murray, South Australia supports high densities of zooplankton during a low flow year, and therefore food does not appear to be a limiting factor for larval survival.

5.1 INTRODUCTION

Larval fish survival is intricately linked to their surrounding environment. The early life period is a time of exceptionally high mortality (90-99%) (Kelso and Rutherford 1996; Trippel and Chambers 1997) which is commonly attributed to either starvation or predation. Starvation commonly occurs during the shift from endogenous to exogenous feeding following the depletion of the yolk-sac, referred to as the *critical period* (Horjt 1914; May 1974). Cushing (1990) suggested that in marine systems, larval growth and survivorship relies upon the degree of temporal overlap between the fish spawning season and zooplankton occurrence, which is reflected in the *match/mismatch* hypothesis (Figure 1.1, Cushing 1990). This model has been adapted for riverine systems, with many authors suggesting that floodplain inundation is necessary to produce the required high densities of prey (Welcomme 1985; Junk *et al.* 1989; Harris and Gehrke 1994; Schiller and Harris 2001).

Models of recruitment variability in fishes typically assume that food availability is the key limiting factor during the larval phase, and that other factors such as growth and predator avoidance are inherently linked to prey resources (Cushing 1990; Jobling 1995; Houde 1997). It is this relationship that has formed the basis for many of the recruitment models currently in use, including the *flood pulse concept* (Junk *et al.* 1989). The flood pulse concept suggests that the lateral flux of nutrients from the floodplain during overbank floods is integral in providing the necessary pulse in primary and secondary productivity, ensuring that larvae and juvenile fish have access to an abundance of food and habitats (Junk *et al.* 1989). The assumption here is that main channel habitats do not provide adequate prey densities for larval survival. The *low flow recruitment hypothesis* predicts that in the absence of floodplain inundation, prey occur in higher densities into specific main channel habitats (embayments, backwaters and still littoral zones), and densities are high enough to support the larvae and juveniles. King (2004a) established in the Broken River that abundant prey are present throughout all main channel habitats (including the mid river channel) during a low flow period, and therefore this may not be influencing habitat use by larvae (King 2005).

As fish grow and develop their physiology and morphology changes, allowing them to be more selective in their choice of food and habitat. This change with development in ecological niches and resources is described as ontogenetic niche shift (Werner and Gilliam 1984). A number of Murray-Darling Basin fish species have been documented to exhibit ontogenetic dietary changes (Gehrke 1992; King 2005). Dietary compositions for larval stages of some Murray-Darling Basin species have been investigated. The majority of these have been conducted under laboratory or hatchery conditions (Lake 1967b; Gehrke 1991;

Arumugam and Geddes 1992; Rowland 1992, 1996; Warburton *et al.* 1998; Tonkin *et al.* 2006; Ingram and De Silva 2007), with some field studies in recent years (Gehrke 1992; Lieschke and Closs 1999; King 2005; Kaminskas and Humphries 2009). Furthermore, researchers need to consider that different habitats are likely to result in differing dietary compositions within the same species. For example: in billabongs adult Australian smelt (*Retropinna semoni*) fed predominately in the pelagic zone on zooplankton (Lieschke and Closs 1999). In contrast, King (2005) identified that in riverine populations whilst Australian smelt larvae fed predominately in the pelagic zone, adults moved to surface feeding on terrestrial invertebrates and chironomid pupae.

Relatively few studies have been conducted on the zooplankton composition in the main channel of the Murray-Darling Basin (although see Shiel *et al.* 1982; Shiel 1985; King 2004a). Zooplankton in the Lower River Murray, South Australia are commonly comprised of micro-crustaceans, rotifers, and the early stages of macroinvertebrates, chironomids and trichopterans (Shiel *et al.* 1982). Additionally, pelagic zooplankton have been recorded in relatively low densities (<150 zooplankters per litre) under relatively high flow (discharge) conditions in the main channel of the Lower River Murray (Shiel *et al.* 1982; Shiel 1985). In contrast, many overseas studies have established that zooplankton abundances are higher in low velocity areas of the main channel as a result of increased water residence times, and that this may act as a source of zooplankton to the flowing river channel (Ferrari *et al.* 1989; Pace *et al.* 1992; Thorp *et al.* 1994; Basu and Pick 1996; Reckendorfer *et al.* 1999; Reynolds 2000). King (2004a) found that under flows contained within the channel the pelagic zooplankton density was low (< 500 zooplankters per litre), but the epibenthic zone supported very high densities (> 1000 zooplankters per litre).

Due to the presence of ten weirs the Lower River Murray is no longer representative of the naturally free flowing river it was historically. The river now is a series of slow flowing, deep, weir pools more representative of a series of isolated lakes (Walker 1992; Walker 2006b). Native fish throughout the Murray-Darling Basin have declined in range and abundance following river regulation, with poor survival of early life stages being implicated as the cause, more so than spawning failure (Cadwallader 1978; Gehrke 1991; Gehrke *et al.* 1995; Humphries and Lake 2000; Humphries *et al.* 2002; Koehn and Harrington 2006). However, further research is needed into the factors influencing survival of early life stages throughout the Murray-Darling Basin; but particularly in the Lower River Murray given the uniqueness of the region. Under low flow conditions high abundances of larvae have been recorded in the pelagic zone of the mid river channel (see Chapters 3 and 4), suggesting that prey density may not be a limiting factor. In the Lower

River Murray, the mid river channel is very deep in parts, and therefore larvae are likely to be reliant on prey densities in the pelagic environment rather than the benthic.

The main objective of this study was to describe the pelagic zooplankton composition in the river channel open water habitats within the Lower River Murray, South Australia as potential prey for larvae. The aims were to describe the composition and the density of potential prey items in the river channel environment during a low flow year. Second, to describe the dietary composition of an abundant species in the river, Australian smelt (*R. semoni*) through its larval development; and finally, to compare the dietary composition of Australian smelt with the riverine zooplankton composition. It was predicted that the densities of potential prey items would be relatively low in the main channel environment during a low flow year. The dietary composition of Australian smelt was expected to exhibit changes with ontogeny and to emulate the riverine zooplankton composition in early stage larvae, but that later stages may exhibit some prey selectivity.

5.2 MATERIALS AND METHODS

5.2.1 Study sites

The present study occurred in the main river channel of the Lower River Murray in South Australia, in both the gorge and floodplain regions (Figure 1.4). Sampling was conducted at three sites: in the tailwaters 5 km downstream of Weir 1 (Site 1 34°21.138'S, 139°37.061'E), Weir 5 (Site 5 34°13.246'S, 140°45.0909'E) and Weir 6 (Site 6 33°59.725'S, 140°53.152' E) (Figure 1.4). The area surrounding Site 1 has been described as the gorge region and the area surrounding Site 5 and Site 6 as the floodplain region (see Chapter 1 for a detailed discussion). Despite the surrounding characteristics of the floodplains being different between the sites, the main channel habitat is generally similar, being wide, deep, slow flowing pool habitats.

5.2.2 Sampling regime

Zooplankton sampling was conducted during the spring/summer of 2006/07, during the spawning season and peak larval abundance for most species within the river system (Chapter 3; Humphries *et al.* 2002; Meredith *et al.* 2002). Sampling consisted of eight trips conducted from September through to February (Table 4.1). Each site was sampled once during the day and at then at night of the same day, and all three sites were sampled within a four-day period.

Table 5.1. Sampling trips and dates used in the analyses and graphs[5]**.**

Trip Number	Sampling week	Date used in analyses/graphs
1	25-29 Sept	28 Sep
2	09-13 Oct	11 Oct
3	23-27 Oct	24 Oct
4	20-24 Nov	24 Nov
5	04-08 Dec	07 Dec
6	18-22 Dec	21 Dec
7	22-26 Jan	25 Jan
8	19-23 Feb	21 Feb

Zooplankton were sampled using a four litre Schindler trap (Schindler 1969) in the pelagic zone (top 2 m of water column) of the open water in the centre of the main channel. Three Schindler trap samples were collected below the water surface during the day and at night of the same day; resulting in three day and three night samples for each site during each trip. The water was sieved through 35 µm mesh, washed into a storage jar, using filtered water, and preserved using 70 % ethanol *in sit*u and returned to the laboratory for sorting and identification.

5.2.3 Sorting and identification

Zooplankton samples were decanted into a 200 ml glass measuring cylinder, the bottle flushed with 70% ethanol to remove adhering plankters and then the sample volume was measured and recorded. The cylinder was capped with Parafilm®, inverted three times to distribute the contents evenly, and a 1 ml Gilson Pipetman® auto pipette used to extract a 1 ml aliquot from the approximate centre of the agitated volume. This 1 ml aliquot was run into a Graticules (UK) Sedgewick-Rafter (S-R) counting chamber, and the contents enumerated and identified. Species were identified using keys in Shiel (1995) and references therein. The total count of plankters in 1 ml was multiplied by the number of millilitres in the original sample volume to provide an estimate of density in the four litre trap volume. The accuracy of this method had previously been established to be ± 2 % by taking triplicate aliquots and calculating standard deviation and standard error. Zooplankton data were standardised to density per litre, the CPUE defined as the abundance of zooplankters per litre of water, using the combined catch from one day and one night sample (n=3 per site and trip).

5.2.4 Dietary analysis for Australian smelt

Validation that larvae were consuming the zooplankton recorded in the water was performed for Australian smelt (*Retropinna semoni*). Australian smelt was selected as a representative for dietary analysis as it is a highly abundant generalist species in the river

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[5] Note all sampling days were used for analyses and but for aesthetics in graphs only one date from this period was presented.

channel at these sites (see Chapter 3). Fish were collected only during trip four (20-24th of November) in 2006, following the methods outlined in Chapter 3. Following identification and staging 10 fish from each developmental category (preflexion, postflexion and metalarvae) were selected for dietary analysis; damaged fish or fish with empty guts were not included. The standard length of each fish was recorded via an eyepiece graticule (fish < 10 mm length) or with vernier callipers (fish > 10 mm). Maximum mouth gape (greatest width when viewed ventrally) was also recorded.

The gut (defined as the entire gastrointestinal tract) was extracted and opened with fine needles. The gut content volume was assigned values ranging from one, less than 30% full, to three when it was full (Carassou *et al.* 2009). Prey items were identified to family level using published guides (Williams 1980; Shiel 1995; Hawking and Smith 1997) and consultation with Russel Shiel (University of Adelaide, pers. comm.). The abundance of families present in the gut was recorded as percentage contribution.

5.2.5 Data analysis

Zooplankton data were analysed at the family level. Bray-Curtis similarity measures were used for analysis of the riverine zooplankton composition (Bray and Curtis 1957). The seasonal and spatial patterns in the riverine zooplankton composition were examined using non-metric multidimensional scaling (NMS) ordination (McCune and Mefford 1999). Prior to analysis the zooplankton data were tested for normality and homogeneity of variance. Given that very few variables met the assumptions, data were analysed using permutational analysis of variance (ANOVA, Anderson 2001b). A two-way design was used to detect any differences among *site* and *trip*; both were treated as random factors. Total zooplankton density was analysed using a univariate analysis. The riverine zooplankton assemblage was square-root transformed to reduce the influence of highly abundant species on the similarity measure (Clarke 1993). A two-way multivariate analysis was used for the riverine assemblage data. Unrestricted permutations of data were performed with 999 permutations to detect differences at α=0.05 (Anderson 2001b). Where significant differences were detected pairwise analysis was performed to determine which sites and or trips differed.

To examine ontogenetic changes in diet, an NMS ordination (using Bray-Curtis similarity measures) was conducted on the percentage contribution data for the prey items, excluding unidentified material and rare prey items (<1% contribution). A one-way multivariate PERMANOVA was also conducted to determine if differences in diet between stages were significant, *stage* was treated as a fixed factor.

The riverine zooplankton samples collected during November at Site 5 were used to determine if Australian smelt were demonstrating prey selectivity. The percentage contribution of zooplankton to the riverine composition was calculated for comparisons with the dietary composition. A one-way multivariate PERMANOVA was conducted to determine if differences between the diet for each stage and the river composition were significant, s*tage/river* was treated as a fixed factor. Where significant differences were detected pairwise tests were performed to identify where the differences occurred. A similarity percentages (SIMPER) analysis was also performed to compare the diet composition of each developmental stage to the riverine composition, a 90 % cumulative cut-off was applied.

5.3 RESULTS

5.3.1 Riverine zooplankton composition

Catch summary and total density

In total, 40 family groups, consisting of 8 classes, were collected from all sites during the study. Rotifera was the most dominant phyla overall (70% at Site 1, 78.8% at Site 5 and 75.6% at Site 6; Table 5.2). Within the Rotifera phylum, the families Brachionidae and Synchaetidae dominated. Trochosphaeridae and Conochilidae also contributed at Sites 5 and 6, respectively and at Site 1 the Difflugiidae (Rhizopodea) were also a significant contributing family (Table 5.2). The riverine zooplankton composition was dominated by fauna less than 1 mm in size (98.36% at Site 1, 99.24% at Site 5 and 99.17% at Site 6; Table 5.2). The 1- 3 mm and 3 mm size classes contributed less than 2% at each site (Table 5.2).

The patterns in total density of zooplankton were different between sites and trips, as indicated by the site x trip interaction (Table 5.3). The total density of zooplankton changed throughout the study period (Figure 5.1). Although there were varied patterns between sites, densities during the early trips were lower than the mid and late season (Figure 5.1; Table 5.3). There were differences in the total density between sites, but differences were not consistent between trips (Table 5.3).

Table 5.2. Total percentage contribution to each site of each taxonomic group to the riverine zooplankton composition. Data are presented as the total percentage collected at Sites 1, 5 and 6 in all trips. The size class of zooplankton is also indicated.

Phylum/ Sub phylum	**Class**	**Order**	**Family**	**Size class (mm)**	**Site 1**	**Site 5**	**Site 6**
ARTHROPODA							
Crustacea	Cladocera	Anomopoda	Bosminidae	< 1	1.38	3.28	6.02
			Chydoridae	< 1	0.00	0.05	0.00
			Daphniidae	< 1	0.04	0.11	0.00
			Daphniidae	1-3	0.00	0.00	0.06
			Ilyocryptidae	< 1	0.27	0.05	0.06
			Moinidae	< 1	0.21	0.22	0.25
	Copepoda	Calanoida	Centropagidae	< 1	3.93	6.37	7.75
			Centropagidae	1-3	0.49	1.54	1.40
		Cyclopoida	Cyclopidae	< 1	1.43	0.48	0.58
	Macroinvertebrate	Tubificoidea	Naididae	< 3	0.00	0.06	0.00
	Ostracoda	Ostracoda	Ostracod	< 1	0.00	0.09	0.13
PROTOZOA							
Ciliophora	Ciliatea	Ciliophora	Colepidae	< 1	0.00	0.00	0.06
		Euplotida	Euplotidae	< 1	0.04	0.00	0.00
		Haptorida	Didiniidae	< 1	0.04	0.00	0.00
		Heterotrichida	Stentoridae	< 1	0.04	0.00	0.00
		Peritrichida	Epistylidae	< 1	0.96	1.96	1.01
			Vorticellidae	< 1	2.32	1.83	2.78
		Rhabdophorina	Tracheliidae	< 1	2.64	0.38	0.19
Sarcomastigophora	Rhizopodea	Rhizopoda	Arcellidae	< 1	0.00	0.05	0.00
			Centropyxidae	< 1	0.25	0.60	0.33
			Cyphoderiidae	< 1	0.16	0.00	0.25
			Difflugiidae	< 1	16.09	4.02	3.30
			Euglyphidae	< 1	0.00	0.00	0.07
			Lesquereusiidae	< 1	0.04	0.00	0.12
			Rhizopoda	< 1	0.00	0.00	0.07
	Dinophycea	Dinophyta	Ceratiaceae	< 1	0.00	0.06	0.00
ROTIFERA							
	Eurotatoria	Bdelloidea	Habrotrochidae	< 1	0.04	0.00	0.00
			Philodinidae	< 1	0.12	0.32	0.23
	Monogononta	Flosculariacea	Conochilidae	< 1	1.02	5.38	14.03
			Flosculariidae	< 1	0.04	0.00	0.00
			Hexarthridae	< 1	3.39	3.56	2.22
			Testudinellidae	< 1	0.04	0.25	0.00
			Trochosphaeridae	< 1	5.88	8.18	9.34
		Ploima	Asplanchnidae	< 1	0.16	0.51	3.51
			Brachionidae	< 1	27.62	17.54	11.84
			Dicranophoridae	< 1	0.00	0.00	0.06
			Epiphanidae	< 1	1.13	0.62	0.80
			Gastropodidae	< 1	0.19	0.05	0.00
			Lecanidae	< 1	0.07	0.10	0.05
			Notommatidae	< 1	0.00	0.00	0.11
			Synchaetidae	< 1	22.42	34.95	25.86
			Trichocercidae	< 1	7.37	6.96	7.29
		Unknown	Indet. Contr. rotifer	< 1	0.16	0.42	0.24
			Total raw number of individuals		**69,280**	**53,238**	**44,977**

Table 5.3. Two-way univariate PERMANOVA for differences among sites and trips for the riverine zooplankton density. Bold text indicates significant value.

Source of variation	*df*	MS	*p*
Site	2	1511.60	**0.021**
Trip	7	283.94	0.590
Site x Trip	14	465.38	**0.001**
Residual	48	67.21	

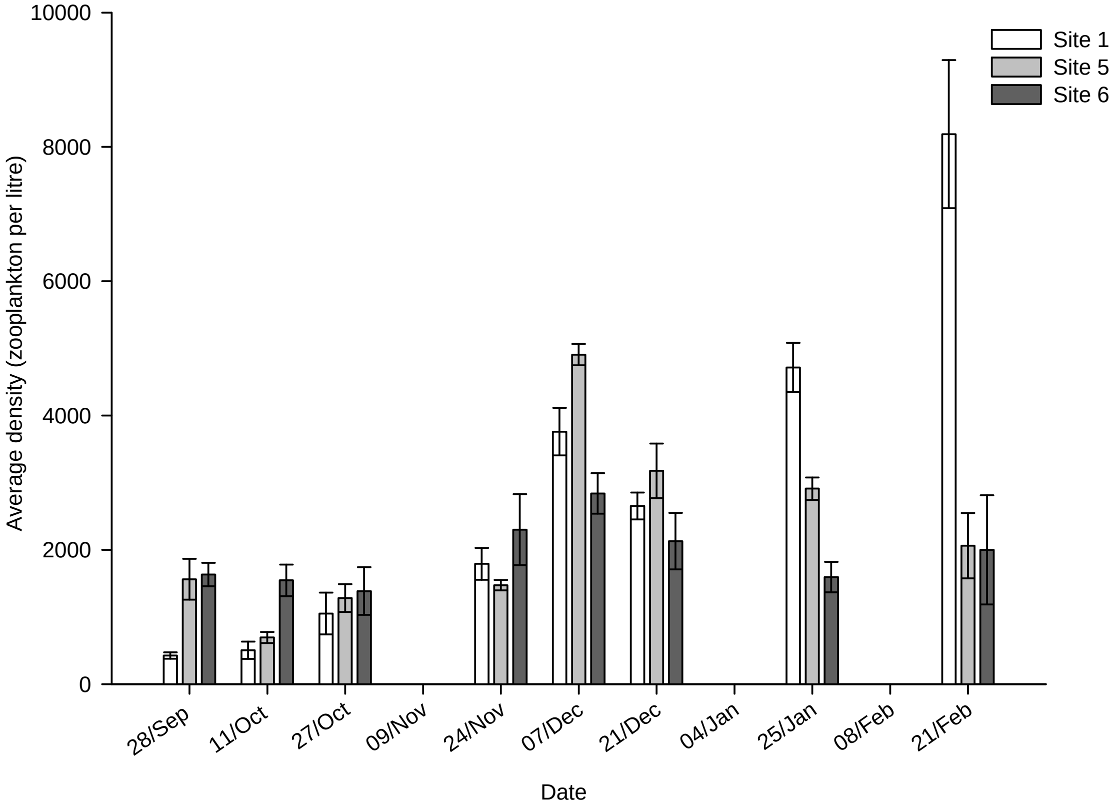


Figure 5.1. Average total density of zooplankters per litre ± standard error recorded during each trip at Sites 1, 5 and 6 throughout the sampling period. No sampling was conducted on the 9th Nov, 4th Jan and 8th Feb.

Spatial and temporal variation in riverine zooplankton composition

The riverine zooplankton composition varied through space and time (Figure 5.2; Table 3.6). The riverine zooplankton composition at Site 1 was generally different from Sites 5 and 6. Site 1 had a much broader dispersion of samples, while Sites 5 and 6 grouped more closely together and have less within-site variation (Figure 5.2a). There were significant differences between trips for all sites, and although the dispersion of samples within trips was broad, a visual progression from trip one through to trip eight indicated that temporal differences followed a stronger pattern than spatial differences (Figure 5.2b).

Table 5.4. Two-way multivariate PERMANOVA for differences among sites and trips for the riverine zooplankton composition. Bold text indicates significant value.

Source of variation	*df*	MS	*p*
Site	2	4186.40	**0.009**
Trip	7	4404.90	**0.001**
Site x Trip	14	1497.20	**0.001**
Residual	48	454.01	

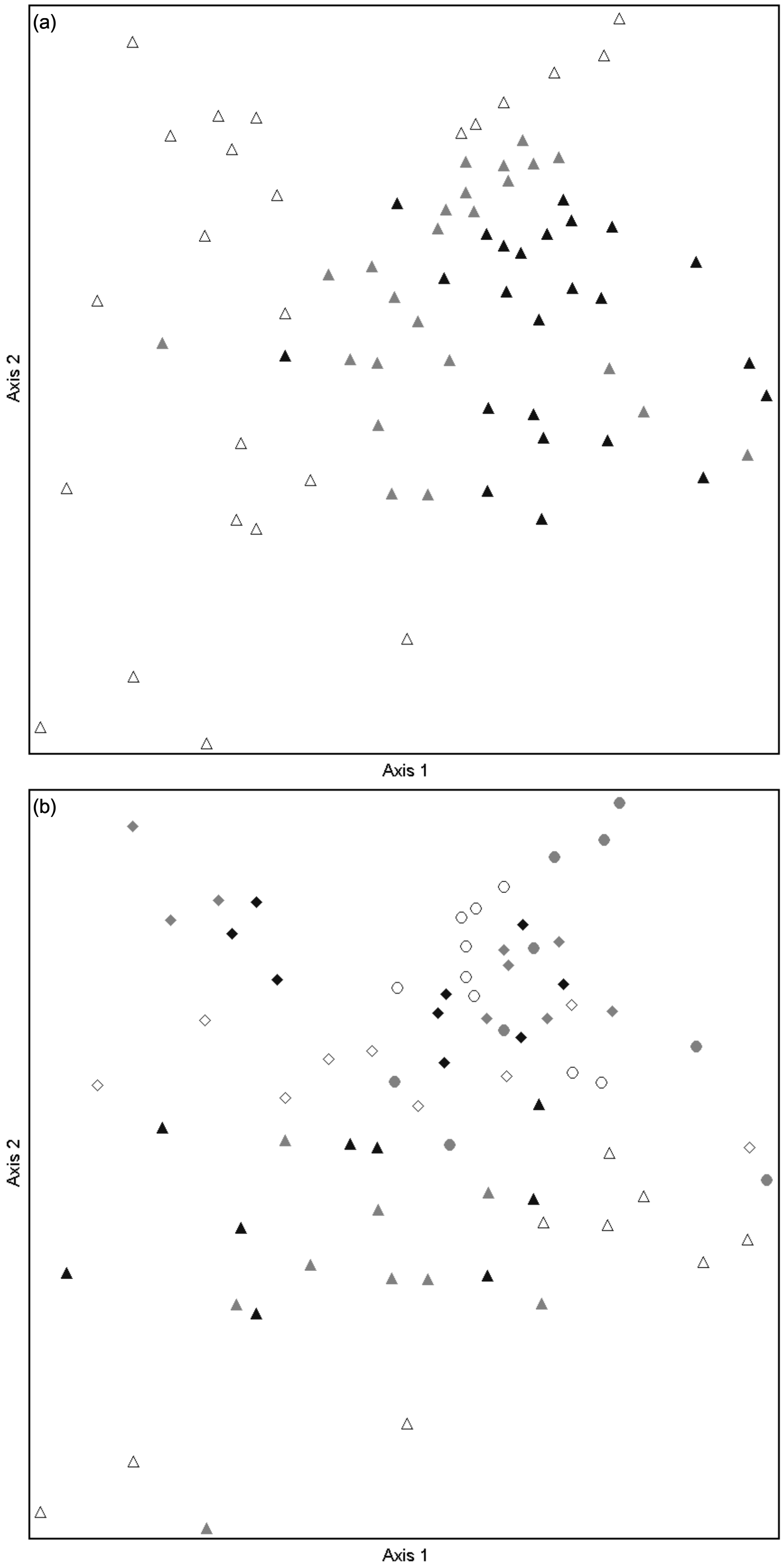


Figure 5.2. Two dimensional NMS ordination (stress 0.19) of the riverine zooplankton composition (a) showing the spatial variation between Site 1 (△), Site 5 (▲) and Site 6 (▲) and (b) the seasonal separation (trip △1, ▲ 2, ▲ 3, ◇ 4, ◆ 5, ◆ 6, ○ 7 and ●8).

5.3.2 Dietary composition of larval Australian smelt and comparison to riverine zooplankton assemblage

Although empty guts were excluded from the analysis, percentage gut fullness increased with development for Australian smelt larvae (Table 5.5). With the exception of the unidentified matter which decreased with each developmental stage, the most common prey items were rotifers (Rotfiera), bosminids (Cladocera) and centropagids (Copepoda) (Table 5.5).

Table 5.5. Range and mean ± standard error of standard length, mean ± standard error gape size and gut content volume (1= 5-50%, 2= 50-99% 3 = full) and mean percentage contribution of dominant zooplankton to dietary composition for each developmental stage of Australian smelt (only the most dominant items were included). (n = 10 for each developmental stage).

		Preflexion	Postflexion	Metalarvae
Standard length (mm)	**Range**	5.5-8.9	9.1-12.5	16.5-19.3
	Mean (± S.E.)	7.29 (0.25)	11.01 (0.40)	17.88 (0.39)
Gape mean (± S.E.)		0.36 (0.02)	0.55 (0.02)	1.33 (0.06)
Percentage of fish in each gut content volume category	**1**	90	40	10
	2	10	60	10
	3	0	0	80
DIETARY COMPOSITION				
Rotifera		26.8	25.9	6.0
Arthropoda				
Crustacea				
Cladocera				
Bosminidae		0	4.6	16.5
Chydoridae		0	0	3.9
Daphniidae		0	0	1.9
Copepoda				
Copepod nauplii		1.3	3.4	0
Centropagidae		2.9	9.1	9.7
Cyclopidae		1.1	5	3.2
Insecta				
Diptera				
Chironomidae		0	0.2	0.6
Chironomidae pupae		0	0	3.3
Terrestrial invertebrate		0	0	4.4
Algae		6.6	4.0	0
Unidentified matter		25.4	16.8	10.7

The diet of Australian smelt varied significantly with developmental stage (d.f. = 2, MS = 13218, p = 0.001; Figure 5.3). In general the diet of preflexion larvae was dominated by rotifers, algae and centropagids (Figure 5.3). These prey items were also characteristic of the postflexion larval diet, although the composition of the diet became more diverse (Figure 5.3). The diet of metalarvae shifted to be dominated by bosminids and centropagids, and included new items such as insects and terrestrial invertebrates (Figure 5.3).

The riverine zooplankton composition was dominated by rotifers, centropagids and ciliates and rhizopods (Protozoans) (Figure 5.3). The presence of ciliates and rhizopods in the river zooplankton composition was a characterising feature that appeared to be absent from the diet of Australian smelt (Figure 5.3). The riverine zooplankton composition was significantly different from the dietary composition of each of the developmental stages for Australian smelt (d.f. = 3, MS = 10934 p = 0.001; Figure 5.3). Percentage composition of rotifers, ciliates and rhizopods were higher in the riverine composition, and contributed greater than 70% to the observed differences between the diets and the riverine composition (Table 5.6). Additionally, each developmental stage had individual characteristics (Table 5.6). The preflexion diet was characterised by a lower percentage contribution of centropagids and bosminids, and a higher contribution of algae (Table 5.6). The postflexion diet was further characterised by higher percentage composition of centropagids, cyclopoids and algae (Table 5.6). The metalarval diet had a higher percentage composition of bosminids, centropagids, terrestrial invertebrates and chydorids (Table 5.6).

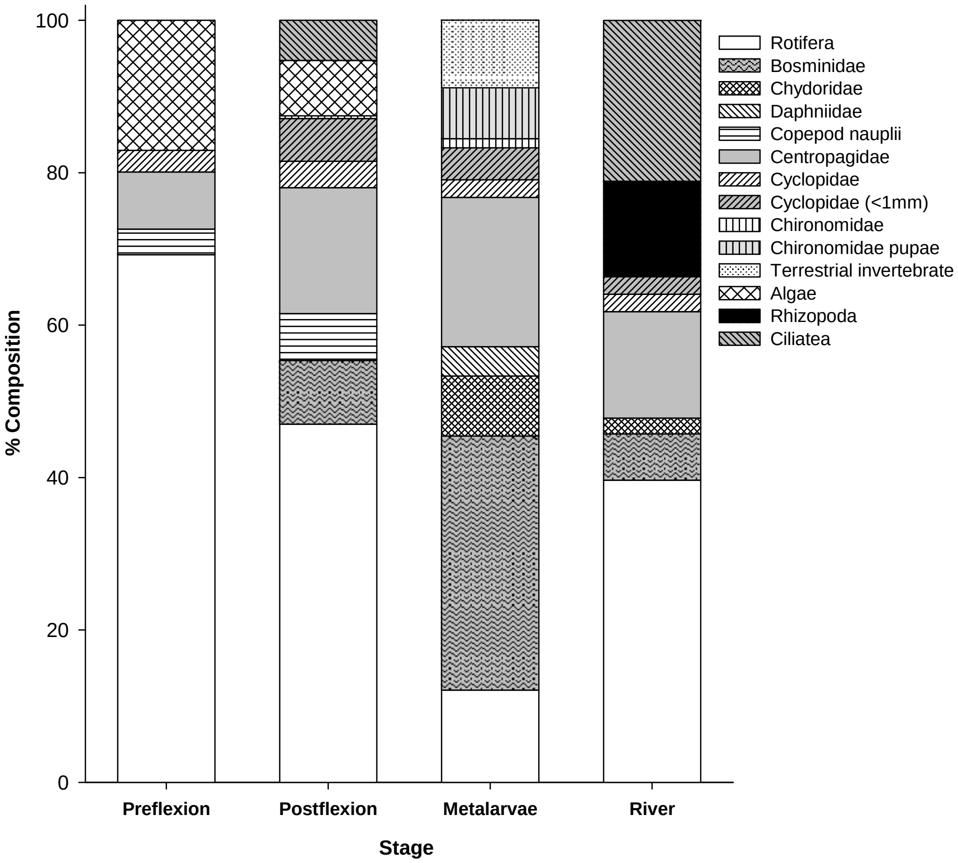


Figure 5.3. Mean percentage composition of zooplankton in the diet of each developmental stage of Australian smelt, and the River Murray samples. Note fish with empty guts and unidentified matter was excluded. (n=10 for each stage).

Table 5.6. SIMPER analysis for the comparison between the riverine zooplankton composition and the diet of preflexion, postflexion and metalarvae of Australian smelt. Results are based on percentage contribution data. CR (consistency ratio) indicates zooplankton distributions between diet and river, with larger values indicating greater consistency. The contribution (%) indicates the proportion of difference between diet and river (shown by PERMANOVA) attributable to individual zooplankton families. Mean dissimilarity is expressed as a percentage ranging from 0% (identical) and 100% (totally dissimilar).

Taxa	Mean percentage composition		CR	Contribution (%)	Cumulative contribution
	Preflexion	**River**		**Mean dissimilarity = 57.46**	
Rotifera	26.8	66.3	3.25	50.74	50.74
Ciliatea	0.0	11.7	1.28	14.8	65.54
Rhizopoda	0.0	7.0	3.84	8.88	74.42
Algae	6.6	0.0	0.8	7.44	81.86
Centropagidae	2.9	7.7	1.59	6.58	88.44
Bosminidae	0.0	4.7	1.46	5.92	94.36
	Postflexion	**River**		**Mean dissimilarity = 53.89**	
Rotifera	25.9	66.3	3.47	50.31	50.31
Ciliatea	0.0	11.7	1.28	14.39	64.71
Rhizopoda	0.0	7.0	3.79	8.64	73.34
Centropagidae	9.1	7.7	1.5	7.26	80.6
Cyclopidae	5.0	1.3	1.06	5.47	86.07
Algae	4.0	0.0	0.77	4.42	90.49
	Metalarvae	**River**		**Mean dissimilarity = 77.52**	
Rotifera	6.0	66.3	5.34	52.8	52.8
Ciliatea	0.0	11.7	1.28	10.2	62.99
Bosminidae	16.5	4.7	1.39	10.13	73.12
Rhizopoda	0.0	7.0	3.78	6.12	79.24
Centropagidae	9.7	7.7	1.82	5.77	85.01
Terrestrial invertebrates	4.4	0.0	0.77	3.61	88.62
Chydoridae	3.9	0.0	1.11	3.18	91.8

5.4 DISCUSSION

The key assumption of the critical period and match/mismatch hypotheses is that the early life stages must encounter high abundances of appropriately sized prey to ensure larval survival and successful recruitment (Hjort 1914; May 1974; Cushing 1990). Many fishes are poorly developed at hatch, with limited swimming ability and are gape-limited predators; therefore high densities of appropriately sized prey are required to sustain larval growth and survival (Houde 2002). In riverine systems, it has been proposed that only the inundated floodplain provides high enough prey densities, and that in the absence of floodplain inundation densities of prey in the main river channel will not be adequate to support developing fish larvae (Welcomme 1985; Junk *et al.* 1989). This leads to the assumption that during low flow years larvae will not encounter high enough densities of prey, which will be reflected as low recruitment (Arumugam and Geddes 1987; Geddes and Puckridge 1989; Harris and Gehrke 1994; Schiller and Harris 2001). However, this relationship has also been questioned by a number of larval studies (e.g. Humphries *et al.* 1999; King 2004a; Humphries *et al.* 2006; Zeug and Winemiller 2008).

Despite the proposal that in the absence of flood plain inundation primary and secondary production in the main river channel is low (Junk *et al.* 1989), zooplankton have been documented in high densities in rivers under low flow and velocity conditions (Ferrari *et al.* 1989; Pace *et al.* 1992; Thorp *et al.* 1994; Basu and Pick 1996; Reckendorfer *et al.* 1999; Reynolds 2000). However, in the River Murray and surrounding tributaries low densities (<150 zooplankters per litre) of pelagic zooplankton have been recorded in a range of main channel habitats (Shiel *et al.* 1982; Shiel 1985; King 2004a). In comparison, the results from this study demonstrate that high densities (>500 zooplankters per litre) of pelagic zooplankton occur in the openwater in the Lower River Murray during low flow conditions.

The prey densities required for the survival of larval Murray-Darling Basin fishes are largely unknown, but proposed densities in marine systems range between 100 and 1000 zooplankters per litre (May 1974; Bone *et al.* 1995). Aquaculture studies on Murray cod larvae have also documented no change in survival rates of larvae with densities between 250 and 5000 zooplankters per litre; however if the timing of the initial feeding was delayed survival rates decreased between 250 and 3000 zooplankters per litre (Rowland 1992). Timing of zooplankton occurrence is critical to the survival of larvae, fish must encounter sufficient densities of suitable prey during the shift from endogenous to exogenous feeding, termed the critical period (Hjort 1914; May 1974). Some Murray-Darling Basin fishes commence first feeding prior to the full absorption of the yolk sac (e.g. Murray cod *Maccullochella peelii peelii*, Australian smelt and carp *Cyprinus carpio*), while others used all of their yolk sac before pursuing prey (King 2005). The overlap between endogenous and exogenous feeding may reduce the chance of mortality as a result of starvation during the critical period; indeed King (2005) suggested that Murray cod may not have a critical period.

The timing of zooplankton occurrence was consistent with the occurrence of preflexion larvae for all recorded species in the main channel environment (see Chapter 4). The pelagic zooplankton composition in the mid river channel was therefore sufficient to support survival and growth of larvae. However, this study focused on the pelagic zone of the mid river channel and did not sample other main channel habitats (e.g. still littoral zones, backwaters, inshore bays or embayments). Nor did this study consider microhabitats such as the epibenthic zone, woody debris or emergent vegetation beds, therefore it is likely that the total zooplankton densities were significantly underestimated (King 2004a).

This further suggests that food is unlikely to be a limiting factor for growth and survival of larval fish in the main river channel of the Lower River Murray.

Macro- and micro-invertebrates are important dietary components for adult and larval Murray-Darling Basin fishes (Kennard *et al.* 2001). Many are opportunistic carnivores and a few are considered omnivorous (King 2005; Lintermans 2007). Larvae of Murray-Darling Basin fishes have been documented to feed predominately on algae, rotifers, copepods, cladocerans, cyclopoids and chydorids (Gehrke 1992; Vilizzi 1998; Lieschke and Closs 1999; King 2005; Kaminskas and Humphries 2009). Australian smelt are a dominant species within the Lower Murray, occurring in high abundances (Chapters 3 and 4) and therefore were used for dietary analysis. Larval Australian smelt in the Lower Murray fed predominately on algae, rotifers, cladocerans and copepods. These results are consistent with the diet identified for larval Australian smelt in the Broken River (King 2005). However, King (2005) also identified that benthic zooplankton formed a significant part of the diet for larvae of many Murray-Darling Basin fishes. It appears that benthic items did not constitute a dominant prey source for Australian smelt in the Lower River Murray; only metalarvae were identified to potentially consume benthic prey items such as chironomid pupae, however, chironomid pupae are not solely benthic as many rise to the surface to hatch and have been documented to occur amongst vegetation (Hawking and Smith 1997). However, the examination of the diet for Australian smelt in the current study was spatially and temporally restricted, and additional research is needed into dietary associations of a range of species in the Lower River Murray using larger sample sizes.

Protozoans are significant contributors to the diets of some marine species, however they are rarely recorded in gut contents as these soft bodied species are often quickly digested (Fukami *et al.* 1999; de Figueiredo *et al.* 2007). Protozoans (rhizopods and ciliates) were not recorded in the gut contents of Australian smelt, despite the high percentage contribution to the riverine zooplankton community. It may be that these are an important food source for developing larvae but may have contributed to the unidentified material in the guts. Protozoans would appear to be a suitable prey source given their high abundances in many systems, small size and slow swimming speed (von Herbing *et al.* 2001). Many studies may have underestimated the availability of prey for developing larvae by excluding protozoans (Fukami *et al.* 1999). Fatty acid and stable isotope analyses have identified protozoans in the diet of krill (Schmidt *et al.* 2006) and therefore, this may be a useful tool for larval gut contents analysis to avoid underestimating easily digestible prey items in future studies.

Ontogenetic shifts in diet commonly occur in fishes (Werner and Gilliam 1984; Winemiller 1989a; Garner 1996; Vilizzi 1998; Mol *et al.* 2000; King 2005; Tonkin *et al.* 2006). Some

Murray-Darling Basin fishes have been shown to exhibit dietary changes with ontogeny, with diets shifting from smaller prey items (namely rotifers) to include larger prey such as copepods, cladocerans and macroinvertebrates as the fish develop (Gehrke 1992; King 2005). In addition, most of these species show a relationship between mouth gape at feeding and size of the largest prey items (King 2005). An ontogenetic shift in diet was identified for Australian smelt from smaller prey items to a more diverse diet including larger prey and the inclusion of some insects and terrestrial invertebrates in later larval stages, although there was evidence of some dietary overlap between the stages. These results are consistent with those from King (2005), where Australian smelt fed predominately on pelagic zooplankton as larvae, and moved to surface feeding as juveniles and adults.

Starvation is thought to be one of the main causes for mortality during the larval phase (Hjort 1914; May 1974; Cushing 1990). Therefore it may be more advantageous for developing larvae to indiscriminately consume all available prey items (Govoni *et al.* 1986; Pryor and Epifanio 1993). However, both field and laboratory studies indicate that fish actively select prey items (Govoni *et al.* 1986; Pryor and Epifanio 1993; King 2005; Tonkin *et al.* 2006; Islam and Tanaka 2009). Australian smelt appeared to have been exhibiting prey selectivity throughout larval development as the contribution of prey to the diet did not reflect the composition of the riverine zooplankton assemblage. This was particularly driven by the absence of protozoans from the diet, which may be an artefact of easy digestion.

In conclusion this study, although temporally and spatially restricted, has demonstrated that during a low flow year an abundant prey source does exist in the main river channel in the Lower River Murray. This suggests the assumption that 'in the absence of floodplain inundation developing larvae do not have adequate access to food' (derived from Junk *et al.* 1989; Harris and Gehrke 1994; Schiller and Harris 2001) may not be applicable for Australian smelt in this lowland temperate system. Furthermore the prey were abundant in the pelagic zone of the mid river channel, not concentrated around/in micro- or meso-habitats. The decline in native fish throughout Australia has been attributed to the poor survival of early life stages, more so than spawning failure for many species (Gehrke 1991; Humphries and Lake 2000; Humphries *et al.* 2002; Koehn and Harrington 2006). However, this study suggests that food may not be a limiting factor in low flow years, at least for some species. Given the decline of many species is frequently linked to prolonged low flow conditions, further research into the growth, habitat associations and competition and predation pressures during low flow periods are needed for understanding larval fish survival.

CHAPTER 6: HABITAT ASSOCIATIONS OF FISH LARVAE IN THE MAIN CHANNEL OF A HEAVILY REGULATED RIVER SYSTEM, AUSTRALIA

ABSTRACT

Habitat availability is a critical component for the survival of early life stages of fish. The assumptions of key floodplain river recruitment models suggest that in the absence of floodplain inundation the main river channel does not provide adequate larval and juvenile habitat, however recent studies have questioned this theory. The aims were to determine if characteristics of three main channel habitat types (open water, still littoral and backwater habitats) differed, and if positioning around a weir affected the physical characteristics of these habitats; then to determine if the larval assemblages differed between the three habitat types and if individual species demonstrated specific habitat associations. Larval fish sampling was conducted in the Lower River Murray during the spring/summer of 2008 during a low flow year. The physical characteristics differed between habitat types; backwater and still littoral habitats had lower water velocity, were shallower and more structurally complex than open water habitats. Furthermore, the characteristics varied in relation to positioning around a weir; lower pool habitats were representative of modified conditions distinctive of regulated regions, whereas tailwater habitats were more representative of natural features. There were variations in the larval assemblage and individual species abundances between the three habitats surveyed, and in some instances, positioning of the weir also affected suitability of habitats. Still littoral and backwater habitats were important main channel environments for developing larvae of three species, even in a heavily regulated environment where the main river channel has relatively low velocities. Important larval and juvenile habitats are often diminished in modified rivers; the results from this study suggest that suitability of habitats may also be influenced by the positioning around regulatory structures, which may have significant implications for the restoration and management of habitats for early life stages of fishes.

6.1 INTRODUCTION

Food, habitat, predation and competition are the key factors that determine the level of mortality during the early life history of fish (Hjort 1914; May 1974; Houde 1997; Trippel and Chambers 1997; Houde 2002). Habitat availability is a critical component for the survival of larvae as food resources, levels of competition and predation are inherently linked to habitat characteristics (Werner 2002). Many fishes move between habitat types throughout ontogeny with larval habitats often being distinct from habitats where juveniles and adults of the same species are found (Scott and Nielsen 1989; Copp 1990; Werner 2002; Rosenberger and Angermeier 2003; King 2004b; Ortiz and Tissot 2008). Habitats where developing larvae are found presumably have suitable conditions for survival and growth. These are often structurally complex areas that provide shelter, access to high prey concentrations and refuge from high water velocity and predation (Haines and Tyus 1990; Tyus 1991; Sempeski and Gaudin 1995; Watkins *et al.* 1997; King 2004b).

Beck *et al.* (2001) defined nursery habitats as areas that have a disproportionally higher contribution of individuals recruiting into the adult population when compared with other areas, making them distinct from other larval/juvenile habitats. This nursery-role hypothesis was developed for marine and estuarine systems, but could also be extrapolated for riverine systems. Under this definition, the term nursery habitat is frequently applied to areas without validation of the contribution of these habitats to the adult population. Therefore, in the current study the term nursery habitat will not be applied, but rather larval habitats will define areas with high larval abundances.

Inundated floodplains are often considered important spawning and larval/juvenile habitats for many riverine fishes (Welcomme 1985; Turner *et al.* 1994). The major assumption of many currently accepted recruitment hypotheses (e.g. *flood pulse concept* and *flood recruitment model*) is that the main river channel does not provide adequate food and habitat for growth and survival of larvae and juveniles (Welcomme 1985; Junk *et al.* 1989; Harris and Gehrke 1994). However, the broad acceptance and application of these models to management of all fish species and all floodplain rivers has been questioned, particularly in Australian systems (Humphries *et al.* 1999; King *et al.* 2003).

The *low flow recruitment hypothesis* was developed for Australian temperate rivers, and proposes that shallow, slow flowing or still littoral habitats (including slackwaters, backwaters, inshore bays and embayments) are important larval habitats for some species particularly during low flow periods (Humphries *et al.* 1999). This model has been supported, with many studies documenting spawning and recruitment within the main channel during low flow periods (Chapter 3; Humphries and King 2003; Humphries *et al.*

2006; Zeug and Winemiller 2008). Furthermore, the significance of main channel habitats has been established world wide, with many studies documenting the use of shallow, still, littoral zones in the main channel as important larval habitats (Haines and Tyus 1990; Tyus 1991; Jurajda 1995; Sempeski and Gaudin 1995; Watkins *et al.* 1997; King 2004b). However, the importance of these habitats requires validation in heavily regulated systems where the main river channel is vastly modified.

The Lower River Murray, South Australia has been severely impacted by intensive river regulation; originally a free flowing river, this region is now virtually a series of discontinuous lakes (Walker 1992; Walker 2006b). Compounding this effect in recent times is the occurrence of one of the most severe hydrological droughts recorded in this region (MDBC 2007; Murphy and Timbal 2008). This has resulted in reduced flows to South Australia since 2001, however due to the extensive regulation water levels in the upper weir pools have not decreased significantly. Low flow conditions are expected to continue for a number of years as a result of the combination of drought and current water resource patterns (Lintermans and Cottingham 2007; MDBC 2008). If low flow conditions persist it may result in a further decrease in water levels and potentially important main channel habitat features (e.g. still littoral zones, slackwaters, backwaters and embayments) may become disconnected from the main river channel. Given the current critical conditions and future predictions, a more detailed understanding of what comprises suitable larval habitat within the Lower River Murray is required.

The overall objective of this study was to compare larval habitat use between three main channel habitats (open water, still littoral and backwater habitats) under low flow conditions in the Lower River Murray. Specific aims were to determine if the characteristics of the three habitat types differed, and if positioning around a weir either upstream (lower pool region) or downstream (tailwater region) resulted in differences in the characteristics of these habitats, to determine if the larval assemblages differed between the three habitat types and between lower pool and tailwater regions, and if individual species demonstrated associations with particular habitats. It was predicted that fish would be in greater abundance within still littoral and backwater habitats compared to the open water, but that given the low velocity of the river channel there would be no significant differences between the still littoral zones and the backwaters. Furthermore, it was predicted that low flow recruitment specialists would have specific associations with backwaters and still littoral zones.

6.2 METHODS

6.2.1 Sampling regime

The study was conducted during the spring/summer of 2008 under typical low flow conditions in the floodplain region of the Lower River Murray in South Australia (see Chapter 1 for a detailed discussion). The regulation in this region is substantial with little of the natural flow pattern retained. Sampling was conducted at 12 sites in the weir pools, three sites in the lower pools (upstream) and three in the tailwaters (downstream) of each of Weirs 5 and 6 (Figure 6.1) (terminology follows Walker 2006b). Sites were randomly selected within each region, although each site had to contain all three habitat types.

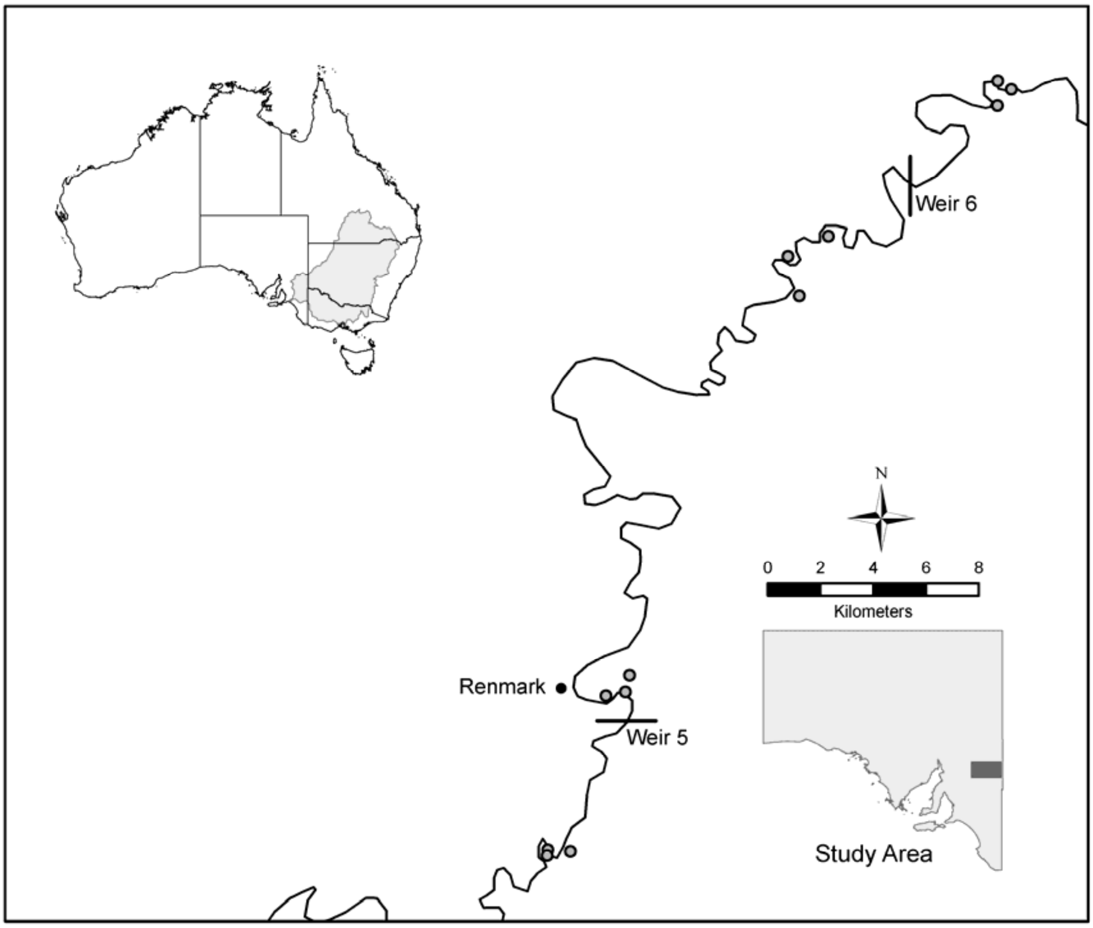


Figure 6.1. Location of the study sites surrounding Weirs 5 and 6. Sites are marked by (○).

Three sampling trips (September, October and November) were conducted during the peak spawning season and peak period for larval abundance for the small- medium bodied native species (see Chapter 4). Each site was sampled during the day and all sites were sampled within a four-day period each month. Sampling occurred such that three replicate samples were taken within each of the three habitat types: open water, backwater and still littoral (Figure 6.2). Open water habitats were in the centre of the main channel; these areas are wide, deep and slow flowing with very little structural habitat. Backwaters were areas with a restricted entry point connected to the main channel. Backwaters were generally between 30 and 60 m wide, shallow, with little to no flow and comprised complex vegetative and structural habitats. Still littoral habitats were the edge of the main channel; these areas differed from backwaters as they did not have a restricted entry point and allowed open connection to the open water.

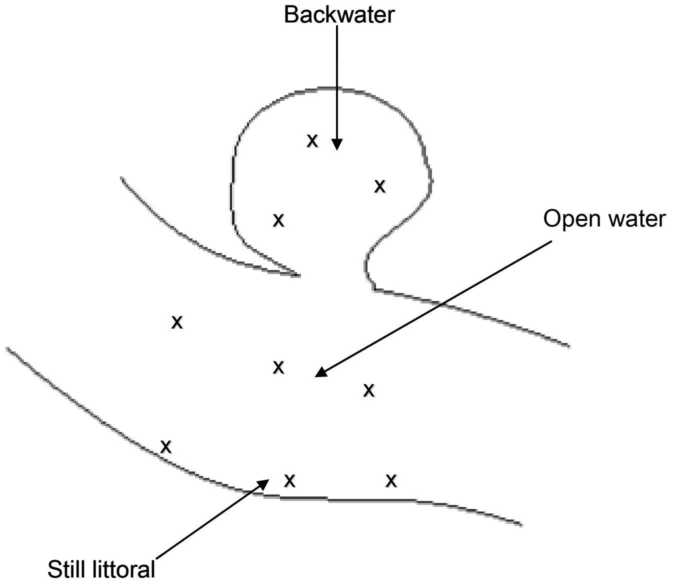


Figure 6.2. A schematic of a section of a sampling site in the river showing the three habitat types, x indicates where replicate samples were taken.

The sampling technique needed to sample both open water and shallow structurally complex habitats, therefore hand trawls and light traps were trialed (Chapter 2). A pilot study indicated that hand trawls were the most suitable method for sampling across the habitat types (KJC unpublished data), therefore this method was employed for sampling. Hand trawl nets were 0.5 m in length with a 0.3 m diameter opening which tapered to a removable jar and were constructed of 500 μm mesh. These were thrown the full length of the attached 5 m rope, and pulled quickly just below the surface of the water. Three replicate samples were taken within each habitat (Figure 6.2); each replicate sample consisted of 15 pooled 5 m throws. The volume of water filtered through each net was determined using a General Oceanics™ flow meter fixed in the centre of the mouth of the net. All larval data were standardised to fish per 10 m^3 of water using the recorded volume of water sampled during each hand trawl replicate.

During each trip a range of habitat characteristics were recorded at each of the sites to allow for comparison of habitat characteristics. Categorical data were used to allow for more consistency between observations. Depth was measured using a graduated pole and recorded as one of nine categories: (1) 0-0.25 m, (2) 0.25-0.50 m, (3) 0.5-0.75 m, (4) 0.75-1 m, (5) 1-2 m, (6) 2-3 m, (7) 3-4 m, (8) 4-5 m and (9) >5 m. Cover of woody debris, submerged vegetation and emergent vegetation was estimated visually, and recorded as one of five categories: absent, present (5%), sparse (5-25 %), medium (25-50%) and dense (>50%). In each habitat three replicate *in situ* measurements (using a TDS water quality meter) were taken 0.3 m below the surface for temperature (ºC), conductivity (μS/cm @ 25 °C), dissolved oxygen (mg/L), and pH. Turbidity (depth mm) was determined using a secchi disc. Water velocity was recorded using Marsh-McBirney Flow Mate™ portable velocity meter (Marsh McBirney Frederick, Maryland, USA) and a hand held depth gauge within each of the habitat types. Additional notes were taken on the dominant vegetation species and the types of woody debris present within each site to define any structural differences.

6.2.2 Preservation and identification of larvae

Each sample was washed into separate buckets where fish were euthanased using high concentrations of clove oil. Samples were preserved in 95% ethanol *in situ* and returned to the laboratory for sorting and identification. Larvae were identified to species level, where possible, using published descriptions (Lake 1967b; Puckridge and Walker 1990; Neira *et al.* 1998; Serafini and Humphries 2004), with the exception of carp gudgeon (*Hypseleotris* spp.) and hardyhead (*Craterocephalus* spp.). Each of these genera were treated as a species complex due to close phylogenetic relationships and very similar morphologies making identification to species level difficult (Bertozzi *et al.* 2000; Serafini and Humphries 2004). Each fish was categorised according to developmental stage, as preflexion larvae (no curvature at tip of notochord), postflexion (upward flexion of notochord, caudal fin rays developing), metalarvae (caudal fins rays developed and pelvic fins forming) or juvenile/adult (rays in all fins fully developed) (Kelso and Rutherford 1996). Only larvae from Australian smelt, carp gudgeon and hardyhead were collected in sufficient numbers for individual species analysis. Juveniles and adults were collected in low abundances and therefore were not included in the analyses.

6.2.3 Data analysis

Frequency distributions were used to describe the differences in habitat characteristics (depth, woody debris, submerged vegetation and emergent vegetation) among each of the lower pool and tailwater regions and the three habitat types.

Fish were patchily distributed and in low abundances, therefore the number of fish was averaged across the three replicates taken within each habitat at each site, resulting in 12 replicates for each habitat and trip. The same procedure was also implemented for the environmental variables.

Prior to analysis the environmental variables, assemblage composition (relative abundance per 10 m^3 for each developmental stage of all species) and individual species abundances (larvae per 10 m^3) were tested for normality and homogeneity of variance. Given that very few variables met the assumptions, data were analysed using permutational analysis of variance (PERMANOVA; Anderson 2001a). Environmental variables and larval data were examined using normalised Euclidean distance, and Bray-Curtis similarity measures, respectively.

To determine if differences could be detected between habitats (backwater, open water and still littoral) and/or regions (lower pool and tailwater), environmental variables and the larval assemblage, Australian smelt and carp gudgeon abundances were examined using a three-way design (*region*, *habitat* and *trip*). Whilst, a two-way design was employed for hardyhead abundances (*region* and *habitat*), as this species was only collected in sufficient numbers during trip three. Multivariate analysis was performed on the larval assemblage data, and univariate analyses were performed for the environmental variables and individual species. *Region* and *habitat* were treated as fixed factors, while *trip* was treated as a random factor. Unrestricted permutations of data were performed for all analyses, with 999 permutations for the test, to detect differences at $\alpha=0.05$ (Anderson 2001a). Where significant differences were detected pair wise comparisons were performed to identify where the differences occurred.

An index of habitat association (I_{HA}) was used to examine patterns of habitat use and change in habitat use through development. The I_{HA} follows the procedure described by King (2004b) and Bult *et al.* (1998). This was calculated using:

$$I_{\mathrm{HA}} = \log_{10}(O_r+1) - \log_{10}(R_r+1)$$

Where:
O_r = is the ranked total number of fish collected within each habitat category of the observed data.
R_r = the average randomised ranked total number of fish within each habitat category.

The observed data were ranked due to the high variance within individual habitat types; this prevented individual samples from skewing the results. *R* was generated using a randomisation procedure, where the observed number of fish per sample were randomly rearranged, and the total number of fish within each habitat type was then calculated. A total of 1000 randomisations were performed. The I_{HA} is not confined to any range; a positive value indicates a positive association, and negative value indicates a negative association. To calculate the significance of the I_{HA} the rank of each of the observed values was established within the generated randomised data (Potvin and Roff 1993). The ranking and randomisation procedures were performed using Visual Basic™ (Excel 2003™) scripts.

6.3 RESULTS

6.3.1 Habitat descriptions and environmental variables

The physical characteristic of the three habitat types (open water, still littoral and backwater) all differed (Figure 6.3). Backwaters were generally shallow (< 0.75 m deep) whilst still littoral and open water habitats were deeper (1-3 m and 2-4 m, respectively). The open water habitats had almost no structural complexity. Woody debris, submerged vegetation and emergent vegetation were almost exclusively restricted to backwaters and still littoral habitats. Backwaters generally had a higher structural complexity than the still littoral habitats.

There were variations in the habitat characteristics between the lower pool and tailwater regions (Figure 6.3). Backwaters had similar depth profiles, but on average were deeper in the tailwater regions (Figure 6.3a). Woody debris was on average greater in the lower pool regions; many of these habitats were created by flooding of low land areas following the installation of the weirs. Open water and still littoral habitats were deeper in the lower pool regions (Figure 6.3b & c). Still littoral habitats had more woody debris in the lower pool region, while percent cover of emergent vegetation was similar (Figure 6.3c). However, the structure of emergent vegetation was different; willows (*Salix* spp.) almost exclusively dominated lower pool habitats, whereas tailwaters were dominated by common native reeds (*Typha* spp. and *Phragmities australis*). Still littoral zones in the tailwater regions had a greater percentage of submerged vegetation, consisting of *Vallisneria spiralis*, *Potamogeton crispis* and *Potamogeton tricarinatus*.

Velocity was low in all habitats and varied between regions and habitats (Figure 6.4a; Table 6.1). The velocity of the backwater habitats in the tailwater region was significantly higher than in the lower pools. In the lower pools velocity differed between all habitats, with greatest velocity occurring in the open water habitats, followed by still littoral and lowest in the backwaters. In the tailwaters, open water habitats generally had higher velocity, but this was only significant during September (Figure 6.4a). Conductivity was significantly higher in backwaters than the open water or still littoral habitats in the lower pool region; no differences were identified in the tail water regions (Figure 6.4c; Table 6.1). Dissolved oxygen and temperature were significantly different between trips; temperature increased significantly throughout the season, dissolved oxygen decreased significantly (Figure 6.4b & e; Table 6.1). No consistent patterns were identified for differences between habitats

(Figure 6.4b & e; Table 6.1). pH varied between regions and trips as indicated by the significant region x trip interaction (Table 6.1), but no consistent patterns could be identified (Figure 6.4d). Turbidity changed throughout the season and visibility was generally higher in the open water habitats (Figure 6.4f; Table 6.1).

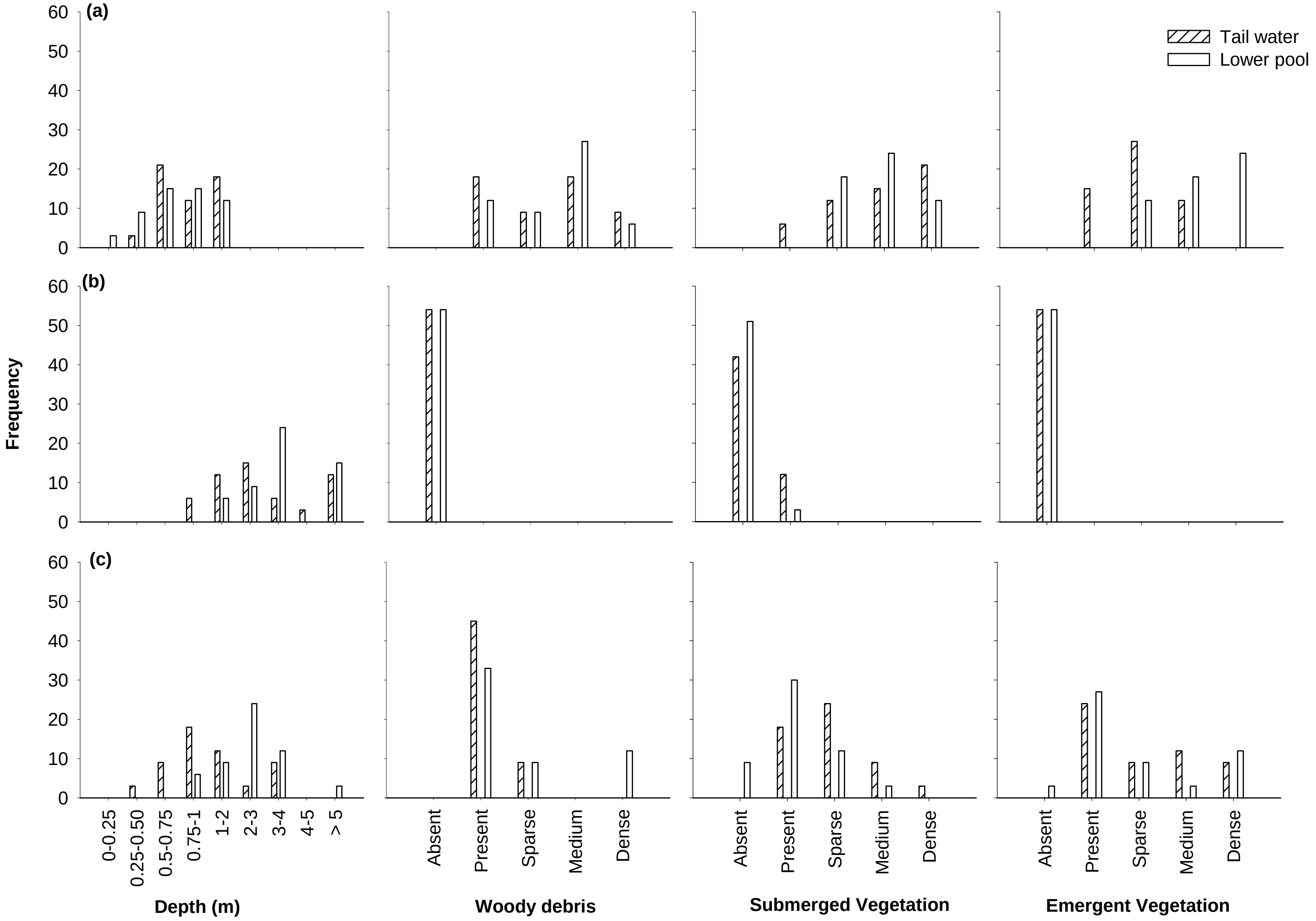


Figure 6.3. Frequency distributions of habitat characteristics: depth (m), woody debris, submerged vegetation and emergent vegetation (percentage cover: absent 0%, present <5%, sparse 5-25 %, medium 25- 50% and dense >50%) in lower pool and tailwater region of each habitat (a) backwater, (b) open water and (c) still littoral.

Table 6.1 Three-way univariate PERMANOVA for differences among trips, region and habitats for velocity, dissolved oxygen, conductivity, pH, temperature and turbidity. Bold text indicates significant value.

		Velocity		Dissolved oxygen		Conductivity		pH		Temperature		Turbidity	
Source of variation	***df***	**MS**	***p***	**MS**	***p***	**MS**	***p***	**MS**	***p***	**MS**	***p***	**MS**	***p***
Trip	2	0.01	0.627	5.66	**0.004**	17.78	**0.001**	2.47	0.077	42.05	**0.001**	10.41	**0.001**
Region	1	0.02	0.413	0.23	0.694	3.06	0.138	1.17	0.611	5.00	0.061	0.29	0.538
Habitat	2	0.29	**0.010**	1.62	0.273	3.60	**0.001**	0.45	0.322	0.06	0.596	8.99	0.083
Trip x Region	2	0.01	0.706	1.37	0.243	0.50	0.431	4.15	**0.015**	0.23	0.286	0.53	0.430
Trip x Habitat	4	0.01	0.922	0.85	0.422	0.06	0.987	0.31	0.863	0.11	0.655	1.76	**0.030**
Region x Habitat	2	0.05	**0.006**	0.00	0.999	2.33	**0.001**	1.40	0.245	0.17	0.204	1.71	0.110
Trip x Region x Habitat	4	0.00	0.988	1.27	0.233	0.01	0.998	0.62	0.626	0.07	0.813	0.43	0.609
Residual	90	0.02	0.627	0.90		0.61		0.95		0.18		0.61	

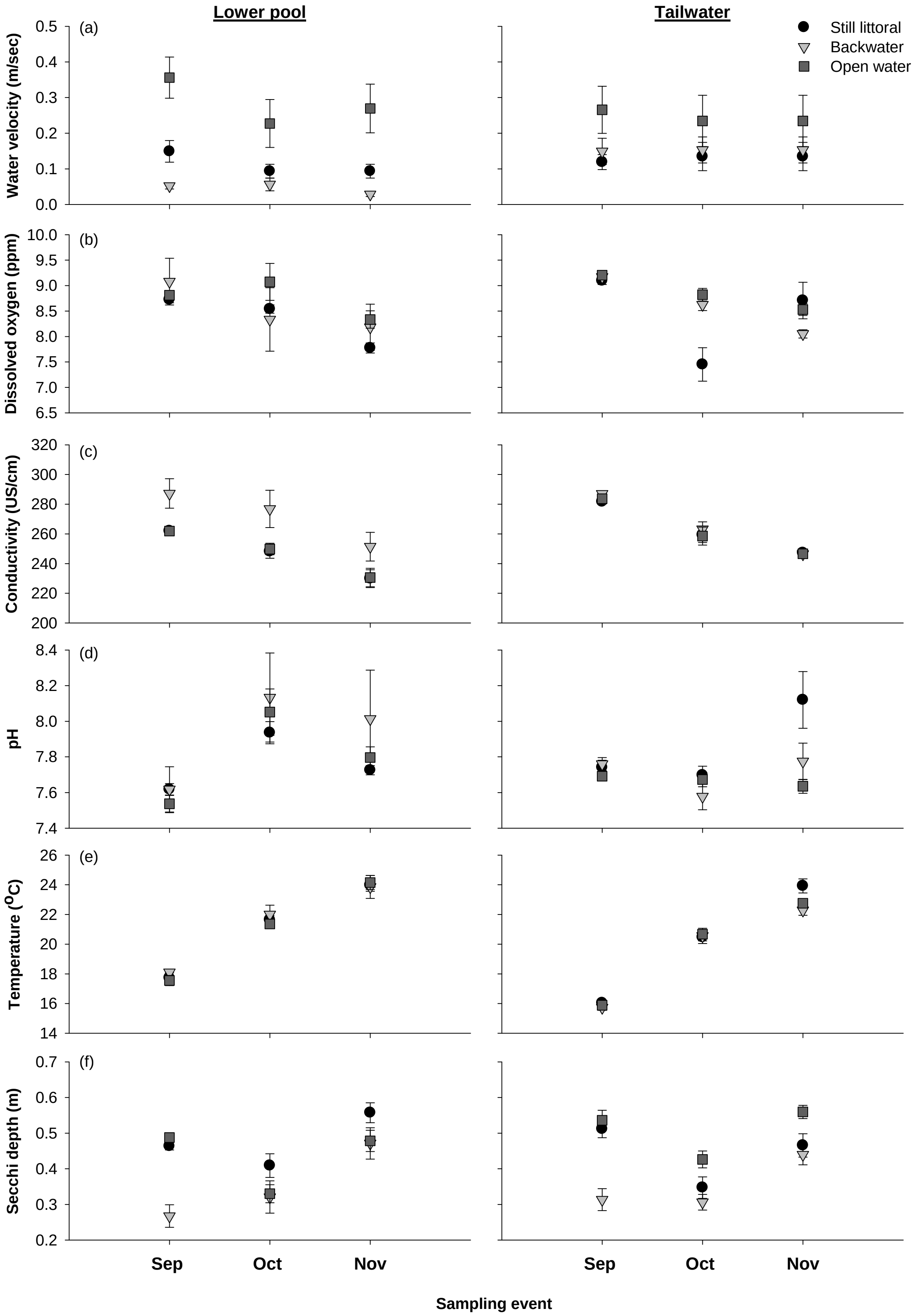


Figure 6.4. Comparison of the mean (± standard error) for water quality variables between habitats (still littoral, backwater and open water) in the lower pool and tailwater regions for a) water velocity (m/sec), b) dissolved oxygen (ppm), c) conductivity (μS cm^{-1} at 25 °C), d) pH, e) water temperature (°C) and f) turbidity (secchi depth m).

6.3.2 Larval assemblages

Nine species were collected throughout the study; the most abundant species were Australian smelt (*Retropinna semoni*), carp gudgeon (*Hypseleotris* spp.) and hardyhead (*Craterocephalus* spp.) (Table 6.2). The assemblage varied between regions and habitats but there were significant interactions between these factors and timing of sampling; as indicated by the trip x region and trip x habitat interactions (Figure 6.5; Table 6.3). Larval assemblages varied between trips, because of species-specific spawning times (see Chapter 4). During trips 1 and 3, the larval assemblage in open water habitats was significantly different from backwater and still littoral habitats. In trip 2, backwater assemblages were significantly different from still littoral and open water assemblages. Furthermore, during trip 3 the assemblages differed between the lower pool and tailwater habitats.

The abundances of carp gudgeon and hardyhead larvae varied significantly among habitats with increased abundances in backwaters (Figure 6.5c & d; Table 6.3). Abundances of Australian smelt differed significantly by habitat, but these differences varied between trips, as indicated by the trip x habitat interaction (Figure 6.5b; Table 6.3). Abundances were significantly greater in still littoral habitats in the tailwater region, and backwater in lower pool regions during trip 1 (Figure 6.5b). During trip 2, abundance of Australian smelt was significantly less in backwaters than still littoral and open water habitats; whereas during trip 3 abundances were significantly lower in open water habitats than backwater or still littoral habitats (Figure 6.5b).

Table 6.2. Species list and total number of larvae collected in each habitat type in lower pool and tailwater regions.

	Lower pool			**Tailwater**			
Common name (*Scientific name*)	**Back water**	**Open water**	**Still littoral**	**Back water**	**Open water**	**Still littoral**	**Species total**
Australian smelt (*Retropinna semoni*)	76	57	51	52	60	219	**515**
Bony herring (*Nematalosa erebi*)	0	3	0	2	0	1	**6**
Carp gudgeon (*Hypseleotris* spp.)	82	7	1	2	0	1	**93**
Flathead gudgeon (*Philypnodon grandiceps*)	11	3	1	4	4	4	**27**
Hardyhead (*Craterocephalus* spp.)	18	0	3	39	0	6	**66**
Carp (*Cyprinus carpio*)	4	0	0	0	0	0	**4**
Redfin perch (*Perca fluviatilis*)	0	0	0	1	0	0	**1**
Gambusia (*Gambusia holbrooki*)	1	0	0	0	0	0	**1**
Habitat total	**192**	**70**	**56**	**100**	**64**	**231**	**713**

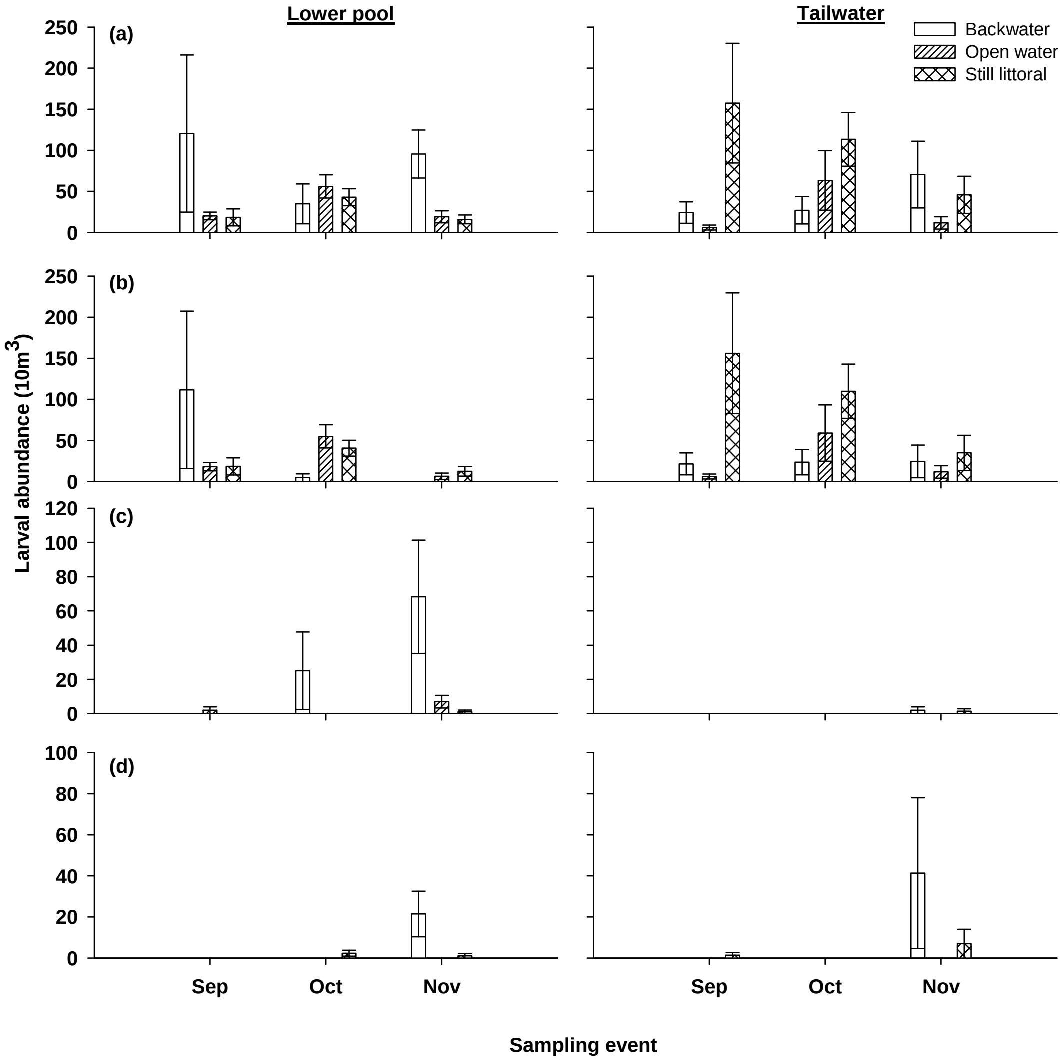


Figure 6.5. Mean relative abundance of larvae (fish per 10 m^3) ± standard error of (a) total larval abundance (b) Australian smelt, (c) carp gudgeon and (d) hardyhead in backwaters, open water and still littoral habitats in the lower pool and tailwater regions.

Table 6.3 PERMANOVA for differences among trips, locations and habitats for the larval assemblage (multivariate) and individual species (univariate). Three-way design was employed for the larval assemblage, Australian smelt and carp gudgeon and two-way design for hardyhead. Bold text indicates significant value.

	Larval assemblage			**Australian smelt**		**Carp gudgeon**			**Hardyhead**		
Sampling trips analysed and number of individuals (n)	**All (713)**			**All (515)**		**2 & 3 (92)**			**3 (63)**		
Source of variation	***df***	**MS**	***p***	**MS**	***p***	***df***	**MS**	***p***	***df***	**MS**	***p***
Trip	2	5843	**0.001**	8329	**0.001**	1	4182	0.231	-	-	-
Region	1	2023	0.419	2499	0.221	1	5770	0.201	1	464	0.602
Habitat	2	9582	0.078	10308	0.124	2	3167	**0.043**	2	5376	**0.002**
Trip x Region	2	2762	**0.029**	919	0.662	1	1559	0.241	-	-	-
Trip x Habitat	4	3063	**0.004**	3577	**0.034**	2	571	0.572	-	-	-
Region x Habitat	2	3143	0.078	4501	0.100	2	2750	0.136	2	826	0.439
Trip x Region x Habitat	4	1101	0.666	1486	0.465	2	742	0.295	-	-	
Residual	90	1302		1592		60	599		30	926	

6.3.3 Habitat associations

Patterns in habitat use within species were examined using I_{HA} for Australian smelt, carp gudgeon, and hardyhead larvae, as species abundances varied significantly with habitat. No positive association was identified with the open water habitats for any species (Figure 6.6); however there was a significant negative association between Australian smelt and open water habitats in the tailwater regions (Figure 6.6a). Still littoral habitats showed a significant positive association with larvae of Australian smelt (Figure 6.6a); and within the lower pool region a negative association was identified for carp gudgeon larvae (Figure 6.6b). Backwater habitats had significant positive associations with carp gudgeon larvae (in the lower pool regions) (Figure 6.6b) and hardyhead larvae (Figure 6.6c), whilst a negative association was identified for Australian smelt (Figure 6.6a). None of the species investigated showed differences in habitat use through larval development (Figure 6.6).

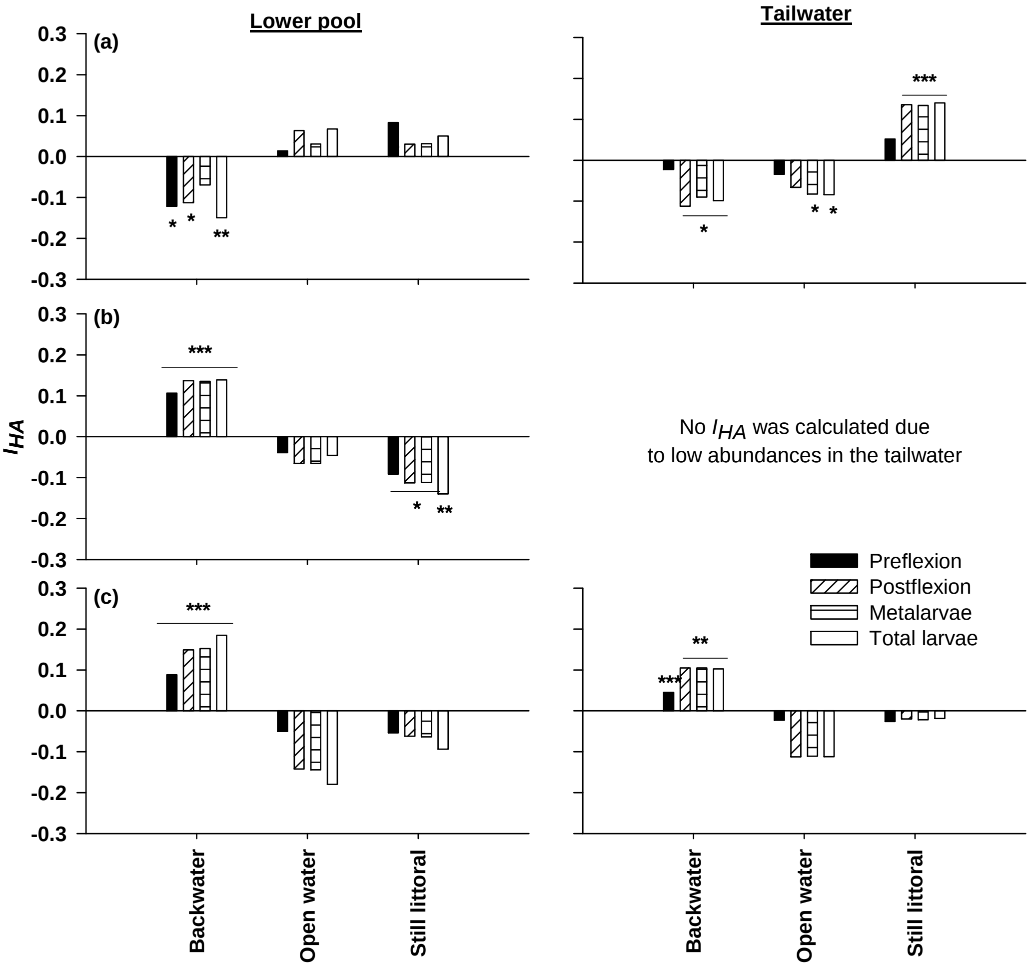


Figure 6.6. Index of habitat association (I_{HA}) for total larvae and each developmental stage of (a) Australian smelt, (b) carp gudgeon and (c) hardyhead in backwaters, open water and still littoral habitats in the lower pool and tailwater regions. * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$.

6.4 DISCUSSION

Differences were identified in the physical characteristics (velocity, depth, woody debris and aquatic vegetation) of habitats between lower pool and tailwater regions in the Lower River Murray, however; larval assemblages did not differ significantly between regions. The characteristics of the lower pool habitats were representative of modified conditions incurred due to river regulation, exotic plants dominated all habitats and most backwaters had high percentage of woody debris, resulting from these areas being formed by the inundation of low-lying areas following the installation of the weirs. Tailwater habitats were more representative of natural characteristics; common native reeds were the dominant emergent vegetation and there was a greater percentage of native submerged vegetation. Patterns of habitat associations did not differ between lower pool and tailwater regions for hardyhead larvae, suggesting that suitability of backwater habitats for developing larvae does not differ around a weir. Contrastingly, Australian smelt showed a significant negative association with the open water in tailwater regions, whilst there was a positive association, although not significant in the lower pool region. This may indicate that the habitat characteristics between lower pool and tailwater regions are potentially influencing the suitability of the open water habitat for Australian smelt larvae. Installation of weirs in the Lower River Murray has resulted in a wide range of impacts on the physical characteristics and ecology of the region. Impacts include alterations to the flow regime, channel morphology, composition of the littoral plant community and increases in salinity, sedimentation and erosion (Thoms and Walker 1993; Walker and Thoms 1993; Maheshwari *et al.* 1995; Blanch *et al.* 2000; Walker 2006b).

The impacts of the installation of regulatory structures and subsequent changes to the riverine environment have consistently been shown to alter fish communities in regulated rivers (e.g. Martinez *et al.* 1994; Gehrke *et al.* 1995; Travinchek *et al.* 1995; Richter *et al.* 1996; Matthews 1998; Penczak *et al.* 1998; Humphries and Lake 2000; Penczak and Kruk 2005). Humphries *et al.* (2006) highlighted that modification of rivers and altered flow regimes have the potential to alter type, frequency and extent of particular habitat patches. In modified rivers, the loss of important larval and juvenile habitats may contribute to recruitment failure (Jurajda 1995; Humphries *et al.* 2002; King *et al.* 2003; King 2005; Humphries *et al.* 2006). The results from this study suggest that the impacts may not be limited to river regulation, but that the positioning of habitats either upstream or downstream of weirs may influence the suitability of habitats for fish. Much attention has focused on changes in the fish community downstream of impoundments, with less on fish communities upstream (Martinez *et al.* 1994; Kruk and Penczak 2003; Penczak and Kruk 2005; Matthews and Marsh-Mathews 2007; Penczak *et al.* 2009). Comparisons of differences in fish assemblages between habitats upstream and downstream of impoundments is an area that requires further research.

Still littoral zones and backwaters are important main channel habitats for developing larvae of these three species examined in this study, even in heavily regulated environments where the main river channel has relatively low velocities. However, none of the species examined were positively associated with open water habitats, despite these areas being slow flowing and food rich (Chapter 5).

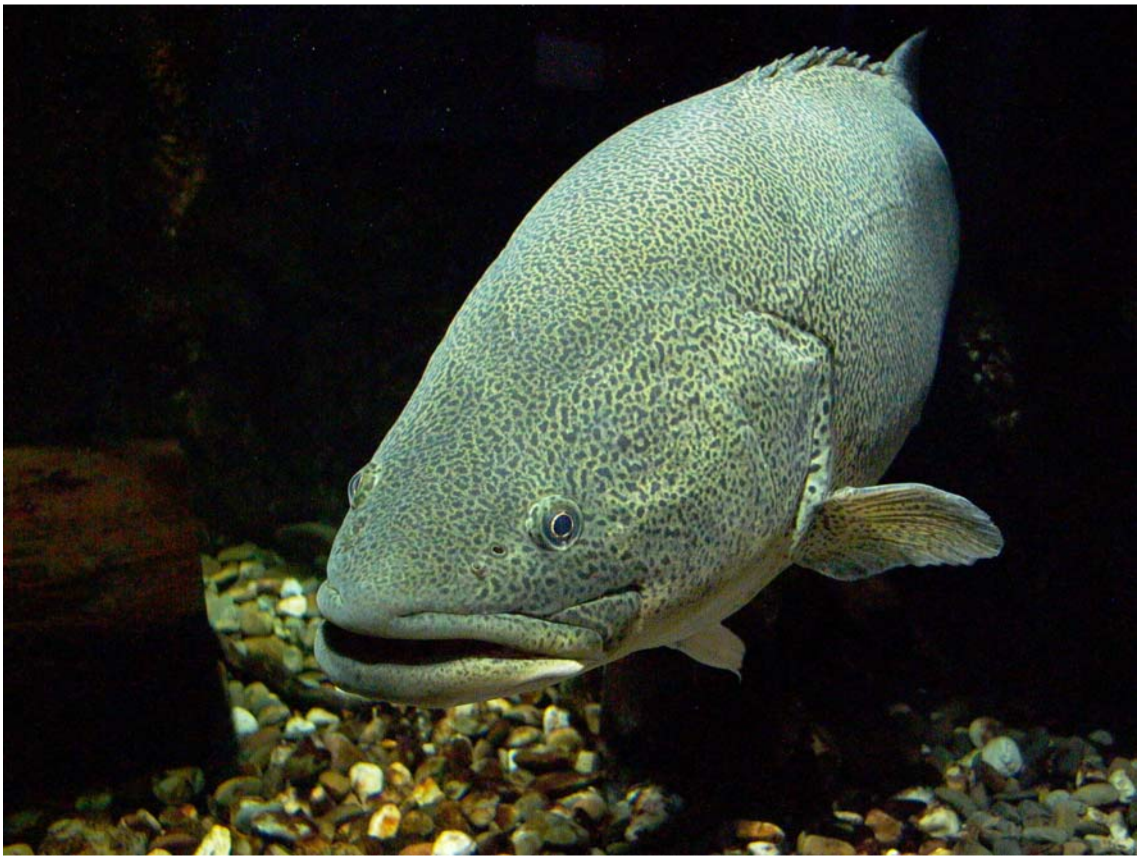
In the current study, Australian smelt larvae were negatively associated with backwaters and positively associated with still littoral habitats. In the Broken River (a tributary of the upper River Murray), King (2004b) found that Australian smelt larvae were predominantly associated with backwaters, and not other low velocity areas including still littoral habitats. Further investigations in the Broken River on the associations of Australian smelt larvae between still-water patches (still littoral, backwater and slackwater habitats) indicated no positive associations (Price 2007). Importantly, however, there were positive associations with moderate snag cover (11-25%) and low instream cover (0-10%) (Price 2007). Given the association with particular habitat characteristics, the differences in habitat association of Australian smelt between the Lower River Murray and the Broken River may be driven by the characteristics of the backwaters. The backwaters surveyed in the current study were on average wider, deeper and had a higher percentage cover of woody debris than reported in the Broken River (King 2004b; Price 2007). Australian smelt are predominantly open water, pelagic, schooling species as adults, fish spawn in the pelagic zone and the eggs sink into demersal habitats attaching to substrate, aquatic vegetation and debris (Lintermans 2007). The open water habitats in the Lower River Murray are very slow flowing, possibly restricting the dispersal of larvae into backwaters, resulting in an increased abundance of larvae in still littoral habitats.

Carp gudgeon and hardyhead larvae were predominately associated with backwater habitats, but their habitat associations have not been investigated elsewhere. Adult hardyhead are predominately associated with littoral habitats and backwaters; aquatic vegetation is a key component of habitats for adults and spawning as the demersal eggs are covered with adhesive filaments that allow them to attach to vegetation, as well as rocks and gravel (Llewellyn 1979; Ivantsoff and Crowley 1996). Adult carp gudgeon are normally associated with still, slow flowing habitats and aquatic vegetation, and their eggs are spawned directly onto submerged vegetation or twigs (Lake 1967a; Allen *et al.* 2002).

Humphries *et al.* (1999) proposed that still littoral and backwater habitats provide refuge from high water velocity, higher densities of food for larvae and warmer temperatures for enhanced larval growth. Water velocity was low in all habitats in the current study as a result of the extensive regulation throughout the Lower River Murray (Walker 2006b), however, the velocity was still significantly lower in backwaters than main channels. Water velocity can affect larval positioning in the water column, ability to escape predation, feeding efficiency and energetic requirements of larvae, therefore preference for lower velocities is likely to partly be a function of poorly developed swimming ability (Facey and Grossman 1992; Heggenes 1996; Flore and Keckeis 1998; Werner 2002). Humphries *et al.* (1999) hypothesised that lower velocity areas such as backwater and still littoral habitats would provide greater concentrations of prey items for fish larvae. Whilst some studies have documented higher concentrations of prey in these low velocity habitats (Thorp *et al.* 1994; Basu and Pick 1996; Reckendorfer *et al.* 1999; Reynolds 2000), other studies have found no difference between low velocity and flowing habitats, and therefore this may not be a driving factor of habitat selection (King 2004a; Humphries *et al.* 2006). Water temperatures were not significantly different between habitats suggesting that in the Lower River Murray temperature may not be a key factor for habitat selection. These results are consistent with King (2004b) who also suggested that larvae were not utilising backwaters to promote faster growth via warmer temperatures.

Larval habitat associations may occur by either active or passive mechanisms. King (2004b) suggested that this may be a function of: 1) larvae being passive drifting particles that are deposited in low flow habitats (such as backwaters and still littoral zones) (passive), 2) spawning adults actively selecting habitats for larvae through spawning sites (passive by larvae/active by adults), 3) larvae actively seeking suitable habitats (active), and 4) larvae being evenly distributed, but survival is higher in particular habitats. The current study suggests that the positive associations with backwater and still littoral habitats do not appear to be driven by either refuge from velocity, food or temperature. The positive association of hardyhead and carp gudgeon larvae with backwater habitats is likely to be a function of adult spawning behaviour. The positive association between Australian smelt and still littoral habitats is likely to be a combination of both adult spawning selection and passive dispersal of larvae. Therefore, it is unlikely to be active selection of these habitats by larvae of the three species, but rather an active selection by adults as spawning sites, or potentially the passive deposition of larvae into these habitats from water currents.

There is a growing body of evidence to suggest that physical modifications (such as river regulation and channelisation) within riverine systems worldwide are responsible for controlling fish diversity and population dynamics rather than biological interactions (Copp 1990; Jurajda 1995; Gaudin 2001; Schiemer *et al.* 2001; Arlinghaus *et al.* 2002). The results from this study are consistent with previous studies, suggesting that backwaters and littoral habitat are important larval and juvenile habitats particularly in heavily modified rivers (Jurajda 1995, 1999; Gaudin 2001; Arlinghaus *et al.* 2002; King 2004b). Restoration of fish populations has primarily focused on adult habitats (White 1996) and flow regimes (Marchetti and Moyle 2001; Brown and Ford 2002; King *et al.* 2008b). While these efforts are integral in restoring native fish populations and will likely provide benefits to larvae, an increased understanding of the requirements of the early life stages is needed, as this is when the greatest mortality is likely, to enable a more targeted restoration of these important habitats. To aid in restoration and management of habitats to promote fish spawning and recruitment studies of species-specific habitat associations will be needed in the future.



'Ponde' Murray cod (*Maccullochella peelii peelii*) (Photo courtesy of Jason Higham.).

CHAPTER 7: GENERAL DISCUSSION

The broad aim of this thesis was to test some of the assumptions of the *flood recruitment model* (Harris and Gehrke 1994) and the *low flow recruitment hypothesis* (Humphries *et al.* 1999), and to determine their applicability for management and restoration of native fish in a heavily regulated river. The thesis described the larval fish assemblages in the Lower River Murray, and examined whether assemblage patterns are similar to previous studies in the Murray-Darling Basin, and how this relates to these two models. Fundamentally, the flood recruitment model and low flow recruitment hypothesis do not contradict one another (Humphries *et al.* 1999). The flow regime is acknowledged in both to be important for fish spawning and recruitment and it is hypothesised that the mid river channel does not provide suitable habitat or abundant food, and therefore, that specific habitats (either the floodplain or backwaters) are required to ensure sufficient growth and survival of larvae (Harris and Gehrke 1994; Humphries *et al.* 1999). Following the concepts outlined in the *flood pulse concept* (Junk *et al.* 1989), the flood recruitment model highlights the importance of high flows to initiate spawning and/or to provide floodplain habitats for spawning and recruitment of some species, and also suggests that floods are important for food production and nutrient cycling (Harris and Gehrke 1994). The low flow recruitment hypothesis emphasises that not all species require a flow stimulus to spawn and that within the main channel environment, still littoral zones and backwaters can provide the necessary conditions for developing larvae (Humphries *et al.* 1999). Importantly, the extent to which these models describe life history strategies and responses for fish has only been tested in a few locations (for example see King 2004b; Zeug and Winemiller 2008); and they remain largely untested in heavily regulated rivers or during periods of drought throughout the world.

The Lower River Murray, South Australia provided an excellent site for testing some of the components of the fish recruitment models, given that it is ecologically dissimilar from the areas where these models were developed (see Chapter 1 for a detailed discussion). In particular, the extensive flow modification in this region contrasts markedly with many areas where previous work has been conducted. The Lower River Murray is now a series of slow flowing, deep, weir pools, which are more representative of a series of isolated lakes than the naturally free flowing river it was historically (Walker 1992; Walker 2006b).

Ecological outcomes

This thesis found that despite the vastly different flow conditions experienced, the life history strategies and spawning seasons of native fish in the Lower River Murray were consistent with previous studies in the mid- and upper River Murray and surrounding tributaries (Chapters 3 and 4). Importantly, while small-medium bodied species were collected as larvae during both low flows and a flow pulse, larvae of some large bodied native species were only collected during the within channel flow pulse.

A number of the small-medium bodied native species (e.g. Australian smelt, bony herring, carp gudgeon, flathead gudgeon and hardyhead) are highly abundant throughout the Basin (Lintermans 2007). They are thought to be opportunistic or generalist species (*sensu* Winemiller and Rose 1992) and are able to spawn successfully under all flow conditions (see Humphries *et al.* 2002; King *et al.* 2003; King 2004b; King *et al.* 2007; Vilizzi *et al.* 2007; King *et al.* 2010). Successful spawning and significant increases in the larval abundances of the small-medium bodied natives occurred in the mid river channel during three years of low flow conditions (Chapter 3). These results provide further support for the low flow recruitment hypothesis (Humphries *et al.* 1999), and demonstrate that some species do not require a flow stimulus to induce spawning in floodplain systems (Humphries and Lake 2000; Humphries *et al.* 2002; King *et al.* 2003; King 2004b; Zeug and Winemiller 2008). However, the small-medium bodied native species are relatively short-lived (1-5 yrs) species, and whilst fishes with short generation times are likely to be most seriously affected by changes to the environment, these species have demonstrated their ability to at least maintain their populations during low flow conditions and during the within channel flow pulse year.

All fish species present in the Lower Murray River spawned during a within channel flow pulse, but importantly, two of the large bodied species, golden perch and silver perch, only spawned during this period (Chapter 3). These results conform with other studies where spawning of golden perch and silver perch only occurred during either within channel flow pulses and/or overbank floods (Lake 1967a; Mackay 1973; Mallen-Cooper and Stuart 2003; Roberts *et al.* 2008; King *et al.* 2009b). Whilst only a low number of larvae of both species were collected in the current study, other spawning events of these species were also recorded in a number of locations throughout the River Murray during 2005, under flood conditions in the Barmah-Millewa region and flow pulse conditions at the Lindsay Island and Chowilla Anabranch systems (see Vilizzi *et al.* 2007; Leigh *et al.* 2008; King *et al.* 2009b; King *et al.* 2010). The flood recruitment model proposes two pathways where increased flows may enhance recruitment in river fish; the first is that flooding directly triggers spawning, and the second is that the inundated floodplain provides food and habitat

(Harris and Gehrke 1994). However, in this study there was some evidence that golden perch and silver perch can spawn without an overbank flood, therefore the applicability of the flood recruitment model for inducing spawning of these species may need to reassess what characteristics of the flow regime (e.g. magnitude, pattern, timing, duration) stimulates spawning. Additionally, previous research suggests that there is little evidence of golden perch and silver perch utilising floodplain habitats as larvae (Humphries *et al.* 1999; King *et al.* 2003; Mallen-Cooper and Stuart 2003). The present study is obviously limited in its conclusions, however, as only one flow pulse year occurred and no overbank flood. Continued long term monitoring of the spawning response through larval assemblage studies in years of higher flows in this type of heavily regulated region is required to more confidently describe their response.

Although temporally and spatially restricted, the results from this study suggest that under low flows, the small-medium bodied native species are not limited by food or habitat within the main river channel in the Lower River Murray (Chapters 3, 5 and 6). The low flow recruitment hypothesis proposes that under low flows, larvae use still littoral and backwater habitats in the main channel to take advantage of higher densities of prey in these habitats (Humphries *et al.* 1999; King 2004a). However, this study identified that a high density of suitable food for larvae occurred in the mid river channel habitat during a low flow period. This is consistent with a number of studies which have found that zooplankton densities are often higher in areas or periods of low flows (Dirnberger and Threlkeld 1986; Pace *et al.* 1992; Basu and Pick 1996; Reckendorfer *et al.* 1999; Reynolds 2000; Sluss *et al.* 2008; Havel *et al.* 2009). Whilst this study identified that sufficient densities of prey for larvae occur during low flow conditions, the assemblage was dominated by very small size classes of zooplankton (such as rotifers and protozoans), which may be a factor of the low flow conditions and isolation from the nutrient rich floodplain. The importance of backwater and littoral habitats for fish larvae of a number of species described in this study is consistent with previous studies, particularly for fishes in heavily modified rivers (Jurajda 1995, 1999; Gaudin 2001; Arlinghaus *et al.* 2002; King 2004b). Previously, low flows were thought to be detrimental to larval survival as the floodplain remains disconnected and therefore, food and habitat are limited. However, the current study (Chapters 5 and 6) and other studies (e.g. King 2004a, b; Price 2007) have demonstrated that habitat and food for larvae are not necessarily limiting factors within the main channel environment of Australian floodplain rivers.

The research presented in this thesis was focussed on investigating changes in spawning (as represented by changes in larval abundances) and the relationships between these changes and the predictions of the recruitment models. However, the current study had significant limitations in relation to the flow regimes that occurred, as it was only able to consider the response of fish during low flow periods and one within channel flow pulse. The role of floodplain inundation in providing habitat, abundant food and nutrient input for the mid channel of the Lower River Murray needs to be investigated during an overbank flood to fully test the principles of the flood recruitment model. Whilst this study has demonstrated successful spawning, survival of larvae, adequate food and suitable habitat all occur during low flows and within channel flow pulses, whether these factors change during overbank floods is yet to be determined. Overbank floods and floodplain inundation have long been suggested to be beneficial for fish spawning and recruitment (Welcomme 1985; Junk *et al.* 1989; Harris and Gehrke 1994; Winemiller 2005). With this in mind, it is possible that the quality of food and habitat available under high flow conditions substantially increases growth, survival and recruitment for some species in comparison to the low flow conditions experienced in this study. Quantitative comparisons of larval and juvenile fish growth rates and body condition during years of varied flows (low flows, within channel flow pulses and overbank floods) may determine the factors that increase recruitment during flood pulses.

Application to native fish management

The high degree of flow modification in the Lower River Murray has resulted in significant environmental change (Walker 1985; Walker and Thoms 1993; Maheshwari *et al.* 1995; Arthington and Pusey 2003), and a subsequent decrease in the range and abundance of native fish (Walker 2006b). For many Murray-Darling Basin fishes the decline is thought to be due to the poor survival of early life stages, rather than a spawning failure *per se* (Gehrke 1991; Humphries and Lake 2000; Humphries *et al.* 2002; Koehn and Harrington 2006). Modification of the hydrology within a system can have cascading effects potentially influencing water quality, energy sources, physical habitat and biotic interactions, resulting in an overall reduction in ecological integrity (Poff *et al.* 1997). In addition, the recent drought, which began in 2002, is believed to be exacerbating the impact of the modified flow regime on the native fish and overall ecological health of the Lower River Murray (Davies *et al.* 2008).

In South Australia, artificial hydrological manipulation, conducted by changing the water levels in weir pools, has been employed in an attempt to improve river health (DWLBC 2000). The water level in the Lower River Murray was artificially raised in 2005 by increasing the height of the weir during a natural increase in flow (Chapter 3). Abundance of Murray cod and freshwater catfish larvae appeared to increase during the water level raising event, although their abundances were very low throughout the entire study. However, it is unlikely that raising the water level alone initiated spawning, as other studies have found that these species do not rely on rising water levels to cue spawning (Lake 1967a; King *et al.* 2003; Humphries 2005; Koehn and Harrington 2006). Unfortunately, during this study it was not possible to isolate the effects of weir pool raising from the natural increase in flow because both conditions occurred only once and at the same time. Isolating which factors of the flow regime (e.g. magnitude, water level height, rate of rise) and environmental variables (e.g. temperature, day length, moon phase) result in successful spawning and then subsequent recruitment is still a key knowledge gap for many Murray Darling Basin species (King *et al.* 2009a).

Providing a more “natural” flow regime in regulated rivers, through environmental water allocations, is one of the potential management options for the restoration of native fish (Marchetti and Moyle 2001; Arthington *et al.* 2006). There are only a few examples of environmental water allocations in rivers throughout the world, and there have been varied levels of success for native fish conservation attributed to the manipulation of flows (e.g. Nesler *et al.* 1988; Travinchek *et al.* 1995; Freeman *et al.* 2001; King *et al.* 2009b; King *et al.* 2010). In the Barmah-Millewa forest on the River Murray, golden perch and silver perch were found to substantially increase their spawning activity, and Murray cod and trout cod increased recruitment during a flood (in 2005), that partly included using environmental water (King *et al.* 2009b; King *et al.* 2010).

The large bodied species collected in this study show positive responses to increases in flow, indeed for golden perch and silver perch, a flow stimulus is required to induce spawning. In the Lower River Murray golden perch and silver perch do not spawn in years of stable regulation (entitlement) discharge, therefore, the current study supports the need for environmental water allocations for management and restoration of native fish species (Chapter 3). The need for environmental water allocations increases during prolonged drought, and yet the availability of water decreases. Therefore, available water (environmental and irrigation) must be carefully managed to achieve the maximum benefits within a system. Throughout much of the world, water in large modified rivers is managed in an effort to meet irrigation and population resource needs, thus water is often released at slow and steady rates at times that reflect the greatest need for irrigation water usage (Ward *et al.* 2001; Bernhardt *et al.* 2005; Nilsson *et al.* 2005; Cowley 2006; Li *et al.* 2009). In such regulated systems, where the rivers act as conduits for irrigation supply it may be necessary to reconsider the way in which water is transported. For example, potentially managing the irrigation water so it is released in a way that the flows and water levels are more representative of the historical variability may also provide benefits for native fish. The results from this study suggest timing the release of water, or water level raising, to coincide with spring/summer and summer spawners will correspond with spawning of the threatened large bodied species, as well as benefiting other native species (Chapter 4). In this context, release of irrigation water or environmental water, could be viewed in an adaptive management framework (Poff *et al.* 2003; Cottingham *et al.* 2005; Richter *et al.* 2006; King *et al.* 2010), and therefore could be utilised as experimental manipulations to better understand the response of fishes and other biota to changes in the flow regime.

However, there are inherent difficulties in conducting large scale manipulative experiments under field conditions, particularly in heavily managed systems (Gillanders and Kingsford 2002). Ecologists are often reliant on monitoring changes within a system over a long period with little control over the range of disturbances, however, forewarned natural or managed disturbances can be treated as experimental manipulations (Underwood 1996). In the River Murray, these disturbances are most often changing flow volumes and raising/lowering water levels, which are prioritised for the management of human enterprise (e.g. agriculture, urban, industry, infrastructure and recreation), above the environment. However, it is essential that in this context monitoring changes in responses of organisms to disturbances should be aimed at identifying causal links (Cottingham *et al.* 2005). Ongoing monitoring, potentially over a decade or more (including years with a variety of flow regimes and varied experimental manipulations) is recognised as essential to properly understand the changes in response of the fish communities (Humphries *et al.* 2008; King *et al.* 2009b; King *et al.* 2010). The low flow conditions throughout the Murray-Darling Basin are predicted to continue for a number of years (Lintermans and Cottingham 2007; MDBC 2008), indicating that opportunities to run field manipulations of flow and water level changes particularly within the main channel of the Lower River Murray may be limited. Therefore, in order to generate sufficient data to properly address many of the future research directions outlined in this thesis, adaptive management principles need to be employed with long term monitoring.

Concluding remarks

This thesis demonstrates that the life history strategies and spawning response of Murray-Darling Basin fishes is not varied across the Basin, indicating that the currently accepted recruitment models are not restricted in their application. Importantly, the current research has provided strong support for the low flow recruitment hypothesis (Humphries *et al.* 1999) and some support for the flood recruitment model (Harris and Gehrke 1994). It suggests that the application of both these models should be used in conjunction for the successful management and rehabilitation of native fish populations in the Murray-Darling Basin. This study has also highlighted that in the heavily regulated, main river channel in the Lower River Murray, adequate densities of food occur during low flow conditions, which suggests that some aspects of these models may need refinement and further investigation in varying flow conditions. Spawning of all species occurred during a within channel flow pulse, however, two of the large bodied species, golden perch and silver perch, only spawned during this period. Whilst this study has limitations, the study suggests that the combination of the river regulation and the drought may have a significant negative impact on some of the flow reliant species.

There is a growing body of evidence to suggest that physical modifications (such as river regulation and channelisation) within riverine systems worldwide is more responsible for controlling fish diversity and population dynamics than biological interactions (Copp 1990; Jurajda 1995; Gaudin 2001; Schiemer *et al.* 2001; Arlinghaus *et al.* 2002). River regulation is one of the major causes of declines in native fishes throughout the world's rivers, as a result of the physical barriers the structures impose and alterations to the flow regimes and habitat (Ward and Stanford 1979; Welcomme 1985; Walker 1992; Matthews 1998; Humphries *et al.* 2006; Walker 2006b; Humphries *et al.* 2008). Dams and weirs have a range of negative impacts on fish; they can limit access to suitable habitats, affect water quality, change the bank structure, act as barriers by obstructing movement of fish, and affect conditions for spawning, recruitment and dispersal of eggs and larvae (Gehrke *et al.* 1995; Jurajda 1995; Humphries *et al.* 2002; Winter *et al.* 2009). In the Lower River Murray, the weir pool environments appear to provide suitable conditions for the spawning and rearing of larvae of some species, particularly small-medium bodied opportunistic or generalist species. Additionally, a number of the large bodied species (with more specific life history requirements) were found to spawn in low densities in the region, particularly under increased flow. This may suggest that the although the main channel of the Lower River Murray is heavily regulated, it could be a suitable region for restoration of the native fish fauna. In particular, to maintain currently declining fish populations, consideration should be given to the application of environmental water, in order to improve the currently debilitated flow regime.

River red gum (*Eucalyptus camaldulensis*) (photo courtesy of Lesley Alton).

REFERENCE LIST

Agostinho A. A., Gomes L. C., Verissimo S. & Okada E. K. (2004) Flood regime, dam regulation and fish in the Upper Parana River: effects on assemblage attributes, reproduction and recruitment. *Reviews in Fish Biology and Fisheries*, 14, 11-19. (doi: 10.1007/s11160-004-3551-y).

Agostinho A. A., Pelicice F. M. & Gomes L. C. (2008) Dams and the fish fauna of the Neotropical region: impacts and management related to diversity and fisheries. *Brazilian Journal of Biology*, 68, 1119-1132.

Allen G. R. (1989) *Freshwater fishes of Australia* (T.H.F. Publications: Neptune City).

Allen G. R., Midgley S. H. & Allen M. (2002) *Field guide to the freshwater fishes of Australia* (Western Australian Museum: Perth, Australia).

Anderson M. J. (2001a) A new method for non-parametric multivariate analysis of variance. *Austral Ecology*, 26, 32-46.

Anderson M. J. (2001b) Permutation tests for univariate or multivariate analysis of variance and regression. *Canadian Journal of Fisheries and Aquatic Sciences*, 58, 626-639.

Appleford P., Anderson T. A. & Gooley G. J. (1998) Reproductive cycle and gonadal development of Macquarie perch, *Macquaria australasica* Cuvier (Percichthyidae), in Lake Dartmouth and tributaries of the Murray-Darling Basin, Victoria, Australia. *Marine and Freshwater Research*, 49, 163-169.

Arlinghaus R., Engelhardt C., Sukhodolov A. & Wolter C. (2002) Fish recruitment in a canal with intensive navigation: implications for ecosystem management. *Journal of Fish Biology*, 61, 1386-1402. (doi: 10.1006/jfbi.2002.2148).

Arthington A. H., Bunn S. E., Poff L. N. & Naiman R. J. (2006) The challenge of providing environmental flow rules to sustain river ecosystems. *Ecological Applications* 16, 1311-1318.

Arthington A. H. & Pusey B. J. (2003) Flow restoration and protection in Australian rivers. *River Research and Applications*, 19, 377-395.

Arumugam P. T. & Geddes M. C. (1987) Feeding and growth of golden perch larvae and fry (*Macquaria ambigua* Richardson). *Transactions of the Royal Society of South Australia*, 111, 59-65.

Arumugam P. T. & Geddes M. C. (1992) Selectivity of microcrustacean zooplankton by golden perch (*Macquaria ambigua*) larvae and fry in laboratory studies. *Transactions of the Royal Society of South Australia*, 116, 29-34.

Backhouse G. N. & Frusher D. J. (1980) The crimson-spotted rainbowfish, *Melanotaenia fluviatilis* (Castelnau 1878). *Victorian Naturalist*, 97, 144-148.

Baker K. D., Devine J. A. & Haedrich R. L. (2009) Deep-sea fishes in Canada's Atlantic: population declines and predicted recovery times. *Environmental Biology of Fishes*, 85, 79-88. (doi: 10.10007/s10641-009-9465-8).

Balcombe S. R., Bunn S. E., Arthington A. H., Fawcett J. H., McKenzie-Smith F. J. & Wright A. (2007) Fish larvae, growth and biomass relationships in an Australian arid zone river: links between floodplains and waterholes. *Freshwater Biology*, 52, 2385-2398. (doi: 10.1111/j.1365-2427.2007.01855.x).

Baron J. S., Poff N. L., Angermeier P. L., Dahm C. N., Gleick P. H., Hairston N. G., Jackson R. B., Johnston C. A., Richter B. D. & Steinman A. D. (2002) Meeting ecological and social needs for freshwater. *Ecological Applications*, 12, 1247-1260.

Basu B. K. & Pick F. R. (1996) Factors regulating phytoplankton and zooplankton biomass in temperate rivers. *Limnology and Oceanography*, 41, 1572-1577.

Batty R. S. (1987) Effect of light-intensity on activity and food searching of larval herring, *Clupea harengus*, a laboratory study. *Marine Biology*, 94, 323-327.

Baumgartner G., Nakatani K., Gomes L. C., Bialetzki A., Sanches P. V. & Makrakis M. C. (2008a) Fish larvae from the upper Parana River: do abiotic factors affect larval density? *Neotropical Ichthyology*, 6, 551-558.

Baumgartner L. J., Reynoldson N. & Gilligan D. M. (2006) Mortality of larval Murray cod (*Maccullochella peelii peelii*) and golden perch (*Macquaria ambigua*) associated with passage through two types of low-head weirs. *Marine and Freshwater Research*, 57, 187-191.

Baumgartner L. J., Stuart I. G. & Zampatti B. P. (2008b) Determining diel variation in fish assemblages downstream of three weirs in a regulated lowland river. *Journal of Fish Biology*, 72, 218-232. (doi: 10.1111/j.1095-8649.2007.01696.x).

Beck M. W., Heck K. L., Able K. W., Childers D. L., Eggleston D. B., Gillanders B. M., Halpern B., Hays C. G., Hoshino K., Minello T. J., Orth R. J., Sheridan P. F. & Weinstein M. R. (2001) The identification, conservation, and management of estuarine and marine nurseries for fish and invertebrates. *Bioscience*, 51, 633-641.

Begon M., Harper J. L. & Townsend C. R. (1996) *Ecology: individuals, populations and communities* (3rd Edn). (Blackwell Science Ltd: Oxford, United Kingdom).

Bernhardt E. S., Palmer M. A., Allan J. D., Alexander G., Barnas K., Brooks S., Carr J., Clayton S., Dahm C., Follstad-Shah J., Galat D., Gloss S., Goodwin P., Hart D., Hassett B., Jenkinson R., Katz S., Kondolf G. M., Lake P. S., Lave R., Meyer J. L., O'Donnell T. K., Pagano L., Powell B. & Sudduth E. (2005) Ecology: Synthesizing US river restoration efforts. *Science*, 308, 636-637. (doi: 10.1126/science.1109769).

Bertozzi T., Adams M. & Walker K. F. (2000) Species boundaries in carp gudgeons (Eleotrididae : *Hypseleotris*) from the River Murray, South Australian: evidence for multiple species and extensive hybridization. *Marine and Freshwater Research*, 51, 805-815.

Bialetzki A., Nakatani K., Sanches P. V., Baumgartner G. & Gomes L. C. (2005) Larval fish assemblage in the Baı´a River (Mato Grosso do Sul State, Brazil): temporal and spatial patterns. *Environmental Biology of Fishes*, 73, 37-47.

Blanch S. J., Walker K. F. & Ganf G. G. (2000) Water regimes and littoral plants in four weir pools of the River Murray, Australia. *Regulated Rivers: Research and Management*, 16, 445-456.

Bone Q., Marshall N. B. & Blaxter J. H. S. (1995) *Biology of fishes* (2nd Edn). (Blackie Academic and Professional Publishers: Glasgow, United Kingdom).

Boulton A. J. (2003) Parallels and contrasts in the effects of drought on stream macroinvertebrate assemblages. *Freshwater Biology* 48, 1173-1185.

Bray J. & Curtis J. (1957) An ordination of the upland forest communities of Southern Wisconsin. *Ecological Monographs*, 27, 325-349.

Brind'Amour A. & Boisclair D. (2004) Comparison between two sampling methods to evaluate the structure of fish communities in the littoral zone of a Laurentian lake. *Journal of Fish Biology*, 65, 1372-1384.

Brodeur R. D. & Rugen W. C. (1994) Diel vertical distribution of ichthyoplankton in the Northern Gulf of Alaska. *Fishery Bulletin*, 92, 223-235.

Brown L. R. & Ford T. (2002) Effects of flow on the fish communities of a regulated California river: implications for managing native fishes. *River Research and Applications*, 18, 331-342. (doi: 10.1002/rra.673).

Bult T. P., Haedrich R. L. & Schneider D. C. (1998) New technique describing spatial scaling and habitat selection in riverine habitats. *Regulated Rivers: Research and Management*, 14, 107-118. (doi: 10.1002/(SICI)1099-1646(199801/02)14:1<107::AID-RRR488>3.0.CO;2-A).

Bye V. J. (1984) The role of environmental factors in the timing of reproductive cycles. In *Fish reproduction: strategies and tactics*. (Eds G.W. Potts and R.J. Wootton). (Acadmeic Press Inc: London, United Kingdom).

Cadwallader P. L. (1978) Some causes of the decline in range and abundance of native fishes in the Murray-Darling River system. *Proceedings of the Royal Society of Victoria*, 90, 211-224. (QY).

Cadwallader P. L. (1977) *J. O. Langtry's 1949-50 Murray River investigations.* Fisheries and Wildlife Paper No. 13, Victoria, Australia.

Cadwallader P. L. (1978) Some causes of the decline in range and abundance of native fishes in the Murray-Darling River system. *Proceedings of the Royal Society of Victoria*, 90, 211-224.

Cadwallader P. L. & Backhouse G. N. (1983) *A Guide to the freshwater fish of Victoria* (Victorian Government Printing Office: Melbourne, Australia).

Cadwallader P. L. & Lawrence B. (1990) Chapter 22: Fish. In *The Murray*. (Eds N.A. McKay and D. Eastburn) pp. 317-363. (Murray-Darling Basin Commission: Canberra, Australia).

Cadwallader P. L. & Rogan P. L. (1977) Macquarie perch, *Macquaria australasica* (Pisces: Percicthyidae), of Lake Eildon, Victoria. *Australian Journal of Ecology*, 2, 409-418.

Carassou L., le Borgne R. & Ponton D. (2009) Diet of pre-settlement larvae of coral reef fishes: selection of prey types and sizes. *Journal of Fish Biology*, Published online by Wiley InterScience. (doi: 10.1111/j.1095-8649.2009.02312.x).

Carter J. G., Lamarra V. A. & Ryel R. J. (1986) Drift of larval fishes in the upper Colorado River. *Journal of Freshwater Ecology*, 3, 567-577.

Chaves H. M. L. & Alipaz S. (2007) An integrated indicator based on basin hydrology, environment, life, and policy: The watershed sustainability index. *Water Resources Management*, 21, 883-895. (doi: 10.1007/s11269-006-9107-2).

Cheshire K. J. & Ye Q. (2008) *Spatial and temporal variations in larval fish assemblages between Locks 1 and 6 in the River Murray, South Australia: with reference to drought intervention monitoring 2007*. South Australian Research and Development Institute (SARDI), Aquatic Sciences, SARDI research report series number: 304, SARDI Aquatic Sciences Publication Number F2008/000685-1, Adelaide, Australia.

Clarke K. (1993) Non-parametric multivariate analysis of changes in community structure. *Australian Journal of Ecology*, 18, 117-143.

Clarke K. & Ainsworth M. (1993) A method of linking multivariate community structure to environmental variables. *Marine Ecological Progress Series*, 92, 205-219.

Clunie P. & Koehn J. D. (2001a) *Freshwater catfish: A resource document.* Final Report for Natural Resource Managment Strategy Project r7002 to the Murray Darling Basin Commision. Arthur Rylah Institute for Environmental Research, Department of Natural Resources and Environment, Melbourne, Australia.

Clunie P. & Koehn J. D. (2001b) *Silver Perch: A Resource Document.* Final Report for Natural Resource Managment Strategy Project r7002 to the Murray Darling Basin Commision. Arthur Rylah Institute for Environmental Research, Department of Natural Resources and Environment, Melbourne, Australia.

Copp G. (1992) Comparative microhabitat use of cyprinid larvae and juveniles in a lotic floodplain channel. *Environmental Biology of Fishes*, 33, 181-193.

Copp G. H. (1989) Electrofishing for fish larvae and 0+ juveniles: equipment modifications for increased efficiency with short fishes. *Aquaculture and Fisheries Management*, 20, 453-462.

Copp G. H. (1990) Shifts in the microhabitat of larval and juvenile roach, *Rutilus rutilus* (L.) in a floodplain channel. *Journal of Fish Biology*, 36, 683-692.

Corbett B. W. & Powles P. M. (1986) Spawning and larval drift of sympatric walleyes and white suckers in an Ontario stream. *Transactions of the American Fisheries Society*, 115, 41-46.

Cottingham P., Quinn G., King A. J., Norris R., Chessman B. C. & Marshall C. (2005) *Environmental flows monitoring and assessment framework.* Cooperative Research Centre for Freshwater Ecology. , Available at: http://freshwater.canberra.edu.au/publications.nsf/60da502789f083dbca256ee40022a54e/b217ed362dcbc90bca256fc7001cf693? (last accessed January 2010), Canberra, Australia.

Cowley D. E. (2006) Strategies for ecological restoration of the Middle Rio Grande in New Mexico and recovery of the endangered Rio Grande silvery minnow. *Reviews in Fisheries Science*, 14, 169-186. (doi: 10.1080/10641260500341619).

Crabb P. (1997) *Murray-Darling Basin Resources* (Murray-Darling Basin Commission: Canberra, Australia).

CRCFE (2003) *Ecological assessment of environmental flow reference points for the River Murray system: Interim Report.* Cooperative Research Centre for Freshwater Ecology Prepared by the

Scientific Reference Panel for the Murray-Darling Basin Commission, Living Murray Initiative, Canberra, Australia.

Cushing D. H. (1990) Plankton production and year-class strength in fish populations: an update of the match/mismatch hypothesis. *Advances in Marine Biology*, 26, 249-293.

Davies P. E., Harris J. H., Hillman T. J. & Walker K. F. (2008) *SRA Report 1: A report on the ecological health of rivers in the Murray-Darling Basin, 2004-2007.* Prepared by the Independent Sustainable Rivers Audit Group for the Murray-Darling Basin Ministerial Council, MDBC Publication No. 16/08, Canberra, Australia.

Davis T. L. O. (1977a) Reproductive biology of the freshwater catfish, *Tandanus tandanus* Mitchell, in the Gwydir River, Australia I. Structure of the gonads. *Australian Journal of Marine and Freshwater Research*, 28, 139-158.

Davis T. L. O. (1977b) Reproductive biology of the freshwater catfish, *Tandanus tandanus* Mitchell, in the Gwydir River, Australia II. Gondal cycle and fecundity. *Australian Journal of Marine and Freshwater Research*, 28, 159-169.

de Figueiredo G. M., Nash R. D. M. & Montagnes D. S. (2007) Do protozoa contribute significantly to the diet of larval fish in the Irish Sea? *Journal of the Marine Biological Association of the United Kingdom*, 87, 843-850.

de Groot S. J. (2002) A review of the past and present status of anadromous fish species in the Netherlands: is restocking the Rhine feasible? *Hydrobiologia*, 478, 205-218.

de Leeuw J. J., Buijse A. D., Haidvogl G., Lapinska M., Noble R., Repecka R., Virbickas T., Wisniewolski W. & Wolter C. (2007) Challenges in developing fish-based ecological assessment methods for large floodplain rivers. *Fisheries Management and Ecology*, 14, 483-494. (doi: 10.1111/j.1365-2400.2007.00576.x).

Denney N. H., Jennings S. & Reynolds J. D. (2002) Life history correlates of maximum population growth rates in marine fishes. *Proceedings of the Royal Society of London. Series B: Biological Sciences*, 269, 2229-2237. (doi: 10.1098/rspb.2002.2138).

Dirnberger J. M. & Threlkeld S. T. (1986) Advective effects of a reservoir flood on zooplankton abundance and dispersion. *Freshwater Biology*, 16, 387-396.

Dudley R. K. & Platania S. P. (2007) Flow regulation and fragmentation imperil pelagic-spawning riverine fishes. *Ecological Applications*, 17, 2074-2086. (doi:10.1890/06-1252.1).

Dufrene M. & Legendre P. (1997) Species assemblages and indicator species: the need for a flexible asymmetrical approach. *Ecological Monographs*, 67, 345-366.

DWLBC (2000) *Flow manipulation using locks and weirs.* Department of Water, Land and Biodiversity Conservation, Fact Sheet 21, Adelaide, Australia.

Facey D. E. & Grossman G. D. (1992) The relationship between water velocity, energetic costs and microhabitat use in 4 North American stream fishes. *Hydrobiologia*, 239, 1-6.

Ferrari I., Farabegoli A. & Mazzoni R. (1989) Abundance and diversity of planktonic rotifers in the Po River. *Hydrobiologia*, 186, 201-208.

Flore L. & Keckeis H. (1998) The effect of water current on foraging behaviour of the rheophilic cyprinid *Chondrostoma nasus* (L.) during ontogeny: evidence of a trade-off between energetic gain and swimming costs. *Regulated Rivers: Research and Management*, 14, 141-154.

Floyd K. B., Hoyt R. D. & Timbrook S. (1984) Chronology of appearance and habitat partitioning by stream larval fishes. *Transactions of the American Fisheries Society*, 113, 217-223.

Forrester G. E. (1994) Diel patterns of drift by 5 species of mayfly at different levels of fish predation. *Canadian Journal of Fisheries and Aquatic Sciences*, 51, 2549-2557.

Fortier L., Ponton D. & Gilbert M. (1995) The match mismatch hypothesis and the feeding success of fish larvae in ice-covered South Eastern Hudson-Bay *Marine Ecology-Progress Series*, 120, 11-27.

Franzen W. G. & Harbight S. M. (1992) Tests of drift samplers for estimating abundance of recently hatched walleye larvae in small rivers. *North American Journal of Fisheries Management*, 12, 396-405.

Freeman M. C., Bowen Z. H., Bovee K. D. & Irwin E. R. (2001) Flow and habitat effects on juvenile fish abundance in natural and altered flow regimes. *Ecological Applications*, 11, 179-190.

Fukami K., Watanabe A., Fujita S., Yamaoka K. & Nishijima T. (1999) Predation on naked protozoan microzooplankton by fish larvae. *Marine Ecology-Progress Series*, 185, 285-291.

Gadomski D. M. & Barfoot C. A. (1998) Diel and distribution abundance patterns of fish embryos and larvae in the lower Columbia and Deschutes rivers. *Environmental Biology of Fishes*, 51, 353-368.

Galat D. L. & Zweimuller I. (2001) Conserving large-river fishes: is the highway analogy an appropriate paradigm? *Journal of the North American Benthological Society*, 20, 266-279.

Gale W. F. & Mohr H. W. J. (1978) Larval fish drift in a large river with comparison of sampling methods. *Transactions of the American Fisheries Society*, 107, 46-55.

Gallagher R. P. & Conner J. V. (1983) Comparison of two icthyoplankton sampling gears with notes on microdistribution of fish larvae in a large river. *Transactions of the American Fisheries Society*, 112, 280-285.

Garner P. (1996) Microhabitat use and diet of 0+ cyprinid fishes in a lentic, regulated reach of the River Great Ouse, England. *Journal of Fish Biology*, 48, 367-382.

Gatz A. J. & Harig A. L. (1993) Decline in the index of biotic integrity of Delaware Run, Ohio, over 50 years. *Ohio Journal of Science*, 93, 95-100.

Gaudin P. (2001) Habitat shifts in juvenile riverine fish. *Archiv fur Hydrobiologie*, 12, 393-408.

Geddes M. C. & Puckridge J. T. (1989) Survival and growth of larval and juvenile native fish: the importance of the floodplain. In *Proceedings of the Workshop on Native Fish Management*. Canberra, Australia. (Ed. B. Lawrence) pp. 105-115. (Murray-Darling Basin Commission).

Gehrke P. C. (1990) Spatial and temporal dispersion patterns of golden perch, *Macquaria ambigua*, larvae in an artificial floodplain environment. *Journal of Fish Biology*, 37, 225-236.

Gehrke P. C. (1991) Avoidance of inundated floodplain habitat by larvae of Golden Perch (*Macquaria ambigua* Richardson): influence of water quality or food distribution. *Australian Journal of Marine and Freshwater Research*, 42, 707-719.

Gehrke P. C. (1992) Diel abundance, migration and feeding of fish larvae (Eleotridae) in a floodplain billabong. *Journal of Fish Biology*, 40, 695-707.

Gehrke P. C. (1994) Influence of light intensity and wavelength on phototactic behaviour of larval silver perch, *Bidyanus bidyanus,* and golden perch, *Macquaria ambigua,* and the effectiveness of light traps. *Journal of Fish Biology*, 44, 741-751.

Gehrke P. C., Brown P., Schiller C., Moffatt D. B. & Bruce A. M. (1995) River regulation and fish communities in the Murray-Darling River System, Australia. *Regulated Rivers: Research and Management*, 11, 363-375.

Gillanders B. M. & Kingsford M. J. (2002) Impact of changes in flow of freshwater on estuarine and open coastal habitats and the associated organisms. *Oceanography and Marine Biology*, 40, 233-309.

Gilligan D. & Schiller C. (2004) Downstream transport of the eggs, larvae and juvenile fish in the Murray River: impact of river regulation on downstream dispersal. In *Downstream movement of fish in the Murray-Darling Basin Workshop*. Canberra, Australia. (Eds M. Lintermans and B. Phillips) pp. 41-50. (Murray- Darling Basin Commission).

Gippel C. J. (2003) *Review of achievements and outcomes of environmental flow initiatives undertaken on the extended River Murray system to August 2002*. Report to The Living Murray Initiative of the Murray-Darling Basin Commission. Fluvial Systems Pty Ltd, Canberra, Australia.

Gotceitas V., Puvanendran V., Leader L. L. & Brown J. A. (1996) An experimental investigation of the 'match mismatch' hypothesis using larval Atlantic cod. *Marine Ecology-Progress Series*, 130, 29-37.

Govoni J. J., Ortner P. B., Alyamani F. & Hill L. C. (1986) Selective feeding of spot, *Leiostomus xanthurus*, and Atlantic croaker, *Micropogonias undulatus*, larvae inthe Northern Gulf of Mexico. *Marine Ecology-Progress Series*, 28, 175-183.

Growns I. (2004) A numerical classification of reproductive guilds of the freshwater fishes of south-eastern Australia and their application to river management. *Fisheries Management and Ecology*, 11, 369-377. (doi:10.1111/j.1365-2400.2004.00404.x).

Habit E., Belk M., Victoriano P. & Jaque E. (2007) Spatio-temporal distribution patterns and conservation of fish assemblages in a Chilean coastal river. *Biodiversity and Conservation*, 16, 3179-3191.

Haines G. B. & Tyus H. M. (1990) Fish associations and environmental variables in age-0 Colorado squawfish habitats, Green River, Utah. *Journal of Freshwater Ecology*, 5, 427-435.

Haldorson L., Prichett M., Paul A. J. & Ziemann D. (1993) Vertical distribution and migration of fish larvae in a Northeast Pacific bay. *Marine Ecology-Progress Series*, 101, 67-80.

Hammer M. & Walker K. F. (2004) A catalogue of South Australian freshwater fishes, including new records, range extension and translocations. *Transactions of the Royal Society of South Australia*, 128, 85-97.

Harris J., H & Gehrke P. C. (1994) Development of predictive models linking fish population recruitment with stream flow. In *Australian Society for Fish Biology Workshop*. Perth, Australia. (Ed. D.A. Hancock) pp. 195-97. (Australian Society for Fish Biology, Sydney).

Havel J. E., Medley K. A., Dickerson K. D., Angradi T. R., Bolgrien D. W., Bukaveckas P. A. & Jicha T. M. (2009) Effect of main-stem dams on zooplankton communities of the Missouri River (USA). *Hydrobiologia*, 628, 121-135. (10.1007/s10750-009-9750-8).

Hawking J. H. & Smith F. J. (1997) *Colour guide to invertbrates of Australian inland waters* (Cooperative Research Centre for Freshwater Ecology: Albury, Australia).

Heggenes J. (1996) Habitat selection by brown trout (*Salmo trutta*) and young Atlantic salmon (*S. salar*) in streams: static and dynamic hydraulic modelling. *Regulated Rivers: Research and Management*, 12, 155-169.

Hernandez F. J. & Lindquist D. G. (1999) A comparison of two light-trap designs for sampling larval and presettlement juvenile fish above a reef in Onslow Bay, North Carolina. *Bulletin of Marine Science*, 64, 173-184.

Hjort J. (1914) Fluctuations in the great fisheries of Northern Europe viewed in the light of biological research. *Rapports Et Proces-Verbaux Des Reunions, Conseil International Pour l'Exploration De La Mer*, 20, 1-228.

Holland L. E. (1986) Distribution of early life history stages of fishes in selected pools of the Upper Mississippi River. *Hydrobiologia*, 136, 121-130.

Holland L. E. & Sylvester J. R. (1983) Distribution of larval fishes related to potential navigation impacts on the Upper Mississippi River, pool-7. *Transactions of the American Fisheries Society*, 112, 293-301.

Horjt J. (1914) Fluctuations in the Great Fisheries of Northern Europe viewed in the light of biological research. *Rapports Et Proces-Verbaux Des Reunions, Conseil International Pour l'Exploration De La Mer*, 20, 1-228.

Houde E. D. (1997) Patterns and consequences of selective processes in teleost early life histories. In *Early life history and recruitment in fish populations.* (Eds R.C. Chambers and E.A. Trippel) pp. 173-196. (Chapman and Hall London, UK).

Houde E. D. (2002) Mortality. In *Fishery Science - the unique contributions of early life stages.* (Eds L.A. Fuiman and R.G. Werner) pp. 64-87. (Blackwell Science: Oxford, United Kingdom).

Humphries P. (1995) Life history, food and habitat of Southern pygmy perch, *Nannoperca australis*, in the Macquarie River, Tasmania. *Marine and Freshwater Research*, 46, 1159-1169.

Humphries P. (2005) Spawning time and early life history of the Murray cod, *Maccullochella peelii peelii* (Mitchell) in an Australian river. *Environmental Biology of Fishes*, 72, 393-407.

Humphries P. & Baldwin D. S. (2003) Drought and aquatic ecosystems: an introduction. *Freshwater Biology*, 48, 1141-1146.

Humphries P., Brown P., Douglas J., Pickworth A., Strongman R., Hall K. & Serafini L. G. (2008) Flow-related patterns in abundance and composition of the fish fauna of a degraded Australian lowland river. *Freshwater Biology*, 53, 789-813.

Humphries P., Cook R. A., Richardson A. J. & Serafini L. G. (2006) Creating a disturbance: manipulating slackwaters in a lowland river. *River Research and Applications*, 22, 525-542.

Humphries P. & King A. (2003) Drifting larvae in Murray-Darling Basin rivers: composition, spatial and temporal patterns and distance drifted. In *Downstream movement of fish in the Murray-Darling Basin Workshop*. Canberra, Australia

Humphries P., King A. J. & Koehn J. D. (1999) Fish flows and flood plains: links between freshwater fishes and their environment in the Murray-Darling River system, Australia. *Environmental Biology of Fishes*, 56, 129-151.

Humphries P. & Lake P. S. (2000) Fish larvae and the management of regulated rivers. *Regulated Rivers: Research and Management*, 16, 421-432.

Humphries P., Serafini L. G. & King A. J. (2002) River regulation and fish larvae: variation through space and time. *Freshwater Biology*, 47, 1307-1331.

Ingram B. A. & De Silva S. S. (2007) Diet composition and preference of juvenile Murray cod, trout cod and Macquarie perch (Percichthyidae) reared in fertilised earthen ponds. *Aquaculture*, 271, 260-270. (doi: 10.1016/j.aquaculture.2007.06.012).

Ingram B. A. & Douglas J. W. (1995) Threatened fishes of the world: *Maccullochella macquariensis* (Cuvier, 1829) (Percichthyidae). *Environmental Biology of Fishes*, 43, 38-38.

Islam M. S. & Tanaka M. (2009) Diet and prey selection in larval and juvenile Japanese anchovy *Engraulis japonicus* in Ariake Bay, Japan. *Aquatic Ecology*, 43, 549-558. (doi: 10.1007/s10452-008-9207-6).

Ivantsoff W. & Crowley L. E. L. M. (1996) Family Atherinidae: silversides or hardyheads. In *Freshwater Fishes of South-Eastern Australia*. (Ed. R.M. McDowall) pp. 123-133. (Reed Books: Sydney, Australia).

Jackson P. D. (1978) Spawning and early development of river blackfish, *Gadopsis marmoratus* Richardsons (Gaopsiformes: Gadopsidae), in the McKenzie River, Victoria. *Australian Journal of Marine and Freshwater Research*, 29, 293-298.

Jobling M. (1995) *Environmental biology of fishes* (Chapman and Hall Publishers: London, United Kingdom).

Johnson D. L. (2000) Preliminary examination of the match-mismatch hypothesis and recruitment variability of yellowtail flounder, Limanda ferruginea. *Fishery Bulletin*, 98, 854-863.

Junk W. J., Bayley P. B. & Sparks R. E. (1989) The flood-pulse concept in river-floodplain systems. In *Proceedings of the International Large River Symposium*. (Ed. D.P. Dodge) pp. 110-27. (Canadian Special Publication of Fisheries and Aquatic Sciences, 106).

Jurajda P. (1995) Effect of channelization and regulation on fish recruitment in a floodplain river. *Regulated Rivers: Research and Management*, 10, 207-215.

Jurajda P. (1999) Comparative nursery habitat use by 0+ fish in a modified lowland river. *Regulated Rivers: Research and Management*, 15, 113-124.

Kaminskas S. & Humphries P. (2009) Diet of Murray cod (*Maccullochella peelii peelii*) (Mitchell) larvae in an Australian lowland river in low flow and high flow years. *Hydrobiologia*, Published online 5 Nov 2009. (doi:10.1007/s10750-009-9973-8).

Karr J. R. (1981) Assesment of biotic integrity using fish communities. *Fisheries*, 6, 21-27.

Karr J. R. (1991) Biological integrity: a long neglected aspect of water resource management. *Ecological Applications*, 1, 66-84.

Kelso W. E. & Rutherford D. A. (1996) Collection, preservation and identification of fish eggs and larvae. In *Fisheries Techniques* (2nd Edn). (Eds B.R. Murphy and D.W. Willis) pp. 255-302. (American Fisheries Society: Marysland, United States of America).

Kennard M. J., Pusey B. J. & Arthington A. H. (2001) *Trophic ecology of freshwater fishes in Australia* (Cooperative Research Centre for Freshwater Ecology: Brisbane, Australia).

King A. J. (2002) *Recruitment ecology of fish in floodplain rivers of the Southern Murray-Darling Basin, Australia*. PhD thesis, Monash University.

King A. J. (2004a) Density and distribution of potential prey for larval fish in the main channel of a floodplain river: pelagic versus epibenthic meiofauna. *River Research and Applications*, 20, 883-897.

King A. J. (2004b) Ontogenetic patterns of habitat use by fishes within the main channel of an Australian floodplain river. *Journal of Fish Biology*, 65, 1582-1603.

King A. J. (2005) Ontogenetic dietary shifts of fishes in an Australian floodplain river. *Marine and Freshwater Research*, 56, 215-225.

King A. J. & Crook D. A. (2002) Evaluation of a sweep net electrofishing method for the collection of small fish and shrimp in lotic freshwater environments. *Hydrobiologia*, 472, 223-233.

King A. J., Crook D. A., Koster W. M., Mahoney J. & Tonkin Z. (2005) Comparison of larval fish drift in the Lower Goulburn and mid-Murray Rivers. *Ecological Management and Restoration* 6, 136-138.

King A. J., Humphries P. & Lake P. S. (2003) Fish recruitment on floodplains: the roles of patterns of flooding and life history characteristics. *Canadian Journal of Fisheries and Aquatic Sciences*, 60, 773-786.

King A. J., Ramsey D., Baumgartner L. J., Humphries P., Jones M., Koehn J. D., Lyon J., Mallen-Cooper M., Meredith S., Vilizzi L., Ye Q. & Zampatti B. P. (2009a) *Environmental requirements for managing successful fish recruitment in the Murray River Valley: review of existing knowledge*. Department of Sustainability and Environment, Arthur Rylah Institute for Environmental Research Technical Report Series No. 197, Heidleberg, Australia

King A. J., Tonkin Z. & Mahoney J. (2007) *Assessing the effectiveness of environmental flows on fish recruitment in Barmah-Millewa Forest.* Arthur Rylah Institute for Environmental Research, DSE, MDBC Project No. BMF 2004.09, Melbourne, Australia.

King A. J., Tonkin Z. & Mahoney J. (2008a) Environmental flow enhances native fish spawning and recruitment in the Murray River, Australia. *River Research and Applications*, Published online by Wiley InterScience. (doi: 0.1002/rra.1209).

King A. J., Tonkin Z. & Mahoney J. (2008b) Environmental flow enhances native fish spawning and recruitment in the Murray River, Australia. *River Research and Applications*, 25, 1205-1218. (doi: 0.1002/rra.1209).

King A. J., Tonkin Z. & Mahoney J. (2009b) Environmental flow enhances native fish spawning and recruitment in the Murray River, Australia. *River Research and Applications*, 25, 1205-1218. (doi: 0.1002/rra.1209).

King A. J., Ward K. A., O'Connor P., Green D., Tonkin Z. & Mahoney J. (2010) Adaptive management of an environmental watering event to enhance native fish spawning and recruitment. *Freshwater Biology*, 55, 17-31. (doi: 10.1111/j.1365-2427.2009.02178.x).

King J., Cambray J. A. & Impson N. D. (1998) Linked effects of dam-released floods and water temperature on spawning of the Clanwilliam yellowfish (*Barbus capensis). Hydrobiologia* 384, 245-265.

Koehn J. D. & Harrington D. J. (2005) Collection and distribution of the early life stages of the Murray cod (*Maccullochella peelii peelii*) in a regulated river. *Australian Journal of Zoology*, 53, 137-144.

Koehn J. D. & Harrington D. J. (2006) Environmental conditions and timing for the spawning of Murray cod (*Maccullochella peelii peelii*) and the endangered trout cod (*M. macquariensis*) in Southeastern Australian Rivers. *River Research and Applications*, 33, 327-342.

Koehn J. D. & Nicol S. J. (1998) Habitat and movement requirements of fish. In *1996 Riverine Environment Forum*. Brisbane, Australia. (Eds R.J. Baens and R. Laehane) pp. 1-6. (Murray-Darling Basin Commision).

Koehn J. D. & O'Connor W. G. (1990) *Biological information for management of native freshwater fish in Victoria.* Government Printer, Melbourne, Australia.

Kruk A. & Penczak T. (2003) Impoundment impact on populations of facultative riverine fish. *Annales De Limnologie-International Journal of Limnology*, 39, 197-210.

Lake J. S. (1967a) Rearing experiments with five species of Australian freshwater fishes: I. Inducement to spawning. *Australian Journal of Marine and Freshwater Research*, 18, 137-153.

Lake J. S. (1967b) Rearing experiments with five species of Australian freshwater fishes: II. Morphogenesis and ontogeny. *Australian Journal of Marine and Freshwater Research*, 18, 155-173. (64).

Lake J. S. (1971) *Freshwater fishes and rivers of Australia* (Thomas Nelson: Sydney, Australia).

Lake P. S. (2003) Ecological effects of perturbation by drought in flowing waters. *Freshwater Biology*, 48, 1161-1172. (doi:10.1046/j.1365-2427.2003.01086.x).

Leigh S. J., Zampatti B. P. & Nicol J. M. (2008) *Spatial and temporal variation in larval fish assemblage structure in the Chowilla Anabranch system: with reference to water physico-chemistry and stream hydrology*. SARDI Research Report Series No. 286. South Australian Research and Development Institute (SARDI) Aquatic Sciences, SARDI Aquatic Sciences Publication No: F2008/000051-1, Adelaide, Australia.

Leis J. M. (1991) Vertical-distribution of fish larva in the Great Barrier Reef Lagoon, Australia. *Marine Biology*, 109, 157-166.

Lemly A. D., Kingsford R. T. & Thompson J. R. (2000) Irrigated agriculture and wildlife conservation: conflict on a global scale. *Environmental management*, 25, 485-512.

Li Y. Q., Chen Y. N., Zhang Y. Q. & Xia Y. (2009) Rehabilitating China's largest inland river. *Conservation Biology*, 23, 531-536. (doi: 10.1111/j.1523-1739.2009.01238.x).

Lieschke J. A. & Closs G. P. (1999) Regulation of zooplankton composition and distribution by a zooplanktivorous fish in a shallow, eutrophic floodplain lake in south east Australia. *Archiv Fur Hydrobiologie*, 146, 397-412.

Lindquist D. G. & Shaw R. F. (2005) Effects of current speed and turbidity on stationary light-trap catches of larval and juvenile fishes. *Fisheries Bulletin*, 103, 438-444.

Lintermans M. (2007) *Fishes of the Murray-Darling Basin: an introductory guide* (Murray-Darling Basin Commission: Canberra, Australia).

Lintermans M. & Cottingham P. (2007) *Fish out of water – lessons for managing native fish during drought*. Murray-Darling Basin Commission, Canberra, Australia.

Llewellyn L. C. (1971) Breeding studies on the freshwater forage fish of the Murray-Darling river system. *The Fisherman*, June, 1-12.

Llewellyn L. C. (1974) Spawning, development and distribution of Southern pigmy perch *Nannoperca australis australis,* Gunther from inland waters in Eastern Australia. *Australian Journal of Marine and Freshwater Research*, 25, 121-149.

Llewellyn L. C. (1979) Some observations on the spawning and development of the Mitchellian freshwater hardyhead *Craterocephalus fluviatilis* McCulloch from inland waters in New South Wales. *Australian Zoologist*, 20, 269-288.

Lloyd L. N. & Walker K. F. (1986) Distribution and conservation status of small freshwater fish in the River Murray, South Australia. *Transactions of the Royal Society of South Australia*, 110, 49-57.

Mackay N. J. (1973) Histological changes in the ovaries of the golden perch, *Plectroplites ambiguus*, associated with the reproductive cycle. *Australian Journal of Marine and Freshwater Research*, 24, 95-101.

Magoulick D. D. & Kobza R. M. (2003) The role of refugia for fishes during drought: a review and synthesis. *Freshwater Biology*, 48, 1186-1198.

Maheshwari B. L., Walker K. F. & McMahon T. A. (1995) Effects of regulation on the flow regime of the River Murray, Australia. *Regulated Rivers: Research and Management*, 10, 15-38.

Mallen-Cooper M. & Stuart I. G. (2003) Age, growth and non-flood recruitment of two potamodromous fishes in a large semi-arid/temperate river system. *River Research and Applications*, 19, 697-719.

Mantel N. (1967) The detection of disease clustering and generalized regression approach. *Cancer Research* 27, 209-220.

Marchetti M. P. & Moyle P. B. (2001) Effects of flow regime on fish assemblages in a regulated California stream. *Ecological Applications*, 11, 530-539.

Martinez P. J., Chart T. E., Trammell M. A., Wullschleger J. G. & Bergersen E. P. (1994) Fish sepcies composition before and after construction of a main stem reservoir on the White River, Colorado. *Environmental Biology of Fishes*, 40, 227-239.

Matthews W. J. (1998) *Patterns in freshwater fish ecology* (Chapman and Hall: Massachusetts, United States of America).

Matthews W. J. & Marsh-Mathews E. (2007) Extirpation of red shiner in direct tributaries of lake texoma (Oklahoma-Texas): a cautionary case history from a fragmented river-reservoir system. *Transactions of the American Fisheries Society*, 136, 1041-1062. (doi: 10.1577/t06-059.1).

May R. C. (1974) Larval mortality in fishes In *The early life history of fish.* (Ed. J.H.S. Blaxter) pp. 3-19. (Springer-Verlag: Berlin, Germany).

McCune B., Grace J. B. & Urban D. L. (2002) *Analysis of ecological communities* (MjM Software: Oregon, United States of America).

McCune B. & Mefford M. J. (1999) PC-ORD. Multivariate analysis of ecological data. (V.5.0.) (MjM Software: Oregon, United States of America).

McDowall R. M. (1988) *Diadromy in fishes; migrations between freshwater and marine environments* (Timber Press: Oregon, United States of America).

McDowall R. M. (1996) Family Retropinnidae: southern smelts. In *Freshwater fishes of south-eastern Australia.* (Ed. R.M. McDowall) pp. 92-95. (Reed Books: Sydney, Australia).

McMahon T. A. & Finlayson B. L. (2003) Droughts and anti-droughts: the low flow hydrology of Australian rivers. *Freshwater Biology*, 48, 1147-1160.

McNeil D. G. (2004) *Ecophysiology and behaviour of Ovens River floodplain fish: hypoxia tolerance and the role of the physicochemical environment in structuring Australian billabong fish communities.* PhD thesis, LaTrobe University.

MDBC (2002) *The Living Murray. A discussion paper on restoring the health of the River Murray.* Murray-Darling Basin Commission, Canberra, Australia.

MDBC (2004) *Native fish strategy for the Murray-Darling Basin 2003-2013.* Murray -Darling Basin Commission, MDBC Publication No. 25/04, Canberra, Australia.

MDBC (2007) *River Murray system drought update number 11.* Murray-Darling Basin Commission, Canberra, Australia.

MDBC (2008) *River Murray system drought update number 13.* Murray-Darling Basin Commission, Canberra, Australia.

Medeiros E. S. F. & Arthington A. H. (2008) Diel variation in food intake and diet composition of three native fish species in floodplain lagoons of the Macintyre River, Australia. *Journal of Fish Biology*, 73, 1024-1032. (doi: 10.1111/j.1095-8649.2008.01959.x).

Meredith S. N., Gawne B., Sharpe C., Whiterod N., Conallin A. & Zukowski S. (2002) *Dryland floodplain ecosystems: influence of flow patterns on fish production.* Final report to AFFA. Murray Darling Freshwater Research Centre, report no. 1/2002, Mildura, Australia.

Meredith S. N., Matveev V. F. & Mayes P. (2003) Spatial and temporal variability in the distribution and diet of the gudgeon (Eleotridae: *Hypseleotirs* spp.) in a subtropical Australian resevoir. *Marine and Freshwater Research*, 54, 1009-1017.

Merrick J. R. & Schmida G. E. (1984) *Australian freshwater fishes biology and management* (Griffin Press: Adelaide, Australia).

Milton D. A. & Arthington A. H. (1983) Reproduction and growth of *Craterocephalus marjoriae* and *C. stercusmuscarum* (Pisces: Atherinidae) in Southeastern Queensland, Australia. *Freshwater Biology*, 13, 589-597.

Milton D. A. & Arthington A. H. (1984) Reproductive strategy and growth of the crimson-spotted rainbowfish, *Melanotaenia spelendid fluviatilis* (Castelnau) (Pisces: Melantaeniidae) in Southeastern Queensland. *Australian Journal of Marine and Freshwater Research*, 35, 75-83.

Milton D. A. & Arthington A. H. (1985) Reproductive strategy and growth of the Australian smelt, *Retropinna semoni* (Weber) (Pisces: Retropinnindae) and the olive perchlet, *Ambassis nigripinnis* (De Vis) (Pisces: Ambassidae), in Brisbane, Southeastern Queensland. *Australian Journal of Marine and Freshwater Research*, 36, 329-341.

Minkley W. L. (Ed.) (1991) *Native fishes of the Grand Canyon region: an obituary? Colorado River Ecology and Dam Management: Proceedings of a symposium May 24-25, 1990 Santa Fe, New Mexico* (National Academy Press: Washington D.C., United States of America).

Mol J. H., Resida D., Ramlal J. S. & Becker C. R. (2000) Effects of El-nino related drought on freshwater and brackish water fishes in Suriname, South America. *Environmental Biology of Fishes*, 59, 429-440.

Munk P. & Kiorboe T. (1985) Feeding behaviour and swimming ability of larval herring (*Clupea harengus*) in realtion to density of copepod nauplii. *Marine Ecology-Progress Series*, 24, 15-21.

Munk P., Kiorboe T. & Christensen V. (1989) Vertical migrations of herring (*Clupea harengus*) larvae in realtion to light and prey distribution. *Environmental Biology of Fishes*, 26, 87-96.

Murphy B. F. & Timbal B. (2008) A review of recent climate variability and climate change in southeastern Australia. *International Journal of Climatology*, 28, 859-879.

Muth R. T. & Schmulbach J. C. (1984) Downstream transport of fish larvae in a shallow prairie river. *Transactions of the American Fisheries Society*, 113, 224-230.

Nabil H., Bailey W. C. & Necibi S. (1999) Beneficial insect attraction to light traps with different wave lengths. *Biological Control*, 16, 185-188.

Neira F. J., Miskieweicz A. G. & Trnski T. (1998) *Larvae of temperate Australian fishes: laboratory guide for larval fish identification* (University of Western Australia Press: Nedlands, WA).

Nelson J. S. (2006) *Fishes of the world* (4th Edn). (John Wiley & Sons, Inc: New Jersey, United States of America).

Nesler T. P., Muth R. T. & Wasowicz A. F. (1988) Evidence for baseline flow spikes as spawning cues for Colorado squawfish in the Yamba River, Colorado. *American Fisheries Society Symposium*, 5, 68-79.

Newman R. (2000) Hydrology of the Lower Lakes and Coorong. In *River Murray Barrages Environmental Flows*. (Eds A. Jensen, M. Good, P. Tucker and M. Long). (Department for Environment, Heritage and Aboriginal Affairs).

Niles J. M. & Hartman K. J. (2007) Comparison of three larval fish gears to sample shallow water sites on a navigable river. *North American Journal of Fisheries Management*, 27, 1126-1138. (doi: 10.1577/m05-207.1).

Nilsson C., Reidy C. A., Dynesius M. & Revenga C. (2005) Fragmentation and flow regulation of the world's large river systems. *Science*, 308, 405-408. (doi: 10.1126/science.1107887).

Oberdorff T., Giulbert E. & Lucchetta J.-C. (1993) Patterns of species richness in the Seine River Basin, France. *Hydrobiologia*, 259.

Ortiz D. M. & Tissot B. N. (2008) Ontogenetic patterns of habitat use by reef-fish in a Marine Protected Area network: a multi-scaled remote sensing and *in situ* approach. *Marine Ecology-Progress Series*, 365, 217-232. (doi: 10.3354/meps07492).

Ostrand K. & Wilde G. R. (2002) Seasonal and spatial variation in a prairie stream-fish assemblage. *Ecology of Freshwater Fish*, 11, 137-149.

Pace M. L., Findlay S. E. G. & Lints D. (1992) Zooplankton in advective environments: the Hudson River community and a comparative analysis. *Canadian Journal of Fisheries and Aquatic Sciences*, 49, 1060-1069.

Penczak T., Agostinho A. A., Gomes L. C. & Latini J. D. (2009) Impacts of a reservoir on fish assemblages of small tributaries of the Corumba River, Brazil. *River Research and Applications*, 25, 1013-1024. (doi: 10.1002/rra.1200).

Penczak T., Glowacki L., Galicka W. & Koszalinski H. (1998) A long-term study (1985-1995) of fish populations in the impounded Warta River, Poland. *Hydrobiologia*, 368, 157-173.

Penczak T. & Kruk A. (2005) Patternizing of impoundment impact (1985-2002) on fish assemblages in a lowland river using the Kohonen algorithm. *Journal of Applied Ichthyology*, 21, 169-177.

Poff N. L., Allan J. D., Bain M. B., Karr J. R., Prestegaard K. L., Richter B. D., Sparks R. E. & Stromberg J. C. (1997) The natural flow regime: a paradigm for river conservation and restoration. *Bioscience*, 47, 769-784.

Poff N. L., Allan J. D., Palmer M. A., Hart D. D., Richter B. D., Arthington A. H., Rogers K. H., Meyers J. L. & Stanford J. A. (2003) Fiver flows and water wars: emerging science for environmental decision making. *Frontiers in Ecology and the Environment*, 1, 298-306.

Porter S. S., Eckert G. L., Byron C. J. & Fisher J. L. (2008) Comparison of light traps and plankton tows for sampling brachyuran crab larvae in an Alaskan Fjord. *Journal of Crustacean Biology*, 28, 175-179.

Potvin C. & Roff D. A. (1993) Distribution free and robust statistical methods: viable alternative to parametric statistics. *Ecology*, 74, 1617-1628.

Pressey R. L. (1986) *Wetlands of the River Murray below Lake Hume*. River Murray Commission, Environmental Report 86/1, Canberra, Australia.

Price A. (2007) *Utilisation of still-water patches by fish and shrimp in a lowland river, with particular emphasis on early-life stages*. PhD thesis, University of Canberra.

Pryor V. K. & Epifanio C. E. (1993) Prey selection by larval weakfish (*Cynoscion regalis*): the effects of prey size, speed and abundance. *Marine Biology*, 116, 31-37.

Puckridge J. T. & Walker K. F. (1990) Reproductive biology and larval development of a gizzard shad *Nematalosa erebi* (Dorosomatinae: Teleostei), in the River Murray, South Australia. *Australian Journal of Marine and Freshwater Research*, 41, 695-712. (56).

Reckendorfer W., Keckeis H., Winkler G. & Schiemer F. (1999) Zooplankton abundance in the River Danube, Austria: the significance of inshore retention. *Freshwater Biology*, 41, 583-591.

Revenga C., Brunner J., Henninger N., Kassem K. & Payne R. (2000) *Pilot analysis of global ecosystems: freshwater systems* (World Resources Institute: Washington DC, United States of America).

Reynolds C. S. (2000) Hydroecology of river plankton: the role of variability in channel flow. *Hydrological Processes*, 14, 3119-3132.

Reynolds L. F. (1983) Migration patterns of five fish species in the Murray-Darling River System. *Australian Journal of Marine and Freshwater Research*, 34, 857-71.

Richter B. D., Baumgartner J. V., Powell J. & Braun D. P. (1996) A method for assessing hydrologic alteration within ecosystems. *Conservation Biology*, 10, 1163-1174.

Richter B. D., Matthews R., Harrison D. L. & Wigington R. (2003) Ecologically sustainable water management: managing river flows for ecological integrity. *Ecological Applications*, 13, 206-224.

Richter B. D., Warner A. T., Meyer J. L. & Lutz K. (2006) A collaborative and adaptive process for developing environmental flow recommendations. *River Research and Applications*, 22, 297-318.

Roberts D. T., Duivenvoorden L. J. & Stuart I. G. (2008) Factors influencing recruitment patterns of Golden Perch (*Macquaria ambigua oriens*) within a hydrologically variable and regulated Australian tropical river system. *Ecology of Freshwater Fish*, 17, 577-589.

Rooker J. R., Dennis G. D. & Goulet D. (1996) Sampling larval fishes with a nightlight lift-net in tropical inshore waters. *Fisheries Research*, 26, 1-15.

Rosenberg D. M., McCully P. & Pringle C. M. (2000) Global-scale environmental effect of hydrological alterations: introduction. *Bioscience*, 50, 746-751.

Rosenberger A. & Angermeier P. L. (2003) Ontogenetic shifts in habitat use by the endangered Roanoke logperch (*Percina rex*). *Freshwater Biology*, 48, 1563–1577.

Rowland S. J. (1983) The hormone-induced ovulation and spawning of the Australian freshwater fish golden perch, *Macquaria ambigua* (Richardson) (Percicithyidae). *Aquaculture*, 35, 221-238.

Rowland S. J. (1992) Diet and feeding of Murray cod (*Maccullochella peelii*) larvae. *Proceeding of the Linnaean Society of New South Wales*, 113, 193-201.

Rowland S. J. (1996) Development of techniques for the large-scale rearing of the larvae of the Australian freshwater fish golden perch, *Macquaria ambigua* (Richardson, 1845). *Marine and Freshwater Research*, 47, 233-242.

Rozas L. P. & Minello T. J. (1997) Estimating densities of small fishes and decapod crustaceans in shallow estuarine habitats: a review of sampling design with focus on gear selection. *Estuaries*, 20, 199-213.

Sabates A. (2004) Diel vertical distribution of fish larvae during the winter-mixing period in the Northwestern Mediterranean. *ICES Journal of Marine Science*, 61, 1243-1252. (doi: 10.1016/j.icesjms.2004.07.022).

Schiemer F., Keckeis H., Winkler G. & Flore L. (2001) Large rivers: the relevance of ecotonal structure and hydrological properties for the fish fauna. *Archiv fur Hydrobiologie* 12, 487-508.

Schiller C. B. & Harris J. H. (2001) Native and alien fish. In *Rivers as ecological systems: The Murray-Darling Basin.* (Ed. W.J. Young). (Murray Darling Basin Commission: Canberra, Australia).

Schindler D. W. (1969) Two useful devices for vertical plankton and water sampling. *Journal of Fisheries Research Board Canada*, 26, 1948-1955.

Schlosser I. J. (1982) Fish community structure and function along two habitat gradients in a headwater stream. *Ecological Monographs*, 52, 395-414.

Schmidt K., Atkinson A., Petzke K. J., Voss M. & Pond D. W. (2006) Protozoans as a food source for Antarctic krill, *Euphausia superba*: complementary insights from stomach content, fatty acids, and stable isotopes. *Limnology and Oceanography*, 51, 2409-2427.

Scott M. T. & Nielsen L. A. (1989) Young fish distribution in backwaters and main-channel borders of the Kanawha River, West Virginia. *Journal of Fish Biology*, 35, 21-27.

Secor D. H., Dean J. M. & Hansbarger J. (1992) Modification of the quatrefoil light trap for use in hatchery ponds. *The Progressive Fish-Culturalist*, 54, 202-205.

Sempeski P. & Gaudin P. (1995) Size-related changes distribution of young grayling (*Thymallus thymallus*). *Canadian Journal of Fisheries and Aquatic Sciences*, 52, 1842-1848.

Serafini L. G. & Humphries P. (2004) *Preliminary guide to the identification of larvae of fish, with a bibliography of their studies, from the Murray-Darling Basin.* Cooperative Research Centre for Freshwater ecology, Murray-Darling Freshwater Research Centre, Albury and Monash University, Clayton, Identification and Ecology Guide No. 48.

Shiel R. J. (1985) Zooplankton of the Darling River system, Australia. *Verhandlungen der Internationalen Vereinigung fur theoretische und angewandte limnologie*, 22, 2136-2140.

Shiel R. J. (1995) *A guide to the identification of rotifers, caldocerans and copepods from Australian Inland Waters.* Cooperative Research Centre for Freshwater Ecology, Albury, Australia.

Shiel R. J., Walker K. F. & Williams W. D. (1982) Plankton of the Lower River Murray, South Australia. *Australian Journal of Marine and Freshwater Research*, 33, 301-327.

Sluss T. D., Cobbs G. A. & Thorp J. H. (2008) Impact of turbulence on riverine zooplankton: a mesocosm experiment. *Freshwater Biology*, 53, 1999-2010. (doi: 10.1111/j.1365-2427.2008.02023.x).

Smith B. B. & Walker K. F. (2004) Spawning dynamics of common carp in the River Murray, South Australia, shown by macroscopic and histological staging of gonads. *Journal of Fish Biology*, 64, 336-54.

Sousa R., G. C & Freitas C. E. d. C. (2008) The influence of flood pulse on fish communtiies of floodplain canals in the Middle Solimoes River, Brazil. *Neotropical Icthyology*, 6, 249-255.

Sparks R. E. (1995) Need for ecosystem management of large rivers and their floodplains. *Bioscience*, 45, 168-182.

Thoms M. C. & Walker K. F. (1993) Channel changes associated with 2 adjacent weirs on a regulated lowland alluvial river. *Regulated Rivers: Research and Management*, 8, 271-284.

Thorp J. H., Black A. R., Haag K. H. & Wehr J. D. (1994) Zooplankton assemblages in the Ohio River: seasonal tributary and navigation dam effects. *Canadian Journal of Fisheries and Aquatic Sciences*, 51, 1634-1643.

Tonkin Z. D., Humphries P. & Pridmore P. A. (2006) Ontogeny of feeding in two native and one alien fish species from the Murray-Darling Basin, Australia. *Environmental Biology of Fishes*, 76, 303-315.

Toth L. A., Melvin S. L., Arrington D. A. & Chamberlin J. (1998) Hydrologic manipulations of the channelized Kissimmee River: implications for restoration. *Bioscience*, 48, 757-764.

Travinchek V. H., Bain M. B. & Maceina M. J. (1995) Recovery of a warm water fish assemblage after initiation of a minimum-flow release downstream from a hydroelectric dam. *Transactions of the American Fisheries Society*, 124, 836-844.

Tredwell S. & Hardwick R. (2003) *Review of habitat associations of native fish in the Murray Darling Basin.* Murray-Darling Basin Commision, Project 2105, Armadale, Australia.

Trippel E. A. & Chambers R. C. (1997) The early life history of fishes and its role in recruitment processes. In *Early life history and recruitment in fish populations.* (Eds R.C. Chambers and E.A. Trippel) pp. 21-32. (Chapman and Hall London, United Kingdom).

Turner T. F., Trexler J. C., Miller G. L. & Toyer K. E. (1994) Temporal and spatial dynamics of larval and juvenile fish abundance in a temperate floodplain river. *Copeia*, 1994, 174-183.

Tyus H. M. (1991) Movements and habitat use of young Colorado squawfish in the Green River, Utah. *Journal of Freshwater Ecology*, 6, 43-51.

Underwood A. J. (1996) Detection, interpretation, prediction and management of environmental disturbances: some roles for experimental marine ecology. *Journal of Experimental Marine Biology and Ecology*, 200, 1-27.

Vannote R. L., Marshall G., Cummins K. W., Sedell J. R. & Cushing C. E. (1980) The river continuum concept. *Canadian Journal of Fisheries and Aquatic Science*, 37, 130-137.

Vilizzi L. (1998) Observations on ontogenetic shifts in the diet of 0+ carp, *Cyprinus carpio*, from the River Murray, Australia. *Folia Zoologica*, 47, 255-229.

Vilizzi L., Meredith S. N., Sharpe C. P. & Rehwinkel R. (2008) Evaluating light trap efficiency by application of mesh to prevent inter- and intra-specific *in situ* predation on fish larvae and juveniles. *Fisheries Research*, 93, 146-153.

Vilizzi L., Wallace T., Fraser P., Ellis I., Conallin A., Meredith S., Engledow K., Sharpe C. & McCasker N. (2007) *Native fish recruitment and flood pulse water quality monitoring on Lindsay Island.* Murray Darling Freshwater Research Centre, Lower Basin Laboratory, Technical Report 07/07, Mildura, Australia.

von Herbing I. H., Gallager S. M. & Halteman W. (2001) Metabolic costs of pursuit and attack in early larval Atlantic cod. *Marine Ecology-Progress Series*, 216, 201-212.

Walker K. F. (1985) A review of the ecological effects of river regulation in Australia. *Hydrobiologia*, 125, 111-129.

Walker K. F. (1992) The River Murray, Australia: a semi-arid lowland river. In *Rivers Handbook*. (Eds P.A. Calow and G.E. Petts) pp. 472-492. (Blackwell Scientific: Oxford, United Kingdom).

Walker K. F. (2006a) Serial weirs cumulative effects: the Lower River Murray, Australia. In *Ecology of desert rivers*. (Ed. R.T. Kingsford) pp. 248-279. (Cambridge University Press: Cambridge).

Walker K. F. (2006b) Serial weirs cumulative effects: the Lower River Murray, Australia. In *Ecology of desert rivers*. (Ed. R.T. Kingsford) pp. 248-279. (Cambridge University Press: Cambridge, United Kingdom).

Walker K. F., Sheldon F. & Puckridge J. T. (1995) An ecological perspective on dryland rivers. *Regulated Rivers: Research and Management*, 11, 85-104.

Walker K. F. & Thoms M. C. (1993) Environmental effects of flow regulation on the Lower River Murray, Australia. *Regulated Rivers: Research and Management*, 8, 103-119.

Warburton K., Retif S. & Hume D. (1998) Generalists as sequential specialists: diets and prey switching in juvenile silver perch. *Environmental Biology of Fishes*, 51, 445-454.

Ward J. V. & Stanford J. A. (1979) Ecological factors controlling stream zoobenthos and emphasis on thermal modification in regulated streams. In *The ecology of regulated streams*.

(Eds J.V. Ward and J.A. Stanford) pp. 33-55. (Plenum Press: New York, United States of America).

Ward J. V., Tockner K., Uehlinger U. & Malard F. (2001) Understanding natural patterns and processes in river corridors as the basis for effective river restoration. *Regulated Rivers: Research and Management*, 17, 311-323.

Watkins M. S., Doherty S. & Copp G. H. (1997) Microhabitat use by 0+ and older fishes in a small English chalk stream. *Journal of Fish Biology*, 50, 1010-1024.

Weaver M. J., Magnuson J. J. & Clayton M. K. (1993) Analyses for differentiating littoral fish assemblages with catch data from multiple sampling gears *Transactions of the American Fisheries Society*, 122, 1111-1119.

Wedderburn S. D., Walker K. F. & Zampatti B. P. (2007) Habitat separation of *Craterocephalus* (Atherinidae) species and populations in off-channel areas of the Lower River Murray, Australia. *Ecology of Freshwater Fish*, 16, 442-449. (doi: 10.1111/j.1600-0633.2007.00243.x).

Welcomme R. L. (1985) *River fisheries*. FAO Fisheries Technical Paper. United Nations, Report no. 262, Rome, Italy.

Werner E. E. & Gilliam J. F. (1984) The ontogenetic niche and species interactions in size structured populations *Annual Review of Ecology and Systematics*, 15, 393-425.

Werner R. G. (2002) Habitat requirements. In *Fishery Science - The Unique Contributions of Early Life Stages*. (Eds L.A. Fuiman and R.G. Werner) pp. 161-182. (Blackwell Science: Oxford, United Kingdom).

White R. J. (1996) Growth and development of North American stream habitat management for fish. *Canadian Journal of Fisheries and Aquatic Science*, 53, 342-363.

Williams W. D. (1980) *Australian freshwater life. The invertebrates of Australian inland waters* (Macmillan: Melbourne, Australia).

Winemiller K. O. (1989a) Onotogenetic diet shifts and resource partitioning among piscivorous fishes in the Venezuelan Llanos. *Environmental Biology of Fishes*, 26, 177-199.

Winemiller K. O. (1989b) Patterns of variation in life-history among South American fishes in seasonal environments. *Oecologia*, 81, 225-241.

Winemiller K. O. (2005) Floodplain river foodwebs: generalizations and implications for fisheries management. In *Proceedings of the second international symposium of the management of large rivers for fisheries*. Phnom Penh, Cambodia. (Eds R.L. Welcomme and T. Petr) pp. 285-312. (Mekong River Commission).

Winemiller K. O. & Rose K. A. (1992) Patterns of life-history diversification in North American fishes: implications for population regulation. *Canadian Journal of Fisheries and Aquatic Science*, 49, 2196-2218.

Winter H. V., Lapinska M. & De Leeuw J. J. (2009) The River Vecht fish community after rehabilitation measures: a comparison to the historical situation by using the River Biebrza as a geographical reference. *River Research and Applications*, 25, 16-28. (doi: 10.1002/rra.1081).

Wright P. J. & Trippel E. A. (2009) Fishery-induced demographic changes in the timing of spawning: consequences for reproductive success. *Fish and Fisheries*, 10, 283-304. (doi: 10.1111/j.1467-2979.2008.00322.x).

Yaron Z. & Sivan B. (2006) Reproduction In *The physiology of Fishes* (3rd Edn). (Eds D.H. Evans and J.B. Claiborne) pp. 343-386. (CRC Press: United States of America).

Ye Q., Cheshire K. J. & Fleer D. (2008) *Recruitment of golden perch and selected large-bodied fish species following the weir pool manipulation in the River Murray, South Australia.* South Australian Research and Development Institute (SARDI), Aquatic Sciences, SARDI research report series number: 303, SARDI Aquatic Sciences Publication Number: F2008/000606-1, Adelaide, Australia.

Young W. J. (Ed.) (2001) *Rivers as ecological systems: The Murray-Darling Basin.* (Murray Darling Basin Commission: Canberra, Australia).

Young W. J., Schiller C. B., Roberts J. & Hillman T. J. (2001) The rivers of the basin and how they work. In *Rivers as ecological systems: The Murray-Darling Basin.* (Ed. W.J. Young). (Murray Darling Basin Commission: Canberra, Australia).

Zampatti B., Leigh S. & Nicol J. (2008) *Chowilla Icon Site- fish assemblage condition monitoring 2005-2008*. South Australian Research and Development Institute (SARDI), Aquatic Sciences, SARDI research report series number: 319, SARDI Aquatic Sciences Publication Number: F2008/000907-1, Adelaide, SA.

Zeug S. C. & Winemiller K. O. (2008) Relationships between hydrology, spatial heterogeneity and fish recruitment dynamics in a temperate floodplain river. *River Research and Applications*, 24, 90-102.

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