Regulated Rivers and Fisheries Restoration Project

- Experimental study of the effects of cold water pollution on native fish -

K.L. Astles, R.K. Winstanley, J.H. Harris and P.C. Gehrke

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Front Cover

Top Photo:	Experimental stream facility
Bottom Photo:	Juvenile silver perch, after being in warm water channels (left) and cold water channels (right) for 6 weeks.

Photos: *Roy Winstanley*
Imaging: *John Matthews*

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SUMMARY

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- Experimental study of the effects of cold water pollution on native fish -

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OBJECTIVES:
(1) To determine the effects of cold water pollution on native fish species downstream of dams.
(2) To provide a sound scientific basis for changes to water release practices from bottom offtake dams.

SUMMARY:

Cold water pollution occurs below large dams that have a bottom valve offtake. Cold water from the bottom of the dam is released into the river downstream, cooling the river water to 10 – 12°C below its natural range. An experimental facility was built on the banks of the Macquarie River just below Burrendong Dam which allowed fish to be exposed to either cold water at river temperatures during releases from the dam, or warm water at ambient temperature from a reservoir below the spillway. Experiments to test the effects of cold water pollution used juveniles of two species of native fish, silver perch and Murray cod. Three experiments were conducted on four aspects of fish biology – growth, survival, re-distribution and activity.

Juvenile silver perch grew substantially more, in both weight and length, in warm water than in cold water. Juveniles in warm water almost doubled in weight over 31 days whilst those in cold water showed little growth. Few silver perch survived in the cold channels, with numbers declining rapidly within the first 16 days of the experiment. In thermal gradient experiments offering a choice of warm and cold water, silver perch exhibited a strong preference for warm water. Furthermore, silver perch tended to be more active in warmer water than cold but this experiment was not conclusive.

Juvenile Murray cod showed no major difference in growth between warm and cold channels. However, this result may have been caused by the fish not feeding properly during the experiment and the onset of disease. Survival was high in both warm and cold channels. Murray cod showed a similar response to silver perch in thermal gradients, exhibiting a strong preference for warm water.

These experiments demonstrated that juvenile native fish were affected by cold water pollution in the areas of growth, survival, distribution and activity. These effects have serious consequences if cold water pollution is widespread in inland rivers. Consequences include reduced recruitment to adult populations, forced re-distribution of fish populations to less favourable habitats and general declines in fish populations because of decreased survival.

The effects of cold water pollution vary depending on the species. Murray cod and silver perch showed some differences in their responses (e.g. survival). Therefore, some species may be more detrimentally affected by cold water pollution than others. Further experiments should therefore be conducted on other native species (such as catfish and smaller fish like gudgeons) to obtain a better understanding of the range of responses by fish to cold water pollution. Such understanding will enable better management plans to be implemented.

Fish communities were sampled downstream of Burrendong Dam on the Macquarie River and nearby Bogan River, which is not affected by thermal pollution. Samples were collected during the irrigation season, when cold water released from Burrendong Dam chills the Macquarie River downstream, and during the non-irrigation season, when both rivers follow natural seasonal temperatures. Water temperature was measured for analysis with fish samples. Only small numbers of fish were caught from both rivers except for one location on the Bogan River. Abundance of fish did not change between non-irrigation and irrigation seasons. Water temperatures increased gradually with increasing distance downstream in the Macquarie River during the irrigation season but there was no corresponding increase in fish abundance. However, the proportion of native fish in the Macquarie River during the irrigation season gradually increased with increasing temperature downstream. Fish communities in the Macquarie River were different from those in the Bogan River. These differences maybe the result of the different thermal and flow regimes between the two rivers. Furthermore, there is strong evidence that the fish community in the Macquarie River is seriously degraded, and is affected by multiple disturbances that provide little opportunity for surviving species to recover during brief periods of warmer water temperatures.

The results of this study provide valuable insights on the effects of cold water pollution on juvenile native fish. However, a number of important knowledge gaps remain to be investigated. For example, it is possible that slow growth in juvenile fish retards their maturation and subsequently reduces recruitment to breeding populations. Reproductive cycles and strategies may also be altered. The existence and value of thermal refugia is not known. If such habitats exist, they may provide opportunities for fish to compensate for retarded growth, maturation and reproduction. The direct effects of thermal pollution on recruitment rates and other aspects of population dynamics are yet to be demonstrated in wild populations. A better understanding of these aspects is needed to protect critical processes and life history stages from adverse effects of cold water releases.

RECOMMENDATIONS:

1. Acknowledge that cold water pollution is a threatening process affecting the conservation status of silver perch.
2. Modification of the outlet works of Burrendong Dam is necessary to raise the temperature of water in the river downstream of the dam during irrigation releases. Consideration should also be given to modifying other dams where cold water pollution has been demonstrated.
3. Acknowledge there are several knowledge gaps in our understanding and management of the effects of cold water pollution on the aquatic environment in inland rivers in NSW. Further studies should be conducted in the following areas:
    - impacts of cold water pollution on other native fish species.
    - the interrelationships between fish, water temperature and aquatic habitats in the Macquarie and Bogan rivers at a range of spatial and temporal scales to identify the scales at which temperature effects occur. This will help determine whether cold water pollution has a blanket infiltration throughout the river, or if there are regions of warmer water that are used by fish as thermal refuges.
    - identify other river systems where cold water pollution might be great and where similar experiments done at Burrendong dam could be repeated.
    - existence of warm tributaries that may provide thermal refuges in rivers affected by cold water pollution, and the potential of such habitats for assisting rehabilitation of affected rivers.
    - effects of cold water on macroinvertebrates, algae and other trophic levels to provide a broader understanding of temperature requirements to maintain river health and to support fish communities.

1. INTRODUCTION

1.1. River alterations and water temperature

Almost all the rivers in inland Australia have been modified by human development. The most common alteration has been the impoundment of these rivers by major dams. Large dams may serve several purposes, such as supplying water for irrigation, industrial or domestic use, flood control, generation of hydroelectricity and human recreation (Gehrke *et al.,* 1999). Impoundments have substantially changed the natural flow regimes of the rivers including their seasonal flow cycles, variability of flows and water quality. For example, on the Murray River the natural seasonal cycle of low flows in winter and high flows in summer has been completely reversed in order to supply water to irrigators and townships (Close, 1990). These changes have inevitably affected the biota of rivers. Recent studies have shown there has been a dramatic decline in native fish populations throughout the rivers in New South Wales (Harris and Gehrke, 1997; Gehrke and Harris, 2000). Numerous factors have contributed to the decline of native fish, and many are linked to the construction and operation of dams, and subsequent changes in flow regimes, water quality and habitat degradation.

One aspect of water quality that has received little attention in Australia until recently is the change in water temperature of rivers below dams, even though the problem was recognised over twenty years ago (Tunbridge, 1978; Walker *et al.*, 1978). The water temperature of lowland rivers usually follows the seasonal cycle of air temperatures. However, releasing water from most Australian dams can result in dramatic changes in seasonal water temperatures (Walker, 1980; Koehn *et al.,* 1996, Harris, 1997). Water in dams often stratifies during summer because of temperature-density differences (Petts, 1984). As water heats up it becomes less dense so that it expands and forms a warmer layer in the upper 3 – 10m of the dam. This is known as the epilimnion layer. Underneath, at the bottom of the dam the water is at a lower temperature and more dense. It therefore stays below the top layer of water. This bottom layer is known as the hypolimnion. The hypolimnion is often 12-15°C below the surface temperature. Water at the bottom of the dam is therefore significantly colder than the natural river temperature before the dam was built. Most of the major dams in New South Wales have off-take valves near the bottom of the dam wall, and can only release the cold hypolimnion layer of about 11-12°C, chilling the river downstream that would normally be about 21-25°C. This clearly violates the Australian and New Zealand guidelines for temperature decreases in freshwater of less than 2°C per hour (ANZECC, 1992). This temperature depression (hereafter called thermal pollution) can last for several hundreds of kilometres downstream in rivers such as the Macquarie and Murrumbidgee (Harris, 1997).

Large unseasonal changes in water temperature have the potential to disrupt biological processes such as aquatic respiration, invertebrate productivity, migration, growth and reproduction of fish. Little experimental work has been done to examine the effects of temperature changes on native fish (Gehrke 1988a). Other studies have made inferences on the significance of temperature suppression based on the known biology of some fish (Harris, 1997, Koehn *et al.* 1996, Doudoroff, 1957). However, before decisions can be made about the need to modify dam outlets or release procedures to ameliorate coldwater pollution, it is necessary to provide evidence of its environmental impacts.

1.2. Water temperature and native freshwater fish

Temperature is considered the most important factor in the development and growth of fish (Doudoroff, 1957; Crawshaw, 1977; Hoar, *et al.*; 1979; Jobling, 1981; Hoar and Randall, 1988;

Wooton, 1990; Rowland, 1998a,b). Metabolism, respiration, feeding, reproduction, larval development and migratory behaviour of native fish are all strongly influenced by temperature (Gehrke 1988a; Gehrke and Fielder, 1988; Mallen-Cooper, *et al.*, 1995; Rowland, 1998a). For example, spangled perch have been observed to cease feeding when temperatures fall below 16°C, and lose weight until water temperature increases above 16°C again (Gehrke, 1988a). Murray cod in the Wakool River have been known to spawn within 10 days of the river water reaching 20°C (Rowland, 1998b). Golden perch and silver perch require a rise in water level and warm water temperatures in spring and summer to induce spawning. However, if suitable conditions do not occur the gonads undergo recrudescence (Lake, 1967; Rowland,1995). Growth is also an important temperature dependent process in fish. Silver perch, for example, grow more rapidly when kept at high temperatures for aquaculture purposes (Shuenn-Der *et al.*, 1995). In wild populations, golden perch and silver perch in warmer, more northerly habitats grow faster than in more southerly regions (Battaglene, 1987;, Mallen-Cooper *et al.* 1995). Fish eggs, larvae and juveniles are critical growth stages that can also be strongly temperature dependent. For example, Murray cod eggs will hatch over a range of different temperatures but the rate of hatching varies depending upon temperature and conditions. Koehn and O'Connor (1990) report from several studies that eggs will hatch in 13 days at 16.5°C and in 8 to 9 days at 20°C. Golden perch hatch in 24 hours at 27 – 31°C but can take up to 50 hours in temperatures below this range.

Therefore, many aspects of the biology of native fish are tuned to warm river temperatures. The decline in native fish populations over recent decades (Harris and Gehrke, 1997; Reid *et al.*, 1997) and the increasing shifts in flow regimes away from their natural state mean that there is potentially a greater threat to the survival of fish communities. Thermal pollution is only one aspect of the changes human activities have caused in rivers. Yet little work has been done on the lower temperature requirements of native fish. This experimental study was initiated to address some of the deficiencies in understanding of the ecological impacts of cold water pollution by conducting manipulative experiments with native fish in artificial streams.

1.3. Use of artificial channels

Artificial streams consist of a constructed channel with a controlled flow of water. Artificial streams have been used in a wide variety of contexts in experiments on fish and other organisms (see Lamberti and Steinman, 1993). There is a broad range of artificial streams that vary in their capacity to mimic natural systems versus their ability to provide a more controlled environment (Lamberti and Steinman, 1993; Palmer *et al.*, 1994). Outdoor channels tend to have certain elements under control (e.g. flow rate) whilst leaving others to vary naturally (e.g. light and temperature). Channels can be located beside natural rivers and draw their water and organisms directly from the river. Indoor channels, on the other hand, can be artificially illuminated or use natural light in a green house. When indoors virtually all aspects of the environment are under the control of the experimenter. Flow circulation for both indoor and outdoor artificial streams may use either flow through or recycling systems.

Because of their ability to be manipulated, artificial streams have been used with increasing frequency in the last 30 years to study organisms and processes in aquatic science. Organisms that have been examined include algae, micro- and macroinvertebrates and fish. Aspects of riverine ecosystems studied include disturbance, animal behaviour, life history, productivity, energetics, physical and chemical properties (see papers in Lambert and Steinman's review, 1993). Numerous aspects of the ecology of fish have been examined using artificial streams (Gelwick and Matthews, 1993). However, careful attention to channel design is needed to meet the requirements of fish, because they are sometimes large and usually active aquatic animals. Gelwick and Matthews (1993) discuss these issues extensively and conclude that the better the channels mimic a whole stream reach, including pool/riffle combinations, instream habitats and flow velocities, the more likely mechanisms and observed effects are to be applicable to the real world.

In unraveling complex ecological questions, artificial streams offer both advantages and disadvantages. McIntyre (1993) pointed out that the major advantage of using artificial streams is that they allow the explicit testing of hypotheses generated from ecological theory by using replicated treatments and controls that is not possible in natural systems.

However, there are also some limitations associated with using artificial streams. The most important limitation concerns the location of the facility away from the natural river environment and the corresponding increase in control of the experimenter. The more the artificial stream environment differs from natural conditions, the more difficult it is to relate results to real-world environments. McIntyre (1993) warns this may lead experimenters to yield trivial or meaningless results. For studies on fish ecology and biology one major limitation is the size the channels can be. Because of constraints of finance and space, artificial streams can sometimes be too small relative to the size of the fish and their natural habitat (Gelwick and Matthews, 1993) limiting the questions that can be adequately addressed and the stage of the fish's life cycle that can be examined (e.g. larvae, juvenile, adult).

Most Australian studies using artificial streams have investigated the effects of pesticides on riverine invertebrates (Pusey *et al*., 1994; Ward *et al*., 1995;). However Mallen-Cooper (1992, 1994) used indoor artificial streams in the form of vertical slot fishways to examine the swimming ability of three native freshwater fish. The present study is the first to use artificial streams to examine aspects of fish behaviour with respect to water quality.

A major consideration in designing artificial streams for this study was to mimic the fish's natural environment as closely as possible. A realistic experimental environment maximises the ability to relate results to the real world, and increases the reliability of conclusions and recommendations about the impact of cold water pollution on native fish. To this end the experimental facility was situated outdoors beside a river affected by cold water pollution, using water from the river to supply a non-recirculating flow-through system of six replicate artificial streams or channels. The channels were made as long as site logistics and budget would allow (see Section 2.1). Natural substrata and instream habitats were provided to mimic the normal environment of the fish. Juvenile native fish were chosen, rather than adults, to minimise confounding effects caused by confining adult fish in small habitats.

1.4. Aims of project

This project had two purposes. The first was to determine whether juvenile native fish are affected by cold water pollution. The second purpose was to document changes in water temperatures and fish communities associated with the irrigation season downstream of Burrendong Dam on the Macquarie and Bogan Rivers. The following hypotheses were tested:

a) Fish growth and survival in cold water is significantly less than in warm water;
b) Fish display a significant preference for warm water when exposed to a thermal gradient; and
c) Fish activity is significantly different between cold water and warm water treatments.

These hypotheses were tested using experiments in artificial streams on juvenile stages of two native fish species – silver perch (*Bidyanus bidyanus*, Terapontidae) and Murray cod (*Maccullochella peelii*, Percichthyidae). These two species were chosen because they have shown a steady decline in natural populations in most inland rivers of New South Wales (Harris and Gehrke, 1997; Reid *et al*., 1997). In particular, silver perch have been declared as a threatened species in New South Wales, in the vulnerable category, with cold water pollution listed as one of the contributing factors. Furthermore, both these species are popular targets for recreational anglers.

2. GENERAL METHODS

2.1. Facility design and construction

2.1.1. Study site

The artificial stream facility was located at Burrendong Dam (32°40’ 149°09’) 25 km south east of Wellington, in the central-west of New South Wales (Fig. 2.1). Built in 1963 on the Macquarie River, Burrendong Dam has a storage capacity of 1,188,000 Ml and a catchment area of 13,900 km^2. Water release from the dam is controlled via a bell tower, which feeds three large outlet valves situated at the base of the main wall. The bell tower design can only access water from the bottom of the dam, where water temperatures can be less than 12°C in mid summer. Cold water released through the valves flows directly into the Macquarie River channel. The dam spillway is located 3 km downstream of the main valve block, situated in a bedrock cutting. The spillway design incorporates a concrete chute with rotating drum gates, and has a discharge capacity of 1,199,000 Ml d^{-1}. The spillway can only operate if water storage is greater than 100% of capacity. At the base of the spillway chute a causeway retains a large pond just 100 m away from the Macquarie River channel. Water from the spillway enters the river via a shallow channel below the causeway. The experimental facility was situated at the junction of this channel and Macquarie River (Fig. 2.1). The site was on a gentle slope under an established stand of *Eucalyptus* trees, which provided shade to the facility. The land is controlled by the Department of Land and Water Conservation and has restricted public access.

2.1.2. Description of experimental facility

The facility is comprised of six artificial streams fed with water of different temperatures, from the same water source (Fig. 2.2). The facility operated on a site with no electric power or mains water.

The design provides for:

- pumping water from the spillway pond to a header tank at 60 m^3 h^{-1};
- passive cooling of pond water to 10°C below ambient (=warm) temperature, using a heat exchanger situated in the river channel;
- supplying the artificial streams with ambient and cold water; and
- returning overflow water to the spillway pond.

Specifications of major components in the facility and maintenance schedules are described below (Fig. 2.3)

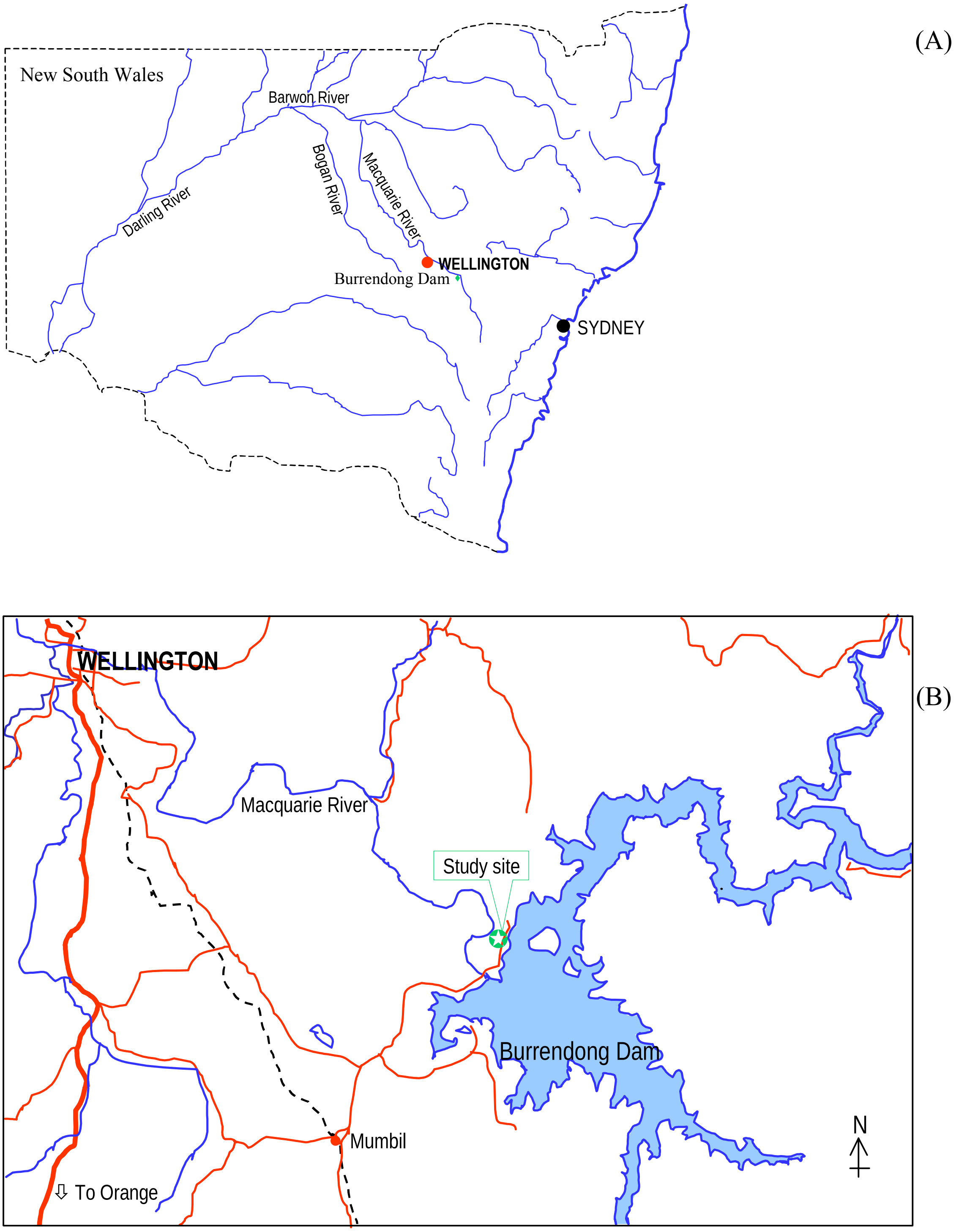

Figure 2.1. The location of (A) Burrendong Dam and (B) the experimental stream facility built on the banks of the Macquarie River below the dam. – – roads, – – rivers or water bodies, – – – railway line.

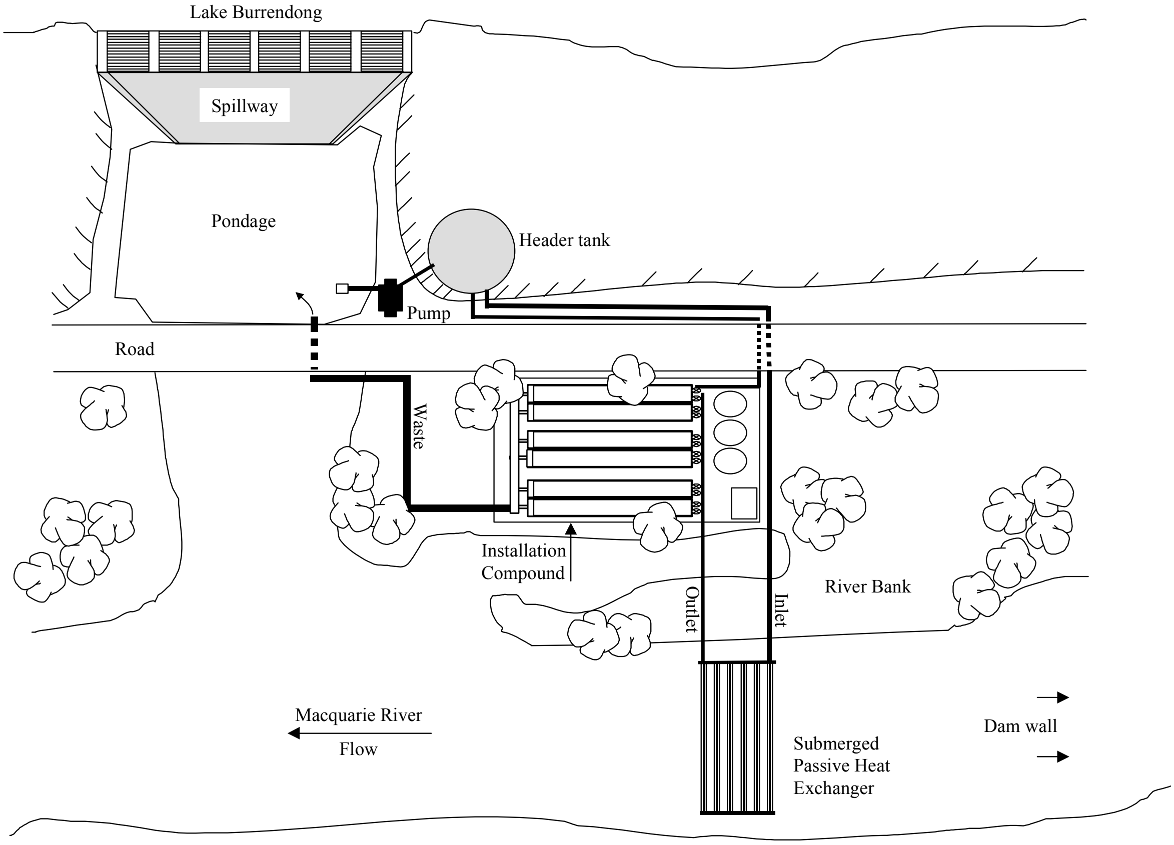

Figure 2.2. Plan view of experimental stream installation (not to scale).

2.1.3. Construction of experimental channels

2.1.3.1. Off-site preparation

Artificial streams were constructed from galvanised steel ducting tube (gauge 0.8 mm). Three tubes (5 m x 0.8 m diameter) were split along their length to make six half troughs. Steel end caps (gauge 1 mm) were welded onto each trough and all welds painted to inhibit corrosion. A weir was created by fitting a false steel wall 40 mm lower and 150 mm from one end. The weirs maintained water depth in the channels at 360 mm, and let water spill to waste. The volume of each channel was approximately 1,662 litres. Three holes were drilled in each channel to take PVC pipe fittings. Hole sizes were 63 mm and 50 mm inlets and 150 mm outlet. To avoid water contamination by zinc leached from the galvanised steel (5 mg l^{-1} when new, declining to 1 mg l^{-1} BHP Coated Steel Australia) the inside of the channels were painted green with Dulux-Amerlock 400®, an epoxy paint system.

2.1.3.2. On-site installation

A front-end loader was used to level a 12 m^2 area of ground on the sloping site. A frame was constructed from treated pine landscape sleepers (3 m x 750 mm x 150 mm) to support channel sections. Sleepers were fixed at the corners with butt joints with fencing wire through pre-drilled holes. Steel fencing star-posts were cut into 1 m lengths and driven in against the frame as support, preventing wall movement and buckling under load. The frame design consisted of 3 large boxes (2 m x 6 m). Each box supported two channel sections, and boxes were separated by 1 m wide walkways.

The channels were bedded on coarse river sand (depth 100 mm) with the inlet end raised 20-30 mm higher to promote water flow. Channels were held in position by straps made of fencing wire and tightened with turnbuckles. The channel boxes were then back filled with river sand to stop channel distortion under the weight of the water.

Channel inlets were fitted with two ball valves (63 mm) to control either ambient (warm) or cooled water through a single inlet nozzle. A second inlet (50 mm) was situated in the channel base at the mid-point. This inlet allowed ambient water to be sprayed uniformly within the channel, directed towards the outlet end. Water overflowing the weir at the end of each channel was returned to the spillway pond via a PVC (150 mm) stormwater pipe. A guard fence made of plastic mesh was fitted on top of each weir to stop fish entering the drain. The flow rate for all experiments was kept constant at an average of 0.167 $l\ s^{-1}$. Each channel was given a substratum of washed river gravel along with six artificial "habitats" made from 90 mm PVC pipe (120 mm long). These were placed along the channel equidistant from each other. For temperature gradient studies a removable baffle was fitted upstream of the mid-channel inlet. This promoted the formation of a thermal gradient along the tank. Semicircular baffles were made from 5 mm aluminum plate (0.4 m radius) with a hole cut in the base, to allow fish passage between cooled and ambient channel sections.

2.1.3.3. Pump unit

The facility specifications required continuous pump operation for up to six weeks with daily air temperatures exceeding 30°C. A Lister (TR1) air-cooled diesel engine coupled to an Indeng Monoflo pump was used to pump water from the spillway pondage to the header tank. The pump had a capacity of 66 m^3/hr with a 24 m head and 6 m suction lift. The pump intake pipe (90 mm by 15 m) along with a foot valve and Y strainer was moored in a deep pool. The unit was fastened on a concrete slab (200 mm thick) next to the spillway pondage. Because the pump was running almost continually for six weeks, regular maintenance was necessary which included changing engine oil and oil, air and fuel filters.

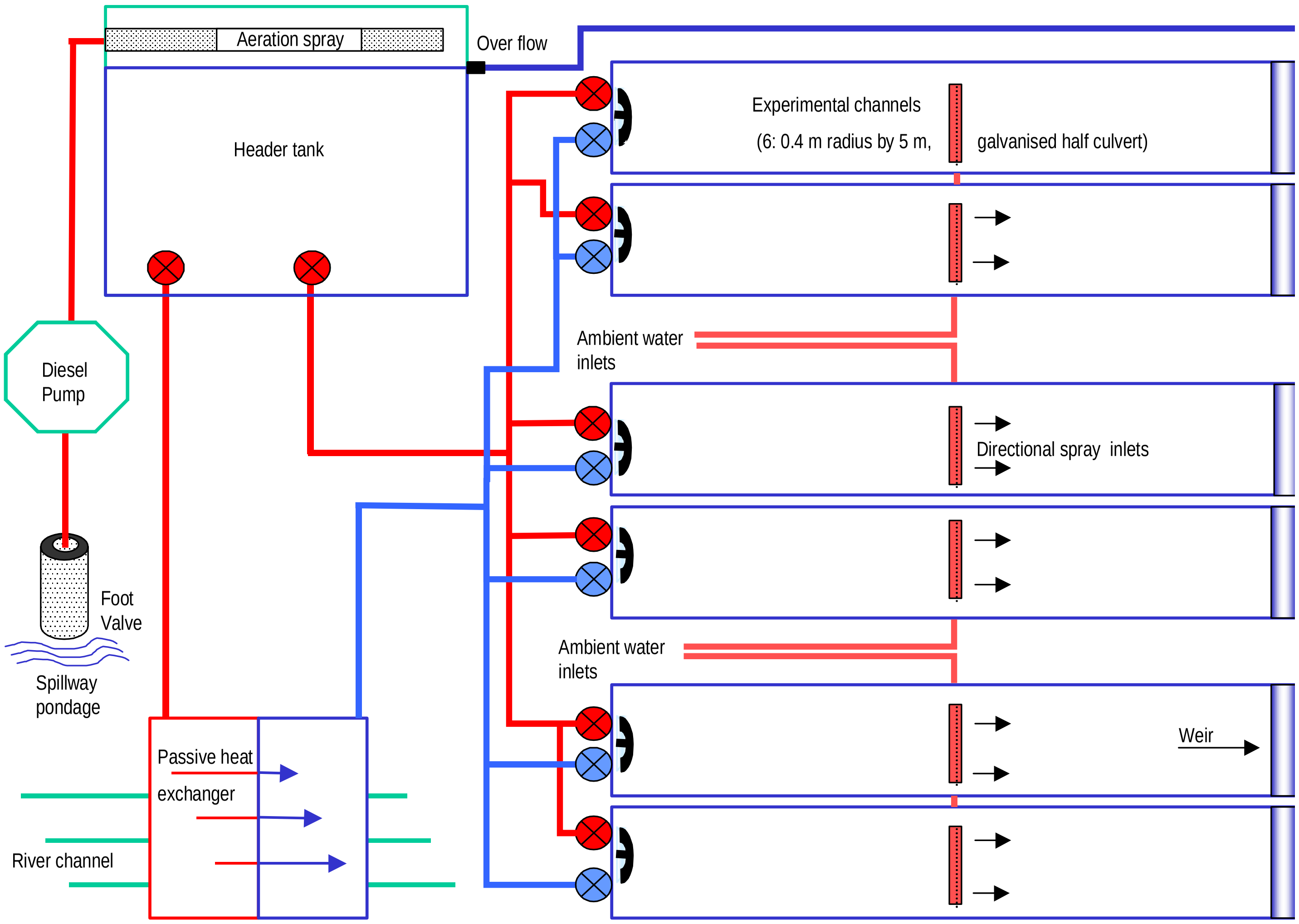

Figure 2.3. Schematic diagram of experimental stream facility at Burrendong Dam, New South Wales.

2.1.3.4. Water storage

Water at ambient temperature was pumped (63 mm PN6.3 pipe) to a header tank installed 21 m above the spillway pond and 17 m above the experimental channels. A polyethylene tank with a capacity of 1,000 l (1.8 m diameter by 2 m high) was embedded on a 200 mm base of course river sand. The river sand enabled the tank to be easily levelled and protected the plastic base from sharp objects. The inlet pipe was fitted through the standard large entrance in the top of the tank. The pipe was fixed to allow water to be sprayed into the tank, thus promoting aeration of stored water. Water level was maintained by an overflow pipe (90 mm PVC) fitted 100 mm below the tank rim. Water that overflowed was directed back to the spillway pond. Two brass gate valves 90 mm and 110 mm were fitted in the base of the tank.

2.1.3.5. Passive heat exchangers

To cool water from the spillway pond (which was at ambient temperature) to within 2°C of the temperature of water released from the dam, outlet valves required four passive heat exchangers. Each heat exchanger consisted of twelve 6.5 m lengths of 50 mm diameter medium black steel-screwed pipe (AS1074 + AS1163) (Fig. 2.4). Threaded elbows joined them in a concertina pattern. All threaded joints were sealed using heavy-duty pink plumbers tape. Because of the weight of the steel tubing (5.44 kg m^{-1}) each heat exchanger was made in modules to allow for ease of transport and assembly on the riverbank. Each module consisted of two pipe lengths and four elbow fittings. Quick-fit tapered flange couplings joined the six modules. Heat exchangers were assembled in pairs and mounted on treated pine logs to form a sled. Steel inlet and outlet pipes (2.5 m long) were fitted vertically to allow HDPE pressure pipe to be attached once heat exchangers were installed in the river. Each sled weighed approximately 1 tonne, and was pulled into the river using an electric powered winch. Heat exchanger inlets were fitted with ball valves (63 mm) grouped in a manifold at the river's edge. The ball valves controlled the number of heat exchangers on line and allowed variable flow rates. A 110 mm (PN8) diameter pipe connected the header tank to the inlet manifold, and a 90-mm (PN8) diameter pipe connected the outlet manifold to the channels.

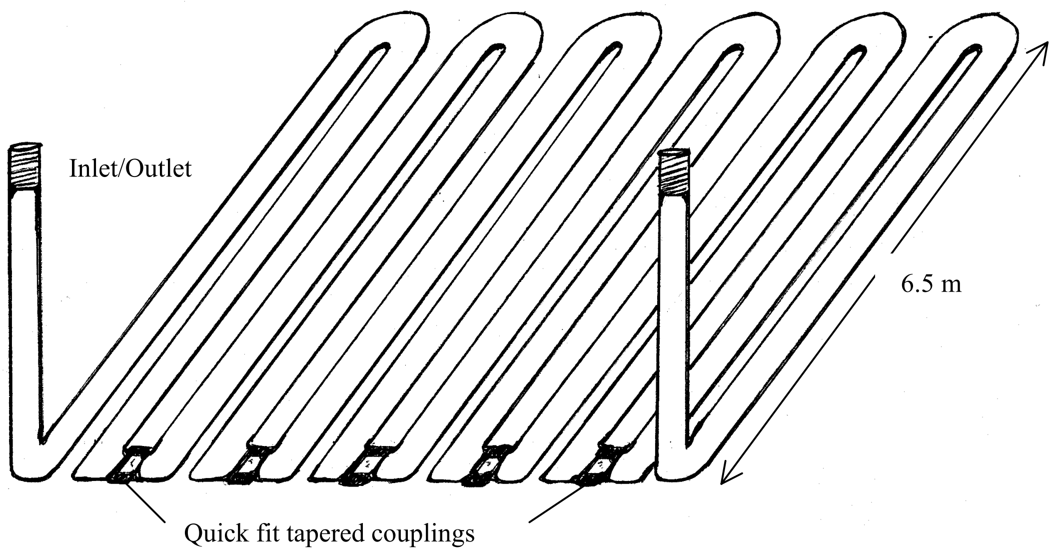

Figure 2.4. One of four passive heat exchanger units used to cool warm water from the spillway pond.

2.1.4. Holding tanks for fish

Three fibreglass open tanks (1.8 m diameter by 1 m high) with a 2,000 l capacity were used to keep fish on-site. The tanks were settled on coarse river sand retained in a frame made of treated pine sleepers (6 m x 2 m x 750 mm), and braced by star pickets (0.5 m long). Inlet water under pressure from the header tank was sprayed into the holding tanks to aerate the water. Flow rate in the tanks was controlled by 25 mm ball valves. A backup oxygen supply in case of engine or pump failure consisted of regulated bottled oxygen supplied to each tank, through aquarium bricks. The system was switched on automatically via a pressure sensitive valve (138 kPa shut-off) on the main water inlet line.

A standpipe incorporating an elbow and press barrel fitting was used to maintain the water level in each tank. The standpipe allowed continuous flow through the tank by spilling water via a flexible hose to the stormwater drain. By swivelling the standpipe through 90° the tank water level could be lowered. Holding tank covers were made from green shade cloth (70% UV reduction) held in position by 6 mm shock cord. The shade cloth minimised fish disturbance and prevented predation by birds.

2.2. Fish maintenance

Juvenile silver perch and Murray cod were obtained from commercial hatcheries. Fish were transported to the study site in cooled saline water and stored in separate holding tanks at 20-25^{o}C until needed for experiments. Silver perch were fed daily (0.05 g per gram body mass) with commercial fish food pellets. Murray cod were fed daily on tubifex worms. Fish kept in the holding tanks were given salt or formalin baths to control fungus and parasite loads. Salt baths were done weekly and formalin baths every third week. This was done by lowering the water level to approximately 500 l for each treatment and adding either dissolved commercial pool salt (1 kg of salt 1000 l^{-1}) for the salt baths or formalin (20 ml of formalin 100 l^{-1}) for formalin baths to each tank. The fish were kept in this solution for 60-80 minutes (salt bath) or 60 minutes (formalin bath) before the tank was refilled. Bottled oxygen was bubbled through the tanks during these treatments as inlet valves were closed to maintain reduced water levels.

2.2.1. Extraction of fish from holding tanks for experiments

When selecting a sample of fish from a large batch for an experiment, care must be taken not to introduce a bias into the sample set (Underwood, 1997). Because fish are highly mobile animals it is possible to only select fish that are the most or least aggressive or the slowest swimmers, depending on the method of capture. Selection bias may be unwittingly reflected in the behaviour, growth or survival of fish in experimental treatments, confounding the results.

To avoid selection bias, the water level in the holding tank was dropped to about 10 cm and 1-2 ml of clove oil was added to mildly sedate the fish and reduce handling stress. The correct quantity of clove oil to sedate juvenile fish was difficult to judge, so a conservative dose was used. After waiting 10 minutes or so for the clove oil to take effect, two shallow rectangular mesh nets were lowered into the tank and rested on the bottom. As fish swam over the nets they were carefully pulled up catching the fish in the nets. The fish were then quickly transferred to a covered bucket. This routine was repeated until sufficient fish had been collected for an experiment. The fish were then placed into each channel randomly using a small hand dip net.

3. GROWTH AND SURVIVAL EXPERIMENTS WITH NATIVE FISH

3.1. Introduction

Riverine water temperatures exhibit a seasonal cycle that follows ambient air temperature, without reaching the same extremes. Much of the biology of fish, particularly breeding and development, is synchronised with this natural cycle (Koehn and O'Connor, 1990). Complex, interacting cycles of water temperature, day-length and flow may be involved in controlling biological cycles, so that de-coupling any one of these environmental controls may disrupt fish physiology (Schiller and Harris, 2000). If colder water temperatures occur out of phase with natural cycles there may be major impacts on the growth, development, migration and survival of native fish.

In the western drainage rivers of New South Wales depression of water temperatures is widespread. Cold water is released from dams during the irrigation season (approximately December to February), which coincides with breeding seasons for many native species and rapid growth periods for juvenile fish (McDowall, 1996; Koehn and O'Connor, 1990). The effect of cold water on the juvenile stages of native fish in the wild are not well understood.

This study examined the immediate effects of cold water pollution on juvenile native fish by comparing growth and survival of fish in cold water (mimicking cold water released from Burrendong Dam) and in warm water (mimicking natural ambient river temperature).

3.2. Methods

3.2.1. Experimental design

Juvenile silver perch (50-65mm total length) and juvenile Murray cod (100-150mm total length) were used for these experiments. Each species was tested at different times (silver perch – 21 January to 24 February 1999; Murray cod – 15 November to 7 December 1999). Fish were transferred from the holding tanks to the channels as described in the general methods. To avoid non-independence of data (see Underwood, 1997) the fish were divided into two subsets. At the start of the experiment, one subset of fish was measured and weighed and returned to a different holding tank whilst the other subset was put in the channels for the experiment without being weighed or measured. The sequence of fish which were measured and those which were not, was randomised in groups of 20 for silver perch and in groups of 15 for Murray cod. For example, the first group of 20 fish may have been measured but the next two groups of 20 were not measured, and so on until there was an equal number of measured and unmeasured fish. The total length and wet weight of each fish was recorded. Treatments of ambient (hereafter called "warm") or cold water were allocated randomly to the channels, with three channels per treatment.

Twenty silver perch were placed in each channel and fed the same quantity of dried food pellets (50g) every day for 31 days. During this period a daily record was kept of any dead fish found in the channels. Dead fish were removed, measured but were not replaced. In addition, qualitative observations of fish behaviour were recorded at the same time each day.

Fifteen Murray cod were placed in each channel and fed the same quantity of tubifex worms each day (10g) for 21 days. Murray cod were slower to start feeding again after they had been transported from Port Stephens to the study site. Different types of food (e.g. live worms, dry pellets) were presented to them in different ways (e.g. deposited near instream habitats, in trays at dusk) in the first week but the fish did not commence feeding until the second week in the channels. Experiments with Murray cod ran for a shorter period (21 days) than the silver perch. The same daily observations and measurements of dead fish were done as for silver perch but no qualitative observations of behaviour were recorded because Murray cod were less active.

Temperature data loggers were set up in each channel, the spillway pond and at the heat exchangers in the river to record hourly water temperature for the duration of the experiments. Unfortunately, during experiments with silver perch only two data loggers in the channels worked, one in a warm channel and one in a cold channel. It was therefore not possible to compare the consistency of temperatures among channels within treatments. All data loggers in all channels functioned properly during experiments with Murray cod.

3.2.2. Statistical analyses

The null hypothesis of no difference in the size of fish between warm and cold water was tested using a two-factor analysis of variance with factors of time period (start/end) and temperature (warm/cold). The deaths of silver perch in the cold channels resulted in an unequal number of fish in this treatment. In order to obtain a balanced design channels within treatments were tested for significant differences. No significant differences were found, so individual channels within treatments were pooled. From the pooled data individual replicates (n=19) within in each treatment (warm and cold) were chosen randomly for the analysis. Student Newman Keul's test was used as a multiple comparison to distinguish among means of significant factors. No statistical analyses were done on the survival data, as patterns in the data were clearly evident.

3.3. Results

3.3.1. Water temperatures during growth experiments

Silver perch: Mean daily maximum and minimum temperatures for the spillway pond, the Macquarie River and the experimental channels are summarised in Table 3.1a. The average temperature difference between the ambient and cooled water channels was approximately 11°C during the experiment. Diel variation in water temperature was greater in the spillway pond and warm channels than in the Macquarie River or cooled water channels (Fig 3.1). The temperature of water drawn from the spillway pond did not increase measurably in the black PVC pipes carrying water to the experimental channels. The mean water temperature of the warm channel was actually cooler than the spillway pond.

Table 3.1. Mean daily maximum and minimum water temperatures (± S.E) logged during growth experiments for a) silver perch (21 Jan - 24 Feb 1999; *N* = 33) and b) Murray cod (15 Nov - 7 Dec 1999; *N* = 21) in the Macquarie River, spillway pond and experimental stream channels.

Location	**Designation**	**Daily maximum water temperature °C**	**Daily minimum water temperature °C**
a) Silver perch			
Macquarie River	Cold: valve release water	13.28 ± 0.04	12.47 ± 0.04
Spillway pondage	Warm: spilled water	28.05 ± 0.24	25.25 ± 0.56
Channel 3	Warm	26.46 ± 0.18	24.16 ± 0.15
Channel 6	Cooled	14.93 ± 0.11	13.31 ± 0.06
b) Murray cod			
Macquarie River	Cold: valve release water	13.54 ± 0.01	12.83 ± 0.01
Spillway pondage	Warm: spilled water	21.50 ± 0.11	20.24 ± 0.08
Channel 1	Warm	21.62 ± 0.08	19.10 ± 0.08
Channel 2	Cooled	15.01 ± 0.03	13.24 ± 0.03
Channel 3	Cooled	15.13 ± 0.03	13.26 ± 0.03
Channel 4	Warm	21.51 ± 0.10	18.77 ± 0.09
Channel 5	Cooled	15.33 ± 0.07	13.03 ± 0.03
Channel 6	Warm	21.95 ± 0.10	19.23 ± 0.09

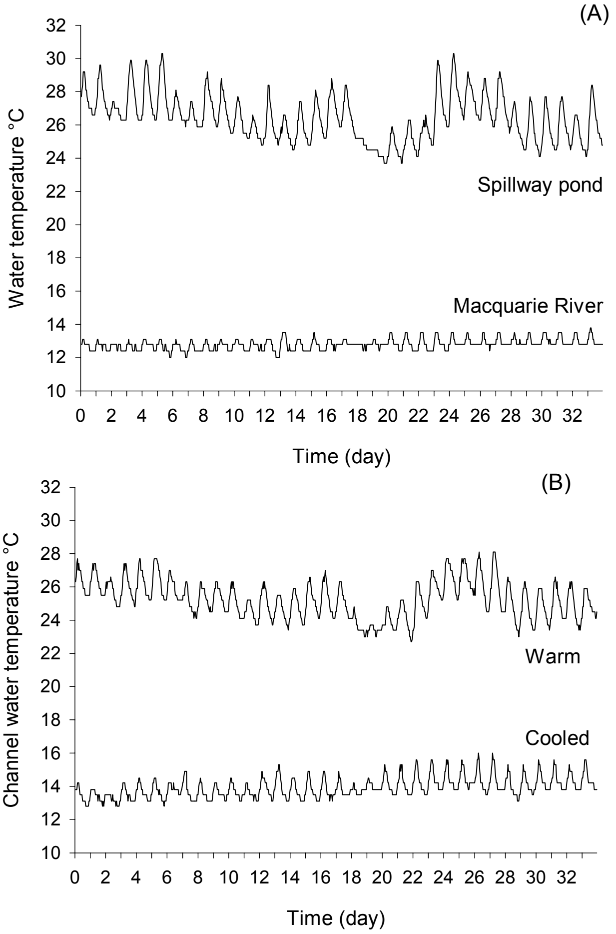

Figure 3.1. Water temperatures logged hourly during the silver perch growth experiment (21 January to 24 February 1999), for (A) spillway pond and Macquarie River, and (B) warm and passively cooled experimental channels.

Murray Cod: The mean daily maximum and minimum temperatures for the spillway pond the Macquarie River and the experimental channels are summarised in Table 3.1b. Variation in the spillway pond water temperature was minimal until the last three days of the experiment, when a sharp increase was recorded (Fig. 3.2). This increase in temperature occurred when the spillway gates closed, creating a static pond. A corresponding temperature increase was recorded in the ambient water channels, whereas the passively cooled water channels showed little variation. The average temperature difference between the ambient and cooled water channels was approximately 6°C over the experiment. Diel variation in water temperature was greater in the experimental channels than in the spillway pond or in the river.

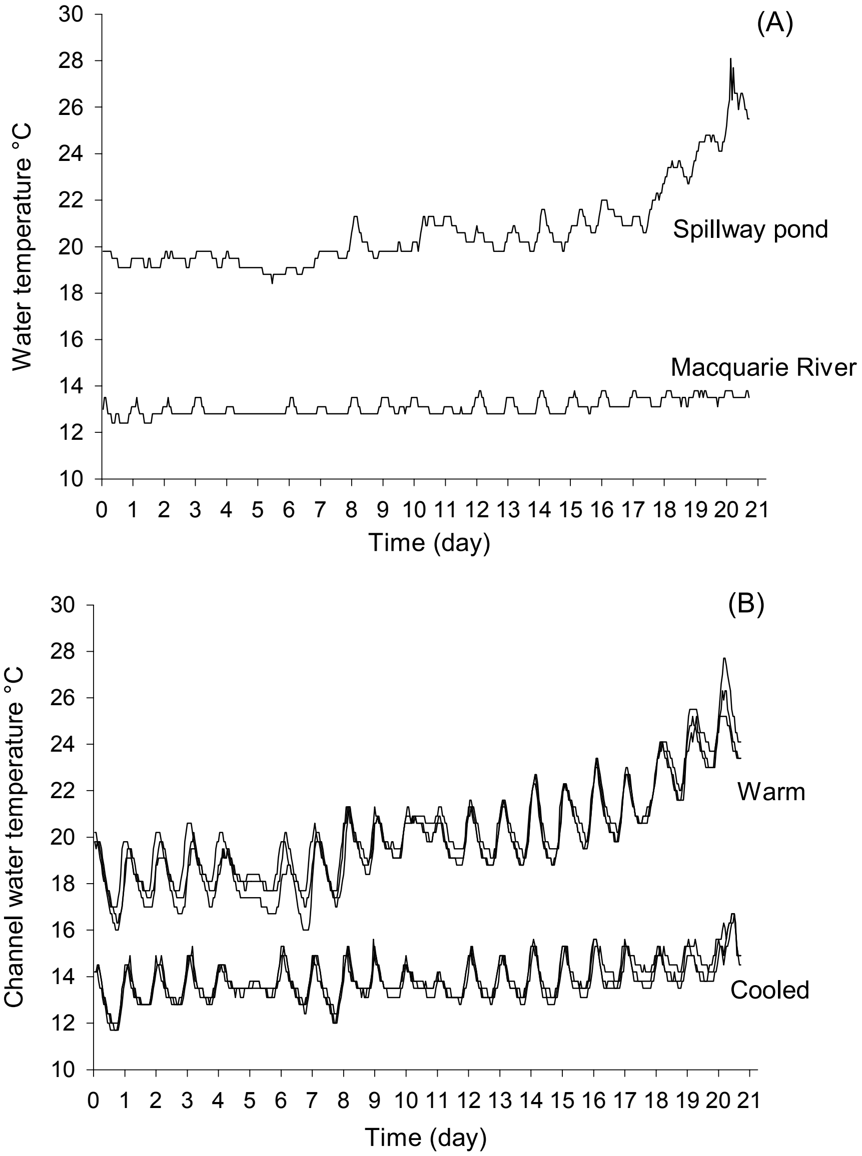

Figure 3.2. Water temperatures logged hourly during the Murray cod growth experiment (15 November to 7 December 1999), for the (A) spillway pond and Macquarie River, and (B) three warm and passively cooled experimental channels.

3.3.2. Silver perch

3.3.2.1. Survival

Only one silver perch died in the warm channels during the experiment. In contrast, a substantial number of fish in the cold channels did not survive (Fig. 3.3a). In two of the three cold channels survival was high until about day 6 after which there was a substantial decline. More fish began to die in all three cold channels after 6-8 days until the days 22 to 25, after which numbers stabilised (Fig. 3.3b). By the end of the experiment survival in the cold channels had been reduced, on average, to less than 50%. The sizes of fish dying throughout the period did not show a trend towards smaller or larger fish.

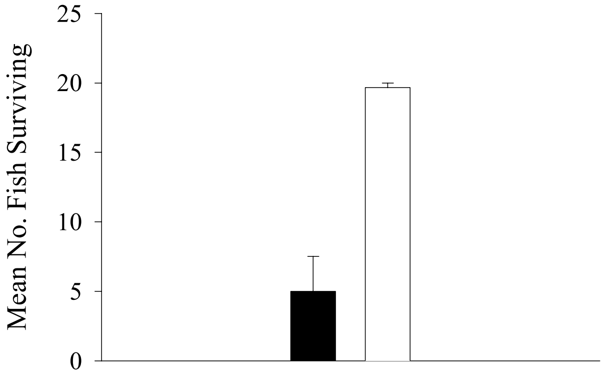

Figure 3.3a. Mean (± SE) number of silver perch surviving over all channels after 31 days. n = 3 channels, ■ Cold channels, □ Warm channels. Initially there were 20 fish per channel.

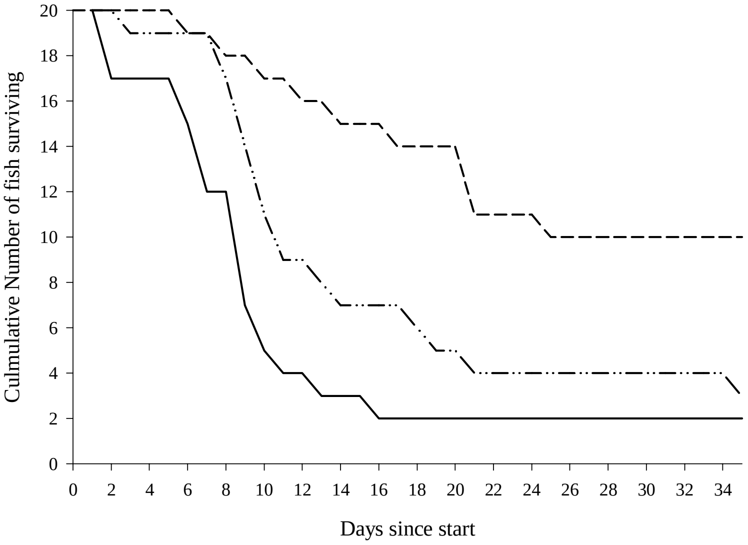

Figure 3.3b. Number of silver perch surviving in each cold channel for the duration of the growth experiment. – – Channel 2, —— Channel 4, – · · – Channel 6.

3.3.2.2. Growth

There was no significant difference in the mean weight and length of silver perch in either warm or cold channels at the start of the experiment (Fig. 3.4, $p > 0.05$). Therefore, all channels initially had similar sized fish irrespective of their treatment. By the end of the experiment fish kept in warm channels were significantly larger in both weight and length than they were at the start (Table 3.2a.b, $p < 0.001$; Fig.3.4). By contrast fish kept in cold channels for the duration showed no significant growth during the experiment (Fig. 3.4, $p > 0.05$). After 31 days fish from warm channels were significantly larger than those in cold channels (Fig. 3.4, $p < 0.001$).

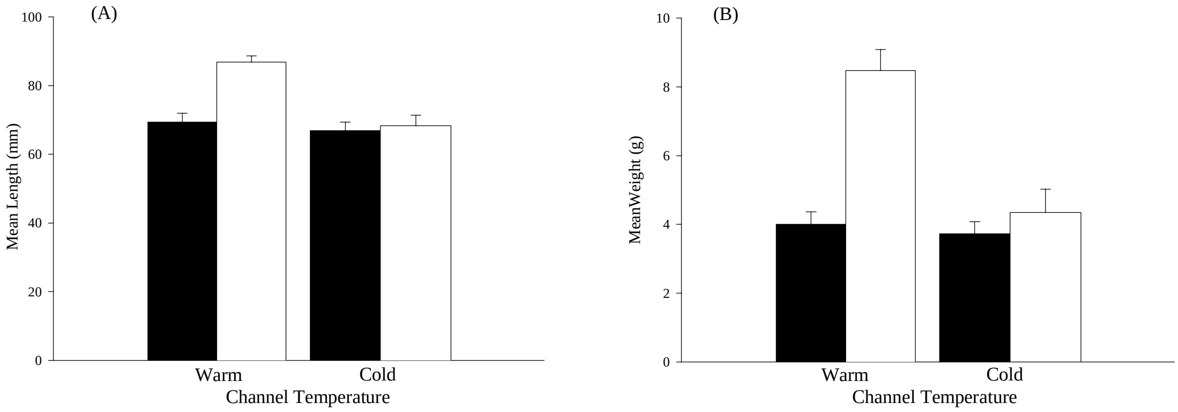

Figure 3.4. Mean length (± SE) (A) and mean weight (±SE) (B) of silver perch in cold and warm channels from start and end of growth experiment. $n = 19$. ■ Day 0, ☐ Day 31; * $p < 0.001$.

Table 3.2a. Summary of results of 2-way analysis of variance for differences in the weight (g) and length (mm) of silver perch kept in cold and warm water channels ($n = 19$). ** $p < 0.01$ and ***$p < 0.001$.

		Length		Weight	
Source	**df**	***F***	***P***	***F***	***P***
Time [T]	1	13.96	0.0004 ***	24.36	0.000005 ***
Warm/Cold [W]	1	17.07	0.00010 ***	18.26	0.00006 ***
T × W	1	10.08	0.0022 **	13.86	0.00039 ***
Residual	72				
TOTAL	75				

Table 3.2b. Summary of results SNK test for differences in mean length and weight of silver perch from the growth experiment in cold and warm water channels. ** $p<0.01$.

Variable	**Warm Channels**	**End of Experiment**
Length	Start < End **	Warm > Cold **
Weight	Start < End **	Warm > Cold **

3.3.3. Murray cod

3.3.3.1. Survival

The majority of fish in both cold and warm channels survived the duration of the experiment. One warm channel (channel 6) had three fish die over the period of 12 days (Table 3.3) whilst one cold channel had two deaths, at day 20. These deaths appeared to have been caused by a disease that started to spread through all channels and some holding tanks. After termination of the experiment many more fish in both the cold and warm channels became sick and eventually died. Mortality appears to be unrelated to water temperature because it occurred in both cold and warm channels. Two fish in warm channels died when they became trapped in the inlet pipe early in the experiment.

Table 3.3. Mortality of Murray cod in cold and warm channels during growth experiments.

Temperature	Channel	No. Deaths	Day of deaths
Warm	1	1	20
	4	1	8
	6	3	8, 11, 19
Cold	5	2	20

3.3.3.2. Growth

There was no significant difference in either the length or weight of Murray cod between warm and cold channels at the either start or conclusion of the experiment (Fig. 3.5, *p* > 0.05). A small change in length and weight on average was evident for fish in the warm channels and a slight decrease in weight for fish in cold channels. But neither of these changes was significant.

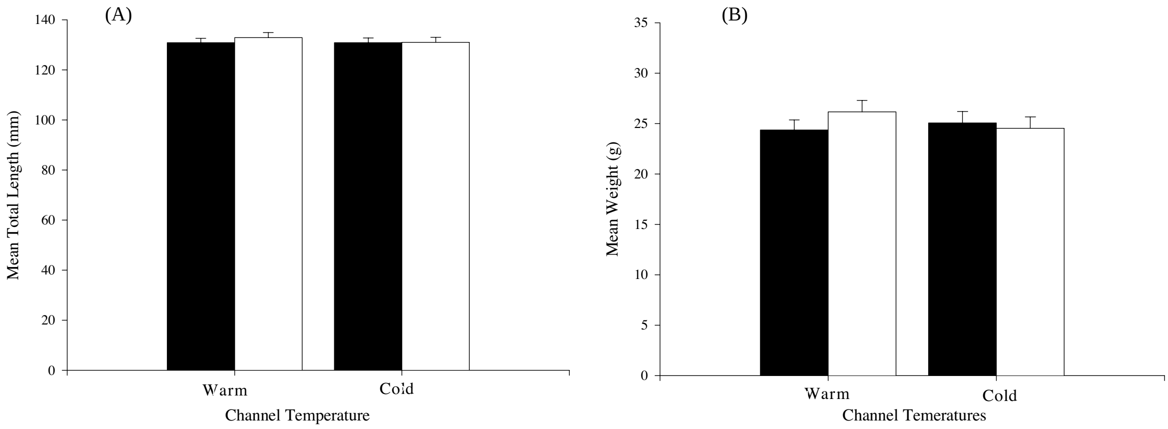

Figure 3.5. Mean Length (± SE) (A) and mean weight (± SE) (B) of Murray cod in cold and warm channels from start and end of experiment. n = 5, ■ Day 0, □ Day 31.

3.4. Discussion

The lack of growth by juvenile silver perch in cold channels could simply be a manifestation of their normal physiological response to cold water. The metabolism of warm water fish is known to slow when they are in cold water (Doudoroff, 1957; Gehrke 1988b). The slower the metabolism the less food is consumed, resulting in little or no growth. There was a substantial amount of uneaten food in the bottom of cold channels which was not present in the warm channels, confirming that the daily food intake in juvenile perch was lower in the cold channels. In contrast, juvenile fish fed and grew well in the warm channels. Other native species of fish show similar patterns. Gehrke (1988a) found that spangled perch under controlled temperatures and in Lake Samsonvale in south-east Queensland ceased feeding when temperatures fell below 16°C and lost weight until water temperature rose above 16°C. In other studies the growth rate of planktivorous fish species was highly correlated with temperature, and daily growth was minimised at temperatures < 14°C (Mooij *et al*., 1994).

In contrast to silver perch, juvenile Murray cod showed no evidence of growth in either warm or cold channels. There are several factors that could have produced this result. Firstly, the fish were not feeding in either cold or warm channels during the early stages of the experiment, and did not appear to have resumed feeding after being transported to the study site from Port Stephens. Therefore, the lack of growth in both treatments could simply reflect a failure to feed during the course of the experiment. A second possibility is that temperature alone is not the dominant factor in determining growth for Murray cod. Rowland (1992) reported rapid growth of juvenile cod in Lake Mulwala during late summer at temperatures as low as 16°C. He suggests that other factors such as habitat type, food type and availability may have a major role in determining the growth of Murray cod. Therefore, lack of some of these elements in the present study, such as different food types, may have contributed more to the absence of growth than temperature differences alone. Considering that there was little indication of growth in either cold or warm channels it is feasible that factors independent of temperature may have been operating. Thirdly, the fish in both channels may have been sick but not manifesting any signs of disease until after the experiment had ceased. Their poor health may have been reflected in their lack of growth.

Juvenile silver perch survived poorly in cold channels but well in warm conditions. However, after about 16 - 24 days the number of fish surviving in two of the cold channels remained steady. Those fish that were not able to tolerate colder temperatures were affected within a short period and died. Therefore, there was some variability amongst individuals in their ability to cope with cold water pollution. The lower temperature limits of silver perch have not been extensively documented. Cadwallader and Backhouse (1983) report that silver perch have a wide temperature tolerance. Whether the survival of the remaining juvenile silver perch could be sustained indefinitely remains to be investigated.

The rapid decline in survival of juvenile silver perch may also be due to thermal shock (Doudoroff, 1957; Crawshaw, 1977) as no period of acclimation was allowed for in the experiment. Fish will vary in their time to acclimate to rapid temperature change but those unable to adjust may not display adverse reactions until a few days later (Doudoroff, 1957). Fish were not acclimated to low temperatures in this study deliberately to mimic the sudden onset of cold water pollution in a river when cold water is released from a dam immediately above it. The results from this experiment indicate that juvenile silver perch will respond poorly to this type of cold water pollution. However, the onset of cold water pollution may become more gradual with increasing distance downstream and the responses of silver perch may be less marked or slower than those observed in this study.

Murray cod survival was high but the results were ambiguous for reasons discussed earlier regarding their feeding problems and the possible onset of disease. Symptoms displayed by affected fish suggest that there may have been an infection of *Chilodonella* that masked underlying temperature effects. The apparent lack of response to the cold water treatment could also be associated with the fact that the Murray cod tested were one year-of-age compared to the 3 month age of silver perch. If juvenile Murray cod have a temperature-sensitive stage within the first year, then they may have passed this critical age before being used in experiments.

3.4.1. Conclusions

These experiments demonstrate that cold water has a negative effect on growth and survival of juvenile silver perch. Major cold water pollution (i.e. large volumes of water released) in western drainage rivers usually occurs during summer from December to February for the irrigation season. Since silver perch spawn during early to mid summer their hatching and early larval stages (Koehn and O'Connor, 1990) directly coincide with the timing of cold water releases. Therefore, juvenile silver perch in rivers affected by cold water pollution may experience high mortality, whilst surviving individuals are likely to suffer severely inhibited growth. Whilst adults are known to live in quite cold water (Cadwallander and Backhouse, 1983; Mallen Cooper *et al*., 1995) this is the first study that examines experimentally effects of cold water on juveniles. The results point to a potential long term consequence for silver perch in inland rivers. Growth is essential for juveniles to recruit to adult populations (Matthews, 1998). Slow growth places juveniles at a greater risk of predation, possibly in poor condition, and can result in high mortality and reduced recruitment (Hoar and Randall, 1988; Matthews, 1998), potentially contributing to population declines that are reported as widespread throughout the Murray Darling Basin (Gehrke and Harris, 2000).

Further study of juvenile Murray cod is required to draw conclusions about the effects of cold water pollution on the species' ecology.

4. RESPONSE OF FISH TO A THERMAL GRADIENT

4.1. Introduction

Integrated relationships between fish and their habitats mean that any major change in habitat characteristics may result in a shift in the distribution and abundance of fish populations. Changes in fish distribution patterns may occur as a result of fish avoiding adverse habitats if they have the opportunity to do so. The relationship between water temperature and fish is a tightly knit one as fish may regulate their metabolism by changing their location in response to thermal heterogeniety (Reynolds and Casterlin, 1982). Fish in the lowlands of western drainage rivers in New South Wales are almost all adapted to warm water. Therefore, cold water pollution is likely to have a major impact on their distribution if they respond by moving away from its influence. Other species of fish are known to respond to adverse water temperatures, although studies have mainly been on species that move to cooler water to prevent over heating during summer (e.g. Peterson and Rabeni, 1996; Biro, 1998; Torgersen, *et al*., 1999). Torgersen *et al*. (1999) found a strong positive correlation between the distribution of adult chinook salmon and cool water areas in a stream during summer when water temperatures were greater than 25°C. Similarly, Biro (1998) found a larger disproportionate density of young brook trout in the coldest water of a stream in mid summer. These studies clearly demonstrate the behavioural thermoregulatory ability of fish in the wild in response to adverse temperature conditions.

This set of experiments examined how juvenile native fish respond to a thermal gradient that included water temperatures below their normal range, in order to understand behavioural responses of fish to changed temperature regimes. The experiments specifically tested the null hypothesis that there would be no significant difference in the proportion of fish between the cold and warm water ends of a thermal gradient.

4.2. Methods

A thermal gradient was created in the channels by releasing cold water from the upper inlets and warm water from the mid-channel inlets. Baffles were placed just upstream of the mid-channel inlets to prevent mixing of cold and warm water and therefore stabilised the gradient. A semicircular hole (130mm diameter) was cut at the bottom of each baffle to allow fish to move freely between the two channel sections (Fig. 4.1a). Baffles were placed in all channels. Channels were left overnight for temperatures to stabilise before the fish were added. Two treatments – cold/warm water and controls, warm/warm water - were allocated at random among channels at each run of the experiment. Water temperatures were measured to the nearest degree Celsius using glass thermometers each hour during the experiments at the upper, mid and lower sections. The upper section extended from the upper inlet to the baffle, the mid section included the area from the baffle to 0.5m down stream (Fig. 4.1b) and the lower section extended from 0.5m downstream of the baffle to the end of the channel.

Experiments were run with juvenile silver perch and Murray cod on separate occasions (silver perch – 11-14 January, 1999; Murray cod – 8-9 December, 1999). Twenty fish were placed in the mid sections of the channels. To avoid confounding, a new batch of fish was used for each run of the experiment. After letting the fish settle for five minutes, the number of fish in each section was counted every 10 minutes in the first hour, then once at three hours after the start and once at six hours after the start. These time intervals were called observation periods. Counts were done within a thirty second time frame by three observers, one stationed along side each section of the same channel. Simultaneous counts reduced the likelihood of counting the same fish twice.

Observers were concealed behind shade-cloth hides to minimise disturbance to the fish. The order in which channels were observed was randomised and observers were allocated randomly to sections within each channel to distribute observer errors evenly among channels. Four runs of the experiment were done for silver perch (1 run per day) but only one, two-day run was done for Murray cod because of the limited number of fish available.

Only two observers were available for the experiment with Murray cod, so fish were counted in only the upper and lower sections. Counts were done in exactly the same way as for silver perch but the time intervals between counts were slightly different. On day one of the experiment, in the first hour and a half, counts were made every 15 minutes and thereafter every 20 minutes for up to 3 hours after the start. On day two of the experiment, counts were made every 20 minutes for up to 2.6 hours.

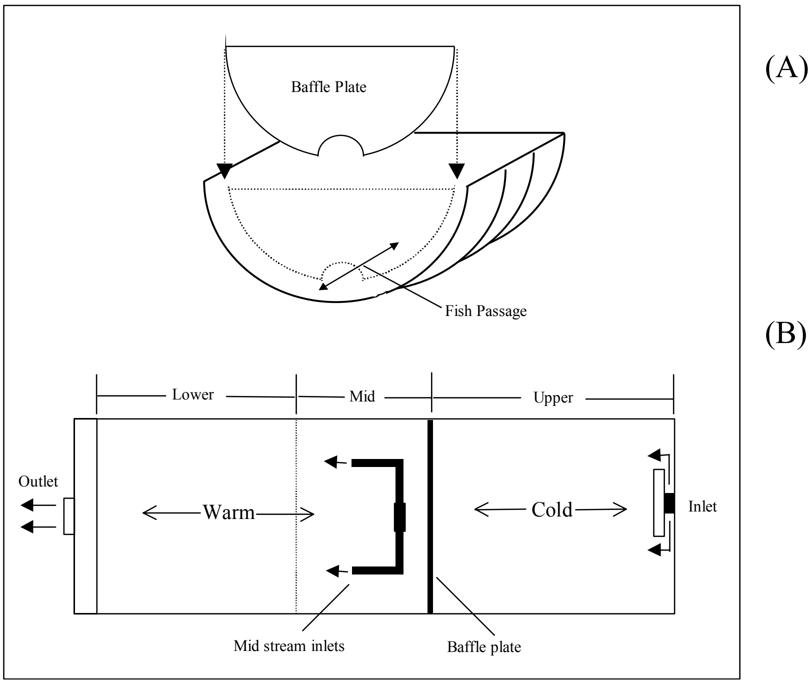

Figure 4.1. Baffle design used to promote a thermal gradient within the experimental channels (A); and areas defined as lower, middle and upper (B) used during behavioural observations; and temperature distribution within a treatment experimental channel.

4.2.1. Analyses

Analyses tested the hypothesis that the proportion of fish at the warmer section of a thermal gradient would be greater than the proportion of fish in the colder section. A three factor analysis of variance was used for silver perch and two factor analysis for Murray cod. For silver perch only, a two factor analysis of variance tested the hypothesis that acclimation of fish to cold water would result in the proportions of fish at either end of a cold-warm thermal gradient being not significantly different after the acclimation period.

The experiments were designed to be analysed using analysis of variance (ANOVA) (Underwood, 1981; Winer *et al.*, 1991). Separate ANOVA's were done for each species. Cochran's test was used to test for homogeneity of variances. Data were transformed by arcsine transformation to normalise proportions and to stabilise variances. Student Newman Keul's test (SNK) was used to distinguish between levels within factors that were significantly different.

One of the basic assumptions of ANOVA's is that the data are independent. There were two sources on non-independence in the data set for these experiments. These were (i) observation periods within the same channel and (ii) sections within the same channel. An independent set of data was obtained by ensuring replicates did not come from the same channel on the same day, and that replicates were not drawn from more than one observation time from the same channel on the same day.

4.3. Results

4.3.1. Temperature separation

Temperature separation in the treatment channels was maintained at an average of 6.7 °C throughout the experiments for silver perch and Murray cod (Fig. 4.2). Warm temperatures in the cold/warm treatment channels (mean temperature = 22.5 °C ± 0.53) did not reach the same maximum as that in the warm/warm control channels (mean temperature = 28.2 °C ± 0.28). However, the results show that this difference between warm and cold in the treatment channels was sufficient to draw a response from the fish under investigation.

4.3.2. Silver perch

There was a significantly larger proportion of fish in the warmer (lower) section of cold/warm treatment than in the colder (upper) section, and no significant difference between sections in the control treatment with no thermal gradient (SNK test, $p< 0.01$) (Table 4.1, Fig. 4.3).

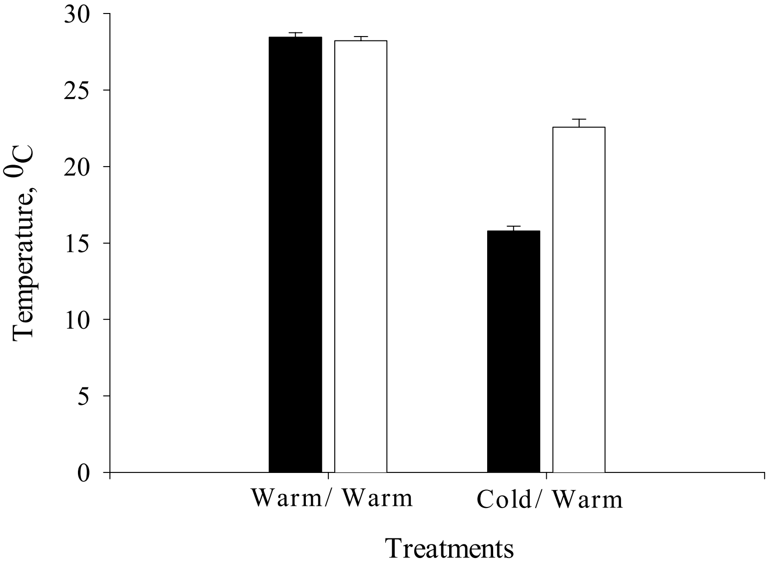

Figure 4.2. Mean temperatures (± S.E.) of channel water achieved in the thermal gradient experiment. ■ upper section, ☐ lower section. $n = 3$.

These results suggest that silver perch responded to the cold water influx by moving toward the warmer ends of the channels. Furthermore, there was a significantly smaller proportion of fish in the upper sections of cold/warm channels after 360 minutes than after 10 minutes and no difference between times in the lower sections of either the warm/warm or cold/warm channels (Table 4.2, Fig. 4.4). Thus the fish showed no tendency to become more tolerant of colder water over the 360 minute duration of the experiment.

Table 4.1. Summary of results of ANOVA of the mean proportion of silver perch in warm/warm and cold/warm for upper versus lower sections. Arcsine transformed, $n = 3$. *** $P < 0.001$; Sig = significance; NS = not significant; NT = no test.

Source	df	MS	F	P	Sig
Day	3	233.94	2.62	0.150	NS
Channel	1	0.03	1×10^{-5}	1.000	NS[†]
Section (Ch)	2	2574.08	28.81	0.001	***
D x T	3	174.79	1.96	0.220	NS
D x S(T)	6	89.34	0.45	0.840	NS
Residual	32	197.70			
Total	47				

[†] From a separate analysis

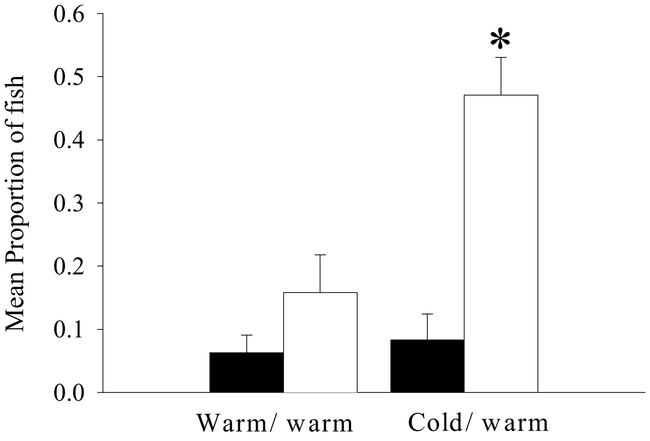

Figure 4.3. Mean proportion (± SE) of juvenile silver perch in upper versus lower sections of channels with a thermal gradient (cold/warm) and without (warm/warm). $n = 6$, * $p < 0.05$, ■ Upper, □ Lower.

Table 4.2. Summary of results of ANOVA of the mean proportion of silver perch occupying upper and lower sections of cold/warm and warm/warm channels after 10 and 360 minutes duration of experiment. n =12, *** $P < 0.001$; NS = not significant.

Source	df	Upper			Lower		
		MS	F	P	MS	F	P
Temperature [T]	1	322.52	12.12	NS	1454.87	31.08	NS
Interval [I]	1	1089.71	11.86	***	458.82	1.33	NS
T x I	1	26.60	0.29	NS	46.81	0.14	NS
Residual	20	91.87			344.72		
Total	23						

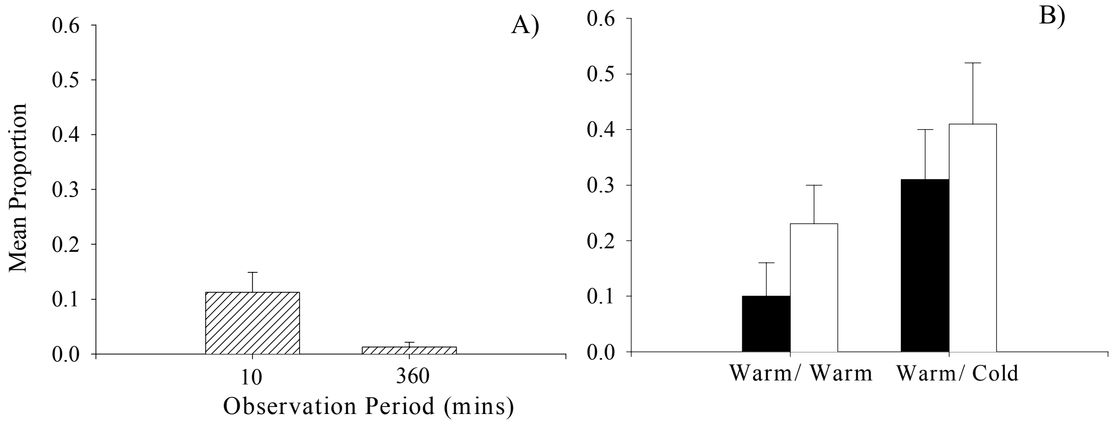

Figure 4.4. Mean proportion (± SE)of silver perch (A) in upper sections over all temperature treatments, and (B) in lower sections of each treatment after 10 and 360 minutes in a thermal gradient. n=12. ■ 10 minutes, ❑ 360 minutes.

4.3.3. Murray cod

Murray cod responded in a similar manner to silver perch to the thermal gradient. There was a significantly larger proportion of fish in the warmer (lower) section than the colder (upper) section in the cold/warm channels and no significant difference in the proportion of fish between the upper and lower sections of the control channels (SNK test $p < 0.01$) (Table 4.5; Fig. 4.5). These results indicate that Murray cod actively avoided cold water by moving toward the warmer end of the thermal gradient.

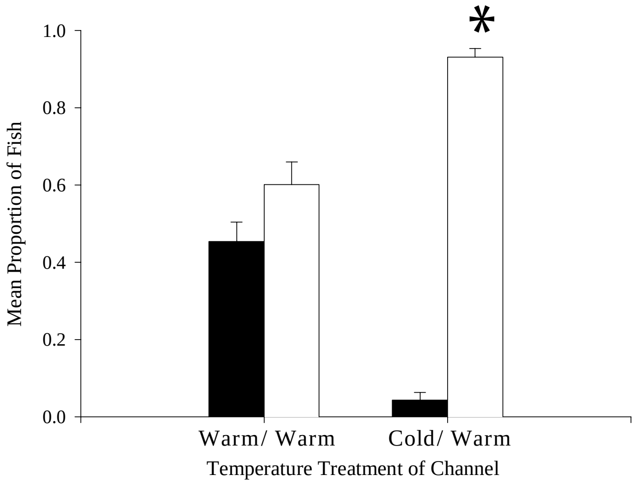

Figure 4.5. Mean proportion (SE) of Murray cod between sections of cold/warm and warm/warm channels after exposure to a thermal gradient. N = 36, * p< 0.005, ■ upper, ☐ lower.

Table 4.3. Summary of results of thermal gradient experiment for Murray cod for differences between upper and lower sections in cold/warm and warm/warm channels (Day 1 = A; Day 2 = B). Transform arcsine. *** $P < 0.001$; Sig = significance.

A. Source	**df**	**MS**	**F**	**P**	**Sig**
Section	1	4.819773	3.867	0.188	NS
Treatment(S)	2	1.246148	39.549	0.001	***
Residual	68	3.15E-02			
Total	71				
B. Source	**df**	**MS**	**F**	**P**	**Sig**
Section	1	1.824086	0.809	0.463	NS
Treatment(S)	2	2.253474	67.251	0.001	***
Residual	44	3.35E-02			
Total	47				

4.4. Discussion

These experiments clearly show that juveniles of two species of native fish are capable of detecting and avoiding cold water**.** Avoidance of cold water occurs rapidly, and can be detected within an hour of exposure to reduced water temperature. This result is not surprising since most fish can only regulate their body temperature by moving to habitats that provide suitable environmental temperatures. The sensitivity of silver perch and Murray cod to temperature differences of 6.8°C (mean range 15.7 to 22.5°C) in this study suggests an even greater ability to detect temperature differences in rivers where the temperature change could be as great as 10-12°C. Peterson and Rabeni (1996) found several species of adult fish responding to a similar range of temperature change (6°C) by moving away from cold water in a main river into a smaller and warmer tributary. Williams (1967) reported that in the Goulburn River in Victoria a 10-15°C reduction in water temperature resulted in a loss of Murray cod which were previously abundant in that section of the river. Therefore, releasing cold water from dams can have almost immediate effects on the behaviour and distribution of juvenile silver perch and Murray cod. It remains to be seen whether adults of these species respond similarly to temperature changes. Thermal tolerances may change during fish development. Adults are often able to tolerate a larger range in temperatures than juveniles (Jobling, 1994). Differences in thermal responses between adults and juveniles are yet to be investigated for silver perch and Murray cod.

Movement of fish away from cold water causes localised depletion of the fish population, with a resultant re-distribution of fish into other habitats either further downstream or in warmer tributaries. Such forced re-distribution may have several negative outcomes. It could result in fish having access to poorer quality food, fewer microhabitats or refuges than were present in the now cold section of the river. Consequently re-distribution of fish may place increased pressure on limited resources. For example, if fish move into tributaries that are smaller than the main channel of river from whence they came, potentially there will be fewer microhabitats available, a smaller food source and an increase in density of fish. Resultant increases in intra- and inter-specific competition may in turn lead to reduced survival of juvenile fish which may not be able to compete successfully with adult fish. Mullan *et al.* (1976) showed that colder water temperatures re-distributed rainbow trout to areas away from their major food source (Calow and Petts, 1994). If redistribution of native fish away from favourable habitat is widespread in western drainage rivers then this could be a factor contributing to the general decline in some native species.

Notwithstanding the negatives highlighted above tributaries may offer some form of refuge to natives fish, especially if conditions are right. For example, fish may be able to exploit different types of habitat and food sources available in tributaries that are not open to them in the main river channel. Juvenile fish therefore may do better under these conditions than if they had remained in the main channel. Peterson and Rabeni (1996) found that a number of species of fish moved into a warmer tributary of the Jacks Fork River (Missouri, USA) when the temperature in the main river channel dropped. In addition, the fish in the tributary were significantly larger and fed more frequently than those in the river (Peterson and Rabeni, 1996). Potentially, then some advantages to fish populations may occur as a response to cold water re-distribution if fish are able to take advantage of new conditions. The probability of such good conditions occurring in the tributaries of rivers in New South Wales, however, is small under present circumstances, based on recent surveys of riverine fish communities in New South Wales (Harris and Gehrke 1997).

4.4.1. Conclusions

The results of the experiments clearly indicate that juvenile Murray cod and silver perch are likely to respond by moving away from cold water towards warmer water. Further work is necessary to determine the direct consequences of this redistribution along the riverine thermal gradient. Determining the nature of any negative outcomes will give weight to recommendations to change the way water is released from dams. If positive outcomes are determined, particularly with respect to the use of tributaries as alternative habitats, then this provides options for management and conservation. If fish are unable to locate warmer alternative habitats, then they are likely to suffer reduced growth and survival as described for silver perch in the preceding chapter. Given that modifications to allow dams to release warm water downstream are likely to reduce the downstream extent of cold water pollution rather than eliminate it entirely (Sherman, 2000), encouraging fish to use tributaries could be a complementary management strategy to conserve native fish communities. Clearly there are potentially many complex implications of the responses of fish to cold water pollution. These can only be unraveled by well planned experiments and field studies on aspects of fish responses to cold water pollution.

5. ACTIVITY OF FISH UNDER DIFFERENT THERMAL CONDITIONS

5.1. Introduction

Fish engage in a variety of activities, such as foraging and feeding, seeking refuges, migration, reproduction and spawning. Each of these activities can be affected by changes in water temperature (Petts, 1984). For example, rainbow trout in the Flaming Gorge Reservoir, USA, were found to have moved away from their normal foraging areas, where food was abundant, into areas of least flow velocity where only algae was present, because of lower natural temperatures (Mullan *et al.*, 1976). This movement resulted in poorer feeding and suppressed growth in the new habitat. Thus cold water pollution can potentially have subtle but important changes on fish activity that are detrimental to their ecology.

Silver perch are normally very active, with long dispersal migrations upstream (McDowall, 1996). They are a strong schooling fish but much remains to be learnt about their movements and their interaction with microhabitats. This study examines effects of cold water on the use of microhabitats by silver perch.

5.2. Methods

Channels were randomly allocated either a warm or cold water treatment, with three channels per treatment. In each channel five artificial habitats (10cm diameter x 5-6 cm long PVC pipe) were placed equidistant along the length of the channel. Habitats were orientated at an angle to the flow so that observers could see inside to count fish. Only juvenile silver perch were used in this experiment. Fifteen fish were placed in the upper section of each channel. Observations were based on the number of fish using each of these artificial habitats. Counts were done by three observers at each channel, one stationed in each section of the same channel (upper, mid and lower). These observers made four different counts for their section corresponding to different activities. For the first 30 seconds the number of fish within a 5 cm radius of a habitat (surround 1 (S1)) was counted, for the next 10 seconds the number of fish inside the habitat was counted, in the next 30 seconds the number of fish within 5 cm of the habitat (surround 2 (S2)) was counted again and in the last 10 seconds the number of fish crossing an imaginary line perpendicular to the flow were counted. For each observation period the fish were counted crossing the “line” in one direction only (i.e. up- or downstream) for a channel but this direction was changed for each channel at random every time it was sampled. These activities were chosen because they were considered to be similar to the natural behaviour of silver perch. Counts were made simultaneously by all observers to reduce the likelihood of counting the same fish twice. Observers were concealed from the fish by shade cloth hides, and observed fish through small peep holes or over the top of the hides. The order in which the channels were observed, and allocation of observer’s to sections within each channel were randomised to distribute observer errors evenly among channels. Four runs of the experiment were done (1 run per day) using a new batch of fish for each run. Observations were made every 5 minutes for the first 2 hours and thereafter every 20 minutes for up to 4 hours.

5.3. Results

The proportion of fish observed using the artificial habitats was small (< 10%) in both cold and warm channels. Therefore, the planned activity measures did not record activity of most of the fish in the channels. Most of the silver perch were in small schools in both warm and cold channels. In the warm channels the fish tended to swim rapidly from one end of the channel to the other and then back again after a pause, ranging from approximately 30 seconds to several minutes. In contrast, fish in the cold channels tended to hide under the inlet pipe in the upper section of the channel and few fish were seen swimming rapidly. It was originally expected that the swimming activity of the fish would be measured as the number of fish crossing the imaginary line (as described in the Methods). However, the fish were not active enough to record swimming activity. In the warm channels, where the fish were the most active, their swims from one end of the channel to the other were not frequent enough to be picked up during observation periods. Whilst there were insufficient data to analyse, overall there was a trend for more fish to be active in warm channels than cold (Fig. 5.1). Fish activity for both warm and cold channels varied among runs of the experiment, indicating that there was natural variability in the way that fish responded to warm and cold water. Fish in the cold channels tended to be more sedentary than those in the warm channels.

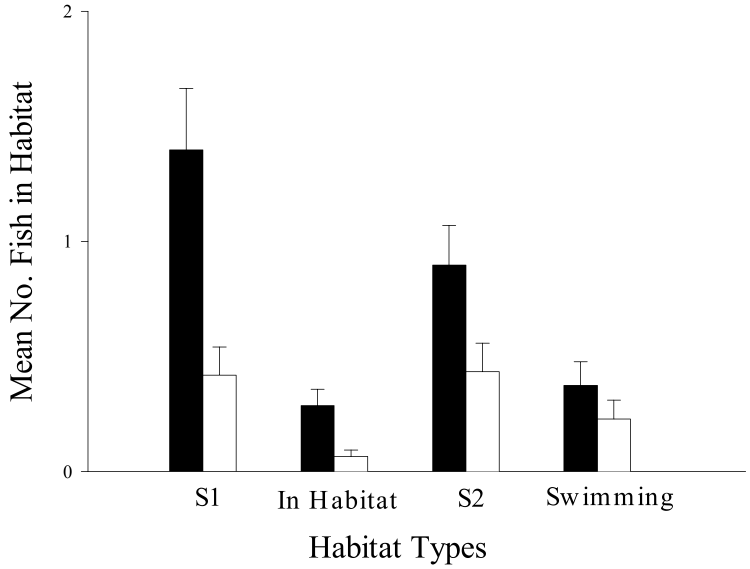

Figure 5.1. Mean ± S.E. number of silver perch using habitats in cold and warm channels over all days of the activity experiment. *n* = 3 channels. S1 – surround 1and S2 – surround 2 were counts of fish within a 5 cm radius of a habitat. ■ Warm, ☐ Cold.

5.4. Discussion

Interpreting fish activity in artificial environments, such as experimental channels, will always carry the problem of whether what is observed is "natural" or contrived as a result of being in such an environment (Gelwick and Matthews, 1993). Extrapolating findings from artificial habitats to the real world therefore needs to be done with great care. The data obtained from this experiment were inadequate to determine whether the activity of silver perch differed between cold water and warm water treatments. However, the results do suggest that fish in cold water become less active than fish in warm water. Reduced activity may suppress the ability of fish to search for food and habitat and also reduce their ability to escape predators. In addition, Gehrke (1988a) found that the metabolic energy requirement of fish in cold water is reduced which in turn reduces their need to ingest food and hence foraging activity.

5.4.1. Conclusions

No definitive conclusions can be drawn from these experiments about the effects of cold water pollution on the activity of juvenile silver perch. However, the limited evidence available suggests that silver perch activity is suppressed sufficiently by low temperatures that further investigation is warranted.

6. TRENDS IN FISH COMMUNITIES DOWNSTREAM OF BURRENDONG DAM

6.1. Introduction

Establishing and describing patterns of relationships between fish and water quality variables is a fundamental step for any study investigating effects of human induced ecological change (Underwood, 1990). However, for thermal regimes there has been little work done in Australia that directly describes the relationship between fish abundance and distribution and water temperature in rivers (Koehn and O'Connor, 1990; Mallen-Cooper *et al.*, 1995). Peterson and Rabeni (1996) in the USA examined the seasonal use of tributaries, by several species of fish when temperature in the main river channel (Jacks Fork River, Missouri, USA) dropped near or below their lower limits. They found that fish used the tributary during cold periods and had a higher biomass than those remaining in the main channel. Scheller *et al.* (1999) used thermal variables to predict the presence and absence of 15 fish species in streams throughout the USA. Richardson *et al*. (1994) found that significant relationships between fish density and water temperature existed for only two species of native fish in New Zealand.

Relationships between biota and water quality is becoming an increasingly important issue, especially in Australia. Many biological processes depend on good water quality and because human activities has had such a negative impact in many places it is important to develop a greater understanding of the relationships between biota and water quality. Gehrke (1991) found that dispersal patterns of golden perch larvae corresponded with gradients in water quality between pond and floodplain habitats. Temperature is one aspect of water quality that has a close relationship with biota. Fry (1971) gives an extensive review of the effects of temperature on fish physiology and behaviour.

Harris (1997) documented changes in water temperature along the length of the Macquarie River but did not include any simultaneous fish sampling. There is a need to build on this work and investigate relationships between fish community composition and temperature regime within the Macquarie River. The relationship between fish distributions and river temperatures will indicate at a within-river scale the extent to which cold water pollution affects fish communities downstream of Burrendong Dam. The relationship will also show the inter-relationships between native and alien fish species in relation to changes in water temperature.

The aim of this study was to determine if there is a relationship between river temperature and fish abundance downstream of a major cold water release dam (Burrendong) and on a similar river with no cold water release. These relationships are examined at a seasonal temporal scale between irrigation season, when most cold water is released from the dam, and non-irrigation season, when little water is released and the river temperature is closer to natural ambient temperature.

6.2. Methods

6.2.1. Sampling sites

Field survey locations were selected on the Macquarie and Bogan Rivers (Fig 6.1, Table 6.1). Sites were surveyed once in non-irrigation (15 - 19 June 1998) and once during the irrigation season (19 – 23 October 1998). Each location was divided into two sites separated by at least 1-2 km of river.

All navigable habitats within the river channel were sampled by electrofishing using a boat-mounted electrofisher. For each site a standard sample consisted of six 2 minute electrofishing shots. Six shots were considered sufficient based on the results of Faragher and Rogers (1997) who found that for the Macquarie and Bogan Rivers few additional species were added to the catch after six boat electrofishing shots. Fish caught during each shot were identified, counted and measured (fork-length) to the nearest millimetre. Temperature was measured at the water surface (within 1 m of the surface) and on the bottom (within 1 m of the bottom) at each site, using a Horiba U10 water quality meter. Three replicate measurements were taken for surface and bottom temperature at each site.

Table 6.1. Field survey locations on the Macquarie and Bogan Rivers sampled by boat electrofishing in the non-irrigation and irrigation seasons, and the distance of locations from Barwon River junction.

River	Location	Latitude	Longitude	Nearest town	Distance from Barwon River (km)
Macquarie	Burrendong	149°04’04”	32°37’28”	Wellington	592.3
	Dubbo weir	148°35’42”	32°15’02”	Dubbo	486.7
	Gin Gin weir	148°10’11”	31°58’59”	Warren	309.3
	Marebone weir	147°43’07”	31°24’46”	Warren	196.9
Bogan	Peak Hill	148°07’21”	32°43’31”	Peak Hill	449.2
	Nyngan weir	147°10’04”	31°35’38”	Nyngan	276.1
	Gongolgon weir	146°53’44”	30°20’56”	Bourke	87.2

The locations were plotted as a proportional distance from the confluence of each respective river with the Barwon River to allow comparisons between rivers on a common basis. Comparison of locations between rivers without a common basis can lead to erroneous interpretations of the results. For example, differences in abundances of fish between locations furthest downstream could be influenced by the proximity of locations to the confluence with another river. Differences associated with proximity to another river may be mis-interpreted as being related to differences between the thermal regimes of the rivers in question.

Figure 6.1. Field survey locations sampled by electrofishing on the Macquarie and Bogan Rivers.

6.2.2. Boat electrofishing

Electrofishing was done using the 5-m Fisheries Research Vessel (FRV) *Electricus*. The boat is equipped with a Smith-Root 7.5 kW GPP electrofishing system (Generator Powered Pulsators). A petrol-powered generator produces an electrical current that passes through a rectifier unit, creating a pulsed Direct Current (DC) waveform. The current is directed through submerged electrodes establishing an electrical field in the water. The electrical current flows in one direction from the cathode (-) to the anode (+). The initial reaction of fish is to swim towards the anode in a response known as galvanotoxis. As fish approach the anode the electrical field is strong enough to stun them (galvanonarcosis) and in this condition they are easily caught. *Electricus* is operated with by a crew of three people, one person to drive the boat and control the electrofisher while two people dip net stunned fish at the bow. All netted fish were placed in a flow-through live well to recover. Safety procedures followed the National Electrofishing Code of Practice (1997).

6.2.3. Analysis of data

Variation among fish communities within and between rivers in all locations was examined using multivariate procedures described by Clarke (1993). These analyses use a measure of similarity between samples to map their relationships in an ordination using non-parametric Multidimensional Scaling (MDS). Ordination produces a graphical representation of the similarity (and dissimilarity) among samples from all locations sampled. Data were transformed to the fourth root to reduce the influence of abundant species and increase to the influence of rarer species. The Bray-Curtis Similarity measure was used to calculate similarities among samples and used to construct three-dimensional MDS plots. The significance of differences among locations and rivers was determined using the ANOSIM randomisation test Clarke (1993). A similarity percentages analysis (SIMPER, Clarke, 1993) was done to determine which species were contributing most to the dissimilarity between groups.

6.3. Results

Mean river temperatures for surface and bottom readings did not differ significantly for any season, or in any location or river. Hereafter, therefore, only surface temperatures are discussed. Mean river temperatures in non-irrigation season varied little between locations on either river. River temperatures in winter ranged from 9°C to 12.5°C (Fig 6.2). During the irrigation season the water temperatures on the Macquarie River ranged from a mean of 11.6°C at Burrendong to 19.1°C downstream at Marebone, a difference of 7.5°C in 395 km by river between locations. Water temperatures at Burrendong on the Macquarie River were similar during both irrigation and non-irrigation season and were substantially lower than the equivalent location on the Bogan River. River temperatures at Marebone during the irrigation season were still approximately 1°C cooler than the Bogan River at Nyngan, which is a similar distance from the confluence with the Barwon River. In the Bogan River during the irrigation season, water temperatures ranged from 17°C at Peak Hill to 20.8°C at Gongolgon, a rise of 3.8°C in 362 km between these locations. Between seasons the rise in water temperature was greater on the Bogan River for each location than on the Macquarie River (Fig. 6.3).

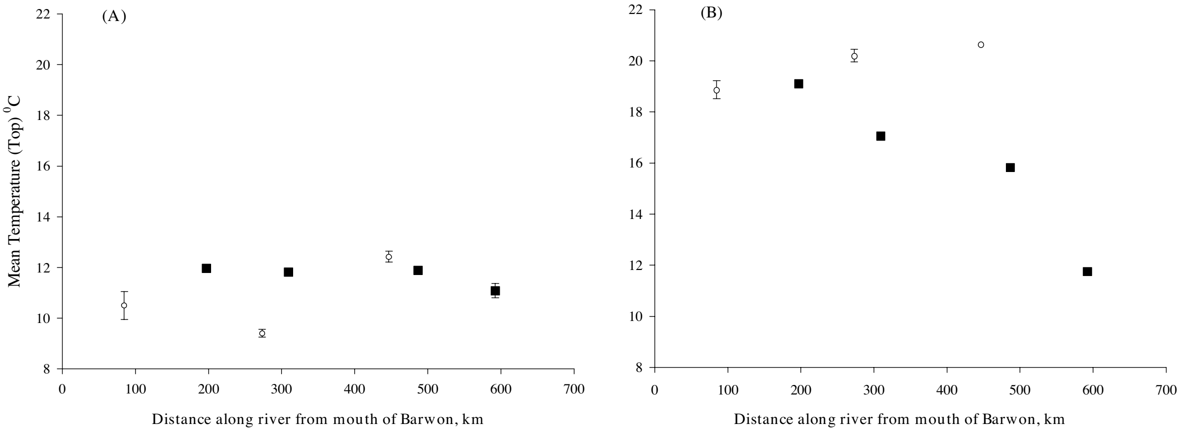

Figure 6.2. Mean surface water temperatures of rivers during two seasons, (A) non-irrigation and (B) during irrigation. ● Macquarie river, ○ Bogan river. N=3.

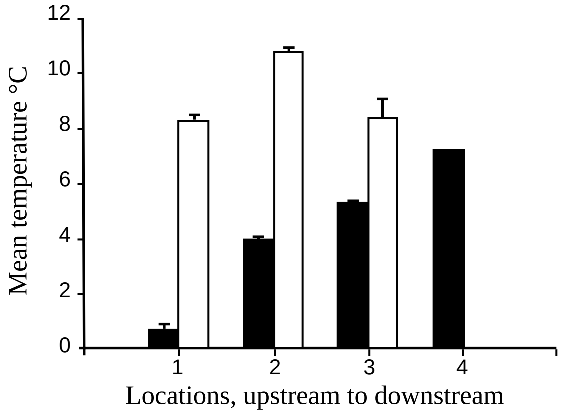

Figure 6.3. Mean temperature differences between irrigation and non-irrigation seasons for each location in the ■ Macquarie and □ Bogan rivers. N=3.

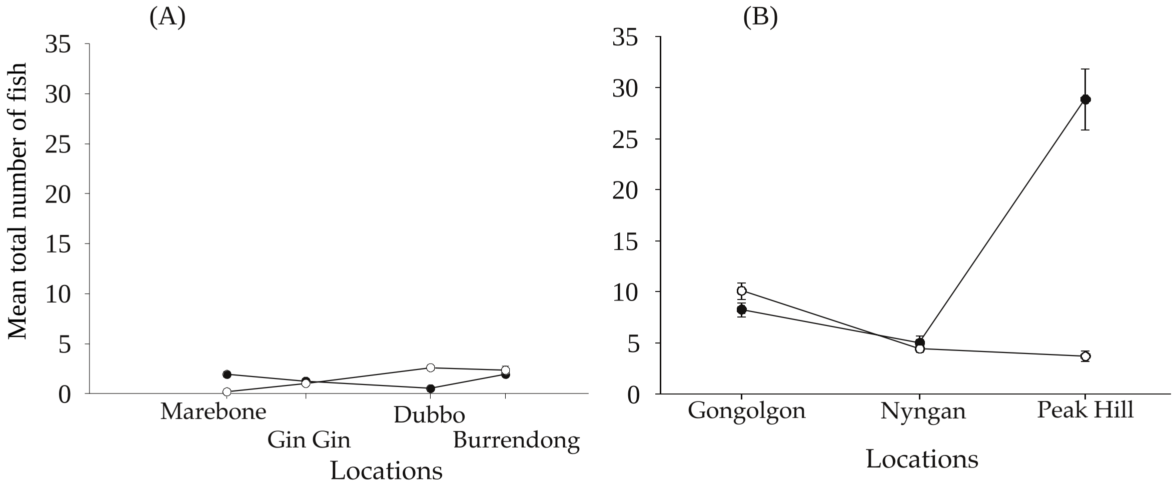

Figure 6.4. Mean number of total fish caught at each location on the (A) Macquarie river and (B) Bogan river, ● non-irrigation season, ○ during irrigation season. N=12. Locations plotted in proportion to distance from the mouth of the Barwon river.

A total of 868 fish was collected, representing 12 species and 9 families. Of the 12 species recorded, 7 were collected from the Macquarie River and 9 from the Bogan River. Five species were common to both rivers (Appendix 1). The Macquarie River had the smallest proportion of native species in the catch in the non-irrigation season (Macquarie – 0.55; Bogan – 0.82) but the largest proportion during the irrigation season (Macquarie – 0.45; Bogan – 0.17). Over both seasons the Bogan River had the largest proportion of native species. The mean number of fish sampled from the Macquarie River (Fig 6.4a) varied considerably between locations. Fish abundance was similar between locations on the Bogan River, except for Peak Hill where large catches were taken (Fig.6.4b). The catch in the Bogan River across all locations and seasons was 82.4% of the total catch for the entire two surveys (Appendix 1).

Fish abundance did not change as river temperature increased from non-irrigation to irrigation season on the Macquarie River (Fig 6.5). For both non-irrigation and irrigation seasons, the mean abundance of fish remained small. Similarly, in the Bogan River during the non-irrigation season there was no pattern of change in fish abundance with river temperature (Fig.6.5). During irrigation, however, fish abundance in the Bogan River showed a slight increase as temperature rose, but this occurred at only one location. In the Macquarie River those locations where temperatures increased during irrigation did not have a corresponding increase in fish abundance.

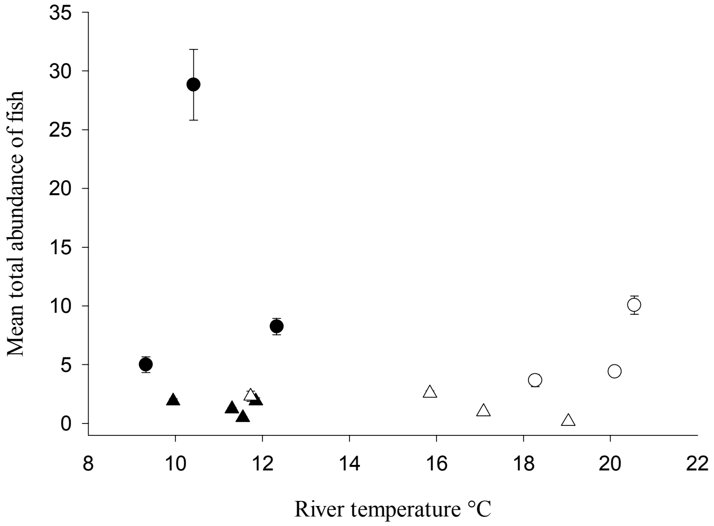

Figure 6.5. Mean fish abundance versus mean water temperature during non-irrigation (filled symbols) and irrigation seasons (open symbols) at each location on the Macquarie (▲,△), and Bogan (●, ○) rivers.

The proportion of native species in the catch at each location had a varying relationship with temperature both within and between seasons. Within the non-irrigation season on the Macquarie River there was no relationship between river temperature and proportion of natives (Fig. 6.6a). However, during the irrigation season the proportion of natives increased with increasing temperature downstream of the dam (Fig. 6.6b). This is consistent with the fact that the intensity of the effects of cold water would dissipate the further locations were from the dam. This pattern therefore, reflects the expectation that if native species are responding to temperature changes there would be an increased proportion of native fish with increased temperature downstream. In the Bogan River, with no cold water releases, the patterns are different. In both irrigation and non-irrigation seasons there is no relationship between temperature changes and the proportion of native fish (Fig. 6.6a,b). This is despite the fact that temperature changes between seasons was greater in the Bogan than that in the Macquarie River (Fig. 6.3).

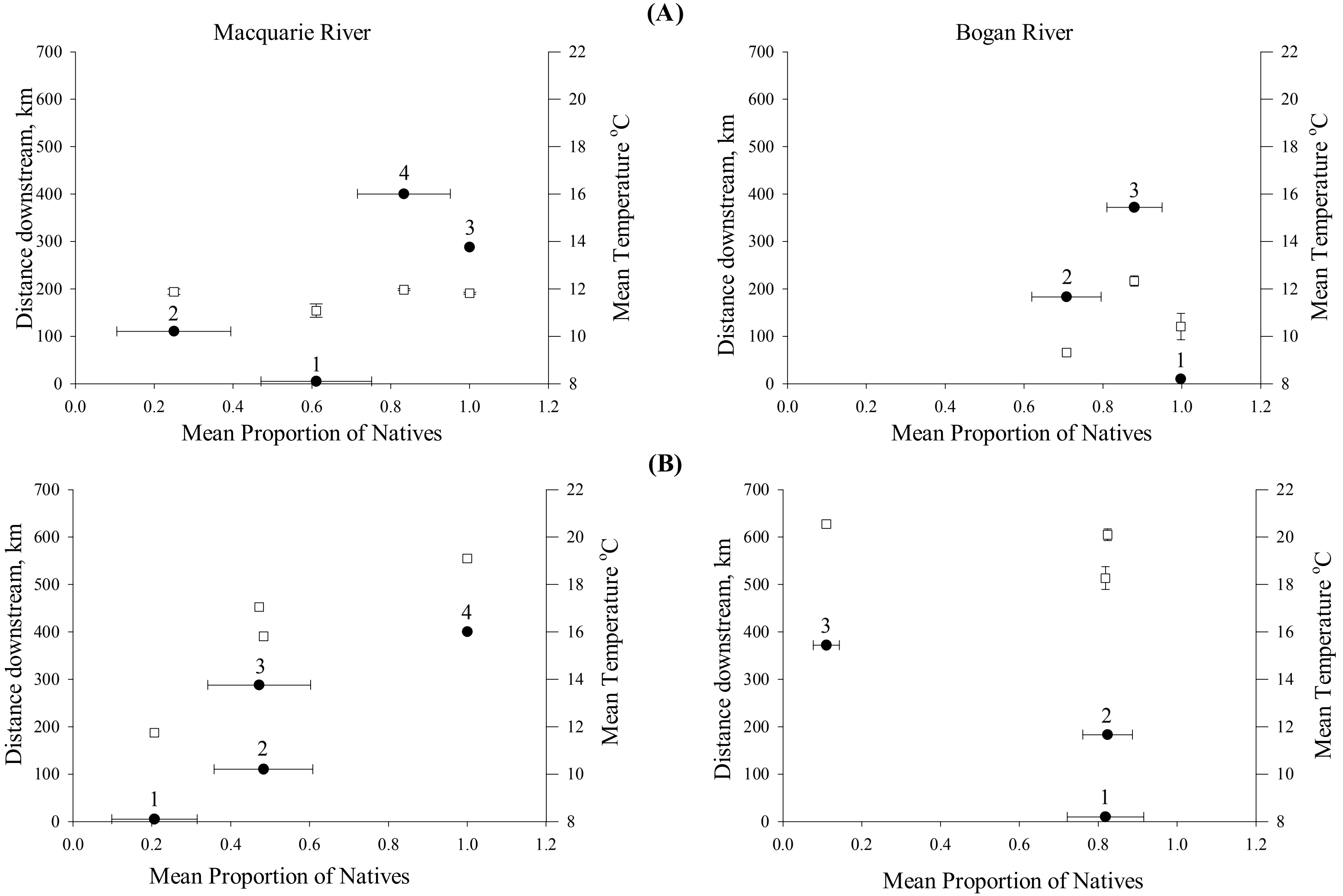

Figure 6.6. Mean proportion of native fish (●) on the Macquarie and Bogan rivers and mean temperature (❑) at each location during (A) non-irrigation and (B) irrigation seasons. Numbers are order of locations from upstream to down stream.

Changes between seasons of the temperature and proportion of natives at each location on both rivers varied considerably. Two locations on the Macquarie River had an increase the proportion of natives with increase in temperature between seasons, whilst the other two locations had a decrease in the proportion of natives with increasing temperature (Fig 6.7a). This pattern occurred irrespective of the location's distance downstream from the dam. A similar pattern occurred on the Bogan River (Fig 6.7b).

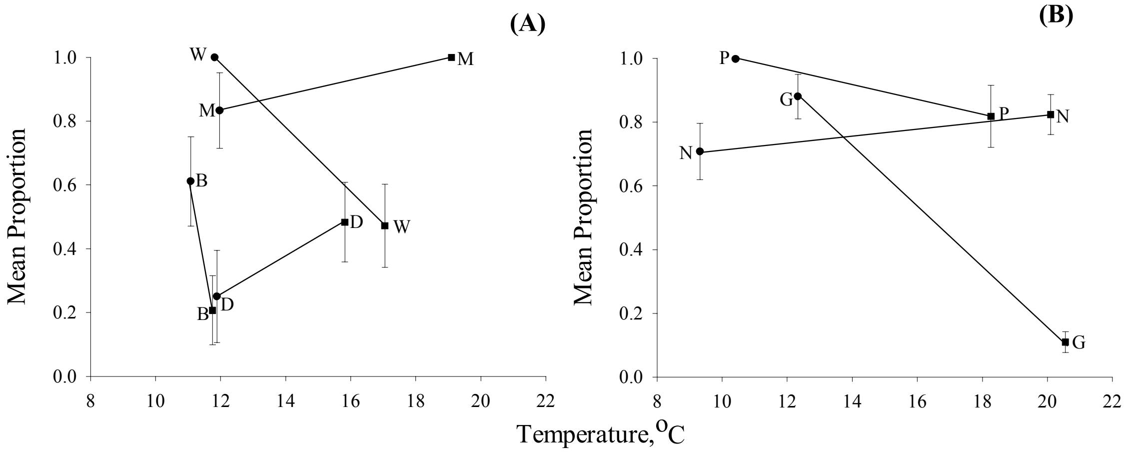

Figure 6.7. Mean proportion of native species in total catch at each location versus river temperature during ● non-irrigation and ■ irrigation season. (A) Macquarie River - B – Burrendong, D – Dubbo, W – Warren, M – Marebone; (B) Bogan - N – Nyngan, P – Peak Hill, G – Gongolgon.

The composition and abundance of fish communities between the two rivers were significantly different from each other (R value = 0.318, $p < 0.0001$) irrespective of irrigation season. Similarly there was a significant difference in the fish communities between irrigation and non-irrigation season across all rivers (R = 0.142, $p < 0.0001$) (Fig. 6.8).

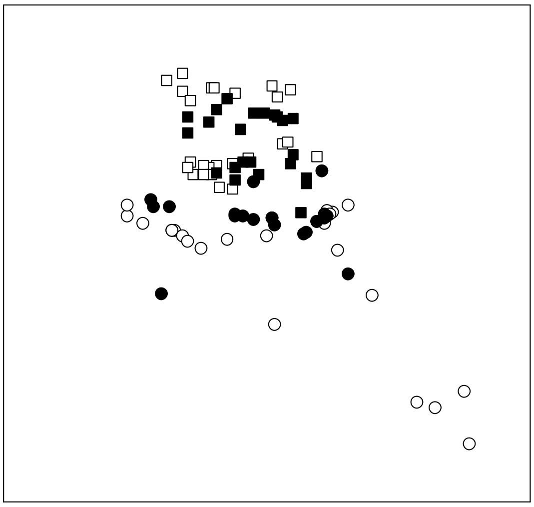

Figure 6.8. Two dimensional MDS of fish communities for irrigation and non-irrigation season in the Macquarie and Bogan rivers. ○ Macquarie, non-irrigation; ● Macquarie, irrigation; ☐ Bogan, non-irrigation; ■ Bogan, irrigation.

Fish communities among locations within each season were also significantly different (R = 0.72, $p < .0001$, non-irrigation; R = 0.50, $p < 0.0001$, irrigation). During the non-irrigation season locations within rivers were clearly separated in the MDS plots indicating that the fish composition within each location was significantly different from other locations (Fig. 6.9a). In particular the most downstream locations on the Bogan River (Gongolgon) and the Macquarie River (Gin Gin and Marebone) were different from the locations furthest upstream. This pattern suggests that fish communities furthest downstream are different from those upstream during the non-irrigation season irrespective of thermal regimes of the rivers. The differences among locations on the Macquarie River were mainly attributable to the greater abundance of *Retropinna semoni* at Gin Gin than other locations and in the Bogan River the difference among locations were due to the presence of *Hypseleotris* spp. and *Retropinna semoni* at Gongolgon. During the irrigation season there are less distinct patterns among locations (Fig. 6.9b).

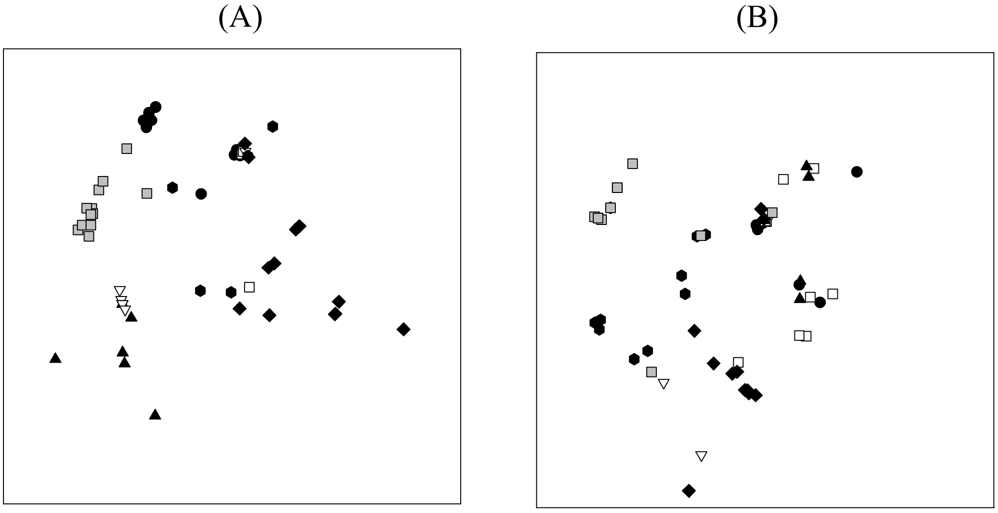

Figure 6.9. Two dimensional MDS of fish communities in each location within the Macquarie and Bogan rivers for (A) non-irrigation and (B) irrigation seasons. (A) Stress = 0.08, (B) Stress = 0.10. Macquarie river: ● Burrendong, ❑ Dubbo, ▲ Gin Gin, ▽ Marebone; Bogan River: ◆ Peak Hill, ⬣ Nyngan, ■ Gongolgon.

6.4. Discussion

The results clearly show that water temperature during the irrigation season in the Macquarie River was lower than that found in the Bogan River. Even at the furthest location from the dam (Marebone) the temperature was lower than at the equivalent location in the Bogan River. This corroborates in part the extrapolations of Harris (1997). The fish communities between these two rivers were also significantly different. This is consistent with the expectation that fish communities in highly regulated rivers such as the Macquarie would be substantially different from unregulated rivers, such as the Bogan. Overall the Bogan River had a larger proportion of native species in the total catch than the Macquarie River which is further evidence that native species are more prevalent in unregulated rivers than in regulated rivers. This pattern is consistent with in the results of the New South Wales Rivers Survey (Harris and Gehrke, 1997; Gehrke and Harris, 2000), which demonstrated a greater mean abundance of native fish in unregulated lowland rivers in the Darling region than in regulated lowland rivers.

The somewhat contrasting responses of the proportion of native fish to changes in temperature suggest that the interaction between fish and temperature is a complex one. It would be expected that, if native fish were adapted to warmer water, there would be an increase at least in the proportion of native species present with increasing temperature. This occurred on the Macquarie River where native fish responded strongly to increasing temperature in the presence of a cold water influence. This observation is even more notable considering the Macquarie River had the smallest proportion of native fish and the smallest abundance of fish. In contrast, on the Bogan River, there was no relationship between proportion of native fish and increase in temperature. This suggests that, in the absence of a cold water influence, other factors are probably having a greater influence on the abundance and distribution of native fish than water temperature alone. At the smaller scale of locations within rivers, the relationship between proportion of native fish and temperature increases between seasons but was also very variable.

The magnitude of fish response to riverine thermal gradients in this study is likely to under-represent the response of a healthy, natural fish community because the fish community in both rivers is degraded. Harris and Gehrke (1997) and Gehrke and Harris (2000) reported a general decline in fish communities for most of the inland rivers in New South Wales including the Macquarie and Bogan Rivers. The decline in fish communities highlights the complicating problem of multiple factors impacting on rivers. In New South Wales rivers have been changed by a variety of human disturbances. Therefore, the apparent poor fish communities sampled here are not the result of just one type of disturbance (i.e. cold water pollution) but rather an accumulation and confounding of several disturbances, such as flow alterations, riparian and in-stream habitat degradation, barriers to migration and alien species (Harris and Mallen-Cooper, 1994; Gehrke *et al*., 1995; Driver *et al*., 1997; Gehrke, 1997). In view of the multiple disturbances acting on the fish communities investigated in this study, the thermal responses detected suggest that water temperature has considerable influence on fish distributions during irrigation releases.

In their currently degraded state, fish communities in the Macquarie River may have a diminished capacity to recover if and when the thermal impacts of cold water releases for irrigation are removed. Confounding effects of multiple disturbances have posed difficulties for other field-based studies of riverine fish communities. For example, coastal fish communities in the Hawkesbury-Nepean River system are affected by nutrient enrichment (Growns *et al.* 1996), bank degradation and removal of riparian vegetation (Growns *et al.,* 1998), and river flow alteration (Gehrke *et al*., 1999), but the causes of specific differences between rivers are difficult to identify because of land use within the catchment, erosion and sand movement within tributaries, and episodic disturbances such as bush fires and storm runoff (Gehrke *et al*., 1999).

Sampling for the present study was conducted at a spatial scale of 1-2 km for each site, whilst locations were several hundreds of kilometres apart. In addition to the large scale changes in fish communities in the Macquarie River reported in this study, some effects of cold water pollution on fish may be manifested at smaller spatial scales. For example, Torgersen *et al*. (1999) examined the distribution of salmon and thermal refugia at several spatial scales. At the smaller scale of pools and riffles they found salmon distribution and cold water temperatures were strongly correlated with reach-levels in warm streams. Pool habitats were preferred by salmon, but riffles were used disproportionately to their availability because they contained cooler water. This example demonstrates the importance of recognising that patterns of abundance and distribution of fish may occur at a number of spatial scales related to habitat use and temperature changes. Future studies of the effects of cold water pollution on fish should therefore consider sampling regimes at several smaller spatial and temporal scales, especially more within-season sampling, related to fish habitat use and temperature change.

Another factor that may influence the ability to detect responses of fish to thermal change is the capacity of fish to recover between irrigation seasons. If cold water pollution reduces survival, spawning and recruitment of fish and causes slower growth then this may result in a reduced ability to respond to improved thermal conditions. Furthermore, the continuous releases from Burrendong Dam for environmental flow purposes, combined with natural decreases in air temperature during winter, allow only limited opportunity to increase water temperatures outside of the irrigation season. Consequently fish in the Macquarie River could be perpetually living in a colder environment than normal. Jobling (1994) reports that fishes' thermal characteristics will change with time when they experience temperature changes over a long period. Acclimation to lower temperatures may take several weeks and fish may gradually loose their tolerance to higher temperatures. If fish in the Macquarie River have become partially acclimatised to the present cold temperature regimes over successive irrigation seasons and depressed temperatures in non-irrigation seasons, then they may acquire marginally lower temperature tolerances than conspecifics from rivers not affected by cold water pollution. This concept is partially supported by the different seasonal responses between fish communities in the Bogan and Macquarie Rivers.

It is predicted that fish communities will move further downstream away from the cold water influx to find warmer water. This should result in an increase in abundance and diversity of fish at the furthest location (Marebone) sampled downstream on the Macquarie River. However, Marebone did not have a significant increase in fish abundance but did have an increase in the proportion of native species. Fish abundance may be reduced as a result of the combined effects of other disturbances within the catchment, which limit the ability of fish to respond to thermal gradients. Low fish abundance also creates a need for increased sampling effort to increase the amount of data on fish distributions and abundance to detect ecologically significant responses.

7. CONCLUSIONS AND RECOMMENDATIONS

7.1. Conclusions

The experiments tested explicit hypotheses relating fish responses to cold water pollution. Juvenile silver perch exhibited clear negative responses to cold water pollution in their growth, survival and re-distribution. There was a negative trend in their activity patterns in cold water, although it was weaker than other responses. Generally, juvenile silver perch grow more slowly and have lower survival in cold water compared to warm ambient temperatures, and avoid cold water when offered a choice of water temperatures. Juvenile Murray cod on the other hand showed less clear negative responses to cold water. Growth and survival did not differ between cold and warm water treatments, but this result may have been caused by the fish failing to feed, the shorter duration of the experiment compared to the silver perch experiments, or the possible onset of disease. However, Murray cod did respond to a thermal gradient in a similar manner to silver perch by avoiding cold water.

Fish communities downstream of Burrendong Dam are seriously degraded, and showed limited responses to changes in water temperature during the irrigation season when unseasonably cold water was released from the dam. Observed responses included an increase in the proportional abundance of native fish downstream during the irrigation season, and a different seasonal response in the fish community between seasons compared to the Bogan River fish community. The limited response was surprising considering that water temperatures in the Macquarie River were substantially lower during the irrigation season at sites within 300-400 km of the dam than at sites in the Bogan River. However, there is strong evidence that the fish community in the Macquarie River is seriously degraded, and is affected by multiple disturbances that provide little opportunity for surviving species to recover during brief periods of warmer water temperatures.

There have been no other studies in Australia which have tested experimentally the effect of cold water pollution on native fish. Although it has been noted before (e.g. Lake, 1967; Koehn *et al.* 1996) cold water pollution has only relatively recently become an issue in Australia (Harris, 1997). Studies done overseas have concentrated more on determining the patterns of thermal changes in rivers and any corresponding changes in fish abundance and distribution (e.g. Scheller *et al*., 1999; Petersen and Rabeni, 1996; Torgersen *et al*., 1999). Ward (1985) reviewed the thermal characteristics of rivers and streams in the Southern Hemisphere and concluded that they were only different from northern hemisphere rivers by degree rather than in kind. He briefly touched on the changes made to thermal regimes for rivers in Australia as a result of human development but did not explicitly note any cold water pollution below impoundments. In Australia Koehn *et al.* (1996) documented the long term changes in the fish and macroinvertebrate fauna communities downstream of Dartmouth Dam. They found that native species of fish had been substantially reduced and the composition of the macroinvertebrate fauna had changed and was dominated by a reduced number of species. Apart from this study little other work has investigated the potential affects of unseasonal cold water influxes into rivers.

The results of this study provide valuable insights on the effects of cold water pollution on native juvenile fish. However, a number of important knowledge gaps remain to be investigated. For example, it is possible that slow growth in juvenile fish retards their maturation and subsequently reduces recruitment to breeding populations. The existence and value of thermal refugia is not known. These habitats may provide opportunities for some fish to compensate for retarded growth and maturation and so remain viable members of the adult population. The direct effects of thermal pollution on recruitment rates and other aspects of population dynamics have been alluded to in this study, but are yet to be demonstrated in wild populations. A better understanding of these aspects is needed to protect critical processes and life history stages from adverse effects of cold water releases.

The temporal and spatial scales at which cold water pollution occurs also require further investigation. This study investigated thermal patterns at one large temporal scale (seasons), and two large to medium spatial scales, i.e. sites (kilometres) and river (hundreds of kilometres). However, releases of water from dams, even during an irrigation season, can undergo large daily and/or weekly changes. For example, water was released over the spillway of Burrendong Dam during the summer of 1999 when the dam was 120% full. These spillway releases were usually greater than the volume released from the valves and would last as little as a few days or as long as a number of weeks. This produced fluctuating water temperatures and flow down the Macquarie River. Fish populations may reflect changes in temperature variability if fish are sensitive to these short term changes. Underwood (1989) pointed out that populations will respond differently to disturbance depending upon the type of disturbance and the resilience of the species to that disturbance. Understanding this variability will enable better management plans to be developed.

This study has demonstrated that cold water pollution has significant effects on juvenile native fish. Furthermore, because physiological temperature – rate relationships of poikilotherms, are relatively constant in both experimental and natural conditions, the responses described are likely to occur in other rivers affected by cold water pollution. It is the degree of response that is likely to differ between experimental and natural conditions because of the combined action of other factors in riverine environments that cannot be experimentally controlled. It is important that these results are kept within their context and scope. Rivers are highly complex environments and human activities have done much in changing them. Most large inland rivers in New South Wales have been impacted by development in numerous ways which encompass such things as riparian habitat degradation, land use alterations, hydrological and water quality changes (Gehrke and Harris, 1996). The importance of cold water pollution in relation to other perturbations in rivers is not known, and combined effects of disturbances are likely to create interactions that may lead to unpredictable outcomes in individual rivers. This point is reinforced by the present study in which only small numbers of fish were collected in the Bogan and Macquarie Rivers. The small catches indicate that fish communities are in decline for reasons in addition to cold water pollution.

It should be realised that in rivers suffering multiple disturbances, reducing only one disturbance may have limited benefits. But the benefits of reducing several disturbances are likely to be multiplicative, so that the cumulative benefits of improved management of multiple disturbances are likely to be greater than the benefits resulting from reducing any single disturbance. Therefore, it would be prudent that a follow up of this study concentrated on issues that expanded and developed the initial findings. These are outlined in the recommendations below.

7.2. Recommendations

1. Acknowledge that cold water pollution is a threatening process impacting on the conservation status of silver perch.
2. Modification of the outlet works of Burrendong Dam is necessary to raise the temperature of water in the river downstream of the dam during irrigation releases. Consideration should also be given to modifying other dams where cold water pollution has been demonstrated.
3. Acknowledge there are several knowledge gaps in our understanding and management of the effects of cold water pollution on the aquatic environment in inland rivers in NSW. Further studies should be conducted in the following areas:
    - impacts of cold water pollution on other native fish species
    - the interrelationships between fish, water temperature and aquatic habitats in the Macquarie and Bogan rivers at a range of spatial and temporal scales to identify the

scales at which temperature effects occur. This will help determine whether cold water pollution has a blanket infiltration throughout the river, or if there are regions of warmer water that are used by fish as thermal refuges.

- identify other river systems where cold water pollution might be great and where similar experiments done at Burrendong dam could be repeated.
- existence of warm tributaries that may provide thermal refuges in rivers affected by cold water pollution, and the potential of such habitats for assisting rehabilitation of affected rivers.
- effects of cold water on macroinvertebrates, algae and other trophic levels to provide a broader understanding of temperature requirements to maintain river health and to support fish communities.

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10. APPENDIX

Appendix 1. Native and alien fish species recorded in the Macquarie and Bogan Rivers pre-irrigation and during irrigation, sampled by electrofishing.

Family	Species	Macquarie River								Bogan River					
		Pre-Irrigation				Irrigation				Pre-Irrigation			Irrigation		
		Marebone	Gin Gin	Dubbo	Dripstone	Marebone	Gin Gin	Dubbo	Dripstone	Gongolgon	Nyngan	Peak Hill	Gongolgon	Nyngan	Peak Hill
Ambassidae	*Ambassis agassizi*	0	0	0	0	0	0	0	0	1	2	0	0	0	0
Atherinidae	*Craterocephalus stercusmuscarum*	0	0	0	0	0	0	0	0	55	34	0	1	22	0
Clupeidae	*Nematalosa erebi*	0	0	0	0	0	0	0	0	29	2	0	6	1	0
Cyprinidae	*Carassius auratus**	0	0	0	0	0	0	3	0	0	8	0	84	3	1
	*Cyprinus carpio**	2	0	5	5	0	5	11	8	16	12	1	24	4	2
Eleotridae	*Hypseleotris spp.*	0	0	0	0	0	0	0	0	0	0	281	4	23	41
	Philypnodon grandiceps	0	0	0	18	0	0	0	0	0	0	0	0	0	0
Melanotaeniidae	*Melanotaenia fluviatilis*	0	5	0	0	0	0	0	0	1	0	0	2	0	0
Percichthyidae	*Maccullochella peelii*	0	0	0	0	1	0	1	0	0	0	0	0	0	0
	Macquaria ambigua	0	0	1	0	1	0	1	0	1	0	0	0	0	0
Percidae	*Perca fluviatilis**	0	0	0	0	0	0	0	1	0	0	0	0	0	0
Retropinnidae	*Retropinna semoni*	21	10	0	0	0	8	15	19	0	2	64	0	0	0
Total Individuals		23	15	6	23	2	13	31	28	103	60	346	121	53	44

* Denotes alien fish species

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