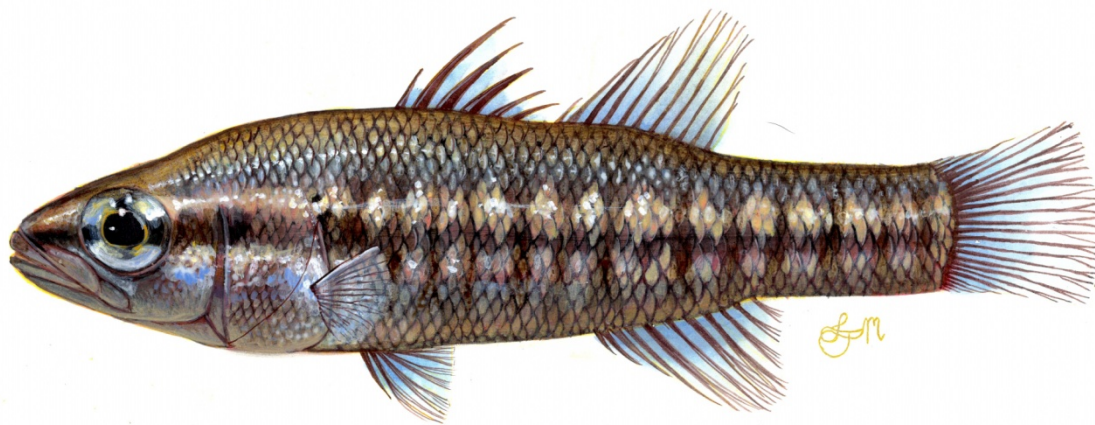


# Migration patterns of fishes of the Blackwood River and relationships to groundwater intrusion



Report to the Department of Water, Government of Western Australia

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**Frontispiece: Balston's Pygmy Perch by Lyndsay Marshall**

## SUMMARY

The Blackwood River catchment is one of two in the Southwest Coast Drainage Division to house all eight freshwater fishes endemic to the region and is therefore of high conservation importance. However, salinisation of the upper catchment has led to substantial range reductions of freshwater species downstream to the largely forested region; where fresh groundwater intrusion by the Yarragadee and Leederville aquifers is greatest. This study represents the only long-term and comprehensive monitoring of freshwater fish populations in the south-west of Western Australia, and consisted of 27 monitoring events between October 2005 and September 2009; which provided information on spatial and temporal movement patterns and identified indicator species of adequate groundwater intrusion.

The overall key implication of this study is that it demonstrates, for the first time, that groundwater plays an important role in maintaining relictual fish fauna in a major river system of this region. This study identifies two species that are appropriate as indicators of river connectivity and in the setting and monitoring of Ecological Water Requirements (EWRs) for this river in light of groundwater extraction, increasing salinisation and reduced rainfall (and thus surface water run-off and groundwater recharge) as a consequence of climate change.

The study specifically identifies Milyeannup Brook (one of two permanently flowing tributaries due to groundwater intrusion) as being of key conservation importance as it houses the only breeding population of the EPBC listed (*Vulnerable*) Balston's Pygmy Perch *Nannatherina balstoni*, and also housed all the other freshwater fishes of the river which were shown to use the system to varying degrees. Microhabitat utilisation by this species within this system during baseflow conditions demonstrated the importance of pool habitats with the Balston's Pygmy Perch found to only occupy the downstream <1600m of permanent habitat during March (baseflow). To maintain this baseflow population, it is crucial that this groundwater discharge is maintained in Milyeannup Brook.

In the main channel of the Blackwood River, the study found a strong relationship between the upstream movement of Freshwater Cobbler *Tandanus bostocki* through riffle zones and discharge during the baseflow period, i.e. March in 2006, 2007, 2008 and 2009. The species was found to undergo large localised movements in the main channel of the Blackwood River that were variable both spatially and temporally. Movements during low flow periods (i.e. highest proportional contribution by groundwater to total flow) were best explained and highly correlated with amount of discharge. It is proposed these movements are probably related to feeding rather than spawning activity as large numbers of small, immature individuals (the study found the females of the species matured at ~172 mm Total Length (TL)) were recorded moving through the riffle zones. Furthermore, by examining the reproductive biology of the species, peak spawning was shown to occur from October to December (i.e. outside the baseflow period).

Subsequent modelling of upstream movements of Freshwater Cobbler over two riffle zones during the driest month (i.e. March) determined that the level of discharge and subsequent riffle depths that would preclude upstream passage by the species were 381.5 l/sec (0.18 m depth) and 101.9 l/sec (0.05 m depth), for the riffles downstream and upstream of the major groundwater discharge zone, respectively. The significance of riffle access to sustaining the population requires further research, however, it is the largest bodied fish of the river and obviously utilises these riffle habitats in large numbers during baseflow. Therefore, if baseflow discharge maintains adequate depth on these riffle zones such that this species is able to access them, then it could be assumed smaller bodied fishes could also access or negotiate them. It

is therefore proposed (along with ensuring the sustainability of the Balston's Pygmy Perch in Milyeannup Brook) that this species should become an indicator of ecological river connectivity during baseflow and be incorporated in monitoring the adequacy of determined EWRs of the river. Furthermore, in terms of an ecological trigger, the rate of future groundwater extraction from the Leederville and Yarragadee Aquifers should not exceed that which will continue to enable this species to access these riffle zones during the baseflow period or lead to a reduction in the baseflow stream length in Milyeannup Brook.

These data represent a comprehensive baseline of fish communities in arguably one of the region's most important river systems, and highlight the value in long term monitoring of a diverse range of aspects relating to the ecology of these fishes. These findings have considerable implication for setting and monitoring Ecological Water Requirements of this and other rivers in this region; particularly in light of regional groundwater extraction pressures and reduced rainfall due to predicted climate change.

### **Recommendations and future research**

- Localised movement patterns of the Freshwater Cobbler through riffle zones in the Blackwood River should continue to be monitored annually during baseflow (March) in order to further validate the models.
- In order to more fully understand the significance of riffle zones to the viability of this species in the Blackwood River, a tracking program (radio tracking and PIT tagging) at key riffle zones should be undertaken in order to determine fine-scale habitat usage of this species during baseflow. Furthermore, the feeding ecology and specific spawning behaviour of the species should also be investigated.
- Following from the above recommendations, Freshwater Cobbler access to riffle zones during baseflow periods could be regarded as a key ecological trigger in monitoring the adequacy of groundwater discharge from the Yarragadee Aquifer.
- The level of groundwater extraction from the Leederville and Yarragadee Aquifers should be set to allow continued access by Freshwater Cobbler to these riffle zones during the baseflow period.
- Milyeannup Brook is the key tributary from a freshwater fish conservation perspective and ongoing biannual monitoring of its fish communities should occur. This monitoring should take place during baseflow (March) to ensure adequate groundwater maintained habitat is available to sustain the *EPBC Act* listed Balston's Pygmy Perch and during the major downstream migration of its juveniles during November to monitor recruitment rates of this crucial population.
- It is recommended that future groundwater abstractions should only occur at levels that do not reduce the amount of baseflow in Milyeannup Brook from its minima of 1600 m from the Blackwood River in 2007.
- Significant decline of the Balston's Pygmy Perch in terms of the baseflow population abundance in Milyeannup Brook (found to be restricted to <1600 m from the confluence of the Blackwood River) or its annual recruitment success should be a key trigger in determining adequate levels of groundwater discharge from the Yarragadee Aquifer.
- Both acute and gradual salinity tolerances of all species of the Blackwood River require determination in order for predictions to be made on the long term viability of these species in this

river and other secondarily salinised systems in south-western Australia in light of predicted changes in salinity levels.

- Further genetic studies should occur on the Balston's Pygmy Perch and Mud Minnow using greater sample sizes. Furthermore, studies are also required for other endemic freshwater fishes of south-western Australia in order to determine the genetic variability between populations as this information has direct management implications in terms of prioritising populations for conservation.

## EXTENSION OF RESULTS

### **Major reports (excluding quarterly progress reports)**

Beatty, S., Morgan, D., McAleer F., Koenders A. and Horwitz P. (2006). *Fish and freshwater crayfish communities of the Blackwood River: migrations, ecology and the influence of surface and groundwater*. Centre for Fish and Fisheries Research, Murdoch University Report to South-west Catchments Council and Department of Water, Western Australia.

Beatty, S.J., McAleer, F.J. and Morgan, D.L. (2008). *Interannual variation in fish migration patterns and habitats of the Blackwood River and its tributaries: annual progress report*. Centre for Fish and Fisheries Research, Murdoch University Report to Department of Water.

### **Oral presentations (conferences, workshops and forums)**

Beatty, S.J.\*, Morgan, D.L., McAleer, F.J., Koenders, A. and Horwitz, P. (2007). Fish and freshwater crayfish communities of the Blackwood River: migrations, ecology and the influence of surface and groundwater. Oral presentation. *Australian Society for Fish Biology, Annual Meeting, Canberra, A.C.T. Australia*.

Beatty, S.J.\*, Morgan D.L. and McAleer F. (2008). Groundwater dependent fish communities in the Blackwood River: understanding migration patterns and the influence of salinisation and river connectivity. Oral presentation. *Australian Society for Limnology. Annual Conference, Mandurah, Australia*.

Beatty, S.J.\*, Morgan, D.L., McAleer, F.J. Fish and freshwater crayfish communities of the Blackwood River: migrations, ecology and the influence of surface and groundwater. *Conservation Council of Western Australia: Scientific forum on the environmental issues surrounding the extraction from the Yarragadee Aquifer*. Perth, May 2006.

Beatty, S.J.\*, Morgan, D.L., McAleer, F.J. Fish and freshwater crayfish communities of the Blackwood River: migrations, ecology and the influence of surface and groundwater. *Conservation Council of Western Australia: Scientific forum on the environmental issues surrounding the extraction from the Yarragadee Aquifer*. Nannup, Western Australia, May 2006.

Beatty, S.J.\*, Morgan, D.L., McAleer, F.J. Fish and freshwater crayfish communities of the Blackwood River: migrations, ecology and the influence of surface and groundwater. *Royal Society of Western Australia. Scientific forum on the environmental studies on the extraction from the Yarragadee Aquifer*. Scitech, West Perth, May 2007.

Beatty, S.J.\*, Morgan D.L., McAleer, F.J. Fish and crayfish communities of the Blackwood River: migrations, ecology, and influence of surface and groundwater. *Western Australia Naturalists Club Lecture Series*. Star Swamp Community Centre September 2007.

Beatty, S.J.\*, McAleer F.J. and Morgan D.L. Overview of the relationships of fish migrations to groundwater and surface in the Blackwood River *Workshop on the Ecology of fish and crayfish in the Blackwood River: relationships to groundwater and surface flows*. Environmental Technology Centre, Murdoch University, Perth, September 2007.

Phillips N.\*, Chaplin J., Morgan D. and Beatty S. The evolutionary significance of Balston's Pygmy Perch and Mud Minnow populations in the Blackwood River. *Workshop on the Ecology of fish and crayfish in the Blackwood River: relationships to groundwater and surface flows*. Environmental Technology Centre, Murdoch University, Perth, September 2007.

McAleer F., Beatty S. and Morgan D. Biology of Freshwater Cobbler in the Blackwood River. *Workshop on the Ecology of fish and crayfish in the Blackwood River: relationships to groundwater and surface flows*. Environmental Technology Centre, Murdoch University, Perth, September 2007.

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## Background

The Southwest Coast Drainage Division has the highest rate of endemism of freshwater fish of any division in Australia (Allen *et al.* 2002). Until relatively recently, there was little information available on the freshwater fishes in the Blackwood River (the largest by discharge in the region), with most fish related research restricted to detailing the estuarine fauna (see Lenanton 1977, Hodgkin 1978, Valesini *et al.* 1997). Morgan *et al.* (1998) provided distributional data (40 sites) on the fishes in the lower reaches of the Blackwood River and a major tributary, the Scott River, and Morgan *et al.* sampled sites from the upper and lower catchment. This research found that the Blackwood River catchment is one of only two to house all eight species of freshwater teleost that are endemic to south-western Western Australia (Morgan *et al.* 1998, 2003). Of the fish species known from the Blackwood River catchment: four are listed on the Australian Society for Fish Biology's List of Threatened Fishes, while one, Balston's Pygmy Perch has recently (2006) been listed as *Vulnerable* under the *EPBC Act 1999*, and, along with the Mud Minnow is also listed as Schedule 1 by CALM under the *Wildlife Conservation Act 1950*.

Endemic fishes of south-western Australia have undergone massive reductions in their overall range over the last 100 years (Morgan *et al.* 1998), largely as a result of modification of habitats, with salinisation of the major catchments a key threatening process. Morgan and Gill (2000) demonstrated that for the various fish habitats in the lower south-west region that there were significant differences in the fish fauna associated with salinised systems, compared to fresh habitats. For example, salinisation throughout both most of the upper catchment and the main channel of the Blackwood River has led to a decline in the range of many of the salt-intolerant fishes and much of the upper catchment and main channel is dominated by salt-tolerant species (Morgan *et al.* 2003). This impact has undoubtedly occurred in rivers in this region as secondary salinisation (associated with wide-scale clearance of vegetation for semi-intensive agriculture) resulting in only ~44% of flow in the largest 30 rivers in the south-west of Western Australia being fresh; the remainder being brackish or saline (Mayer *et al.* 2005). The levels and trends on stream salinity and salt loads are variable by catchment with those rivers with catchments originating in cleared, lower rainfall areas (<1000mm) generally having fresher flow in lower reaches (due to freshwater inputs from forested streams and groundwater) than in their headwaters (Mayer *et al.* 2005).

Potential reductions in river flow in this region may occur as a consequence of reduced rainfall, groundwater extraction and climate change. For example, average annual rainfall in the south-west of Western Australia has declined 10% since 1970, however the non-linearity between rainfall and stream-flow has contributed to a >50% reduction in average flows into public water supply dams (State Water Plan 2007). The ranges of predicted average annual rainfall declines are: by 2030 between 3-22% and 0-22% for the extreme south-west (i.e. the location of the Blackwood River) and the remainder of the region, respectively (Suppiah *et al.* 2007, who summarised the best-performing 15 models performed for the IPCC 2007 4<sup>th</sup> Assessment Report). By 2070, models predict a range of annual average rainfall decline to be between 7-70% and 0-70% for the extreme south-west and the remainder of the region, respectively (Suppiah *et al.* 2007). This has major implications for the long-term viability of freshwater fishes of the region and direct implications for sustainably managing surface and groundwater resources.

### Ecological importance of groundwater and the Blackwood River

Groundwater has been estimated to account for between 30 and 70% of the world's total freshwater, with surface waters such as rivers containing <0.01% (Freeze and Cherry 1979, Petts *et al.* 1999). Many rivers are classified as groundwater-dependent ecosystems, and are further characterised by the degree of

dependency on groundwater (Boulton and Hancock 2006). Groundwater-dependent ecosystems are complex, often support a relatively diverse fauna and may provide refugia for relictual species, however they vary in their degree of dependency on groundwater to maintain their composition and function (Hattons and Evans 1998, Power *et al.* 1999, Murray *et al.* 2003, Humphreys 2006). Localised areas of groundwater water discharge into streams creates a unique environment known as the hyporheic zone. Characteristics of this region are important in maintaining populations of aquatic species, including fish. For example, the hyporheic zone often provides a thermal refuge for aquatic species by buffering against extreme upper and lower lethal temperatures (Power *et al.* 1999, Hayashi and Rosenberry 2002). The hyporheic zone influences water quality by maintaining flows independent of surface runoff, supplying dissolved oxygen, maintaining stream productivity, and providing habitat and maintaining migratory routes. There are a number of specific examples that document the importance of groundwater to particular species in particular systems, however, Sear *et al.* (1999) considered that the nature of the importance of groundwater is difficult to determine at a regional scale, but should be assessed at a local or catchment level.

The Yarragadee Aquifer groundwater currently discharges into the Blackwood River in the reach just downstream of Layman's Brook to just upstream of Milyeannup Brook (Figure 1). Although the Yarragadee groundwater discharge contributes only ~1% of the 940GLyr<sup>-1</sup> of the annual Blackwood River discharge during the dry months, groundwater from the Yarragadee and Leederville Aquifers contribute to between 30-100% of the discharge depending on the amount of summer rainfall (Strategen 2006). This groundwater discharge effectively dilutes the salinity of the river during dry months; when the tributaries of the river contract or dry completely.

Future reduction in groundwater discharge into the main channel of the Blackwood River is predicted due to allocated water extractions from the Yarragadee Aquifer (Hyde 2006) and reduced recharge from lower rainfall expected from climate change in this region (Hughes 2003). In fact, Golder and Associates (2008) recently found a strong relationship between baseflow in the section of the Blackwood River that this study focuses on and the previous 12 months rainfall. Importantly, they found that this relationship has already changed due to rainfall reductions and extraction.

### **Aims of the study:**

Although information existed on the distribution of fishes in the Blackwood River, no information existed on their movement patterns or habitat use and how these relate to environmental variables; particularly fresh groundwater intrusion.

The overall aim of the study was to relate patterns of fish migrations to prevailing environmental variables in the Blackwood River. Specific aims were to:

- Gather a comprehensive seasonal baseline dataset of the patterns of fish and crayfish movements in the Blackwood River to allow ongoing monitoring of potential biotic changes that may result from predicted alterations to the current hydrological regimes.
- Describe the population demographics and migrations of the fish and freshwater crayfish fauna associated within the tributaries of the Blackwood River within the Yarragadee Aquifer discharge zone (i.e. Milyeannup Brook, Poison Gully and Layman Brook) and compare these to adjacent tributaries that are not fed by this aquifer (i.e. Rosa Brook and McAtee Brook).
- Identify relationships between migration patterns, population demographics and the key environmental variables.

- Identify key indicator fish species for determination and ongoing monitoring of Ecological Water Requirements within areas of groundwater discharge in the Blackwood River and its tributaries.

## **Materials and Methods**

### **Study sites**

#### ***Blackwood River main channel***

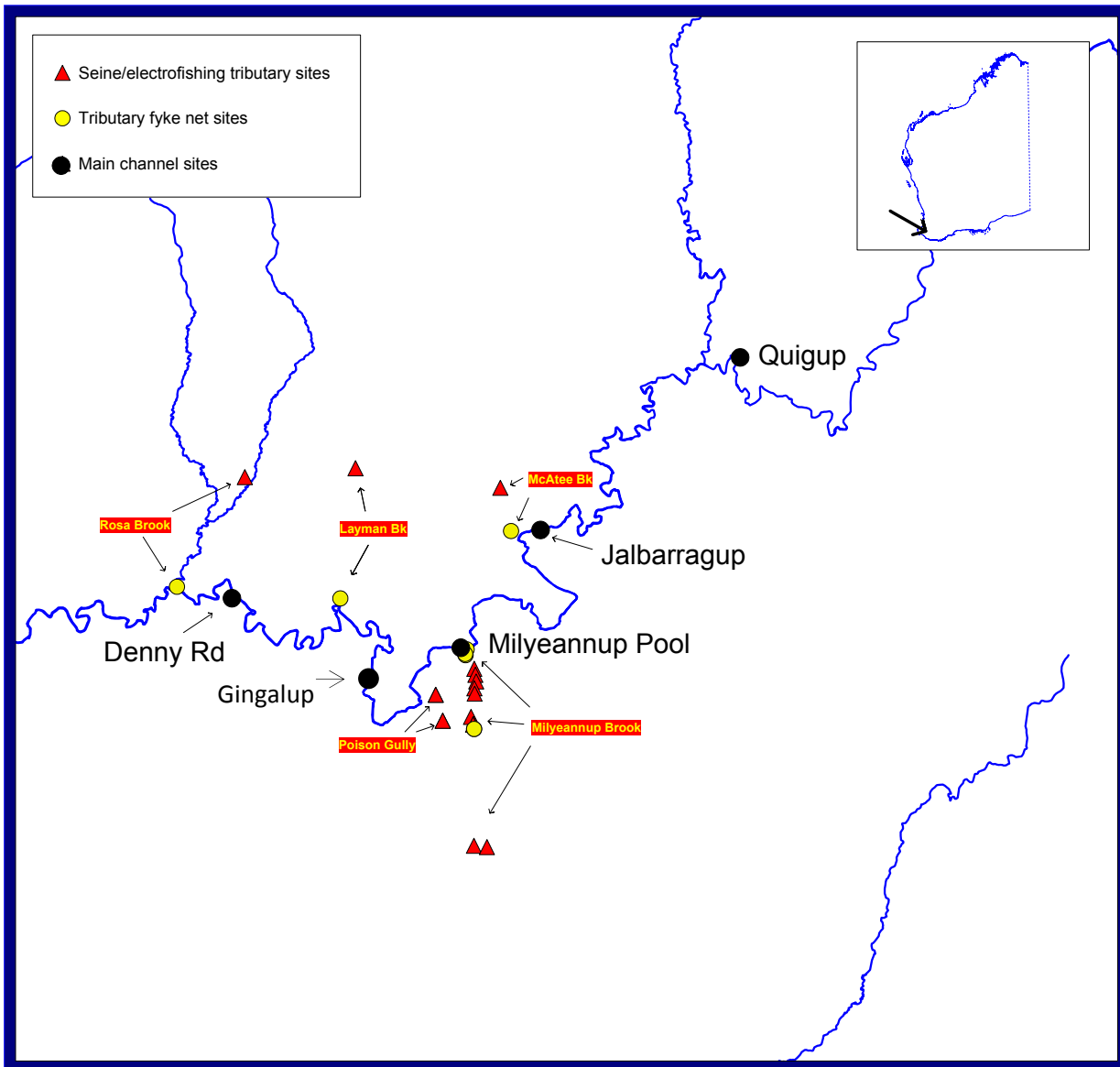
Site selection for determining the temporal changes in population demographics and migrations of the fish and crayfish fauna in the Blackwood River main channel was based on their differing proximities to the major zone of groundwater discharge and to the discharge measurement sites as designated by the Department of Water. For example, in 2005/2006 the fish fauna found within two sites in the Blackwood River main channel that is subjected to the major discharge of ground water (from the Yarragadee or Leederville Aquifers) was compared to two sites upstream of this discharge. In 2007/2008 this was modified to three sites subjected to discharge of groundwater, from either the Yarragadee or Leederville Aquifers, and one site directly upstream of the discharge.

The two sites within the main channel in 2005/2006 that receive groundwater input were immediately downstream of the mouth of Milyeannup Brook (referred to as Milyeannup Pool) (34.0909°S, 115.5661°E) and just upstream of the mouth Rosa Brook (referred to as Denny Road) (34.1081°S, 115.4505°E). Additionally, Gingilup (34.10445°S, 115.51947°E), which is gauged by the Department of Water, was included. The two upstream sites were Jalbarragup Road crossing (34.0421°S, 115.6025°E) and Quigup (33.9736°S, 115.7008°E) in 2005/2006. In 2007/2008 monitoring ceased at Quigup in favour of a compilation of three sites in close proximity; namely Darradup during March 2007, Jalbarragup Road Crossing (as above) from March to November 2007 and Orchid Place in March and April 2008 (see Figure 1).

Sampling was conducted in October, November and December 2005; February, March, June, August and September 2006; March, May, June, August, September, October, November and December 2007 and March and April 2008. Sampling is scheduled to continue until mid 2010.

#### ***Blackwood River tributaries***

A number of tributaries were similarly monitored for comparison of the aquatic fauna with both each other and the main channel. These include Milyeannup Brook, Poison Gully, Layman Brook, Rosa Brook, McAtee Brook and St Johns Brook. Milyeannup Brook and Poison Gully are directly maintained in dry months (summer/early autumn) by groundwater discharge from the Yarragadee Aquifer. Layman Brook receives groundwater discharge from the Yarragadee Aquifer during winter and spring but not summer; when it ceases to flow and usually dries. The temporal changes in the population demographics and migrations of the fish and crayfish fauna within these tributaries were compared with two adjacent tributaries that flow seasonally, i.e. Rosa Brook and McAtee Brook which are within the Leederville Aquifer discharge zone. In addition, St Johns Brook, which is situated within the Leederville Aquifer discharge zone, was seasonally sampled in June and November 2007 and June 2008.



**Figure 1** Sampling sites in the main channel and tributaries of the Blackwood River.

## **Environmental variables**

### ***Water quality monitoring***

The climatic regime of Bridgetown (the nearest centre where long-term data was available) for the study period were obtained for the Australian Bureau of Meteorology. These patterns in rainfall and temperature were compared to long term data to provide indications of variability or consistency of the climate during the sampling period with that typically observed for this region. The prevailing rainfall has particular implications for influencing surface flow regimes of the study sites.

On each sampling occasion at each site, the temperature, conductivity, pH, and dissolved oxygen were measured in the middle of the water column at three locations and a mean ( $\pm 1$  SE) determined. In addition, temperature data loggers (*Tinytag*<sup>TM</sup>) were placed *in situ* in the sampling sites and logged water temperature every three hours. Temperature loggers were downloaded every three months.

### ***Discharge, baseflow riffle profiles and stage-height relationships***

Monthly discharge rates in the Blackwood River during the study period were obtained from the Department of Water, Government of Western Australia gauging stations at Darradup, Gingilup and Hutt Pool (Figure 1). Monthly discharges at the sampling sites were estimated from: the Darradup gauging station for the Jalbarragup site, Gingilup gauging station for the Gingilup site, and those for the riffle sites at Milyeannup and Denny Road from a combination of the instantaneous cross-sectional discharge-depth profiles taken during baseflow months in 2007, 2008 and 2009 (as part of the cross-section profiles, see below), and by interpolating discharges from the other adjacent gauging stations for the remaining sampling months. Instantaneous discharges on each sampling occasion were taken in the following tributaries: McAtee Brook (just upstream of the Yarragadee Aquifer Discharge Zone (YADZ), Milyeannup Brook and Poison Gully (within the YADZ and directly receive Yarragadee discharge), Layman's Brook (within the YADZ but does not receive discharge from the aquifer) and Rosa Brook downstream of the YADZ.

Three baseflow cross-sectional profiles were taken across the riffle zones at Milyeannup and Denny Rd in March and May in 2007, and March and April in 2008; coinciding with sampling of base-flow fish movements at those riffles. The relationships between Water Stage Levels (WSL) and discharge over these riffles were then plotted and a number of models fitted in order to select that which provided the best overall descriptions of the relationships between the variables (i.e. usually both the highest coefficient of determination and significance value).

Using the relationships between discharge and mean upstream movement of Freshwater Cobbler over four consecutive baseflow periods (i.e. March 2006, 2007, 2008 and 2009) at Denny Road and Milyeannup Pool, the discharge and stage heights at each riffle, and the depths across the profiles at different stage heights, estimates of the minimum discharge and riffle zone depths for upstream Freshwater Cobbler movement were determined (see below *Relationship between baseflow hydrology and riffle usage*).

Using the relationships between discharge and mean upstream movement of Freshwater Cobbler over four consecutive baseflow periods (i.e. March 2006, 2007, 2008 and 2009) at Denny Road and Milyeannup Pool, the discharge and stage heights at each riffle, and the depths across the profiles at different stage heights, estimates of the minimum discharge and riffle zone depths for upstream Freshwater Cobbler movement were determined (see below *Relationship between baseflow hydrology and riffle usage*).

## Sampling for fish movements

A number of techniques were employed to examine the fish fauna of the main channel and tributary sites in the Blackwood River. Each method is outlined below, i.e. the use of fyke nets (11.2 m in width, including two 5 m wings and a 1.2 m wide mouth fishing to a depth of 0.8 m, 5 m long pocket with two funnels all comprised of 2 mm woven mesh); seine nets (5, 10 and 15 m nets comprised of 2 mm woven mesh and a 26 m seine net consisting of two 9 m wings of 6 mm woven mesh and an 8 m bunt of 3 mm mesh); 240 and 12 v electrofishers; and crayfish traps during 2005/2006. During the 2007/2008/2009 sampling concentrated on emerging trends in migration patterns and utilised fyke nets.

### ***Blackwood River main channel***

#### *Species migrations*

At the main channel sites (see Figure 1), fyke nets were used to determine temporal trends in species migrations. Fyke nets were set facing upstream, to determine downstream movements of fish, and facing downstream, to determine upstream movements of fish. Each fyke net was set for a period of 72 hours and sampled every 24 hours.

Each fish captured was identified, a sub-sample measured (total length (TL) for fish and orbital carapace length (OCL) for crayfish) to the nearest 1 mm and where possible sexed and released. A subsample of Freshwater Cobbler were retained for analyses of biological indices such as gonadal development and aging as this species was identified as a key indicator species (see Results).

The mean number of each species captured on each occasion was adjusted to account for total number of each species migrating through a section of the river. The total numbers referred to here on in are the actual numbers captured, while the migration figures reflect the adjusted data to show the approximate numbers of fish actually migrating and to thus allow for comparisons between the various riverine reaches.

### ***Blackwood River tributaries***

On each sampling occasion (during stream flow), fyke nets were set over 72h in Milyeannup Brook (two sites). Additionally, Layman Brook, Rosa Brook, McAtee Brook and Poison Gully (Figure 1). At each site, one net was set facing upstream, to capture fish that were moving downstream, while another was set facing downstream (to capture fish moving upstream) and each was checked every 24 hours. As with the main channel site captures, the percent coverage of each set was determined and the catches later adjusted to 100% of the stream width.

The shallow, diffuse nature of Poison Gully required smaller fyke nets. These were used elsewhere, but still consisted of 2mm woven mesh. The same methodology i.e. three, 24h sets was utilised with these modified fykes. Fish species were identified, with a large sub-sample measured for total length (mm TL) before being released. Those not measured were identified and counted to determine total numbers and immediately released. A small sub-sample of native species was retained for biological investigation into the gonadal development (up to ~30 per month) and for genetic analysis (see Phillips *et al.* (2007) for *Nannatherina balstoni* and *Galaxiella munda*).

## Relationships between fish movement and environmental variables

Regression analysis was undertaken for a number key species in the tributaries, and for the Freshwater Cobbler in the main channel (the species identified as an appropriate indicator of river connectivity due to

the apparent relationship to baseflow discharge by Beatty *et al.* (2006)). Freshwater Cobbler movement pattern analysis was also refined via specific sampling of key riffle zones during baseflow and also undertaking complementary hydrological data including cross-sectional profiles and discharges at those monitoring sites (courtesy of the Department of Water, Bunbury). This was aimed at estimating the flow requirements to maintain migratory ability through key riffle zones during base-flows.

### ***Tributaries***

#### *Overall analysis*

The fishes included in analyses were those where adequate migration data were recorded (i.e. utilised the most number of tributaries). These species included the Western Minnow, Western Pygmy Perch and Nightfish. The relationships between the upstream and downstream movement of the above species during the major flow and breeding periods (i.e. the period that all four tributaries were flowing, August to December) and the prevailing environmental variables in Milyeannup Brook, Rosa Brook, McAtee Brook, and Layman's Brook were determined (see Beatty *et al.* (2006) for details of the specific statistical analyses).

#### *Milyeannup Brook baseflow monitoring*

Finer scale monitoring of fish and crayfish populations and environmental variables along Milyeannup Brook was undertaken at those sites previously examined during baseflow conditions in 2006 (see Beatty *et al.* 2006 for the location of the sites in Milyeannup Brook). Sampling took place in March 2007 and 2008 and involved examination of fish and crayfish distributions within habitat types at key sites, and also the measurement of hydrological characteristics within those broad habitat types present (i.e. pool and/or riffles) at each of those sites. This aimed to determine preliminary habitat preferences of each species during baseflow conditions in Milyeannup Brook.

On each sampling occasion at Mil 3 (just upstream of Brockman Hwy), Mil 4, Mil 5, and Mil 6 (limit of surface water in both 2007 and 2008), fish and freshwater crayfish densities were determined in up to three habitat types (pool and riffles where present) at each site using double pass electrofishing with each habitat sectioned off using stop nets. All fish were identified and measured to the nearest 1mm TL (fish) or OCL (crayfish) and a density determined by measuring the area of the habitat between the stop nets. Densities of each species within each habitat in baseflow conditions in 2007 and 2008 were graphically compared.

Depth profiles were taken across three cross-sections and along one longitudinal-section on each occasion. Mean and maximum depths of both section type in each habitat and also each habitat overall was determined and displayed graphically. The instantaneous discharge at each site (where flow was evident) was determined on both occasions via measuring channel width, depths and flows rates (using a hand-held flow meter). Mean water temperature, conductivity, pH, dissolved oxygen, and turbidity were also determined for each site.

In order to determine whether relationships existed between the depth of baseflow habitats in Milyeannup Brook and the density of species occupying those habitats, preliminary linear regression analysis was conducted between these variables and scatter-plots generated.

## **Main Channel**

### *Freshwater Cobbler movements and environmental variables: 2005-2008*

The association between the strength of upstream and downstream Freshwater Cobbler movements and the above key environmental variables were examined. For overall sampling time analysis, all sampling occasions at all sites were included in stepwise regression analysis to determine which of the environmental variables best explained the variation in upstream or downstream Freshwater Cobbler movements in the Blackwood River main channel.

The mean upstream and downstream movements of Freshwater Cobbler in the main channel were determined for the period of lowest flow (i.e. December-April) and their relationship to environmental variables during those periods analysed. Correlation analysis and stepwise multiple regression models were developed to determine which single or combinations of environmental variables during the major movement period best explained the most variation in upstream and downstream migrations of Freshwater Cobbler. For both of the above analyses, Shapiro-Wilk statistical tests for normality were undertaken for each variable and all data were subsequently log-10 transformed prior to analysis. Bi-variate correlations between each environmental variable were calculated (Pearson's correlation coefficient) prior to each stepwise regression analysis to initially examine possible associations between environmental variables. In the subsequent stepwise regression analyses, levels of co-linearity between independent variables were investigated via determining condition indexes and eigenvalues. Mean velocity was consistently highly correlated with discharge and was therefore excluded from the analyses to avoid problems with co-linearity. The more conservative, adjusted co-efficient of determination ( $r^2$ ) were examined in each model as the adjusted values closely reflect the goodness of fit of the model (SPSS, 2005).

### *Relationship between baseflow hydrology and riffle usage*

The above analyses revealed that discharge appeared to be a key variable in influencing upstream Freshwater Cobbler movement during the seasons of lowest flow (i.e. summer and early autumn, see *Results*); the relationship between upstream movement of Freshwater Cobbler over the two riffle zones and hydrology during baseflow was therefore determined. The month with the consistently lowest discharge (i.e. March, see *Results*) was chosen for this analysis in order to provide minimum flow and depth requirements.

The relationship between mean upstream movements of Freshwater Cobbler and discharge at two riffle zones in March 2006, 2007, 2008 and an additional sample in 2009 were determined using regression analysis. Mean upstream movement during each March period was plotted against discharge at those times and a number of models were tested with the highest co-efficient of determination fitted. These models were then rearranged to determine the theoretical discharge level at which zero movement would occur; implying the minimum baseflow required for access to the riffles during baseflow.

Using the zero-movement discharge estimates on each riffle and the WSL and cross-sectional profiles, the minimum depth across each riffle that the Freshwater Cobbler movement would hypothetically cease was determined. This was achieved by first using the non-linear relationships of WSL and discharge at each riffle to determine the stage heights at those zero-movement discharges. These zero-movement stage heights were then plotted on the three cross-sections of each riffle zone to determine the maximum depths (deepest passage) that would be present at those minimum discharges. The shallowest maximum cross-sectional depth was inferred to represent the depth at which Freshwater Cobbler would cease to be able to move upstream over these riffle sections in the Blackwood River.



### *Mark-recapture of Freshwater Cobbler*

In order to investigate whether movements of this species through riffle zones are localised or associated with larger distances, a total of 437 were tagged between October and December 2005 using individually numbered t-bar tags at each main channel site. One each subsequent sampling occasion, site of recapture was recorded for each individual.

### *Reproductive biology*

The spawning period and length at first maturity of Freshwater Cobbler were determined in order to provide evidence of whether movements may be linked to spawning behaviour. The temporal trend in the reproductive biology of *T. bostocki* was determined by retaining monthly samples of females (pooled between sites). These fish were euthanased in an ice slurry, weighed to the nearest 0.01g, their gonads removed, and also weighed to the nearest 0.01g. The monthly gonadosomatic indices (GSI) of mature and immature individuals of both sexes were determined using the equation:

$$\text{GSI}=100(\text{W1}/\text{W2})$$

Where W1 is the wet weight of the gonad and W2 is the wet somatic weight. Ovaries and testes were assigned on macroscopic appearance using the seven stages adapted from Laevastu 1965, namely: I virgin (immature), II maturing virgin/recovering, III developing, IV developed, V mature (gravid), VI ripe (spawning) and VII spent. The lengths at first maturity of female *T. bostocki* were determined by fitting a logistic equation to the percentage contribution made, during the spawning period, by immature and mature fish in each 10 mm length increment.

The logistic equation used to predict length at which females commenced maturation was:

$$P = \frac{1}{1 + \exp[-\ln(19)(L - L_{50})/(L_{95} - L_{50})]}$$

Where  $P$  = the proportion mature,  $L$  = fork length in mm,  $L_{50}$  and  $L_{95}$  are lengths in mm at which 50 and 95% of female fish reach sexual maturity, respectively, and  $\ln$  is the natural logarithm. Females were classified as mature during stages III to VI because they had the potential to spawn, were spawning or had spawned.

## Results and Discussion

### *Water quality and discharge regimes*

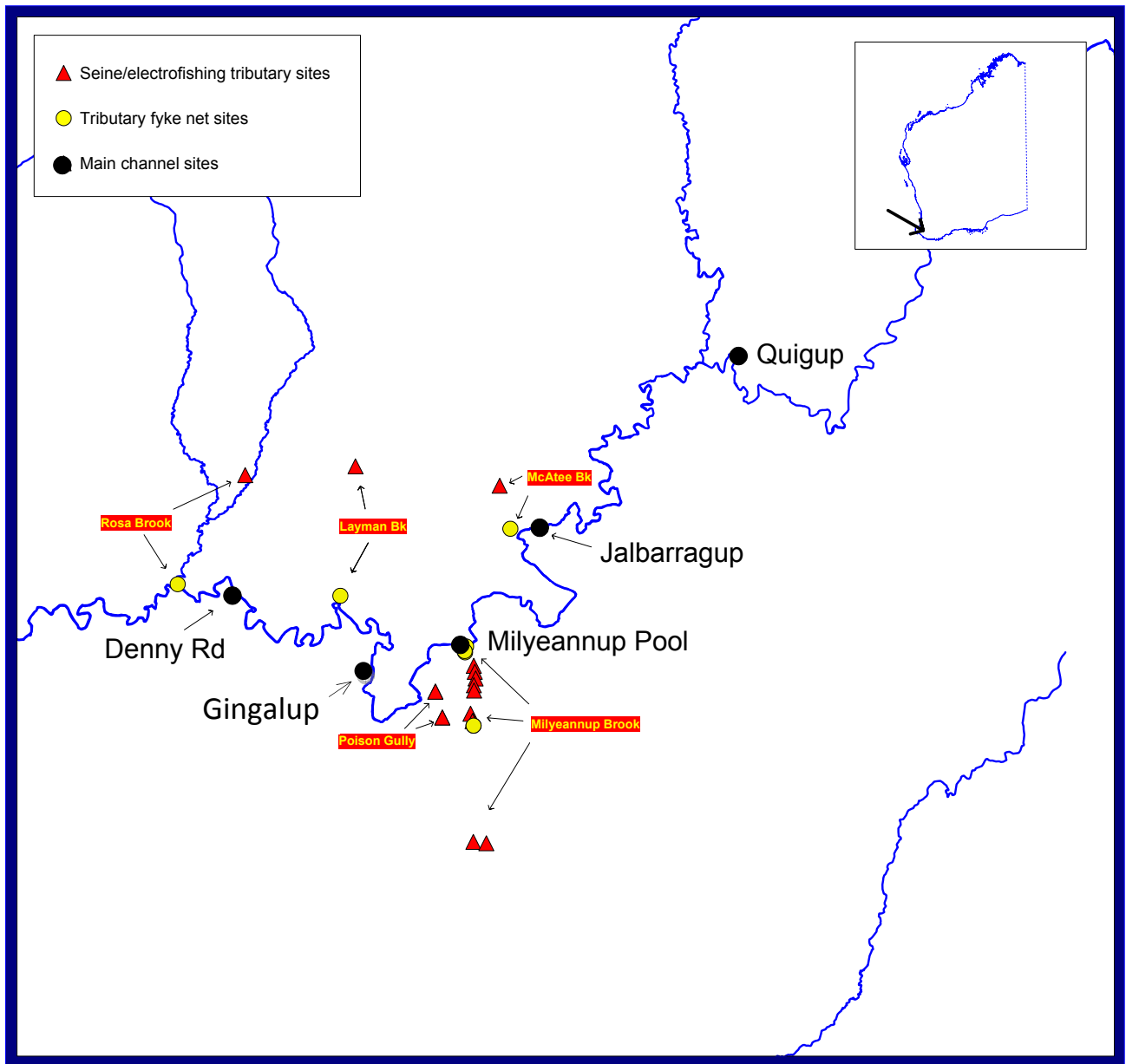
Examination of the climatic conditions in the Blackwood River study area (Figure 1), it is evident that the 2005-2006 sampling period was typified by above average spring 2005 to summer 2006 rainfall with a delayed onset of major winter rainfall in 2006. During the same period in 2006-2007, the reverse occurred with a well below average spring and early summer rainfall followed by variable rainfall in later summer and autumn 2007 (Figure 2). The very low late spring and summer rainfall in the 2007-2008 period was also notable.

The greater discharge during base flow conditions in the main channel sites in early 2006 (i.e. March, see Figures 3-6) compared to those experienced in 2007 and 2008 were probably the result of the above average rainfall between October 2005 and January 2006. This also corresponded with much stronger movement of Freshwater Cobbler during the baseflow conditions in 2006 compared with the subsequent two years (see section on Relationship between Freshwater Cobbler and baseflow discharge).

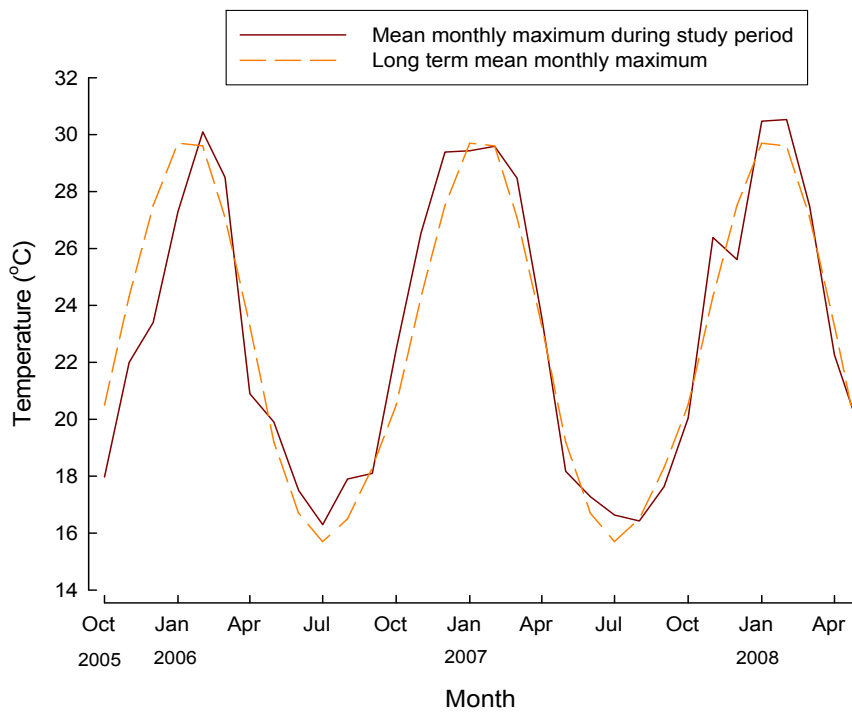
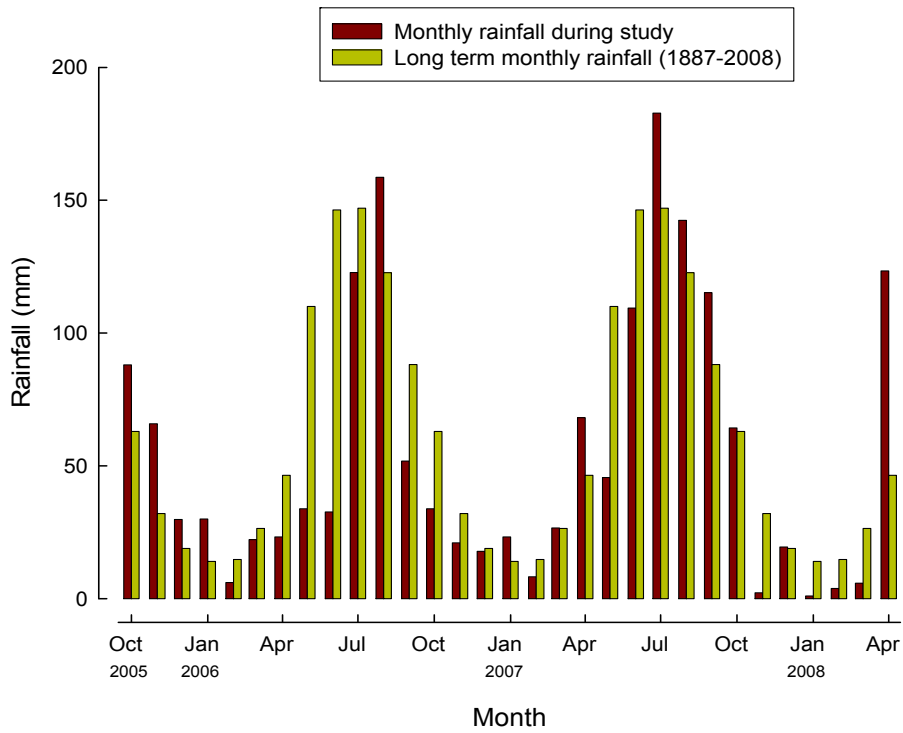
The water temperature traces from the data loggers between 2005 and 2008 clearly indicate that most are highly associated with ambient air temperatures (Figure 8). However, along with the instantaneous water temperatures at the times of sampling in the tributaries (Figure 7), demonstrated that Milyeannup Brook maintained a less variable and lower temperature than the other tributaries (Figures 7 and 8). This is attributed to the input of the Yarragadee Aquifer groundwater that results in the buffering of the water temperature against air temperature fluctuations. That is, this system maintains cooler water temperatures during the baseflow period and is warmer during the winter period (see also section on Milyeannup Brook Baseflow Monitoring). Poison Gully, although also maintaining permanency due to aquifer discharge, does not display this buffering effect; probably due to the streams morphology in that it is a wider, shallower and more diffuse stream line which would increase the influence of air temperature.

Main channel water temperatures upstream and downstream of the major aquifer discharge zone (i.e. Denny Road and Milyeannup Pool, Figures 7 and 8) did not differ during baseflow period as may have been expected. This was probably due to the effect of external heating of the cooler aquifer water as it flows downstream offsetting the lower temperature of the aquifer water.

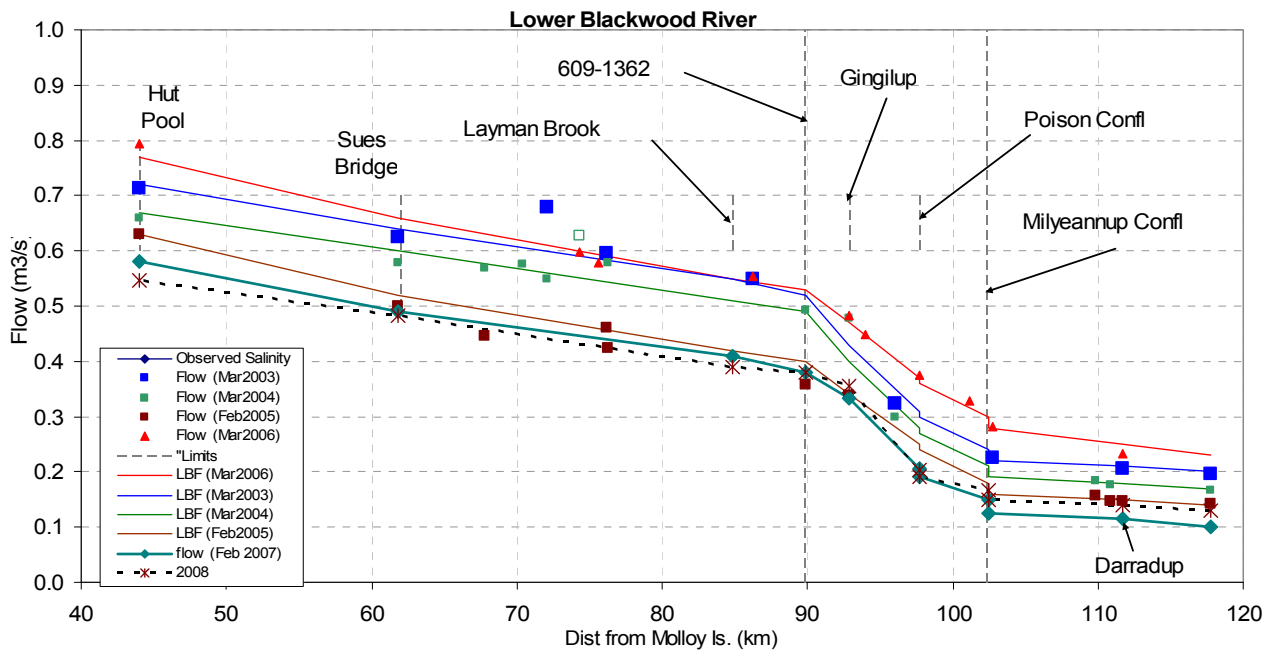
Conductivities in the tributary sites show that Milyeannup Brook and Poison Gully generally remained fresher than the other tributaries in baseflow conditions (see also section on Milyeannup Brook Baseflow Monitoring) (Figure 9). Those tributaries that cease to flow and pool during baseflow (i.e. St John's Brook, Rosa Brook and McAtee Brook) showed elevated salinities during baseflow due to evapoconcentration of salt without fresh groundwater input; however, all remain fresh throughout the year. Although Rosa Brook dries and exhibits elevated salinities at the Denny Road sampling site, further upstream at the confluence of Rosa Brook and Mowen Road permanent flowing water is located. It is recommended that the Mowen Road site is sampled and compared to the Denny Road site as the permanent flow, and presumably lower salinity, provides an important habitat for the Mud Minnow (Morgan *et al.* 2003, 2004).



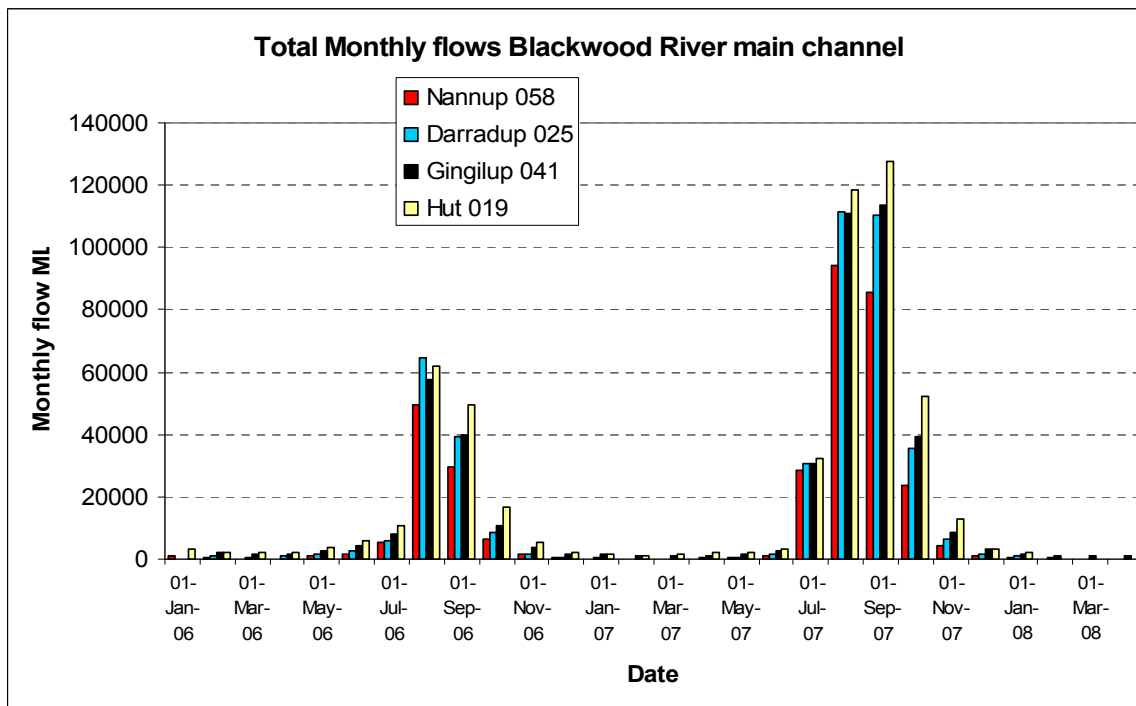
**Figure 1** Sampling sites in the main channel and tributaries of the Blackwood River.



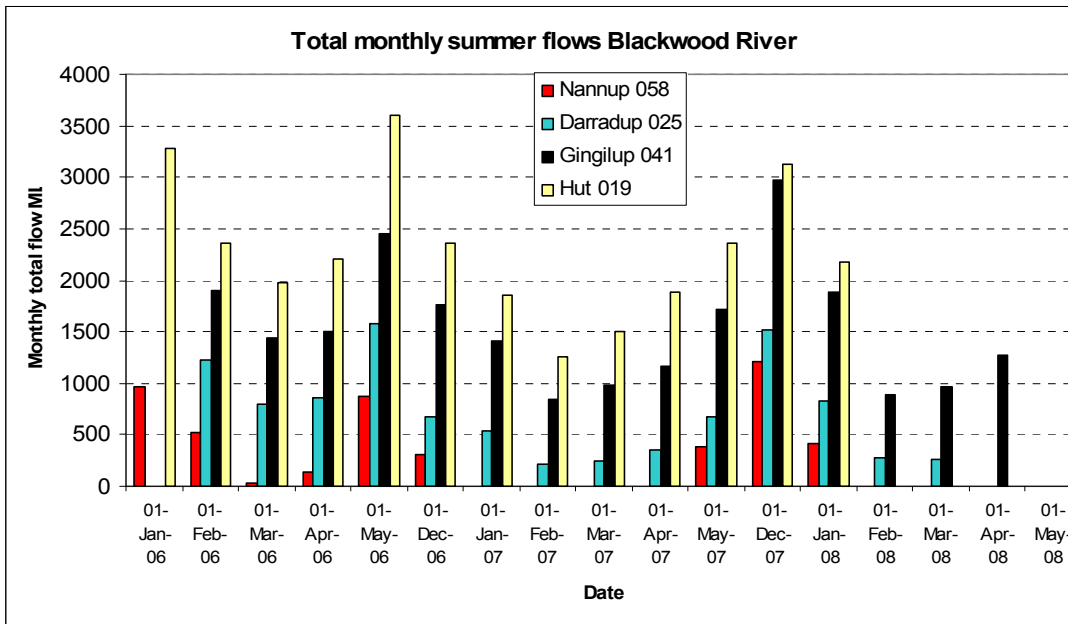
**Figure 2** Mean monthly rainfall (top) and maximum air temperatures (bottom) during the initial period of the study and long term averages (from Bridgetown). Data from Australian Bureau of Meteorology.



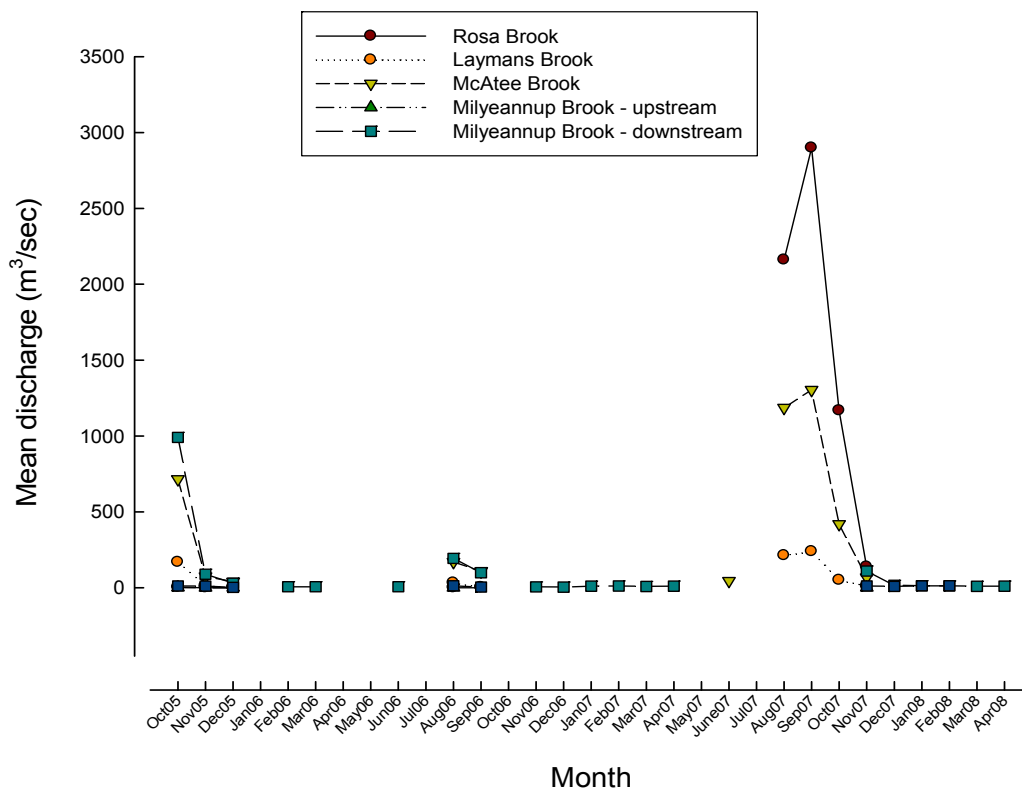
**Figure 3** Consecutive annual baseflow longitudinal discharge traces in the main channel of the Blackwood River in March. N.B. the relatively high baseflow discharge in March 2006 compared to 2007 and 2008. (Figure from Department of Water, Bunbury).



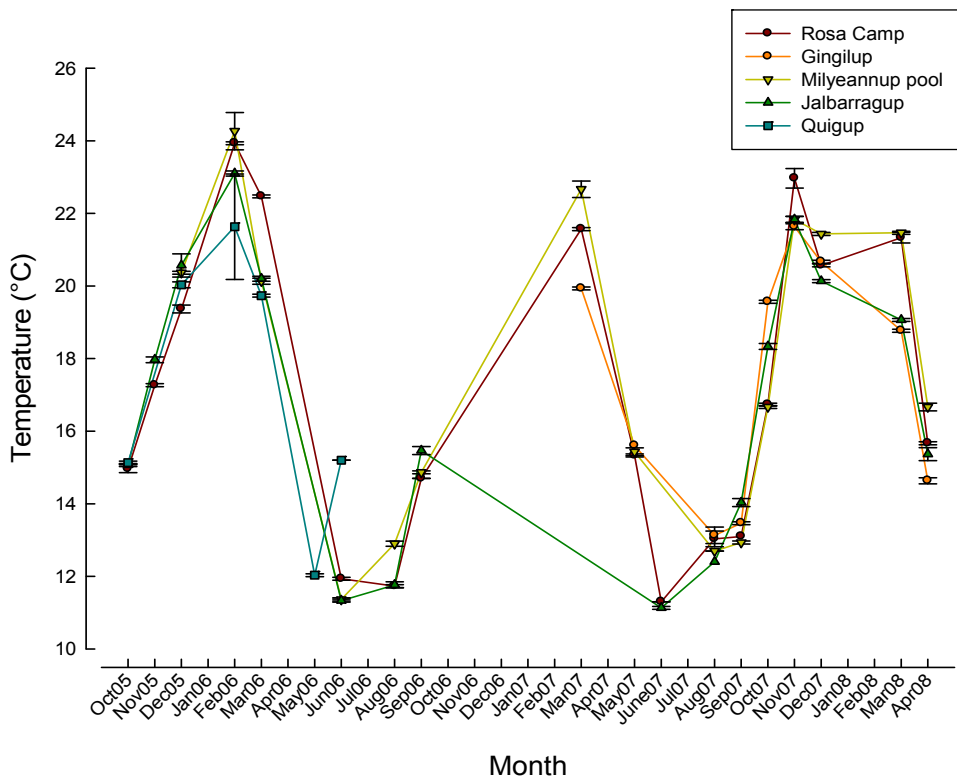
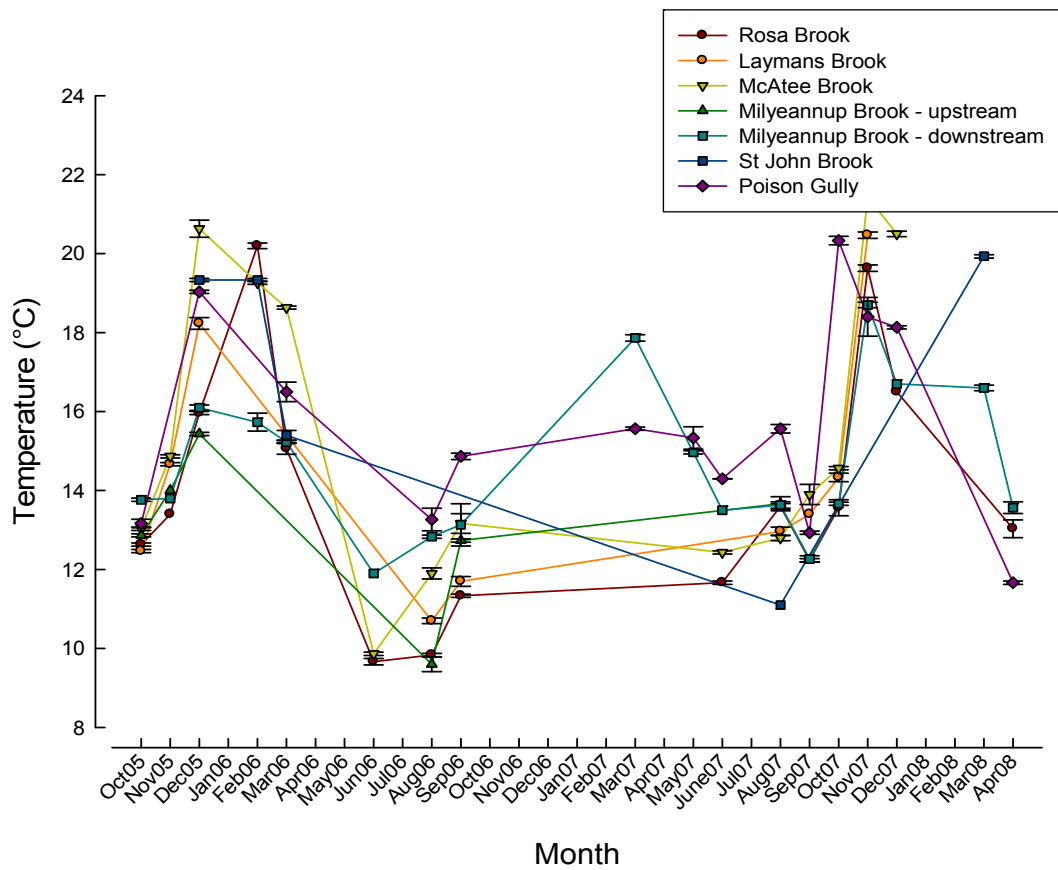
**Figure 4** Total monthly flows at sites in the main channel of the Blackwood River during the majority of the sampling period (Figure from Department of Water, Bunbury).



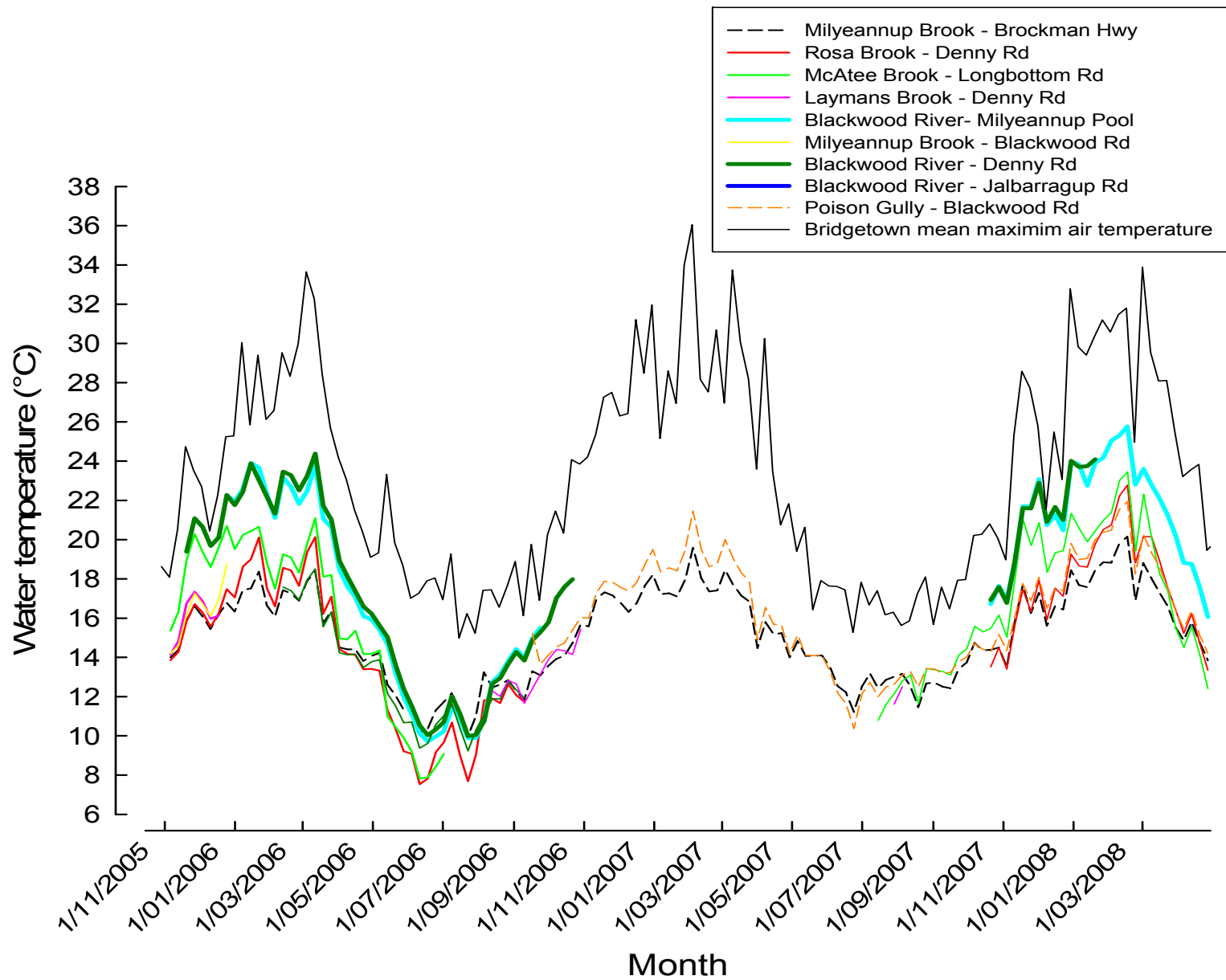
**Figure 5** Total monthly summer flows at sites in the main channel of the Blackwood River (Figure from the Department of Water, Bunbury).



**Figure 6** Mean discharge at tributary sites in the main channel of the Blackwood River.

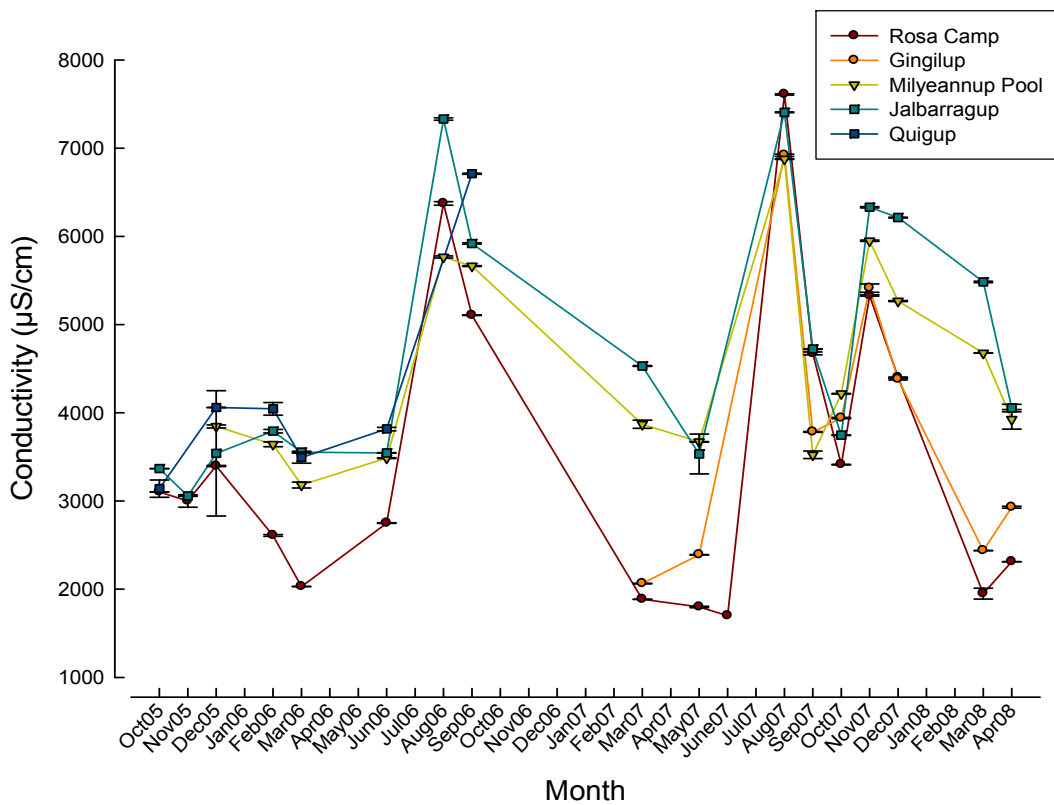
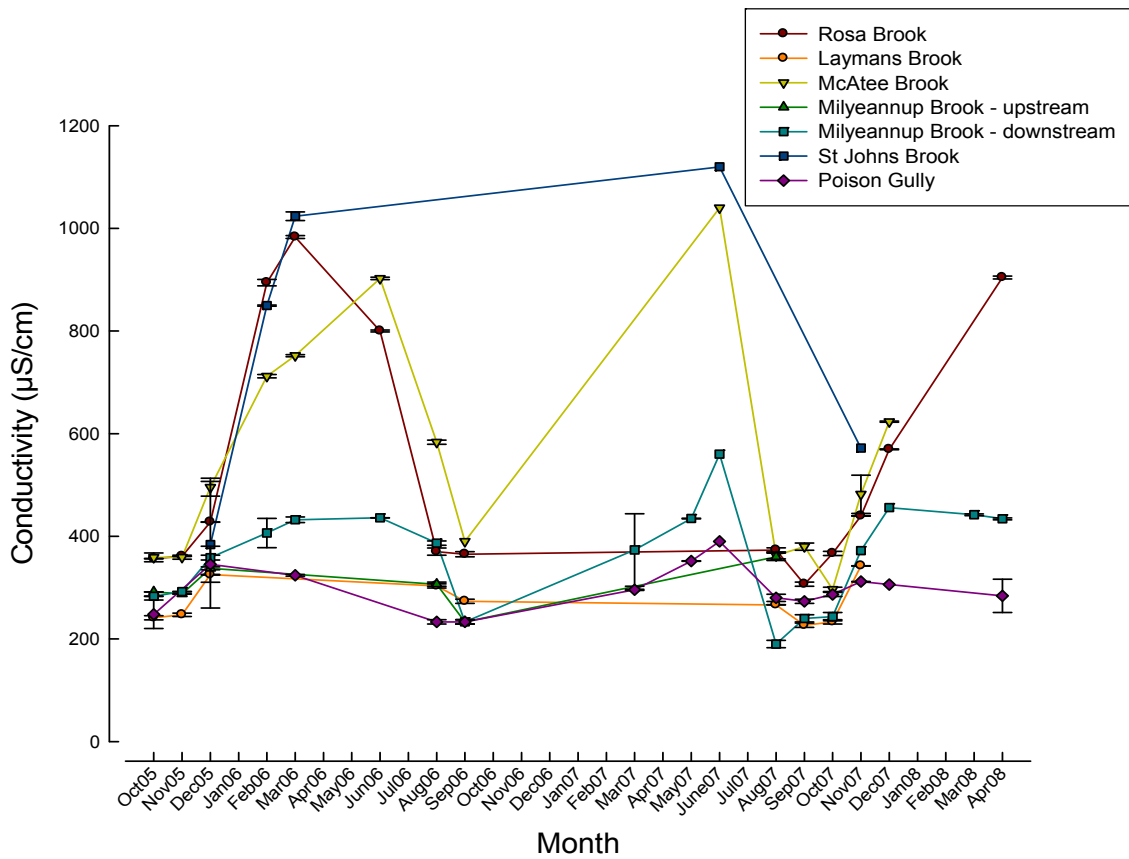


**Figure 7** Mean instantaneous water temperature at sites in the tributaries (top) and main channel (bottom) of the Blackwood River.



**Figure 8** Water temperatures from the data loggers (logged every 3 hours) at sites in the Blackwood River and its tributaries. N.B. the close association of water temperatures in all sites and maximum air temperature (in Bridgetown) and cooler water temperature during summer in Milyeannup Brook.

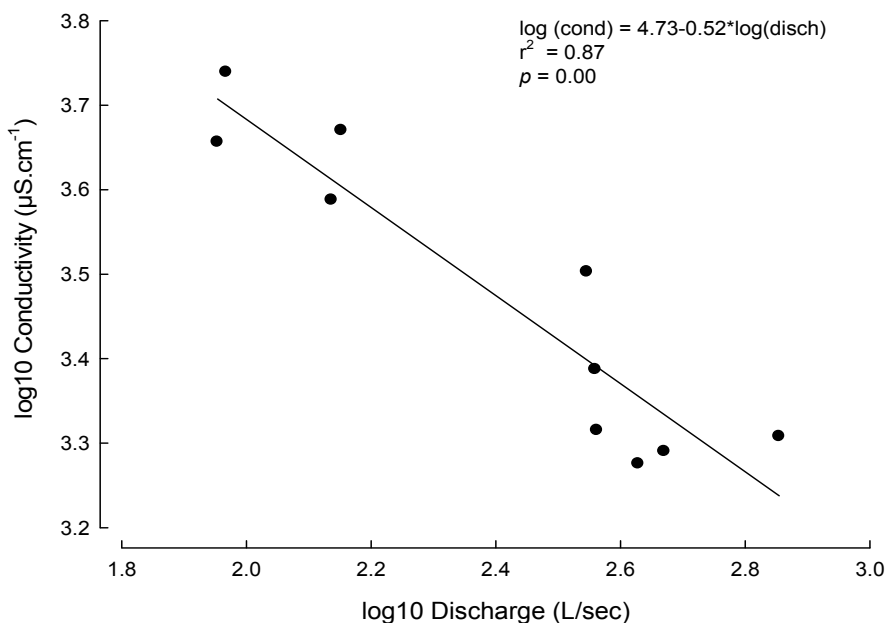




**Figure 9** Mean instantaneous conductivities at sites in the tributaries (top) and main channel (bottom) of the Blackwood River. N.B. fresher conditions at the more downstream sites (i.e. Denny Road and Gingilup) during baseflow periods.

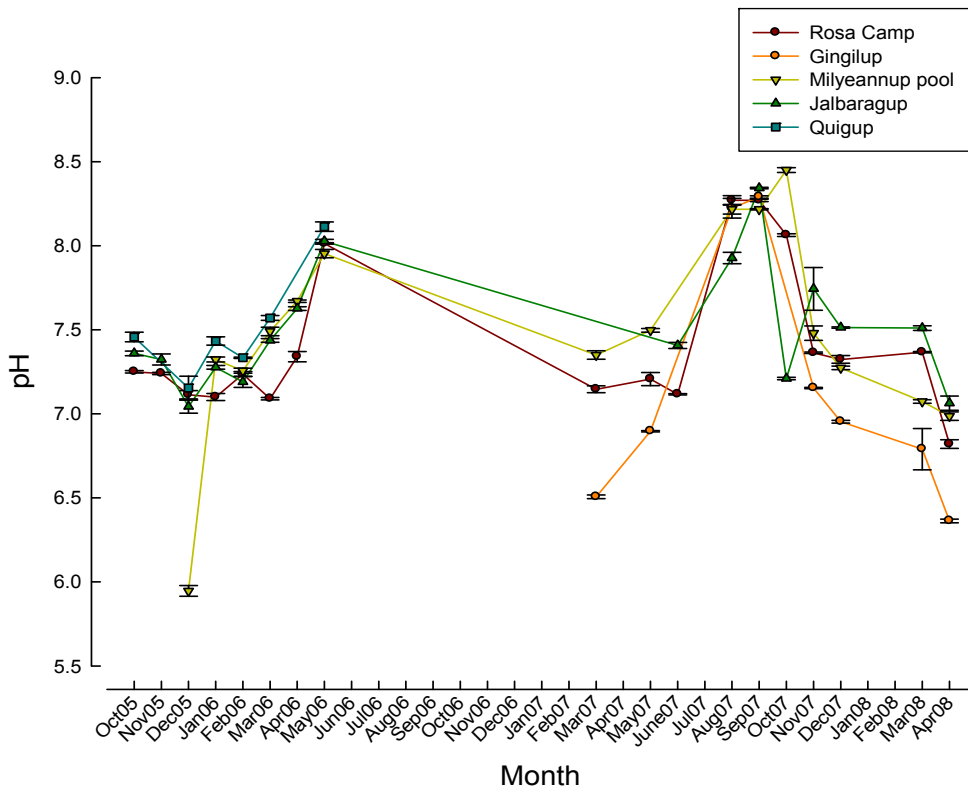
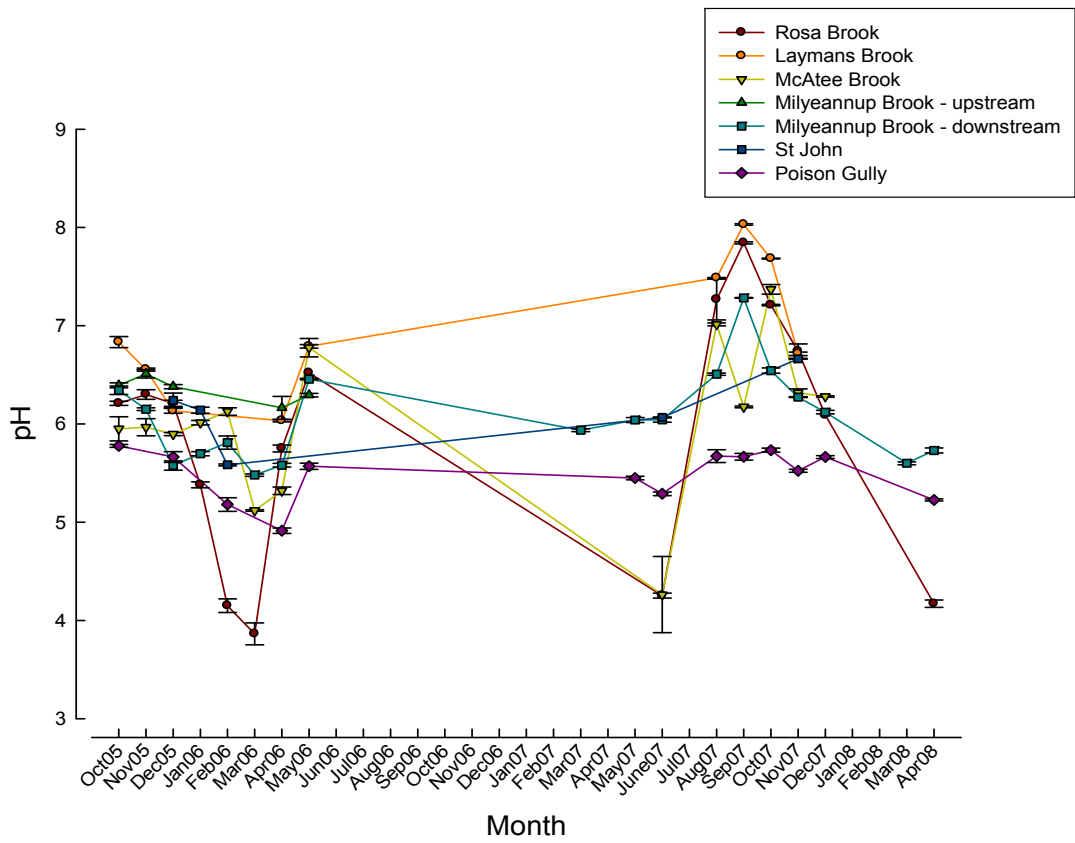
The trends in conductivity of the main channel sites clearly show the effects of the increase groundwater input due to the Yarragadee Aquifer (see Figures 8, 9 and 10) with the Denny Road baseflow conductivity in March 2007 and 2008 ~60% fresher than those upstream of the zone. This difference is even greater than that recorded during the (relatively high discharge) baseflow conditions in 2006 when a ~40% difference was recorded. The relationship between the main channel baseflow discharge and conductivity is significantly negatively related (Figure 10). This clearly demonstrates the importance of groundwater input in reducing the salinity in the main channel of the river during baseflow period when many of the tributaries cease to flow. This has implications for the seasonal habitat usage patterns of salt-intolerant freshwater species in the Blackwood River. For example, the fact that the Mud Minnow is not found in the main channel of the Blackwood River (Beatty et al. 2006, see also Mud Minnow section in the current report) may be due to its inability to tolerate the prevailing conditions in that habitat (particularly salinity). Phillips et al. (2007) demonstrated that the Mud Minnow populations in the various tributaries of the Blackwood River have considerable genetic variability; further supporting this theory of isolation.

The trends in PH are generally to decrease during baseflow conditions compared with high-flow periods (Figure 11). This is probably due to the increased influence of acidic tannins from riparian vegetation and salt water during reduced water levels. Figure 12 shows that dissolved oxygen levels were highly variable between sites and seasons. Generally, dissolved oxygen levels were lower during summer period.

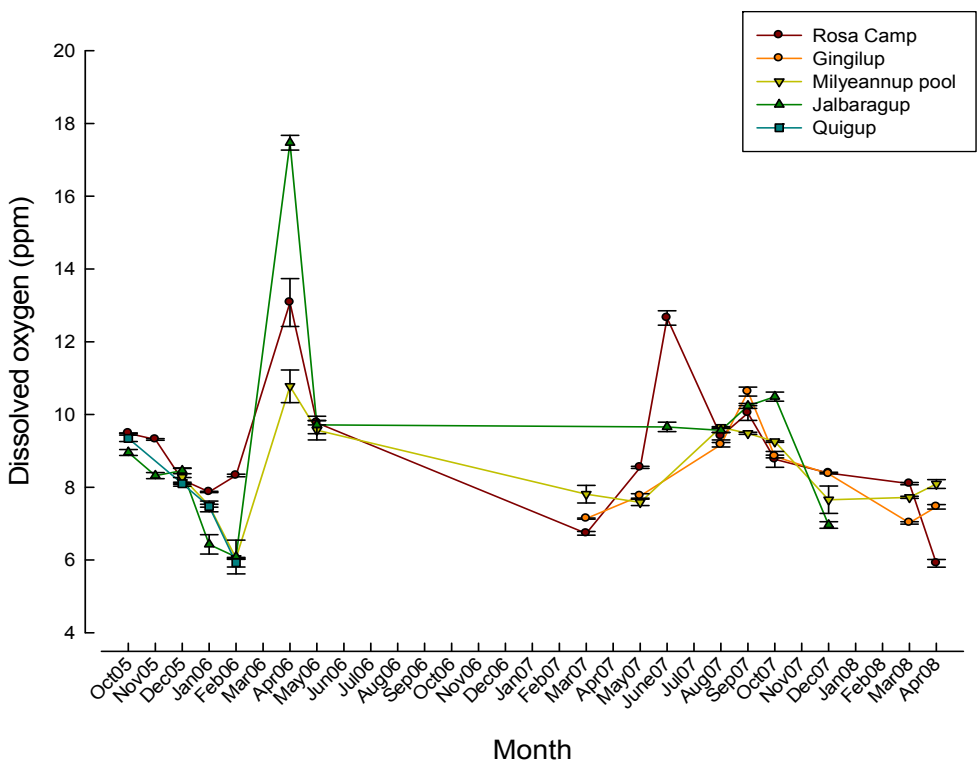
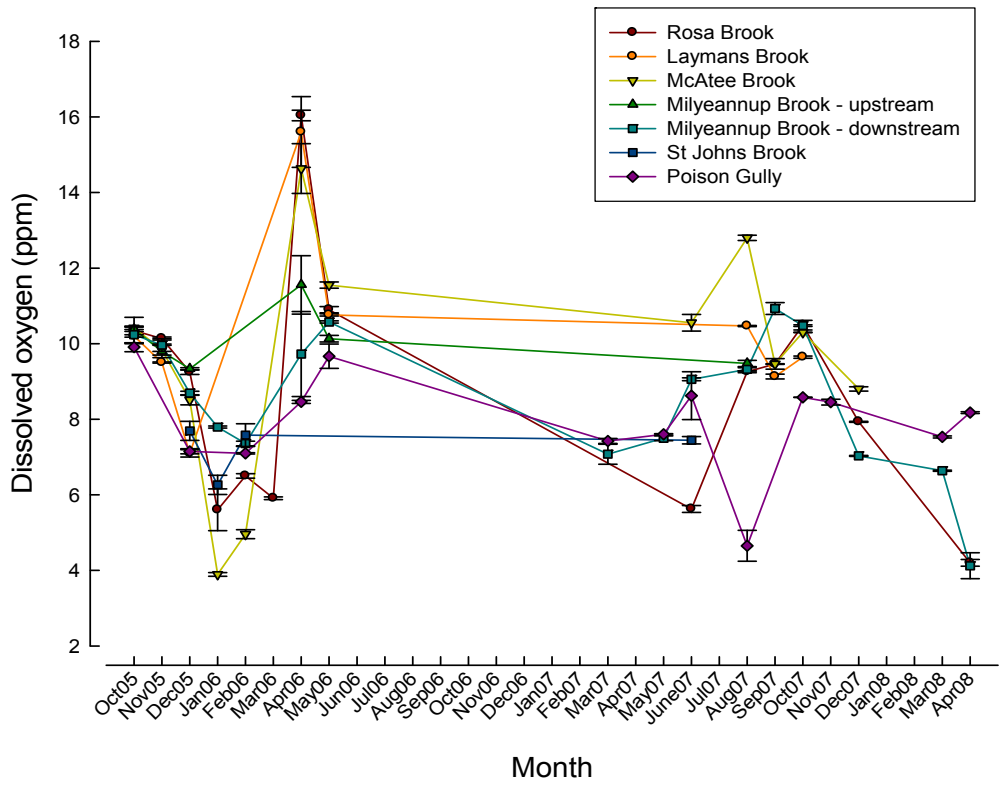


**Figure 10**

Relationship between the baseflow discharges (i.e. in all March samples) at all sites sampled in the main channel of the Blackwood River and the mean water conductivities at those sites.

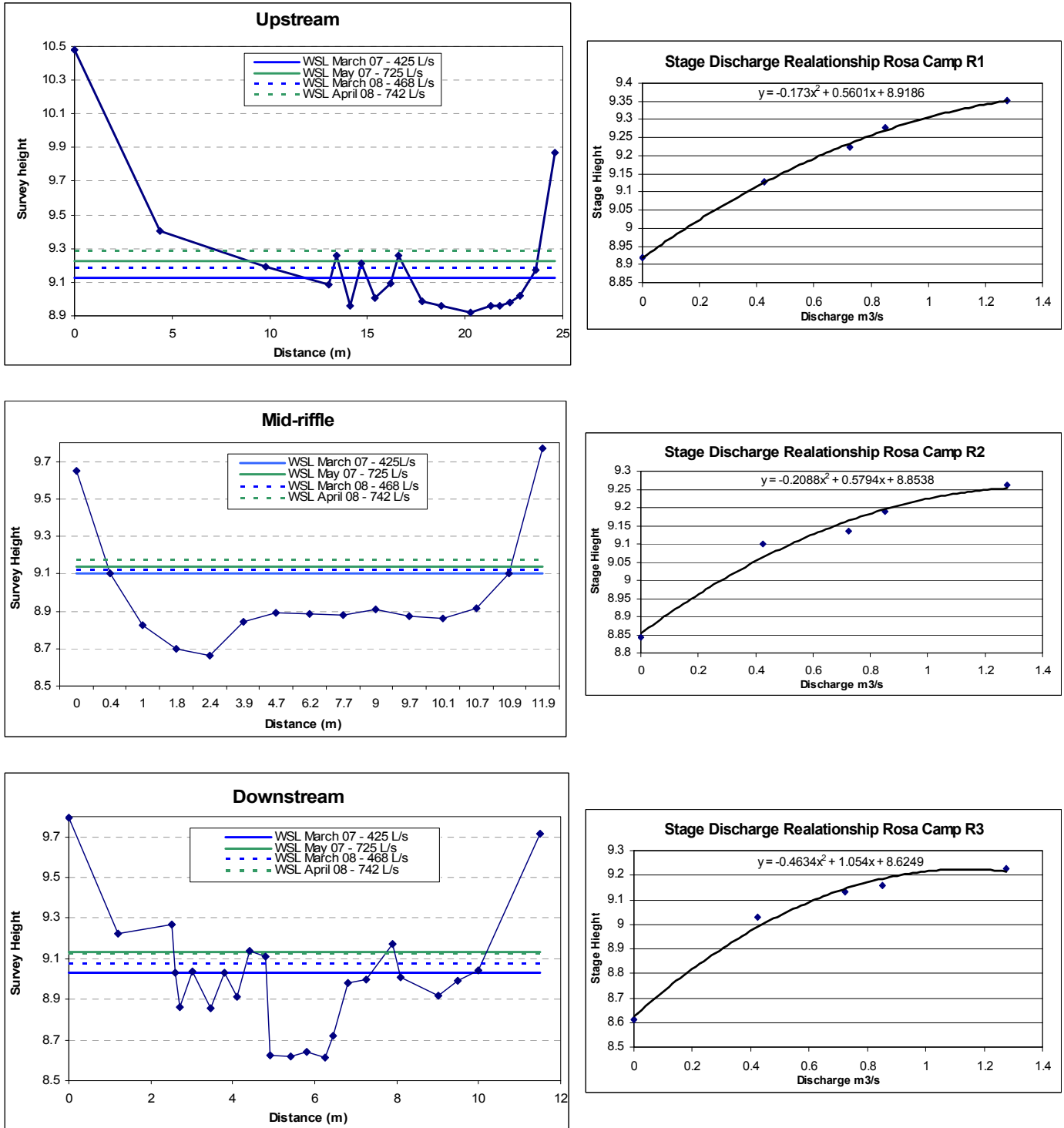


**Figure 11** Mean instantaneous pH at sites in the tributaries (top) and main channel (bottom) of the Blackwood River.

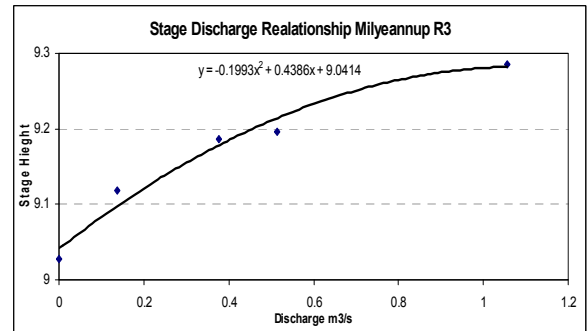
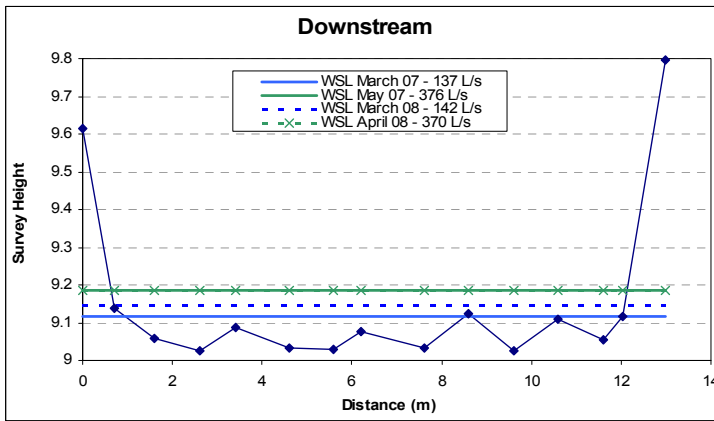
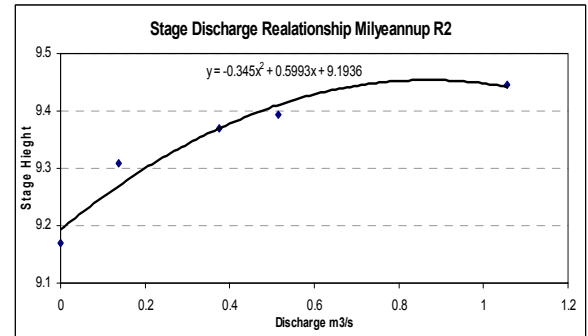
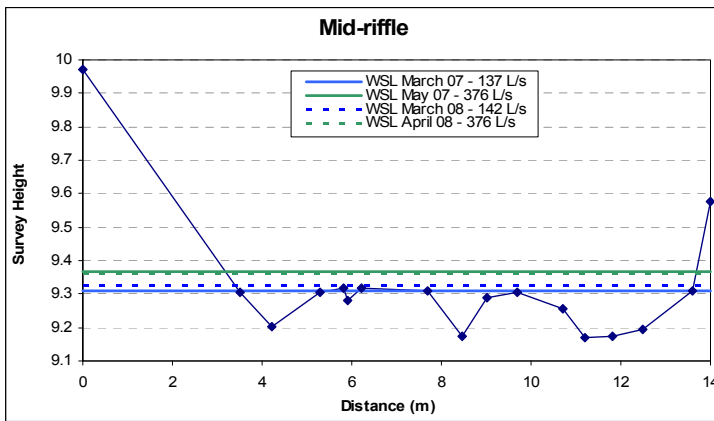
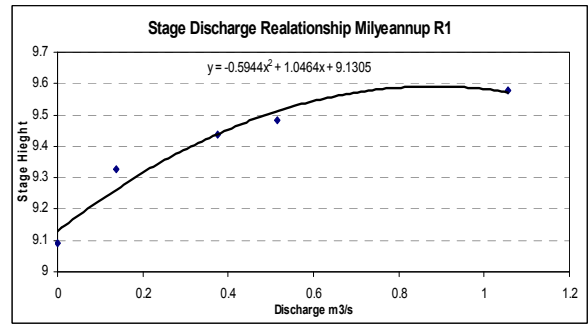
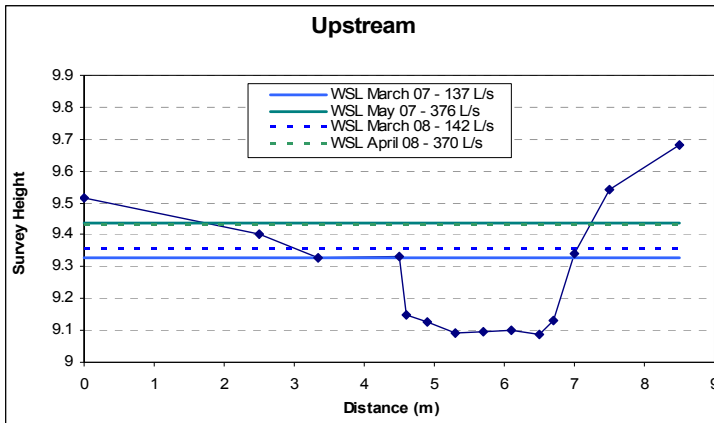


**Figure 12** Mean instantaneous dissolved oxygen at sites in the tributaries (top) and main channel (bottom) of the Blackwood River.

Discharge measurements at the various gauging stations along with cross-sectional discharge and depth profiles at the Denny Road and Milyeannup riffle zones in the main channel suggested that baseflow conditions occurred during March in this section of the Blackwood River (Figures 13 and 14).



**Figure 13** Cross-sectional riffle profiles during base-flow months in 2007 and 2008 at Denny Road in the Blackwood River (Figure from Department of Water, Bunbury).



**Figure 14** Cross-sectional riffle profiles during base-flow months in 2007 and 2008 at riffle downstream of Milyeannup Pool in the Blackwood River.

## Freshwater Cobbler



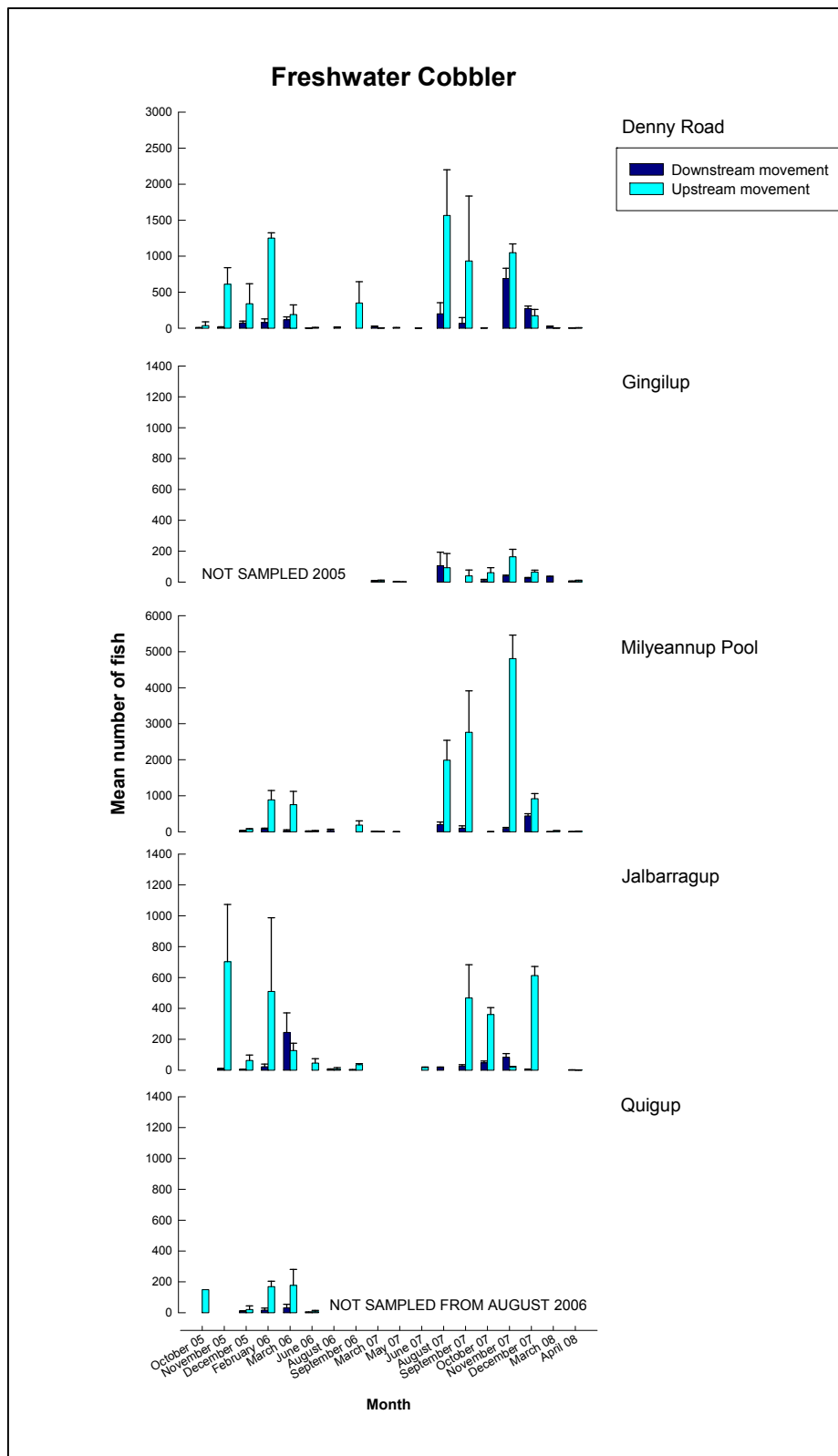
### Spatial and temporal patterns in Freshwater Cobbler migration

A total of 5972 Freshwater Cobbler were captured in the main channel during the study. The movement strength generally peaked in late spring and summer with considerable differences in the temporal movement patterns existing between the sites (Figure 15). The mean upstream movement of Freshwater Cobbler in all sampling months was positively associated with the mean downstream movements during those sampling times ( $r^2=0.32$ ,  $p=0.000$ ) (Figure 16).

Of the 437 tagged Freshwater Cobbler in the main channel of the Blackwood River, a total of 86 (19.7%) were recaptured (Table 1). Of these, 68 were recaptured once, 14 twice, five three times and two were recaptured four times (Table 1). With the exception of one fish, all were caught at the initial tag-capture site.

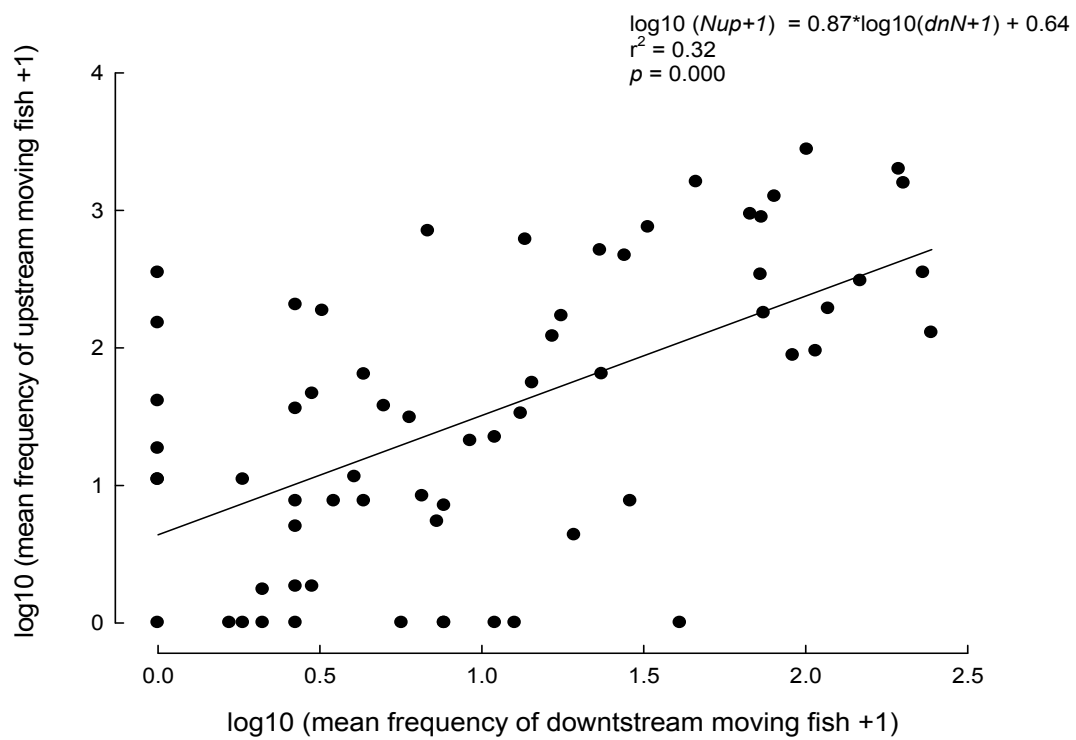
**Table 1:** Freshwater Cobbler tagged and recaptured in main channel sites of the Blackwood River. N.B. Includes % recaptured per site and % total of all recaptures when compared to total number tagged.

SITE	Number Tagged	# Recaptured (%)	# Recaptured Twice (%)	# Recaptured 3 times (%)	#Recaptured 4 Times (%)
Denny Road	215	42 (19.5)	8 (3.7)	1 (0.5)	
Milyeannup Pool	42	7 (16.7)	2 (4.76)		
Jalbarragup	110	17 (15.5)	4 (3.6)	4 (3.6)	2 (1.8)
Quigup	70	2 (2.9)			
TOTAL (%)	437	68 (15.6)	14 (3.0)	5 (1.1)	2 (0.5)



**Figure 15** Upstream and downstream migration of Freshwater Cobbler in the Blackwood River main channel sites.





**Figure 16** Relationship between the mean strength of upstream versus downstream movements of Freshwater Cobbler in the Blackwood River main channel in all sampling months. N.B. Data were log10 transformed and migration number was standardised for effort; see text for details.

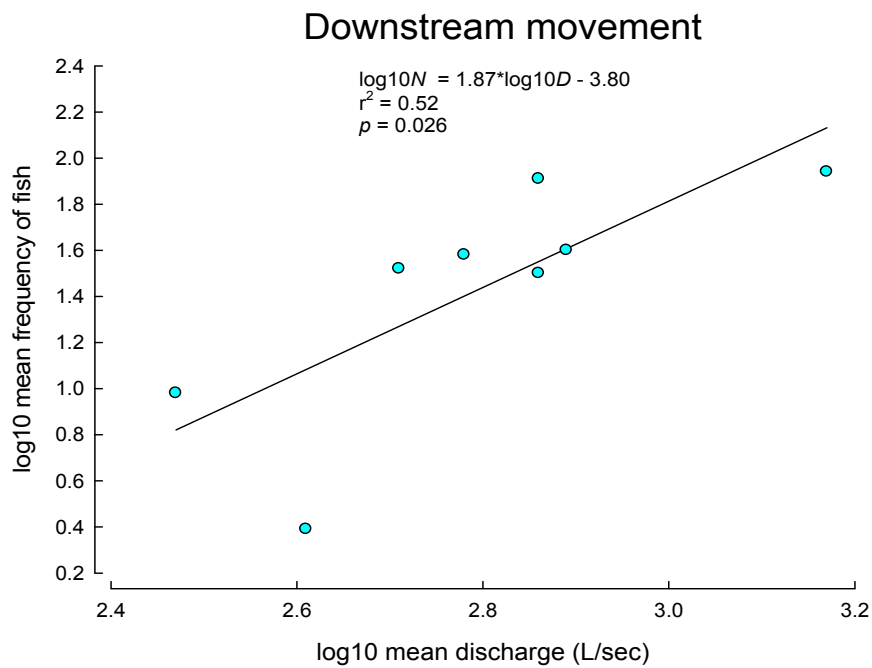
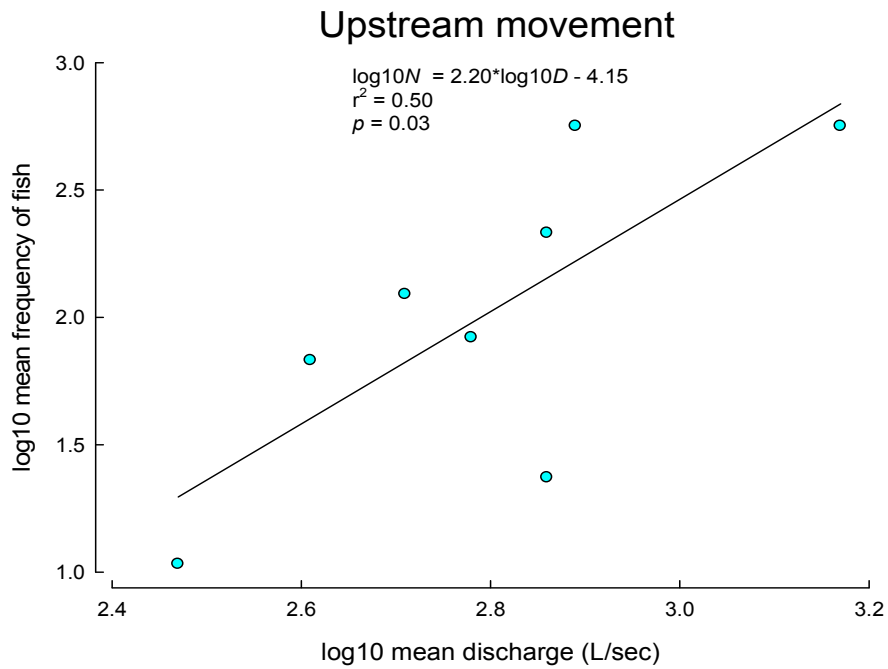
### **Relationships between Freshwater Cobbler migration patterns and environmental variables in the Blackwood River**

Correlation and stepwise multiple regression analysis revealed that the upstream ( $r^2 = 0.50$ ,  $p = 0.03$ ) and downstream ( $r^2 = 0.52$ ,  $p = 0.026$ ) movements of Freshwater Cobbler in all main channel sites during the period of lowest flow (i.e. December to April) were most significantly explained by mean discharge at those sites during those periods (Table 2, Figure 17).

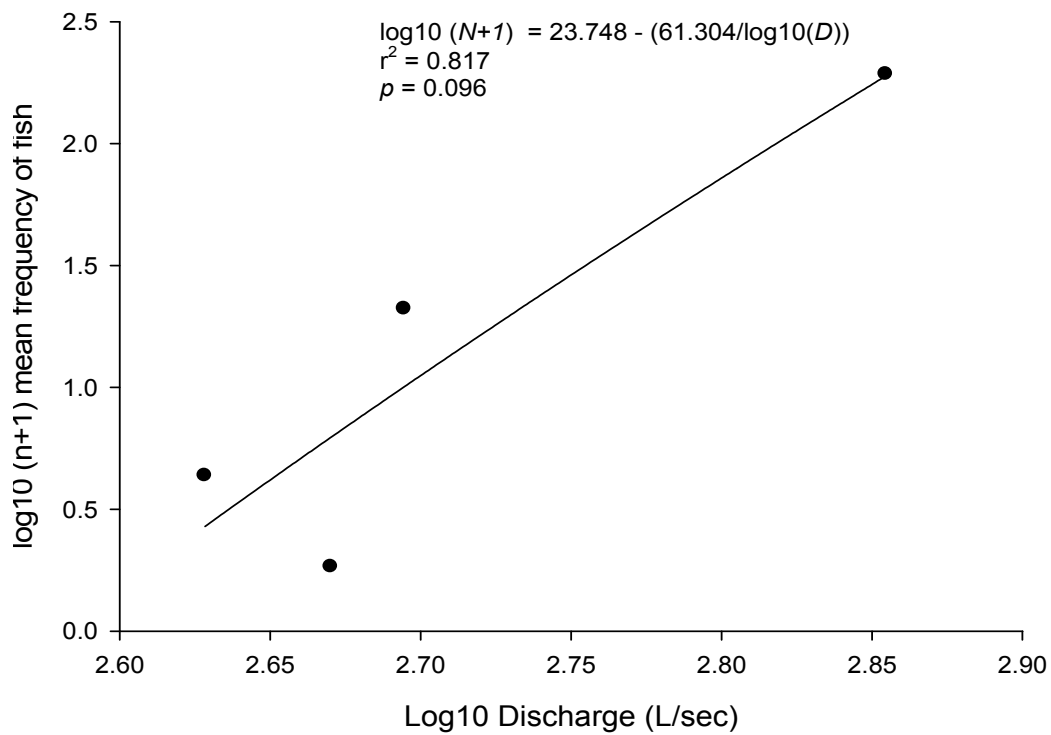
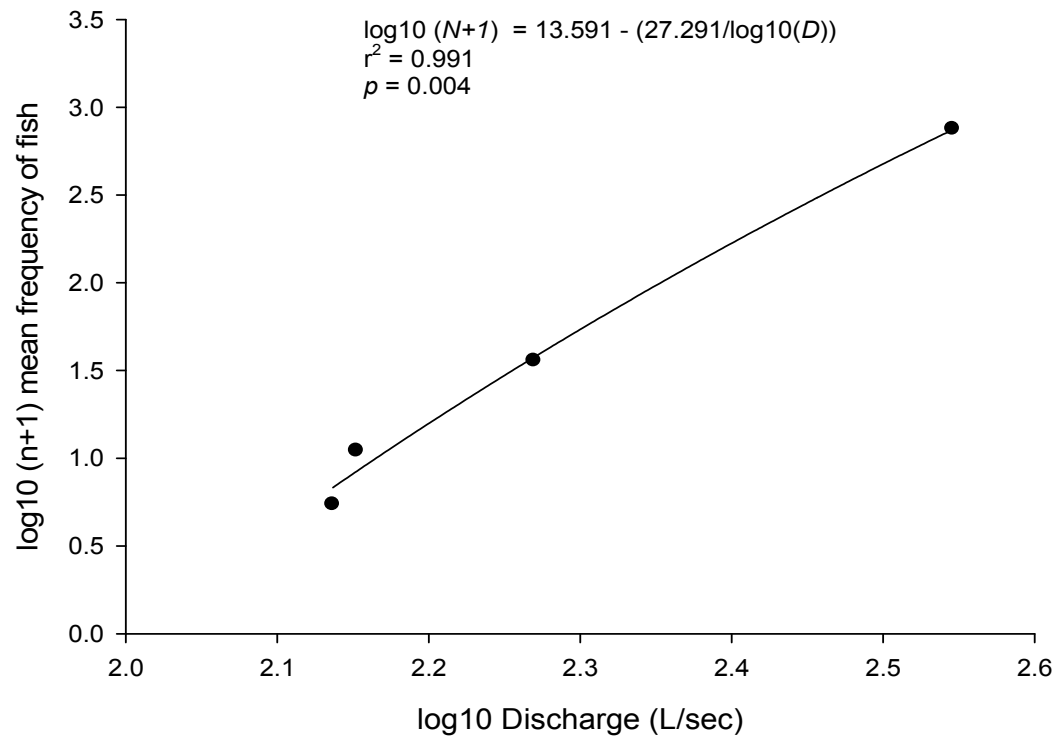
The models that gave the highest coefficients of determination for the relationship between upstream movements of Freshwater Cobbler over the two riffle sites at baseflow (i.e. March) and levels of baseflow discharge in those months were inverse polynomials and these revealed a strong association between these variables at both the Milyeannup Pool ( $r^2 = 0.991$ ,  $p = 0.004$ ) and Denny Road riffles ( $r^2 = 0.817$ ,  $p = 0.096$ ) (Figure 18). Using these models, the minimum discharge that Freshwater Cobbler movement would be totally precluded at the Milyeannup Pool and Denny Road riffles are estimated to be 381.5 L/sec and 101.9 L/sec, respectively. Based on this zero-movement discharge, analysis of the riffle cross-sections and the discharge-stage height relationships (Figures 13 and 14), the depth across riffles when zero movement discharge would occur is 0.18 and 0.05 m for Denny Road and Milyeannup Riffle, respectively. However, those individuals that were captured moving upstream during the March 2007, and 2008 period were much smaller than those captured during 2006; when the greatest baseflow discharge levels were recorded (Figure 19).

**Table 2** Correlations between overall mean upstream and downstream movements of Freshwater Cobber in the main channel sites of the Blackwood River and prevailing environmental variables during the lowest discharge period (December to April). N.B. Data were log10 transformed, \* denotes correlation is significant at the 0.05 level (2-tailed).

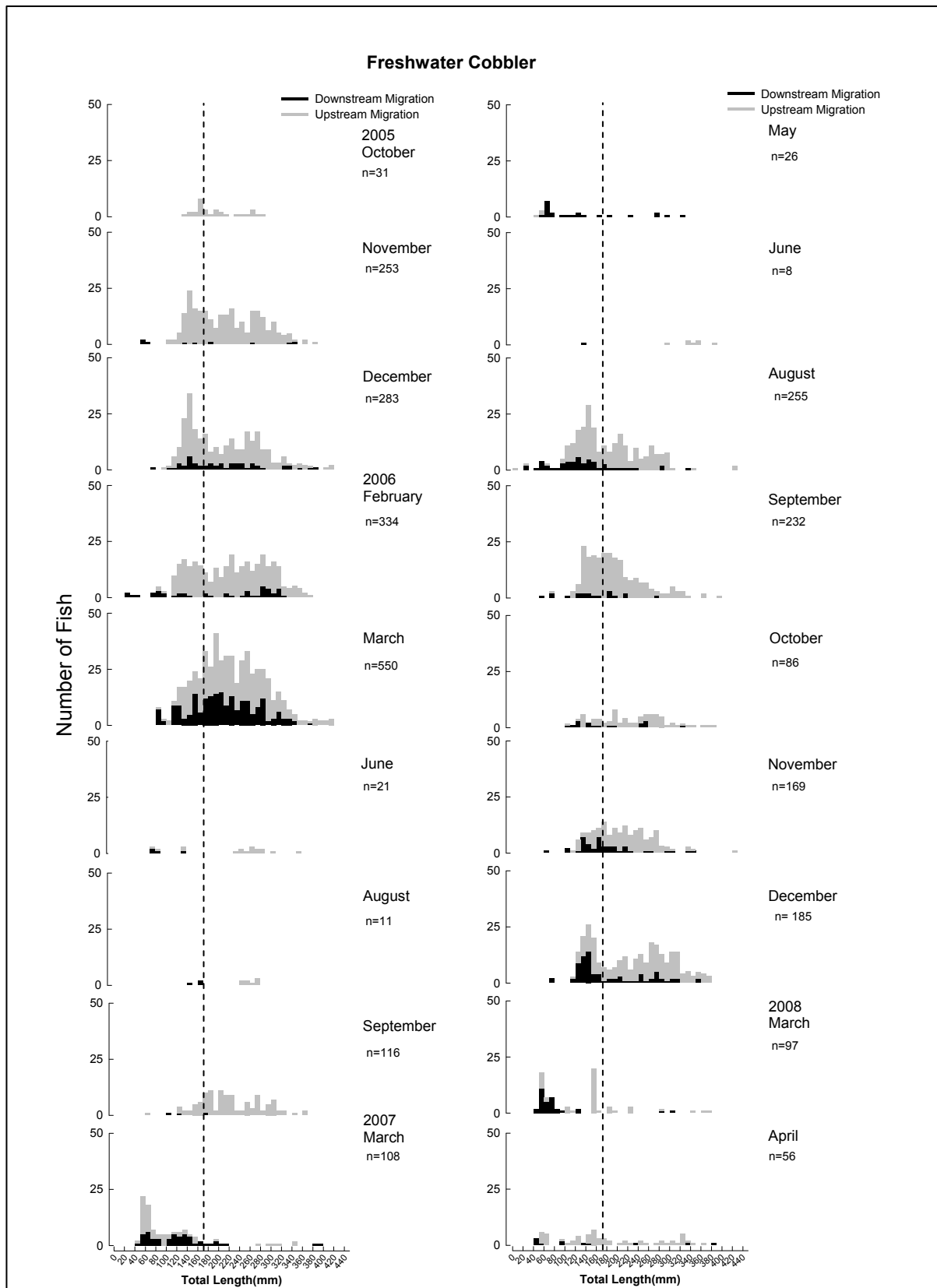
		Downstream	Upstream	Discharge	Temperature	Conductivity	pH	Dissolved O2
Downstream	Pearson Correlation	1						
	Sig. (2-tailed)							
Upstream	Pearson Correlation	.566	1					
	Sig. (2-tailed)	.143						
Discharge	Pearson Correlation	.767(*)	.756(*)	1				
	Sig. (2-tailed)	.026	.030					
Temperature	Pearson Correlation	.455	.851(**)	.673	1			
	Sig. (2-tailed)	.257	.007	.067				
Conductivity	Pearson Correlation	-.747(*)	-.502	-.905(**)	-.366	1		
	Sig. (2-tailed)	.033	.205	.002	.373			
pH	Pearson Correlation	-.193	-.396	-.288	.073	.365	1	
	Sig. (2-tailed)	.648	.332	.489	.863	.373		
Dissolved O2	Pearson Correlation	-.049	.332	.530	.173	-.515	-.318	1
	Sig. (2-tailed)	.909	.422	.177	.683	.191	.443	



**Figure 17** Relationships between the mean strength of upstream movement of Freshwater Cobbler and the mean discharge in the Blackwood River main channel. N.B. Data were log10 transformed and migration number was standardised for effort, see text for details.



**Figure 18** Relationship between mean upstream Freshwater Cobbler movement over the Milyeannup (top) Denny Road (bottom) riffles and the mean discharge at those sites during March 2006-2009.

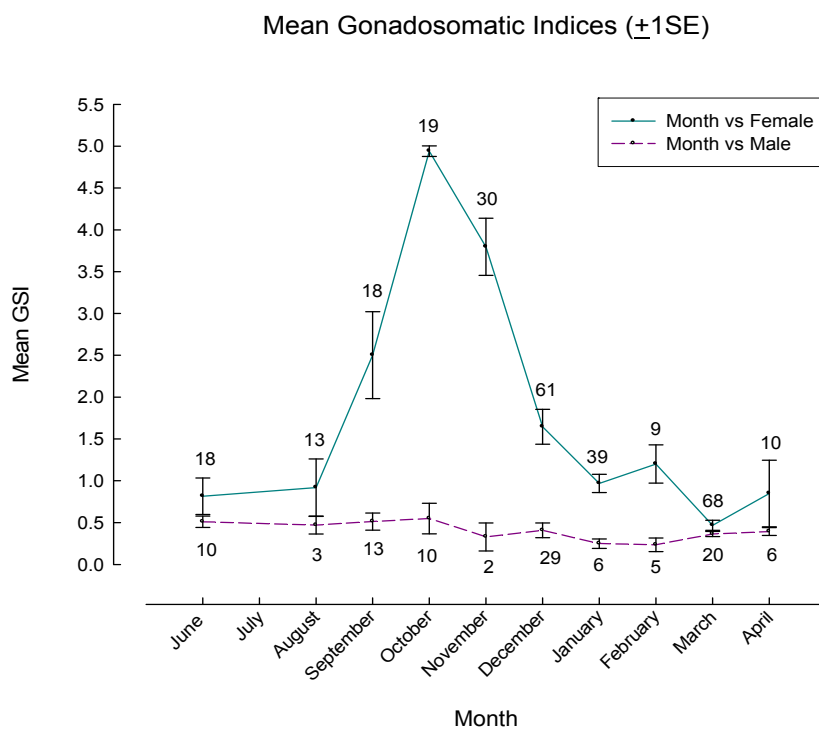


**Figure 19** Length-frequency of Freshwater Cobbler, differentiated by upstream and downstream movement, in the Blackwood River main channel. N.B. the dashed line is the length at maturity ( $L_{50}$ ) of females (172mm TL).

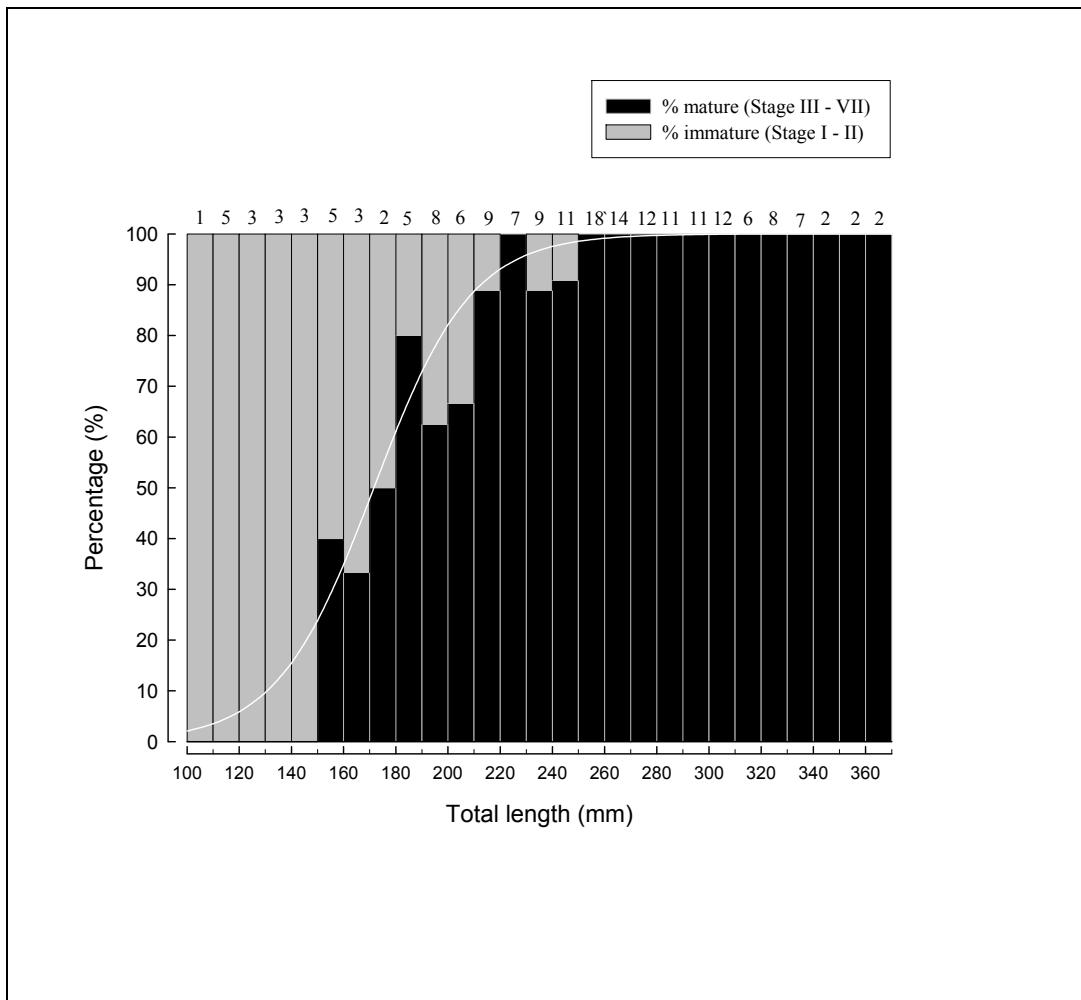
## Reproductive biology

The mean GSI of mature females of Freshwater Cobbler increased sharply between August and October; peaking in the latter month (Figure 20). From October to December the most significant decline occurred dropping from 4.94 to 1.65 between these two months, respectively; suggesting this is the peak spawning time. The GSI of males, while having far less variability than females, also peaked in October at 0.55.

The logistic curve, fitted to the percentage contribution of Freshwater Cobbler with gonads of stages III-VII in sequential 10 mm TL increments during the spawning period, yielded a L50 of ~172 and L95 of 226 for females i.e. lengths at which 50% and 95% of the population is sexually mature (Figure 21).



**Figure 20** Mean gonadosomatic indices (GSI) ( $\pm 1SE$ ) for male and female Freshwater Cobbler in the main channel of the Blackwood River.



**Figure 21** Mean length at first maturity for female Freshwater Cobbler in the Blackwood River.

**Freshwater Cobbler: summary and implications**

This study has demonstrated considerable movements of *T. bostocki* within the main channel of the Blackwood River. The mark-recapture program also suggested these movements were highly localised and suggested a high degree of site (possibly on the scale of individual pools) fidelity. Strengths of movements differed markedly both spatially and temporally; however, upstream movement peaked at all sites in spring and summer. Furthermore, the degree of upstream movement during the period of lowest flow (and greatest proportional contribution of groundwater to total flow) was significantly related to volume of discharge during that period at those sites. The impetus for these considerable localised movements through riffle zones is probably due more to feeding activities rather than spawning as feeding on this habitat type has been shown to occur with other freshwater catfishes (e.g. *Pygodictis olivardis*, Trautman 1981, *Hatcheria macraei* Barriga and Battini 2009) and this study demonstrated a wide size range (including smaller immature individuals) accessed the riffles at those times. However, for this to be confirmed, the feeding ecology of the species needs to be determined along with precise identification of its spawning habitats.



The significance of the considerable upstream movements of Freshwater Cobbler in terms of sustaining the population requires further validation (e.g. by comparing levels of recruitment, feeding and reproductive ecology of this and other populations), however, it is proposed that this relatively large-bodied species could be an appropriate indicator of baseflow river connectivity throughout south-western Australia given (Morgan *et al.*1998). The baseflow models presented here may also be used to set minimum baseflow discharges required for riffle connectivity for this species. For example, the current study suggests that upstream movement of Freshwater Cobbler during March would be prevented at ~102 L/sec (5cm depth) and 381 L/sec (19 cm depth), for the riffles upstream and downstream of the major zone of groundwater discharge, respectively. Based on these lower limits, reductions of ~33% and ~8% reductions from the relatively low March 2007 discharges recorded at Milyeannup Pool and Denny Road riffles, respectively, would prevent any upstream Freshwater Cobbler moving over these riffle zones. As the baseflow discharge rates in this section of the Blackwood River (i.e. Darradup and Gingalup gauging stations) are dependent on the rainfall from the previous 12 month period and abstraction rates (Golder and Associates 2008), the levels of abstraction rates should be set to ensure the above minimum discharge rates are exceeded to ensure riffle access is maintained for this large bodied species.

However, these preliminary minimum estimates are for zero movements and it can be seen from the length-frequencies of the relative few Freshwater Cobbler captured moving over these two riffles in 2007 and 2008 that the sizes were relatively small (dominated by juveniles 40-100 mm TL) compared to the greater number that moved over them in the relatively high-flow event in March 2006 (no fish under 100 mm TL, dominated by mature adults 180-340 mm TL) (Figure 10). This suggests that larger, mature Freshwater Cobbler may be inhibited from upstream movement through these riffle zones at a discharge, and associated minimum depths, considerably greater than the estimates in Table 2.

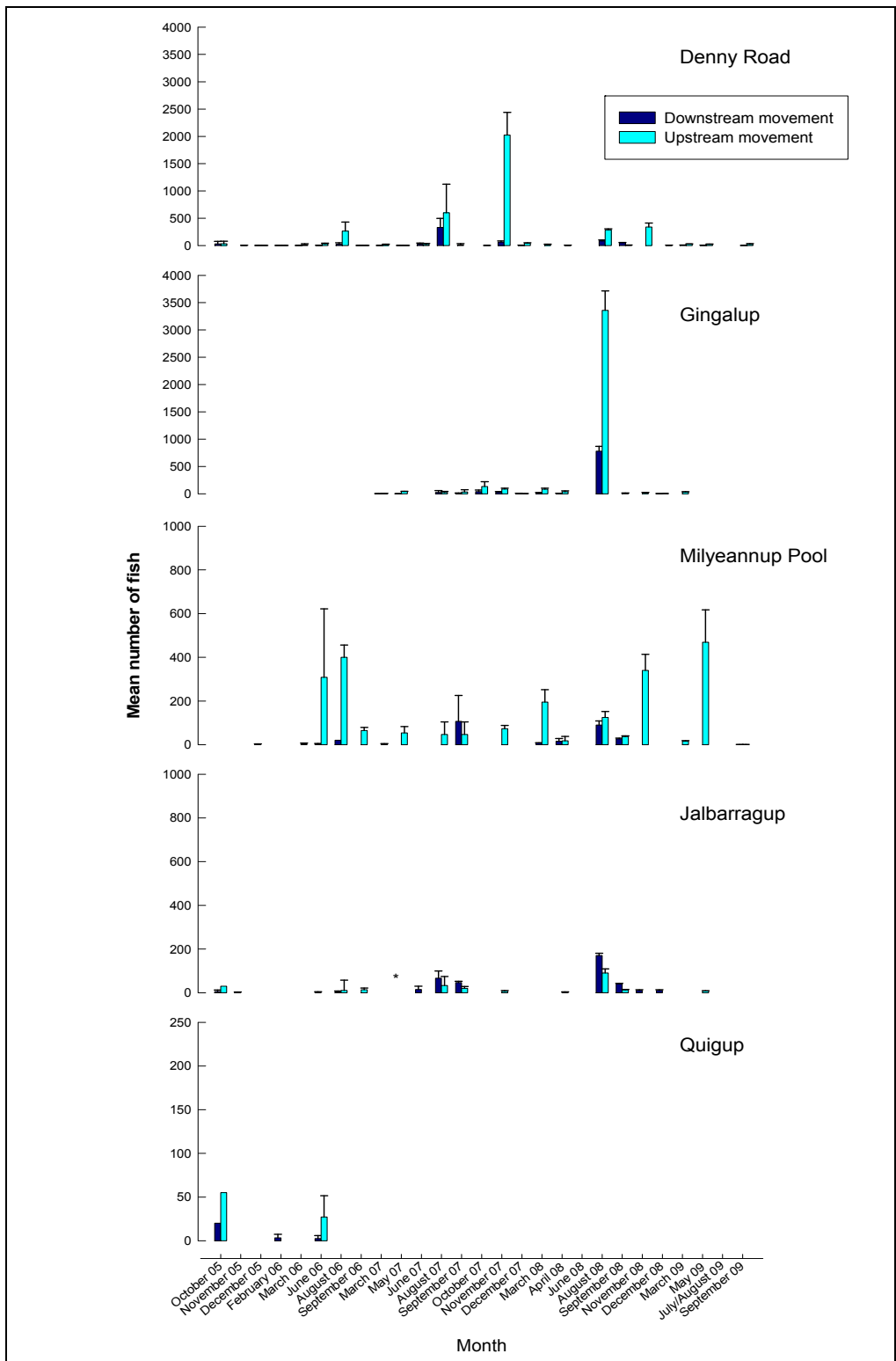
The relationships between Freshwater Cobbler movements through groundwater-maintained riffle zones demonstrated here could be further validated by ongoing monitoring of this population during baseflow and prevailing hydrology at these sites. A tracking program (using acoustic and radio-tracking techniques) of Freshwater Cobbler at key riffle sites would also provide fine-scale information on their habitat use and requirements. Furthermore, understanding spatial and temporal patterns of feeding and reproductive ecology would also enable a better understanding of the purpose of these movements during baseflow conditions and their significance in maintaining populations of this species in the rivers of south-western Australia.

## Western Minnow

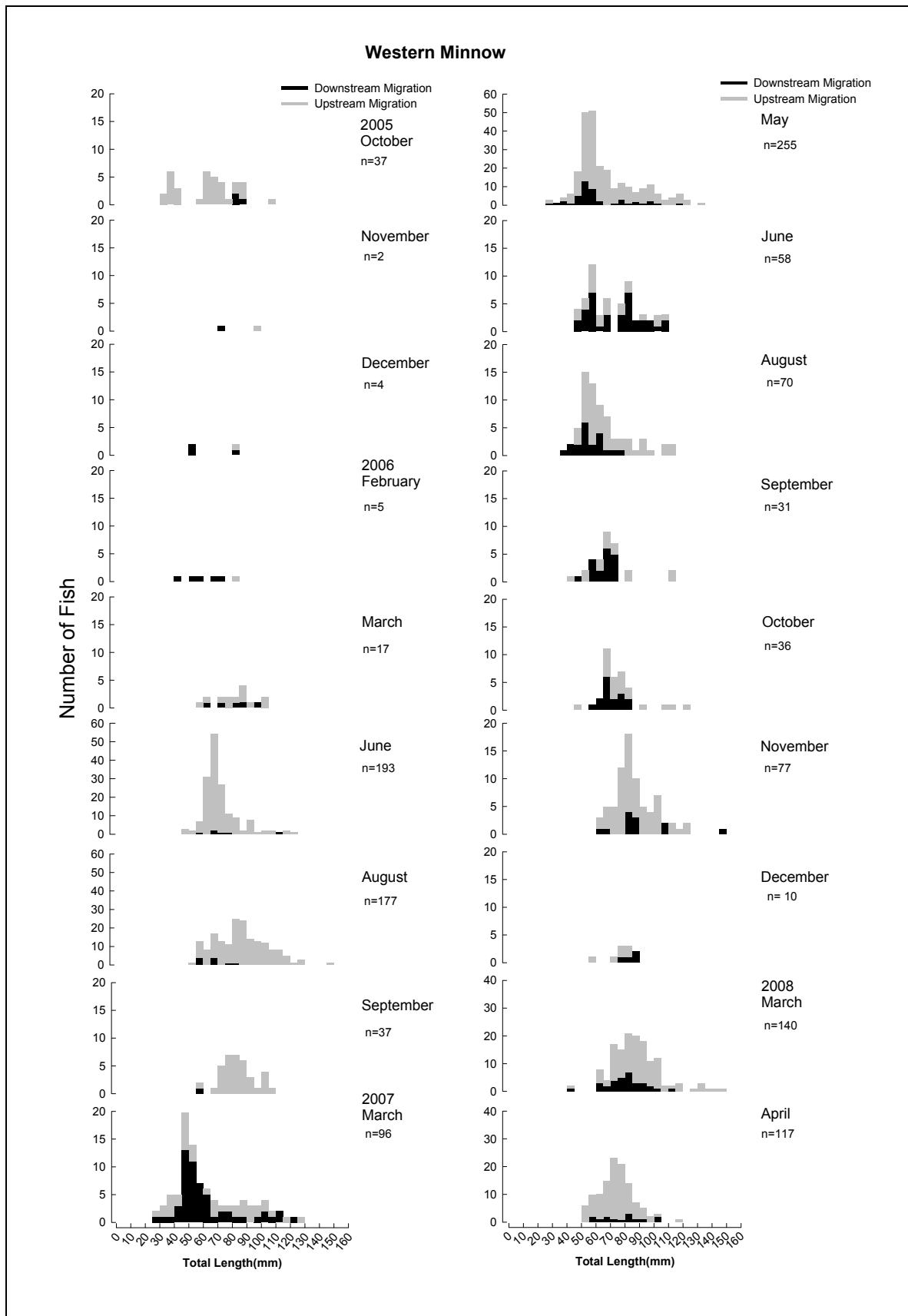


The Western Minnow was captured at the majority of sites sampled on most occasions, with large numbers recorded in both the main channel sites and in the tributaries. This widespread distribution, being present in nearly all habitats sampled, reflects this species tolerance to a wide range of salinities; having previously been recorded in salinities up to ~24 ppt or ~two thirds the salinity of seawater (Morgan and Beatty 2004). However, recent rapid change salinity trials on the species from the Blackwood River have revealed a LD<sub>50</sub> (length at 50% of individuals showed severe stress that would lead to death) at ~14 ppt (Beatty *et al.* 2008). Furthermore, larval stages of the species may have an even lower tolerance to salinity.

There were limited movements of Western Minnow in the most upstream main channel sites (i.e. Jalbarragup and Quigup) compared to the more downstream sites (i.e. Denny Road, Gingilup and Milyeannup Pool) and the upstream movement of Western Minnow was generally strongest during winter of each year (Figure 22). The majority of these fish were large adults (>60 mm TL) that were likely to be moving as a precursor to spawning (Figure 23).



**Figure 22** Upstream and downstream movement of Western Minnow in the Blackwood River main channel.



**Figure 23** Length-frequency histograms, differentiated by upstream and downstream movement, of the Western Minnow in the Blackwood River main Channel.

The migration pattern of the Western Minnow within tributaries exhibited considerable interannual variation (Figures 24-28). Migration within the perennial Milyeannup Brook occurred at least one month each year in than in Rosa Brook (Figure 24). For example, considerable upstream movements occurred in June each year within the perennial Milyeannup Brook that were not yet occurring in Rosa Brook or McAttee at those times. In Milyeannup Brook there were considerable downstream movements in late spring and early summer (Figure 24).

Examination of length-frequency histograms of fish caught in main channel sites compared to tributary sites again revealed that the vast majority of Western Minnows captured less than 40 mm TL were only found within the tributaries (Figures 25-28). This, in combination with the strong upstream migration of adults prior to the known spawning period (i.e. winter/early spring) strongly suggests that breeding takes place within tributaries and that these habitats are therefore vital spawning areas for this species.

The upstream and downstream migration of the Western Minnow in the tributaries were both previously found to be positively correlated with the mean discharge from the tributaries during the major flow period (Beatty *et al.* 2006). With the addition of the 2007 migration period, this relationship was still evident; however, no longer significant at  $p < 0.05$  (although upstream was significant at  $p < 0.1$ , and downstream at  $p = 0.12$ , see Figures 29, 30, Table 3).

The strength of upstream and downstream movement of the Western Minnow during this major migration period in tributaries was positively and significantly related ( $r^2 = 0.71$ ,  $p = 0.009$ , Figure 31). This indicates that upstream movement of Western Minnows into tributaries can be used to predict the subsequent downstream movement of the species; that is, the return of those mature individuals moving back down into the main channel of the Blackwood River and also the subsequent degree of downstream recruitment of juveniles back into the Blackwood River.

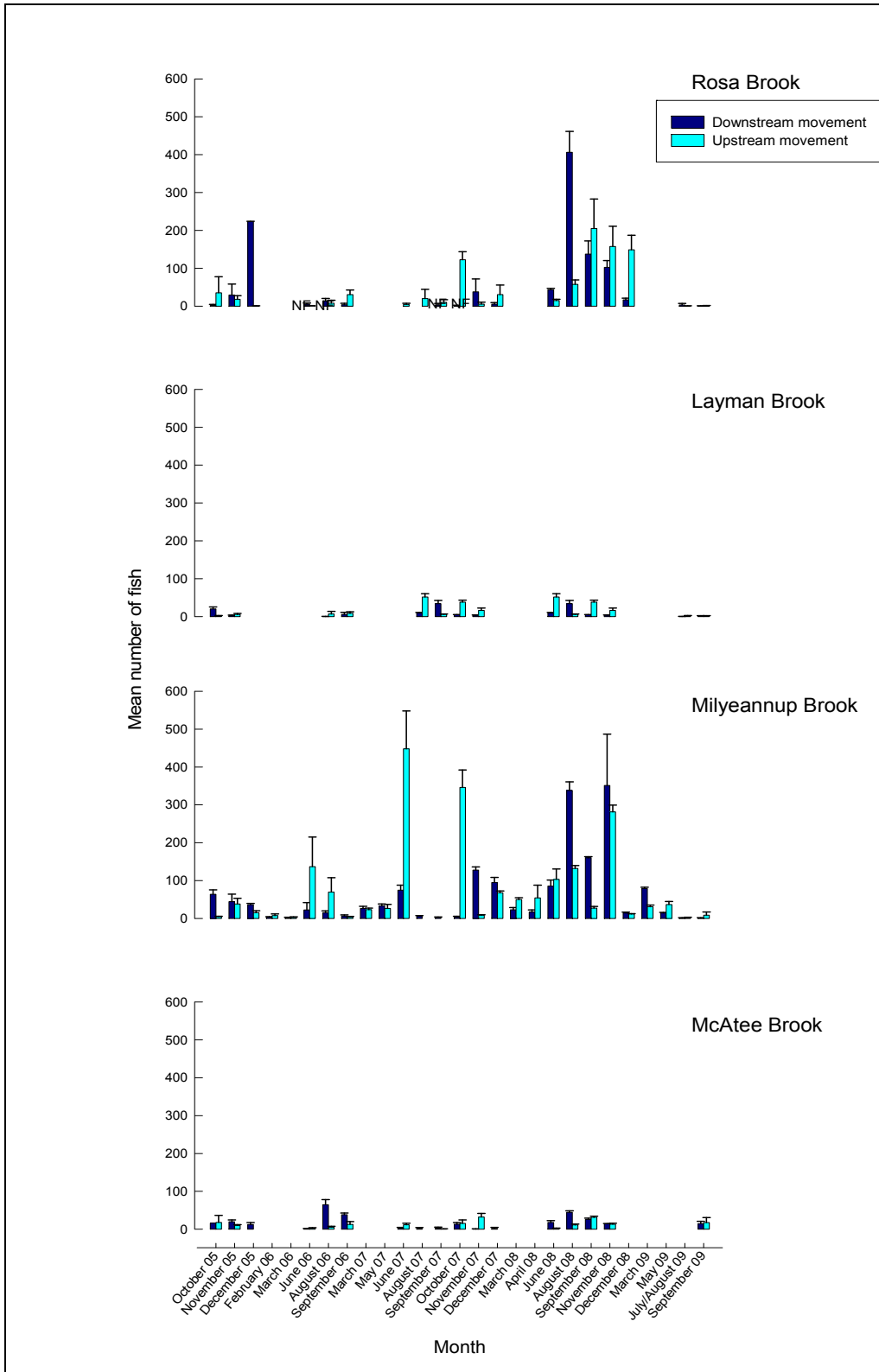
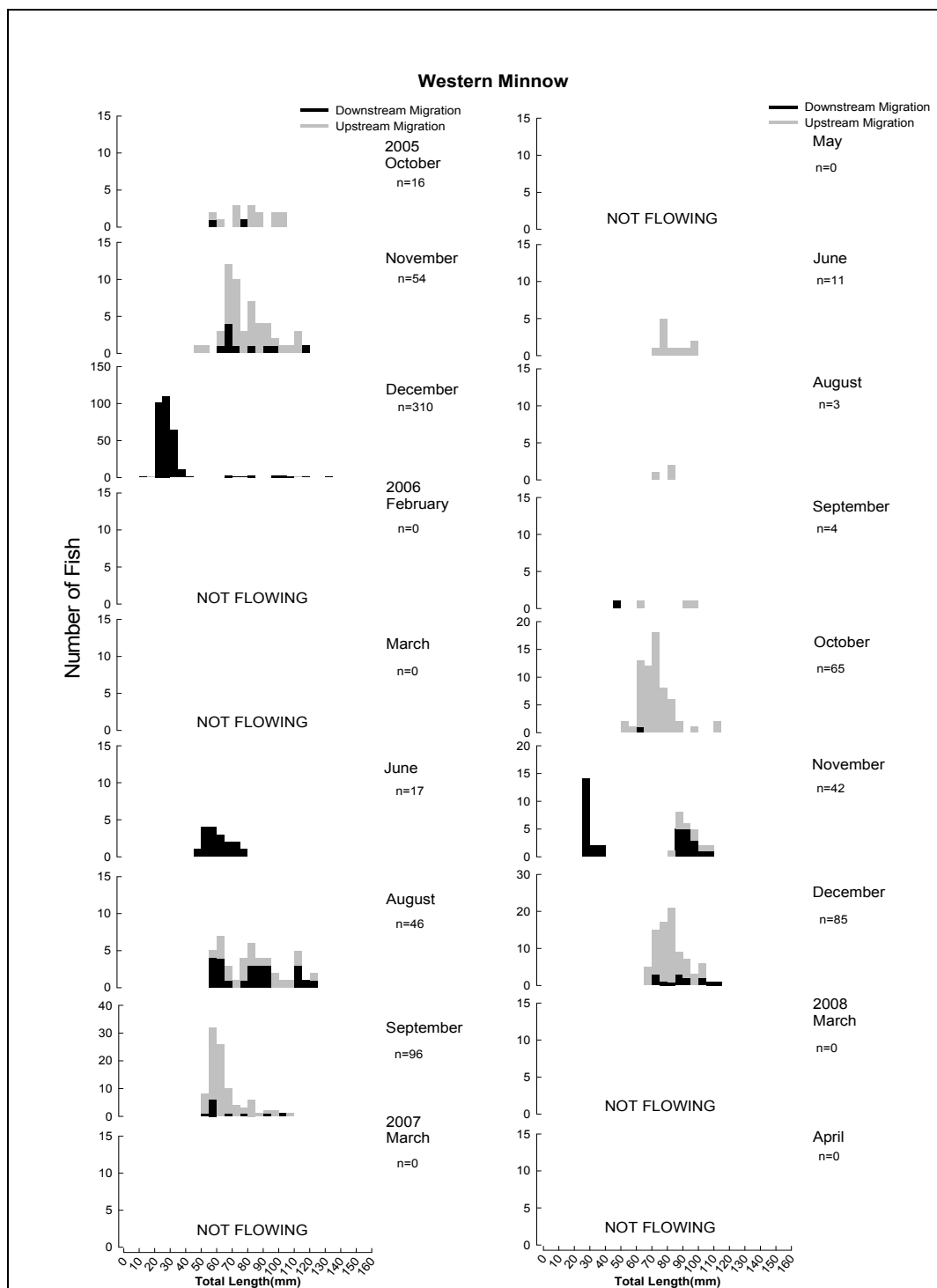
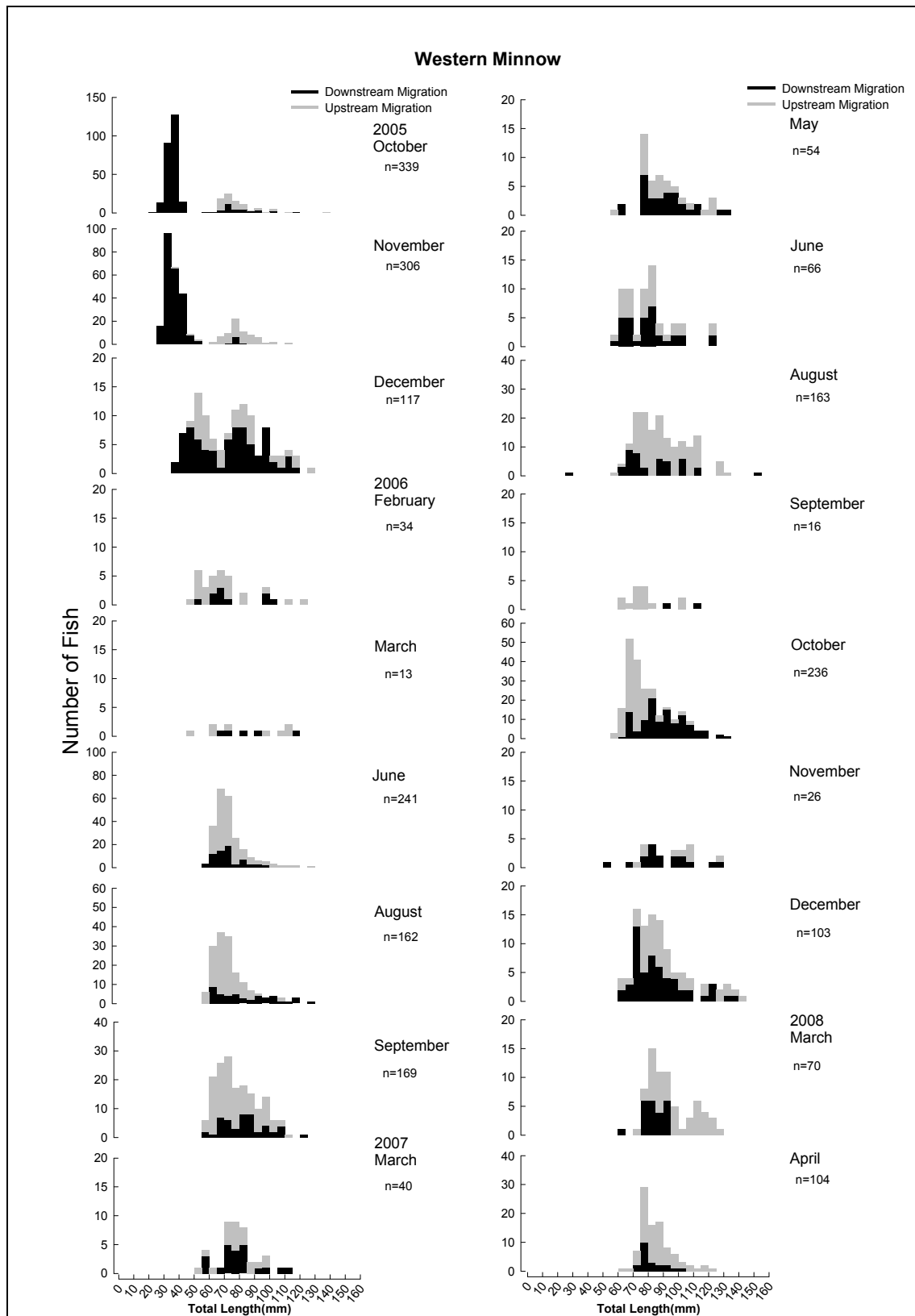


Figure 24

Upstream and downstream movement of Western Minnow in the tributaries.

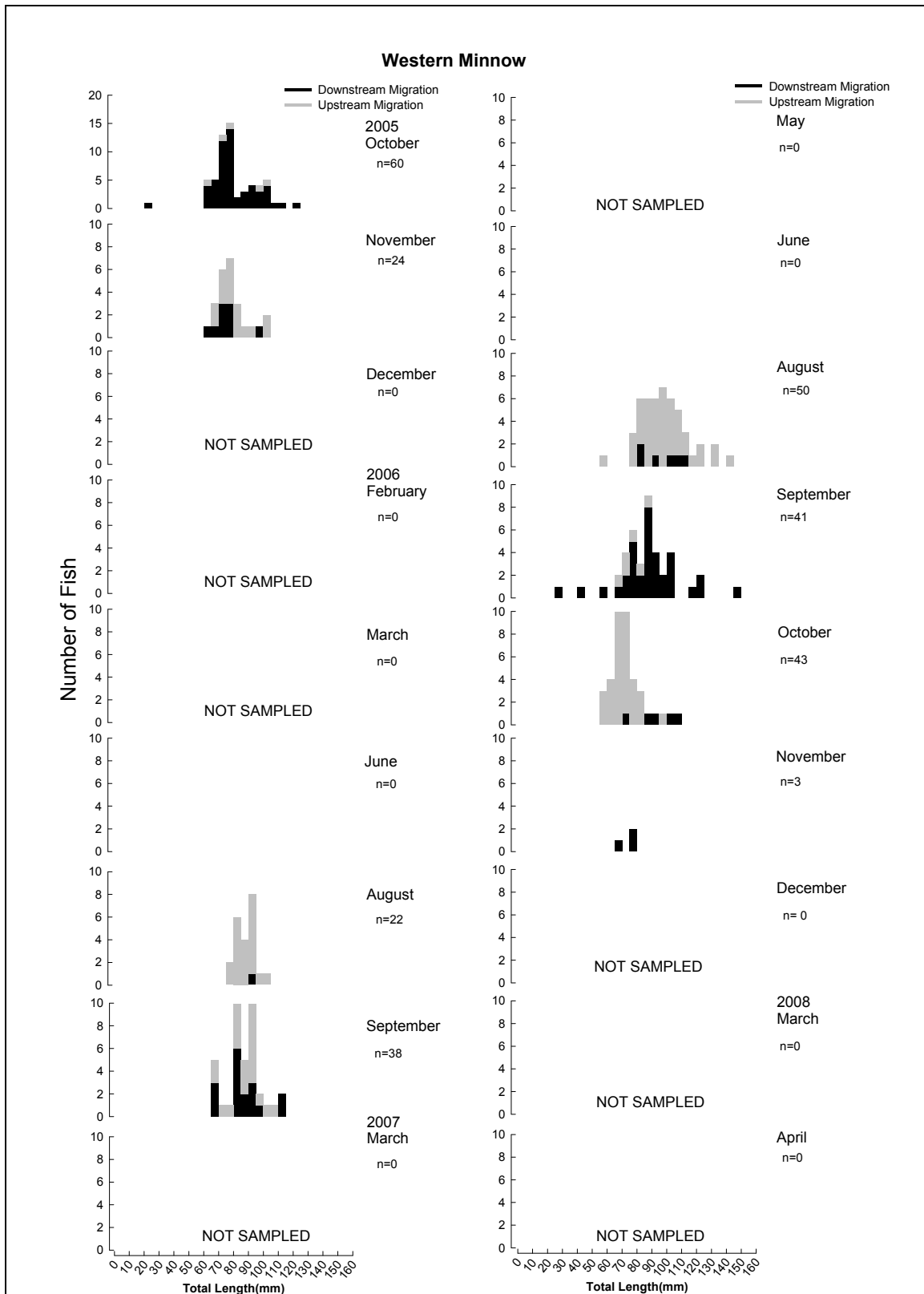


**Figure 25** Length-frequency histograms, differentiated by upstream and downstream movement, of Western Minnow in Rosa Brook.

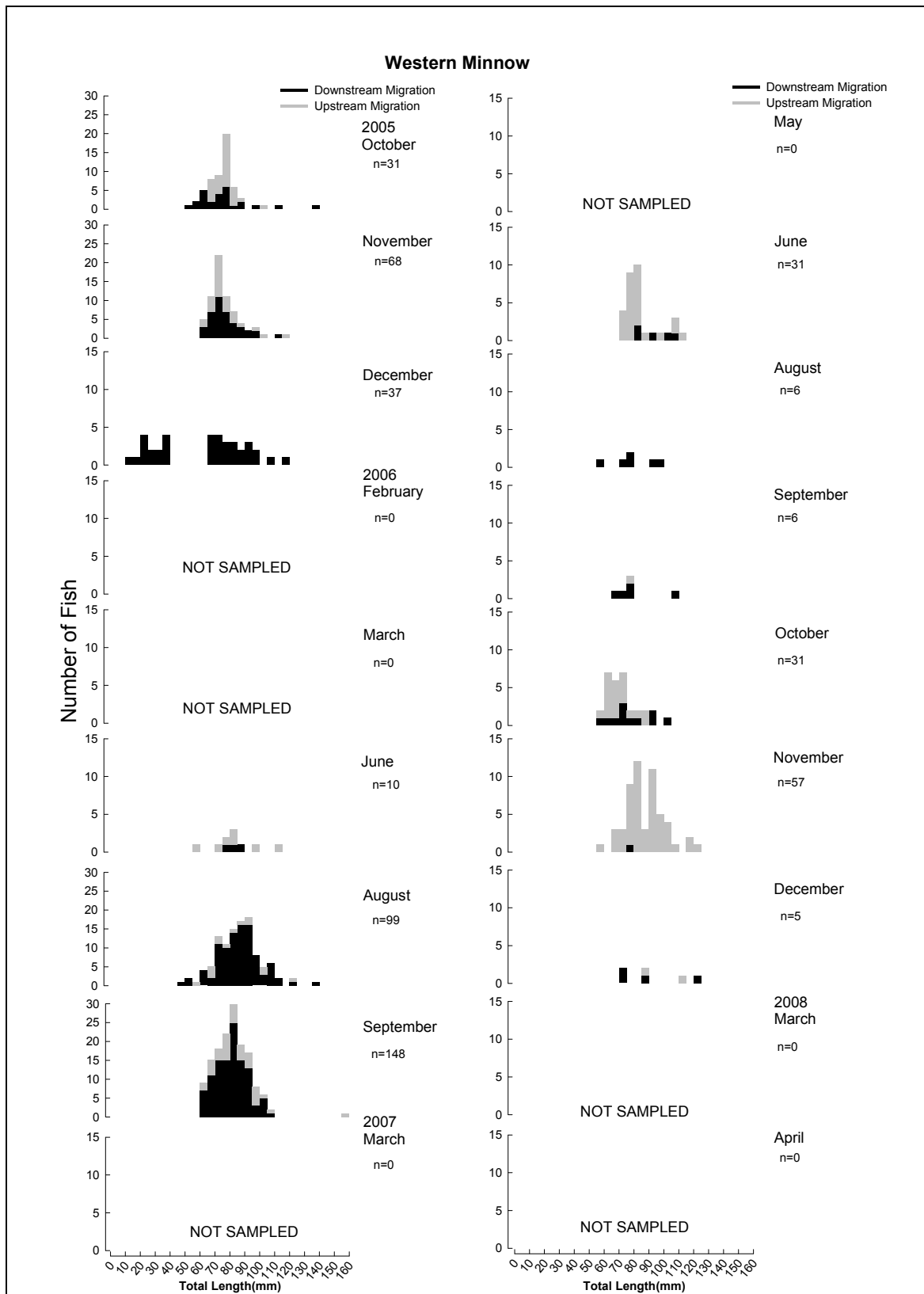


**Figure 26** Length-frequency histograms, differentiated by upstream and downstream movement, of Western Minnow in Milyeannup Brook.

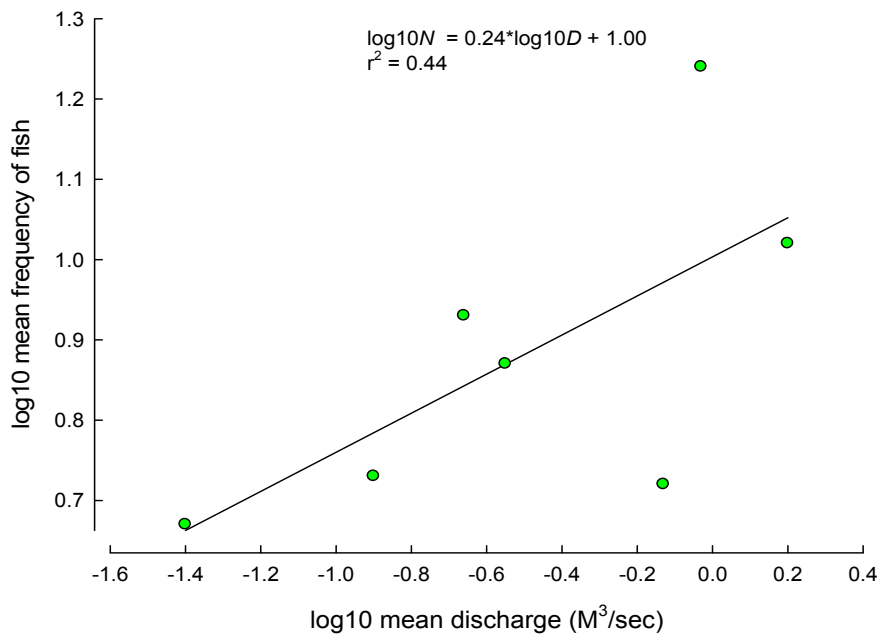




**Figure 27** Length-frequency histograms, differentiated by upstream and downstream movement, of Western Minnow in Layman Brook.

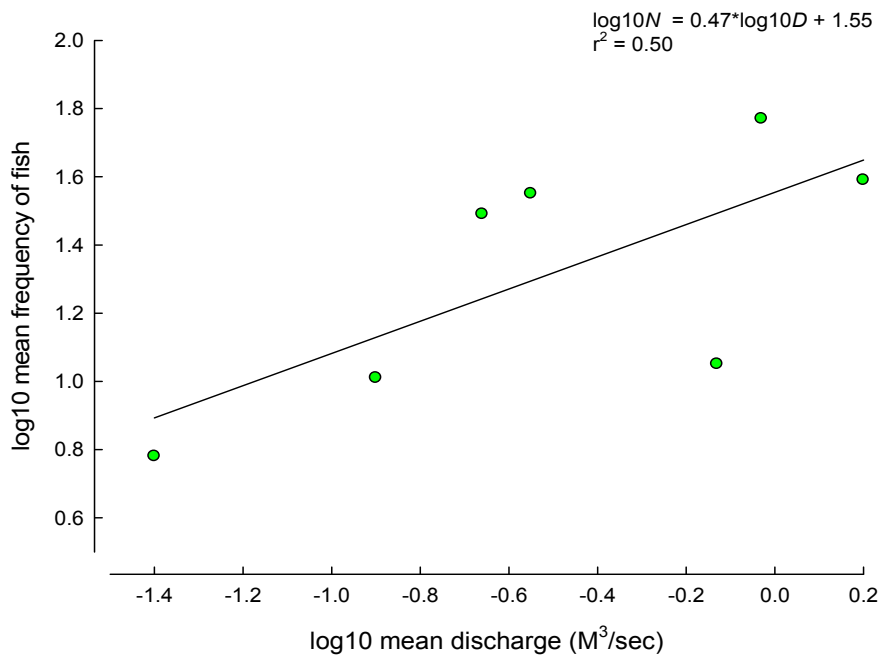


**Figure 28** Length-frequency histograms, differentiated by upstream and downstream movement, of Western Minnow in McAtee Brook.



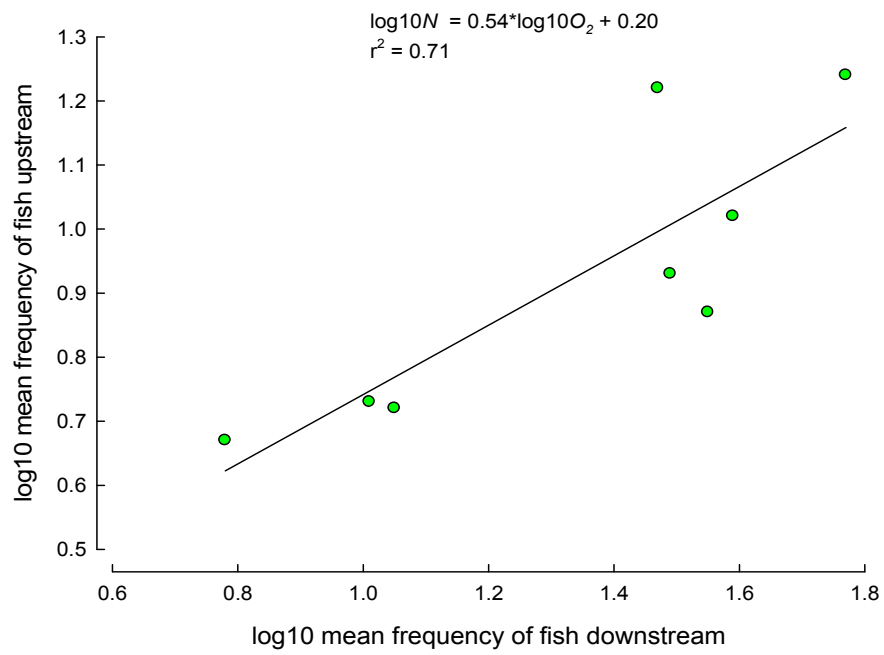
**Figure 29**

Relationship between the mean strength of upstream migration of Western Minnows within the major flow period (August and December 2005/06-2007) and the mean discharge in the four tributaries during that period. N.B. Data were log<sub>10</sub> transformed and migration was standardised for effort, see text for details.



**Figure 30**

Relationship between the mean strength of downstream migration of Western Minnows within the major flow period (August to December 2005/06-2007) and the mean discharge in the four tributaries during that period. N.B. Data were log<sub>10</sub> transformed and migration was standardised for effort, see text for details.



**Figure 31** Relationship between the mean upstream and downstream migration of Western Minnows in the four tributaries within the major flow period (August to December 2005/06-2007) N.B. Data were log10 transformed and migration was standardised for effort, see text for details.



**Table 3** Correlations between upstream and downstream movement of Western Minnows in the tributaries of the Blackwood River and prevailing environmental variables during the major migration period. N.B. Data were log10 transformed, \* denotes correlation is significant at the 0.05 level \*\* at 0.01 level (2-tailed).

		Log temperature	Log conductivity	Log pH	Log O2	Log discharge	Log NTU	Downstream movement
Log conductivity	Pearson Correlation	.560						
	Sig. (2-tailed)	.149						
Log pH	Pearson Correlation	.127	-.311					
	Sig. (2-tailed)	.764	.453					
Log O2	Pearson Correlation	.022	.301	-.580				
	Sig. (2-tailed)	.958	.469	.132				
Log discharge	Pearson Correlation	.140	.731	.022	-.174			
	Sig. (2-tailed)	.765	.062	.963	.709			
Log NTU	Pearson Correlation	.292	.951(*)	-.311	-.033	.999(*)		

Log downstream movement	Sig. (2-tailed)	.708	.049	.689	.967	.023		
	Pearson Correlation	-.251	.453	-.419	.017	.708	.465	
Log upstream movement	Sig. (2-tailed)	.549	.260	.301	.968	.075	.535	
	Pearson Correlation	-.345	.220	-.265	-.016	.666	.045	.841(**)
	Sig. (2-tailed)	.402	.601	.527	.971	.102	.955	.009

\* Correlation is significant at the 0.05 level (2-tailed).

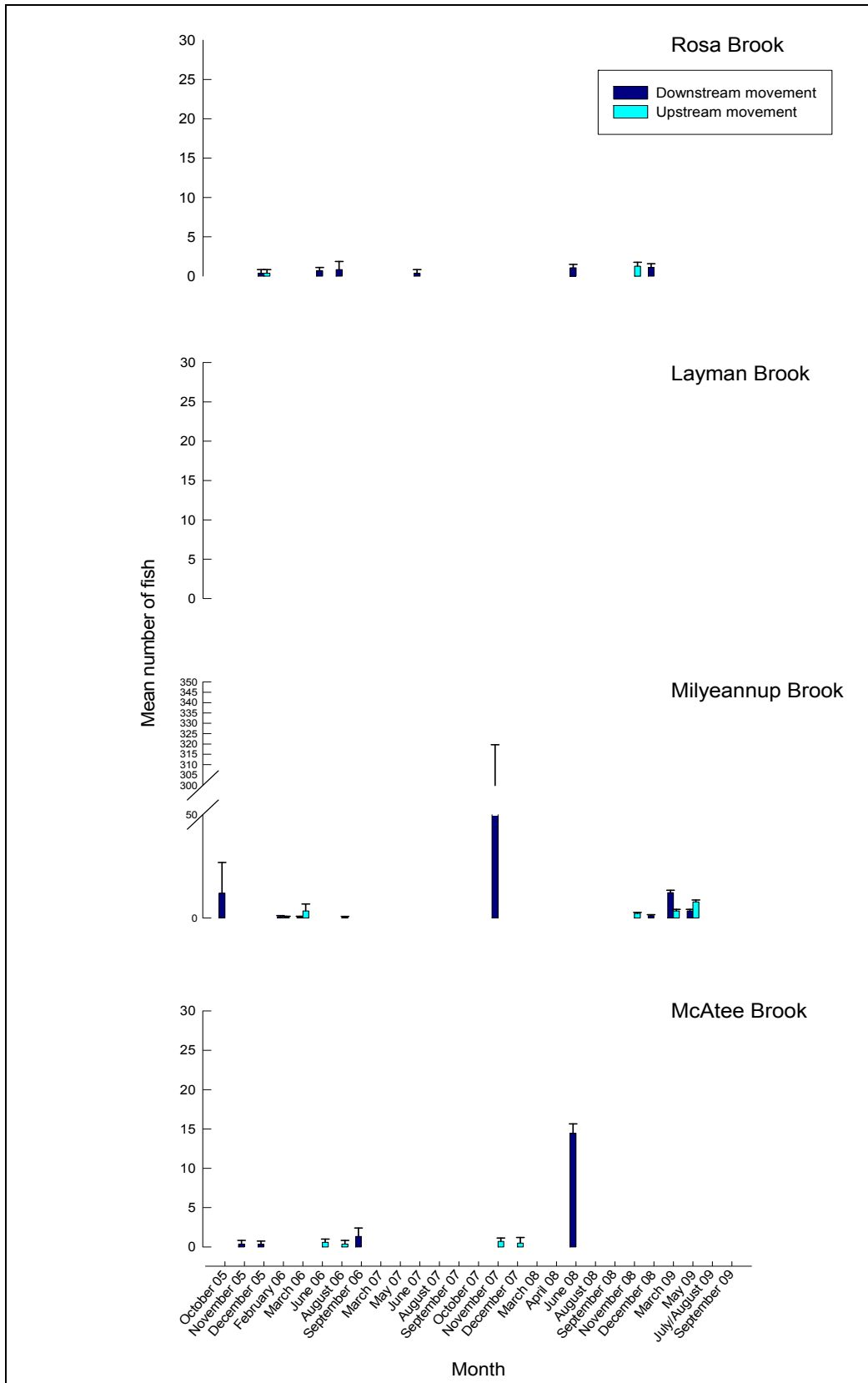
\*\* Correlation is significant at the 0.01 level (2-tailed).

## Mud Minnow



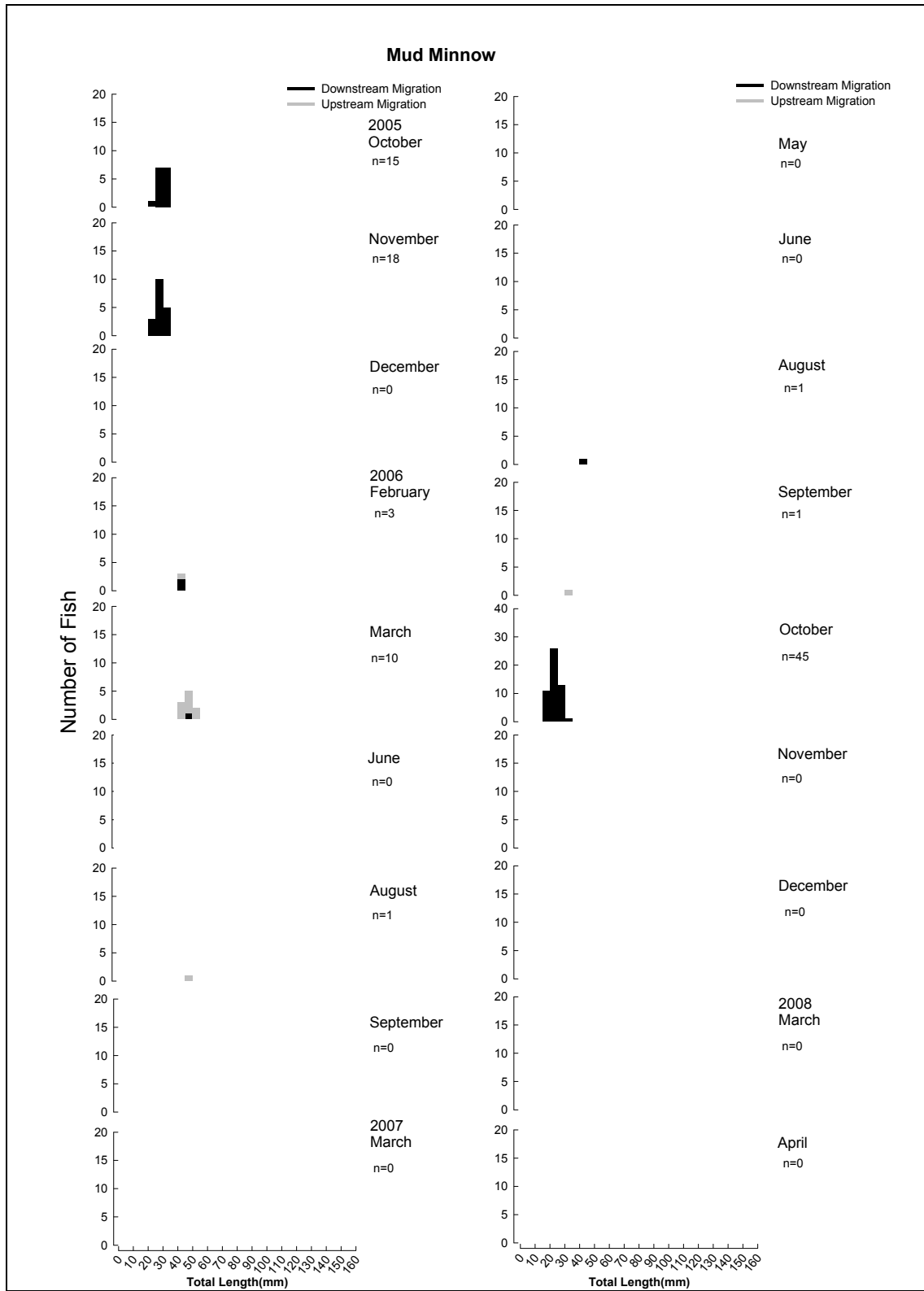
The Mud minnow was captured in relatively low numbers and was only captured in tributary sites (Figure 32). In the current study, it was recorded in Poison Gully, Milyeannup Brook, Rosa Brook and McAtee Brook. It is also known from Rosa Brook and Red Gully (Morgan and Beatty 2005). Phillips *et al.* (2007) found evidence that there are considerable genetic differences between these tributaries suggesting that the populations do not readily mix. They also found that Red Gully had the greatest genetic diversity of any population examined. The current study lends support to those findings and the salinity tolerance and habitat preferences of the species require determination to elucidate the mechanism by which the main channel of the Blackwood River is preventing mixing of those populations.

This species is known to have a one year lifecycle (Pen *et al.* 1991) and in the case of Rosa Brook, the breeding period is between August and October (Morgan *et al.* 2004). While the overall numbers of Mud Minnows captured was low, a large downstream migration of recently metamorphosed juvenile Mud Minnows occurred in Milyeannup Brook during October and November 2005 and again in October 2007 suggesting this to be an important breeding habitat for the species (Figures 32 and 33).



**Figure 32** Upstream and downstream movement of Mud Minnow in the tributaries.





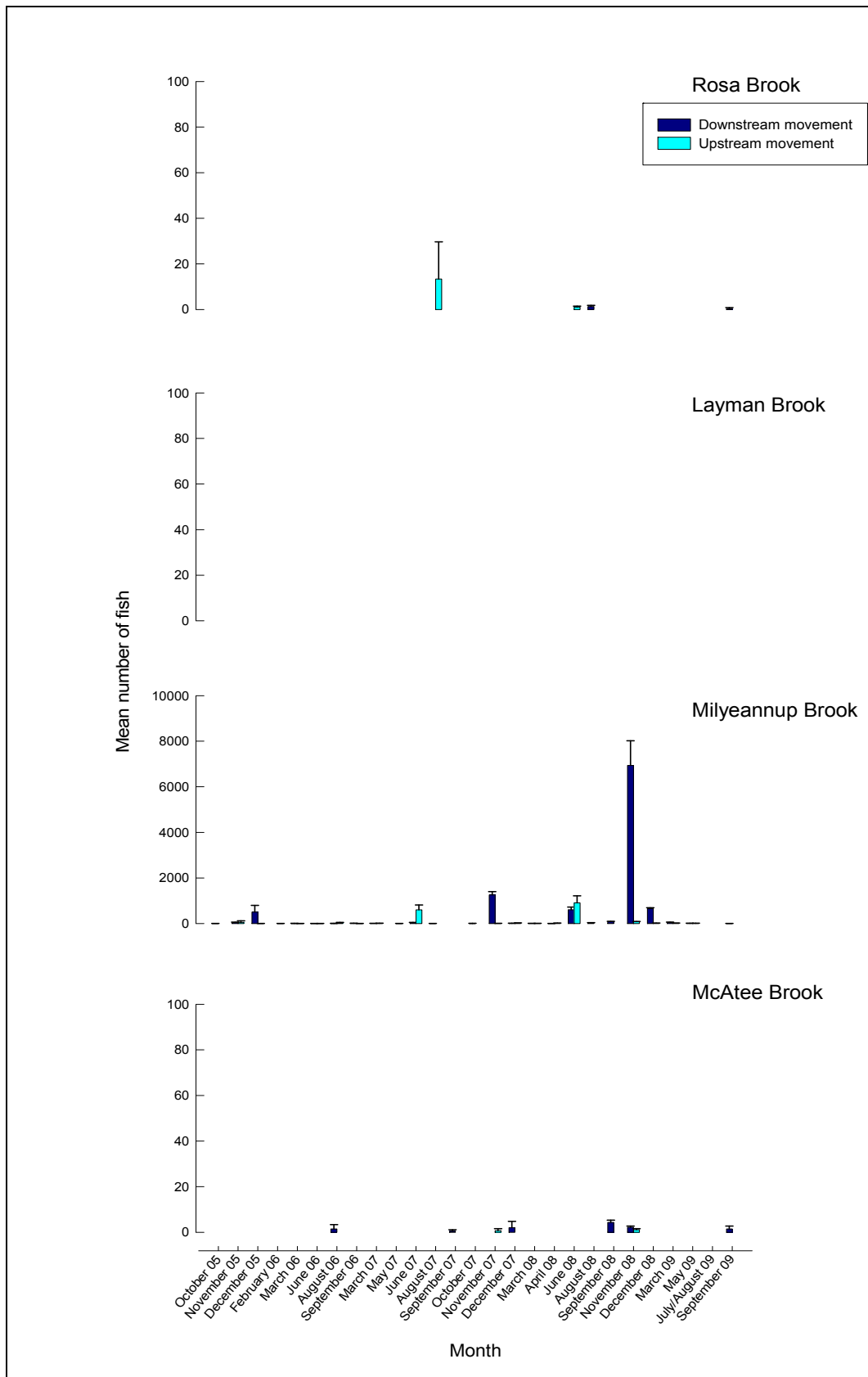
**Figure 33** Length-frequency histograms, differentiated by upstream and downstream movement, of Mud Minnow in Milyeannup Brook.

## Balston's Pygmy Perch

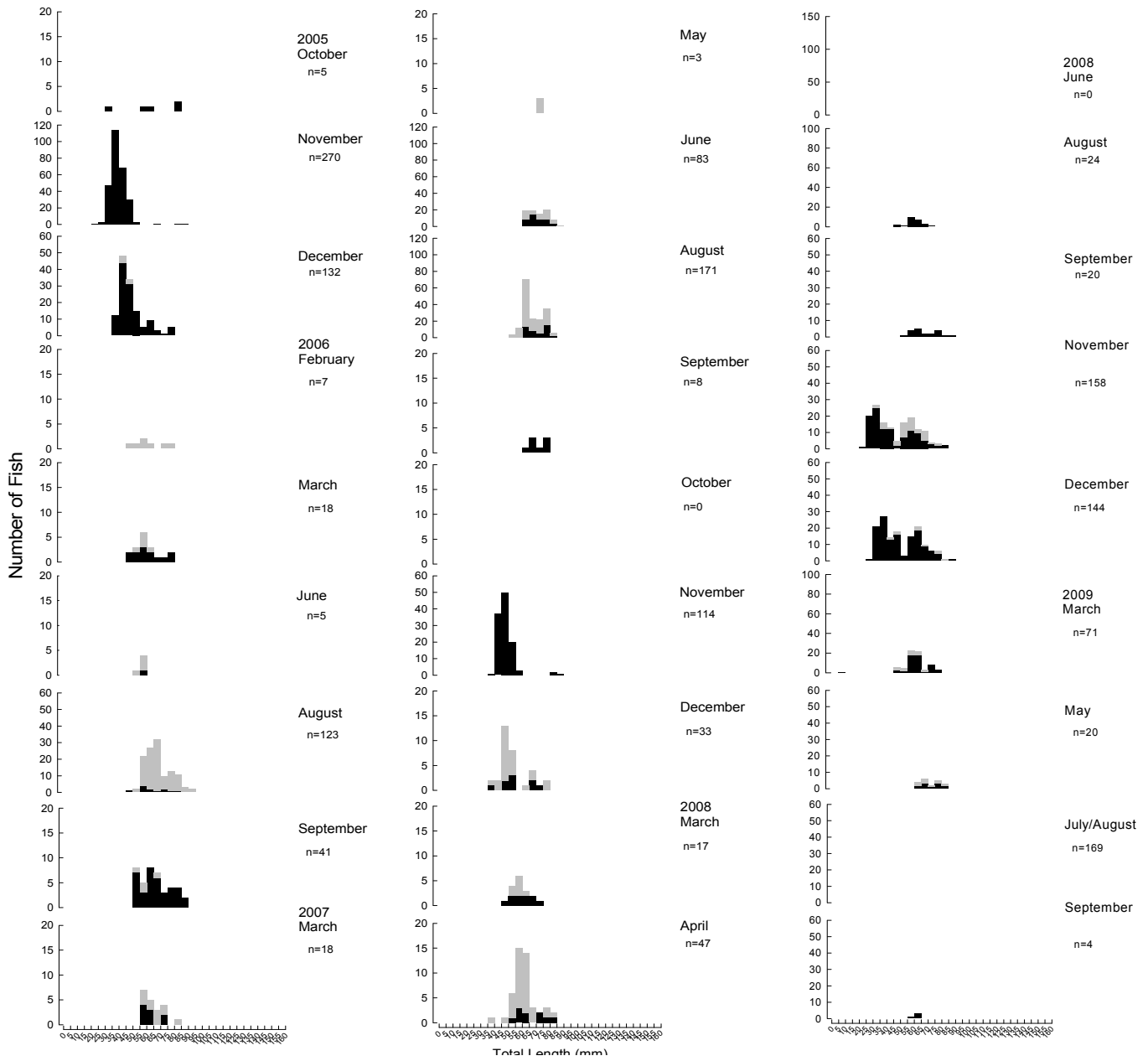


Balston's Pygmy Perch is listed as *Vulnerable* under the *EPBC Act* and listed as Schedule 1 at State level (*Wildlife Conservation Act, 1950*) (Morgan 2009). This study has demonstrated that Milyeannup Brook is the only breeding habitat for this species in the Blackwood River catchment; probably due to the consistency of suitable available habitat facilitated by the permanency of flow due to groundwater discharge in this system. Much of the upper catchment of the Blackwood River is now unsuitable for this endangered species due to secondary salinisation. Acute salinity trials revealed that this species has an acute tolerance of ~8 ppt; compared with ~14 ppt for Western Minnow and Western Pygmy Perch (Beatty *et al.* 2008). Although there are areas upstream of the current study area that have salinities less than 8ppt, the tolerance of earlier life history stages of all of these species (i.e. larvae) is as yet unknown and would probably be lower than the acute tolerance of the adults (Beatty *et al.* 2008).

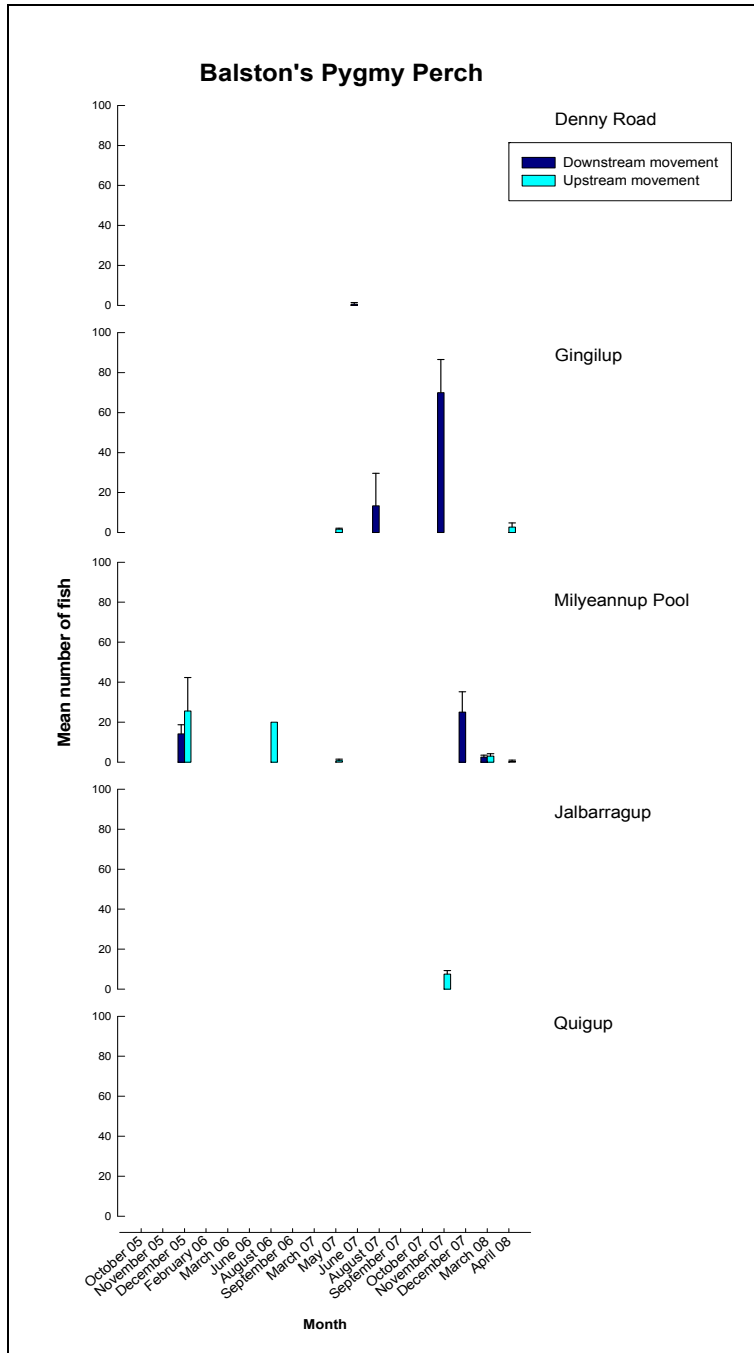
This species was captured on 25 sampling months in Milyeannup Brook. The large downstream movement of juveniles annually only in Milyeannup Brook (Figures 34 and 35) demonstrates this tributary as the breeding habitat for the species. During 2007 and 2008 larger numbers were captured in the main channel than in the previous year. This highlights the importance of the long-term sampling that has been undertaken in this study as it allows quantification of interannual variations in migration patterns. The vast majority of these fish were captured in the zone of Yarragadee Aquifer intrusion into the main channel (i.e. Milyeannup Pool (below the mouth of Milyeannup Brook) and Gingilup) and analysis of the population structure confirms these individuals correspond to the age cohorts moving from Milyeannup Brook into the main channel at those times (Figures 36 and 37). These data show that Balston's Pygmy species utilises the main channel within the major groundwater intrusion zone downstream from Milyeannup Brook.



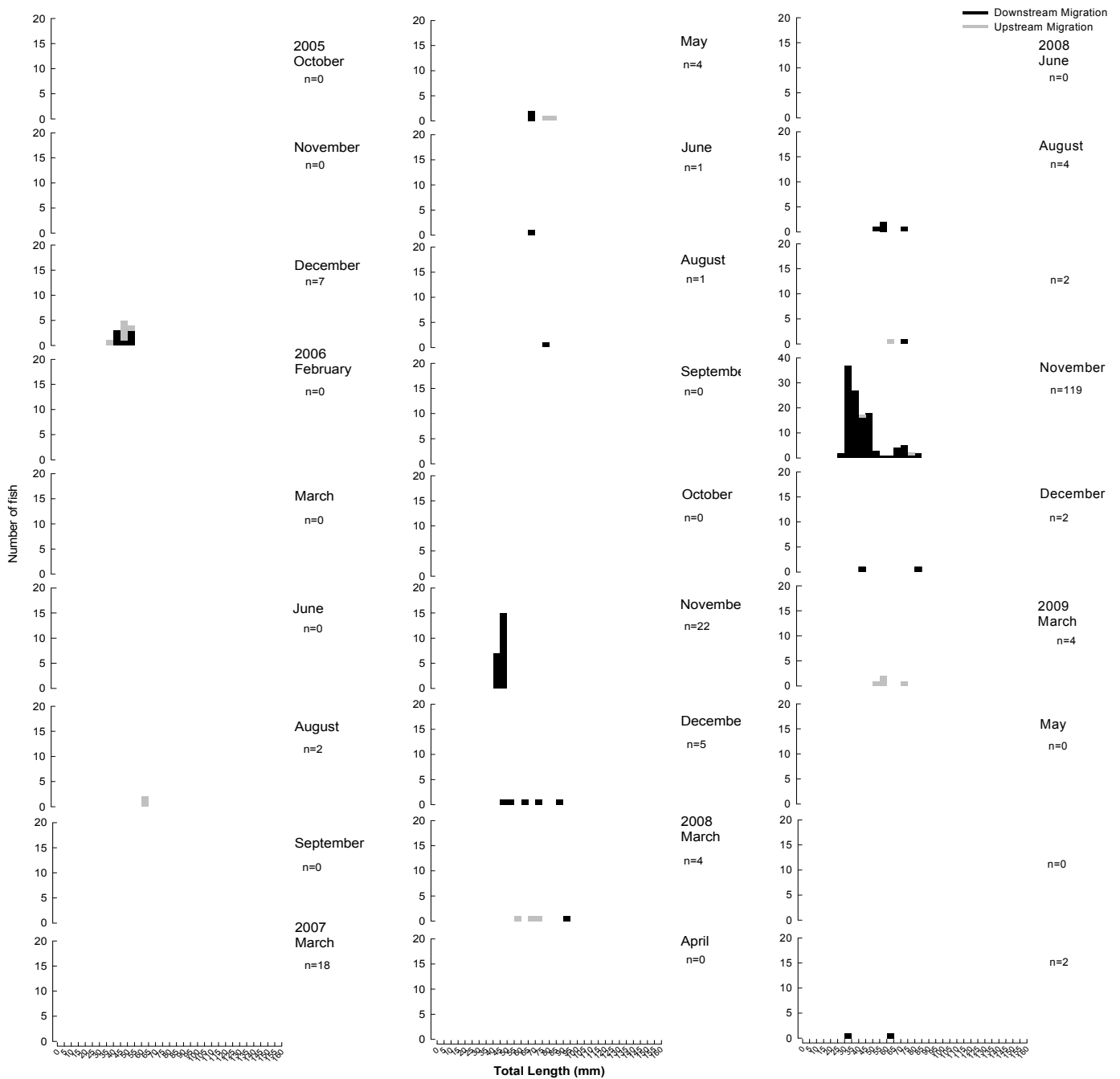
**Figure 34** Upstream and downstream movement of Balston's Pygmy Perch in the tributaries.



**Figure 35** Length-frequencies of Balston's Pygmy Perch, differentiated by upstream and downstream movement, in Milyeannup Brook between October 2005 and September 2009.



**Figure 36** Upstream and downstream migration of Balston's Pygmy Perch in the Blackwood River main channel.



**Figure 37** Length-frequencies of Balston's Pygmy Perch, differentiated by upstream and downstream movement, in the Blackwood River main channel between October 2005 and September 2009.

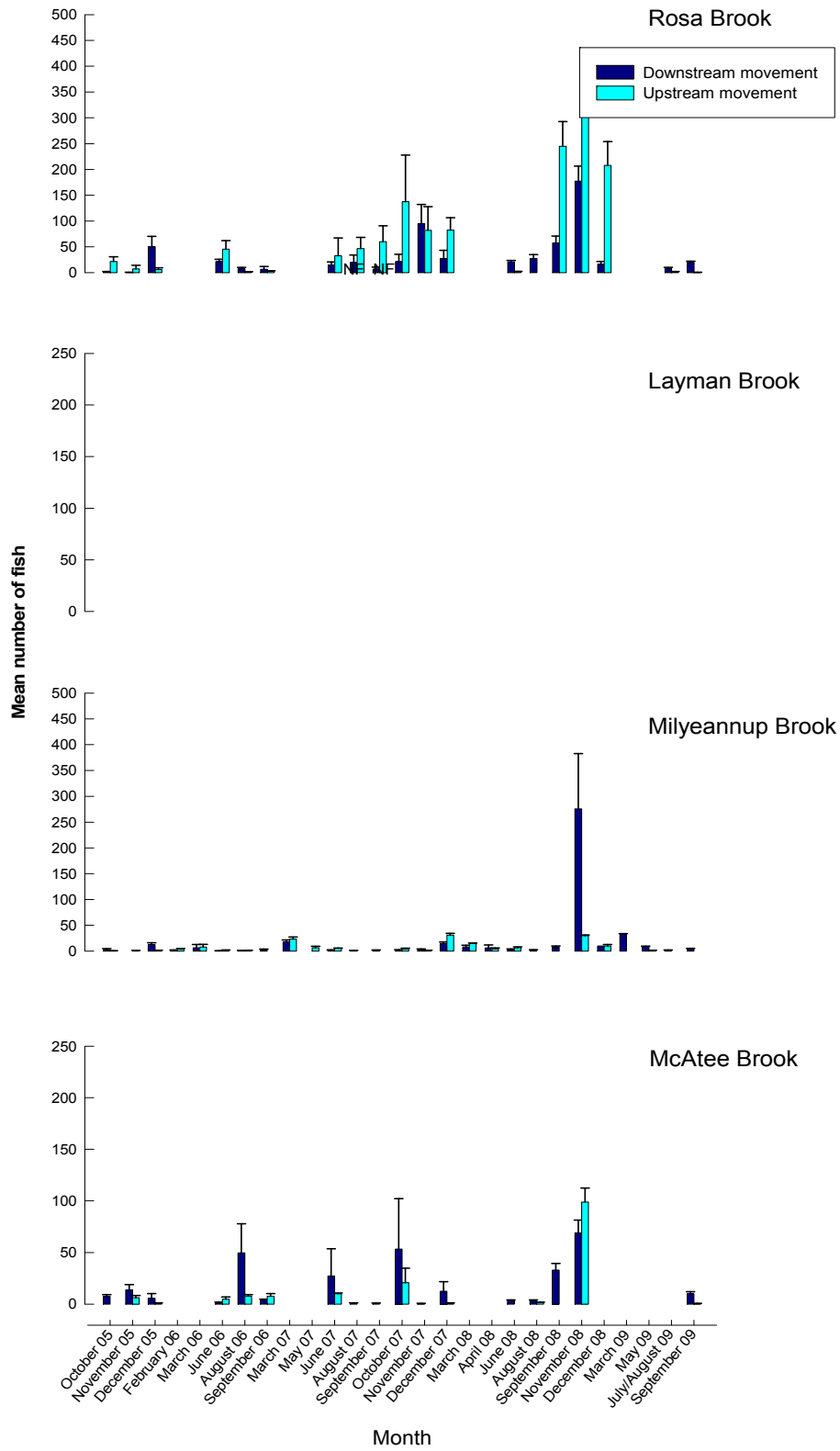
## Western Pygmy Perch



Western Pygmy Perch was recorded in the majority of sampling occasions moving within Rosa Brook, Milyeannup Brook and McAtee Brook and was absent from Layman Brook (Figure 38). The species was also recorded moving in the main channel of the Blackwood River at the more downstream sites in spring and early summer in 2007 and 2008 (Figure 39). This movement activity in tributary sites peaked in late winter and early spring which corresponds to its breeding period and the greatest movement was recorded in spring 2008. Migration strength was greatest within Rosa Brook; which appeared to support the largest population of Western Pygmy Perch of any system. The majority of movement within the main channel was in a downstream direction, however, upstream movement was recorded during the period of low flow in summer 2008/2009. In contrast to other tributaries, within the perennial Milyeannup Brook, there continued to be upstream and downstream migrations throughout summer and early autumn; facilitated by the continuation of flows in this system resulting from direct groundwater discharge (Figure 38).

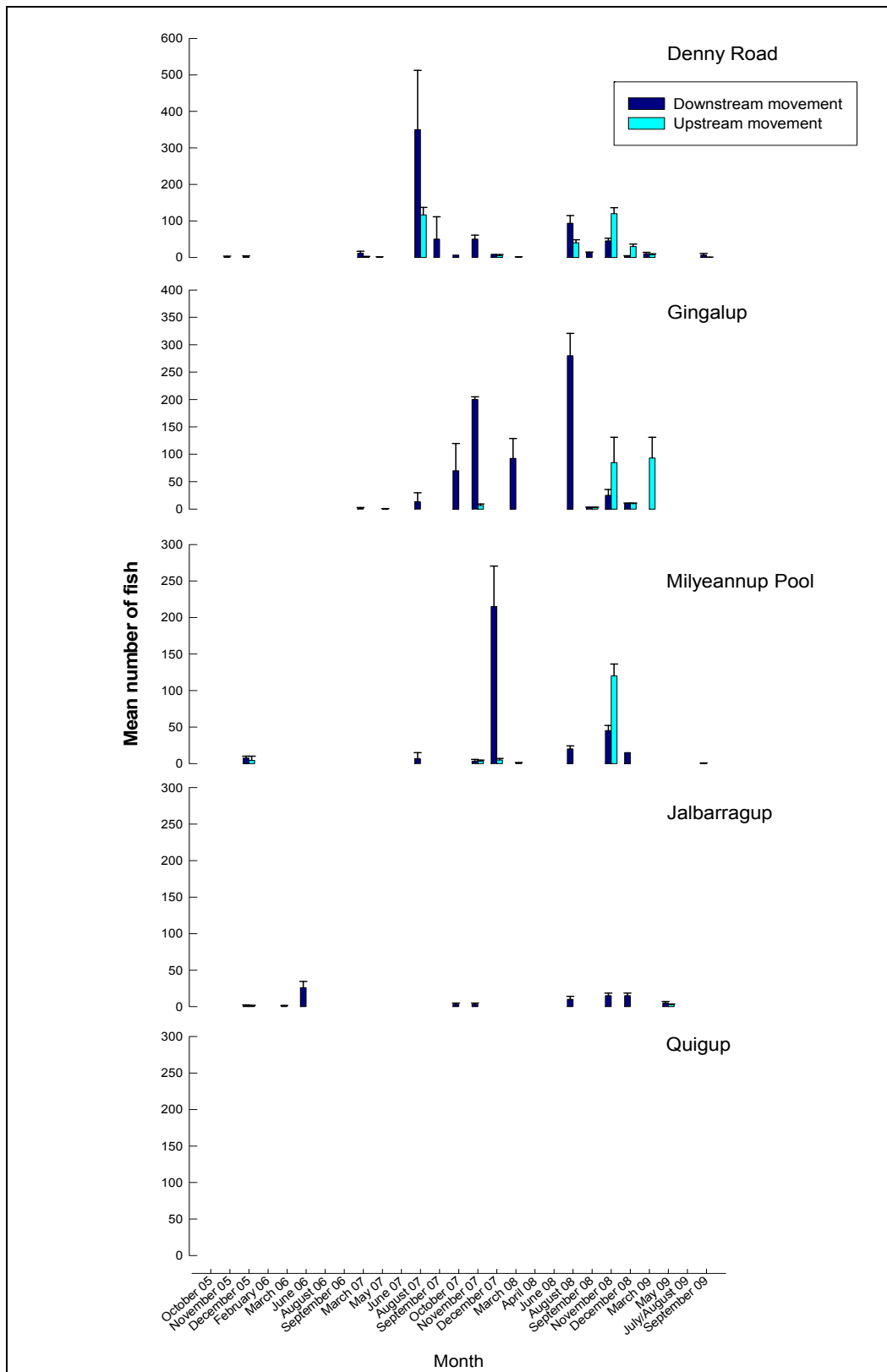
As was found with the Western Minnow, mean upstream and downstream movement of this species during the major migratory periods was significantly ( $p = 0.032$ ) positively related (Figure 40).

From the length-frequency distributions in Rosa, McAtee and Milyeannup Brooks, the species clearly breeds in these systems as evidenced by downstream movements of juveniles in these tributaries but not the main channel (Figures 41-44). Salinity trials (Beatty *et al.* 2008) found that the adults of the species are as tolerant as Western Minnow to sudden changes in salinity ( $LD_{50} = \sim 14$ ppt). The continued presence of the species in the main channel confirms that environmental conditions (particularly salinity) in this section of the river continue to be within its tolerance levels and enables it utilise this habitat; however, it breeds within the tributary habitats.

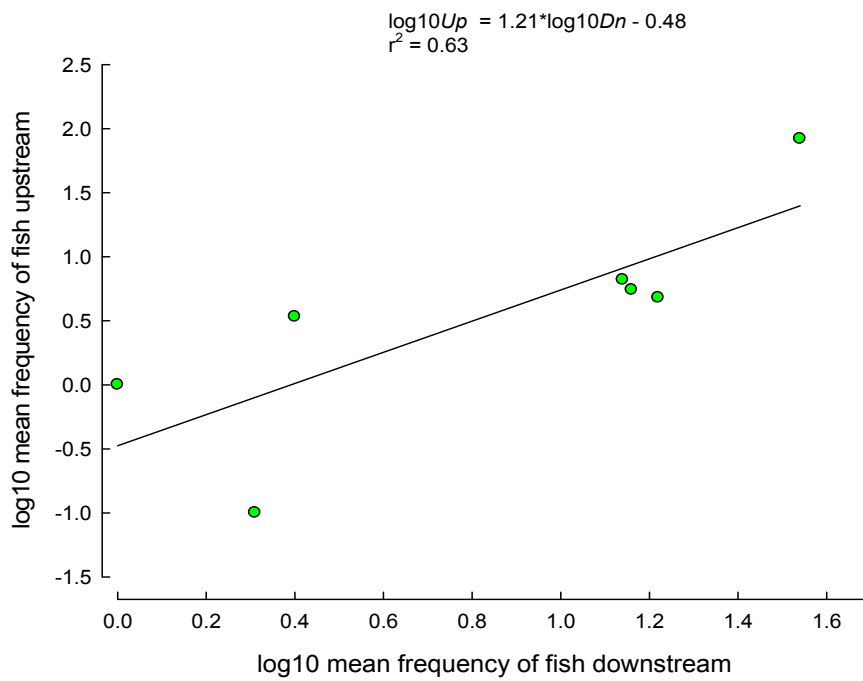


**Figure 38** Upstream and downstream migration of Western Pygmy Perch in the tributaries.

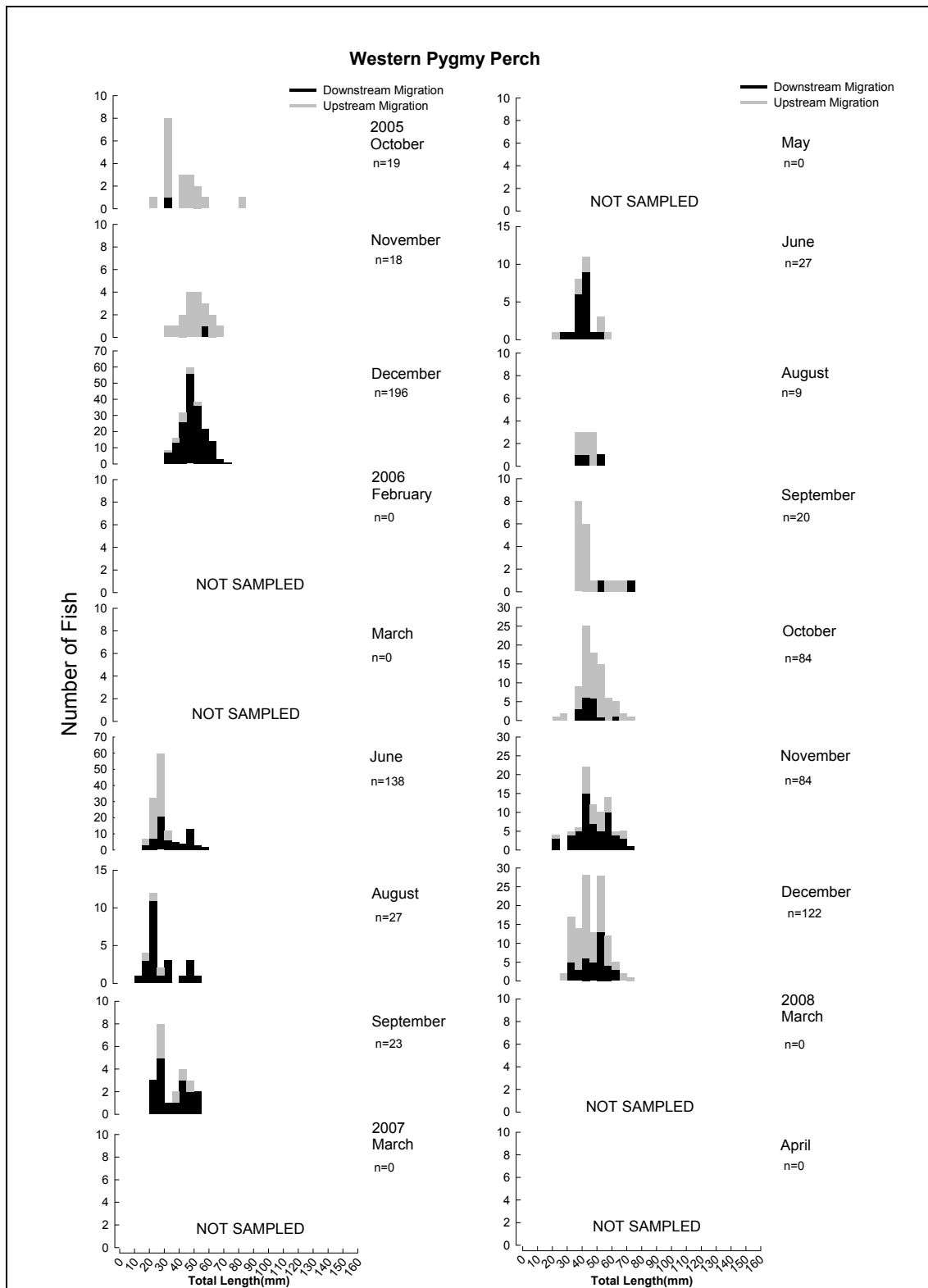




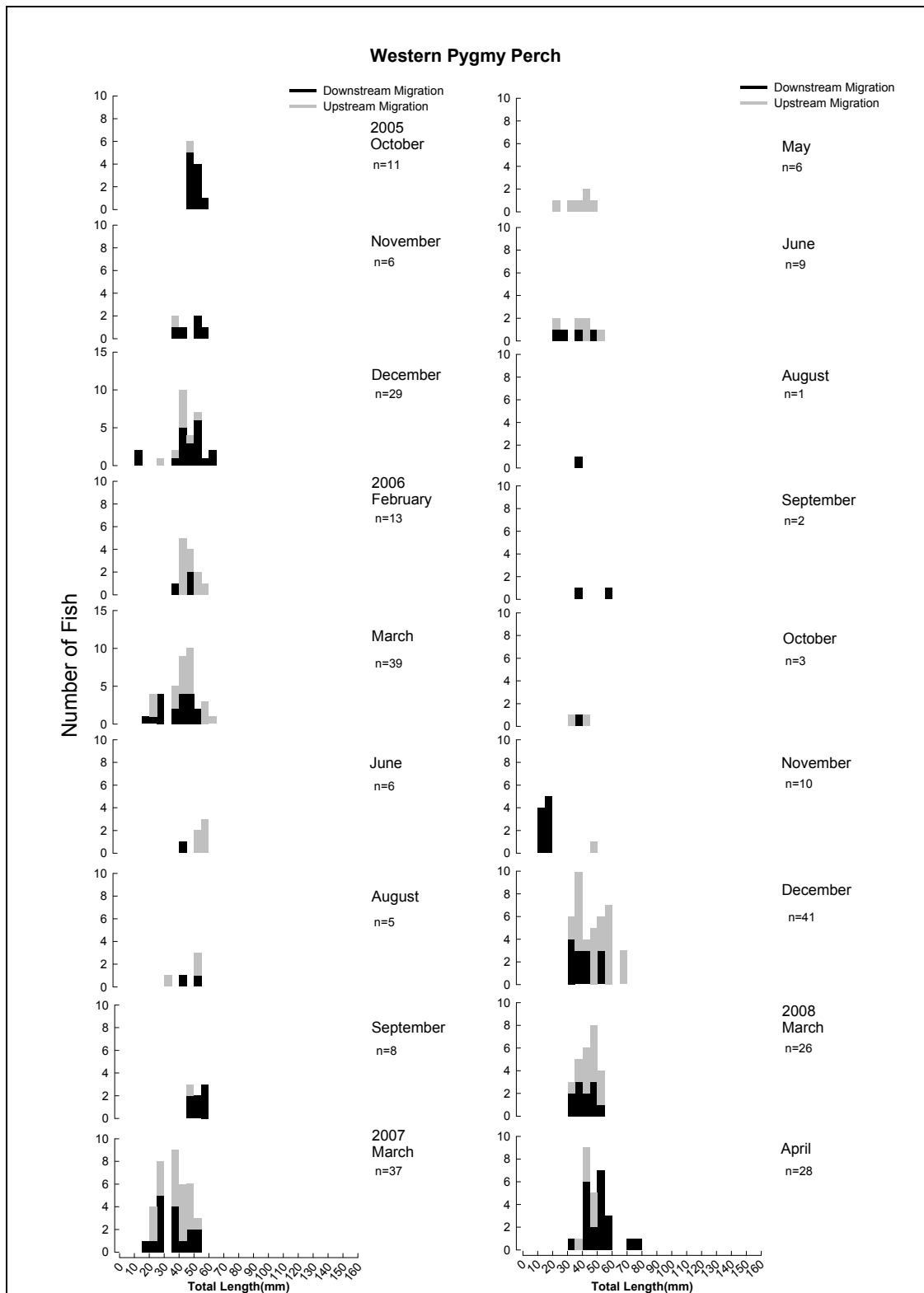
**Figure 39** Upstream and downstream migration of Western Pygmy Perch in the Blackwood River main channel.



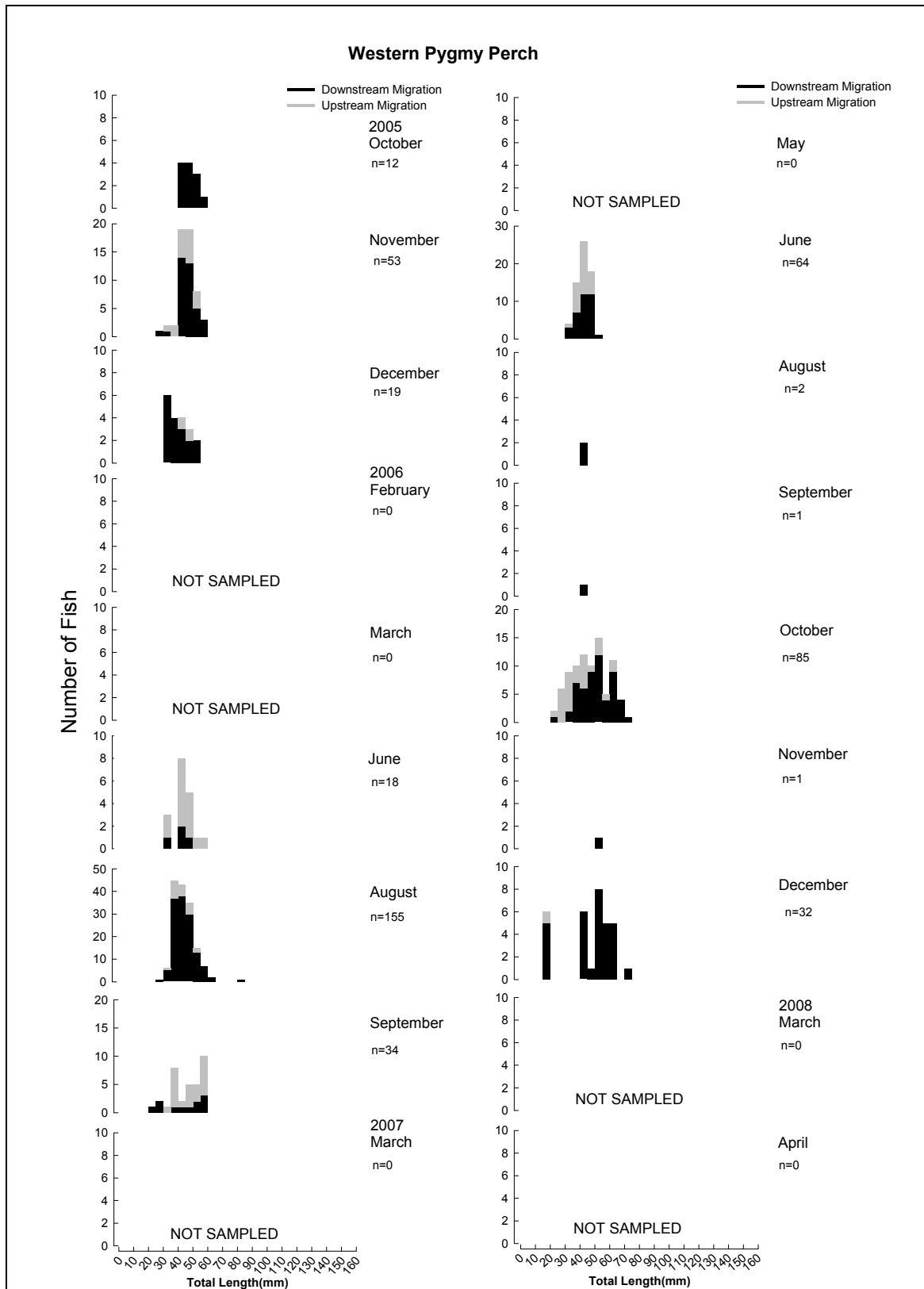
**Figure 40** Relationship between the mean upstream and downstream migration of Western Pygmy Perch in the four tributaries within the major flow period. N.B. Data were log10 transformed and migration was standardised for effort, see text for details.



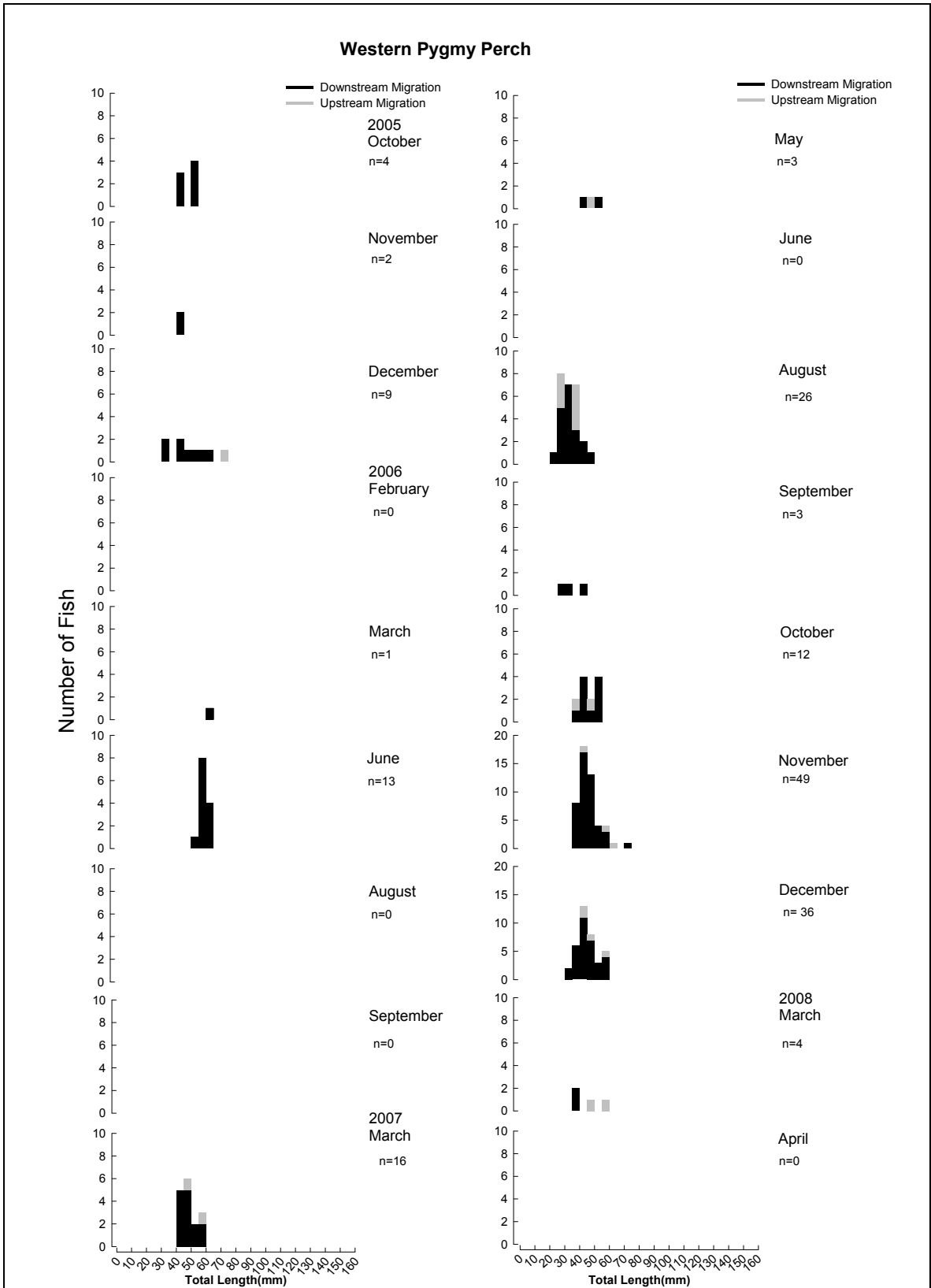
**Figure 41** Length-frequency histograms, differentiated by upstream and downstream movement, of Western Pygmy Perch in Rosa Brook.



**Figure 42** Length-frequency distributions, differentiated by upstream and downstream movement, of Western Pygmy Perch in Milyeannup Brook.



**Figure 43** Length-frequency distributions, differentiated by upstream and downstream movement, of Western Pygmy Perch in McAtee Brook.



**Figure 44**

Length-frequency distributions, differentiated by upstream and downstream movement, of Western Pygmy Perch in the Blackwood River main channel.

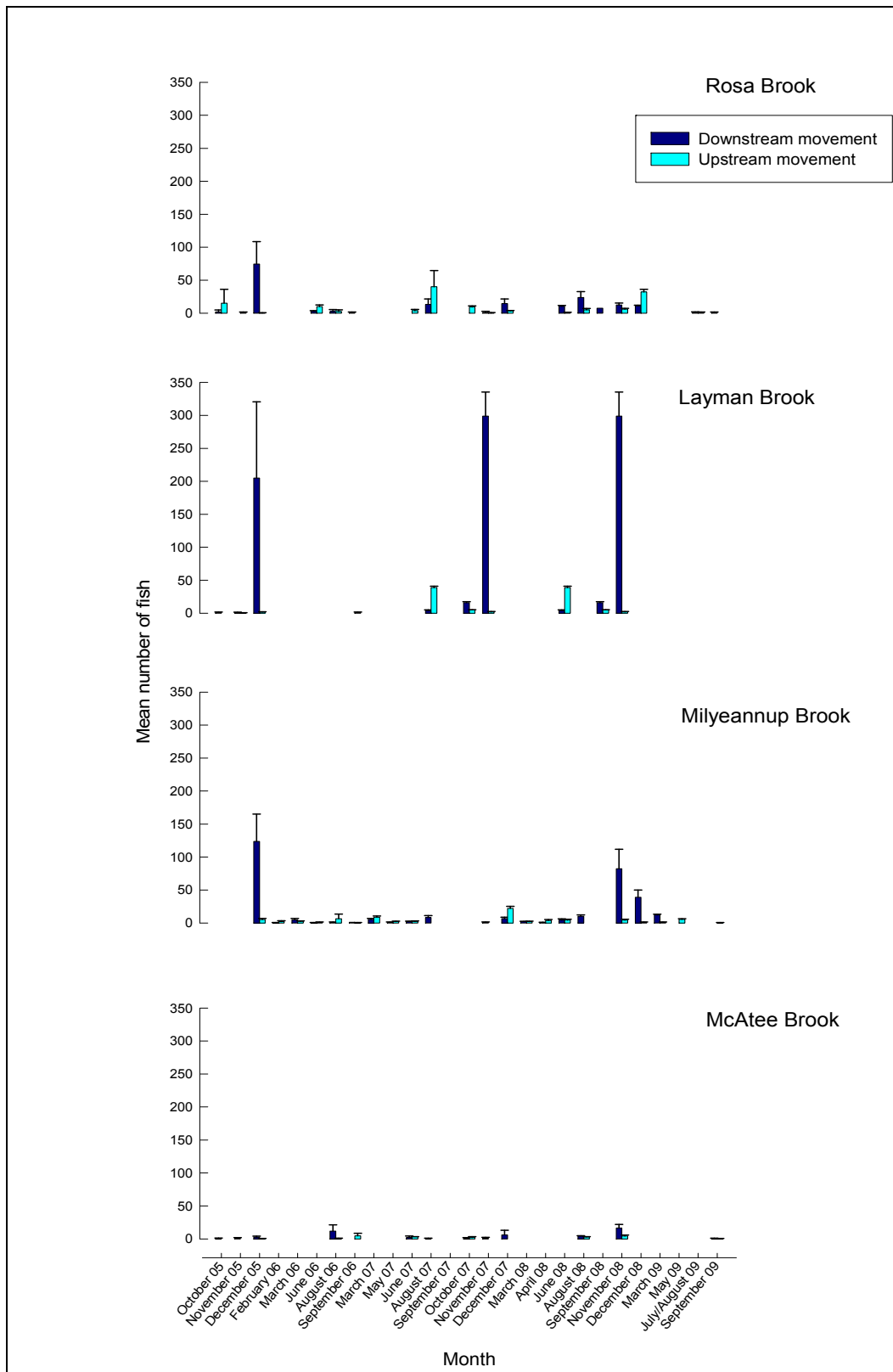
## Nightfish



The Nightfish was captured within all tributaries sampled and also within each main channel site (Figures 45-51). However, the species appears to be reliant on tributary habitats for breeding with juveniles almost exclusively only being recorded from those systems (Figures 46-49).

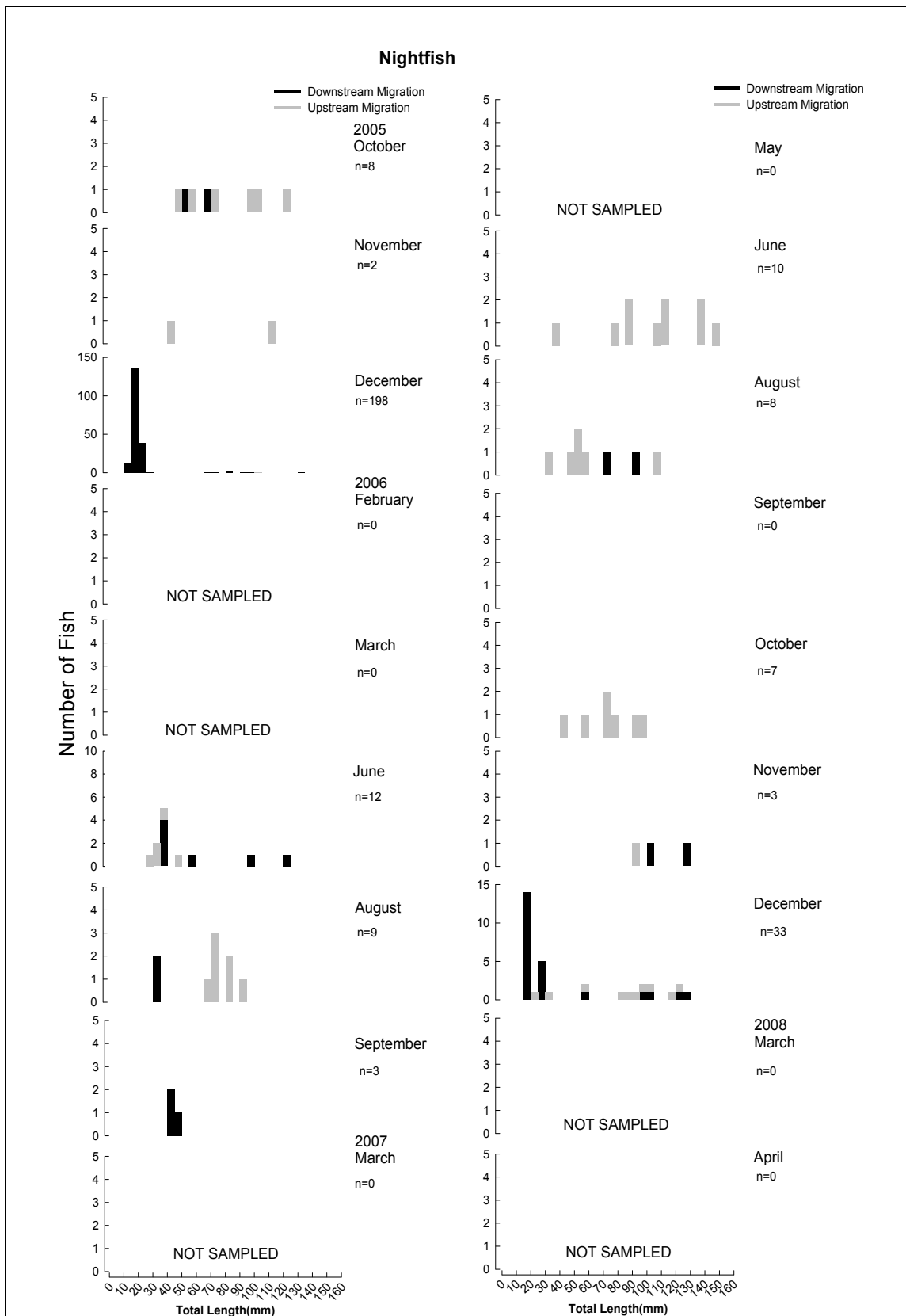
The Nightfish bred annually in Layman Brook as indicated by an annual downstream movement of juveniles in November suggesting a spawning in late winter early spring (Figures 45 and 48). This annual recruitment of juveniles was also recorded to a lesser degree in Rosa, Milyeannup and McAtee Brooks. The downstream juvenile migration also corresponds to a reduction in discharge from these tributaries.

Unlike the Western Minnow and Western Pygmy Perch, the mean upstream and downstream movement of this species during the major migratory periods was not significantly related. This suggests that greater strength of upstream movement of mature fish does not necessarily result in higher downstream movement of recruits (Table 6).

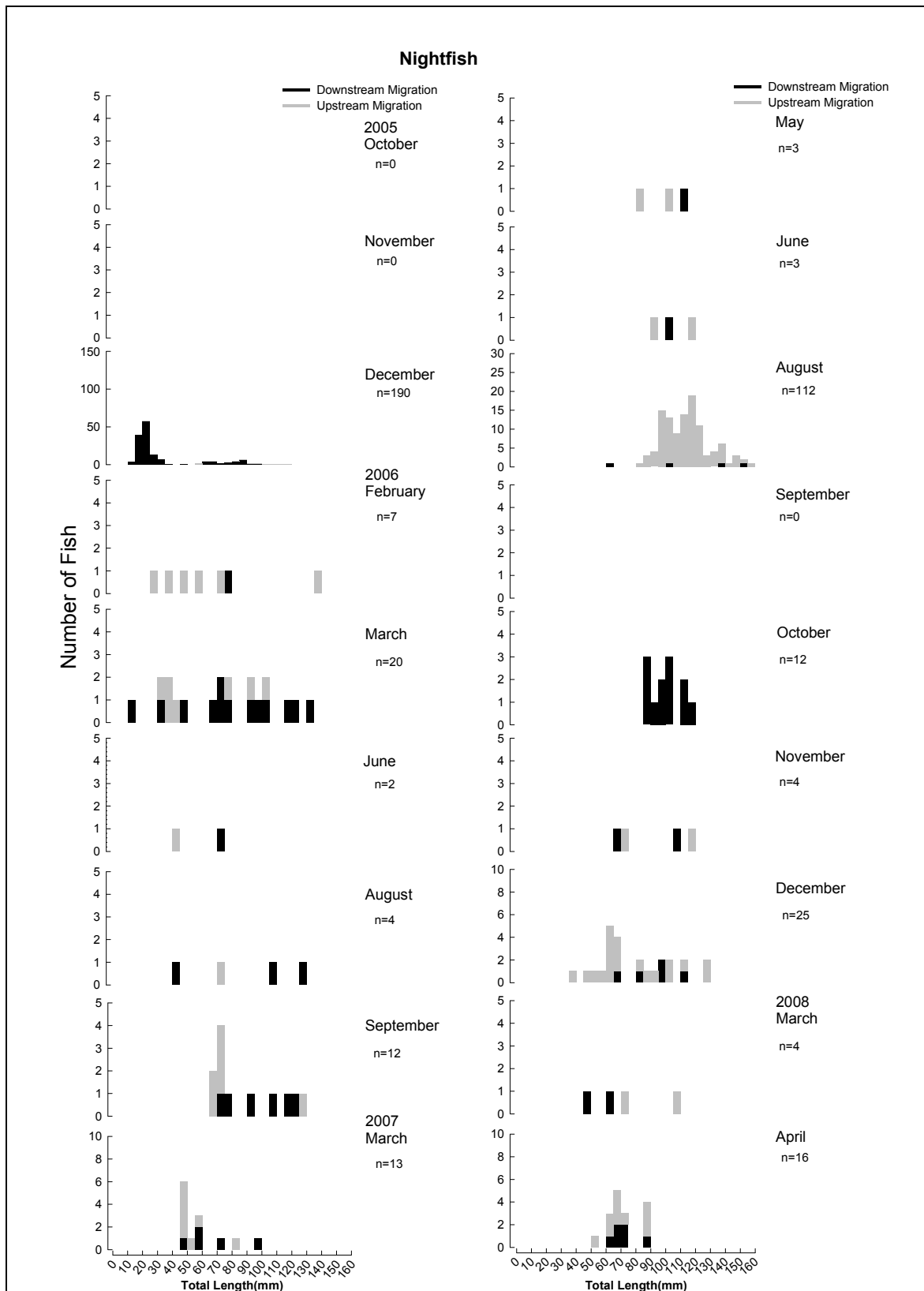


**Figure 45** Upstream and downstream migration of Nightfish in the tributaries.

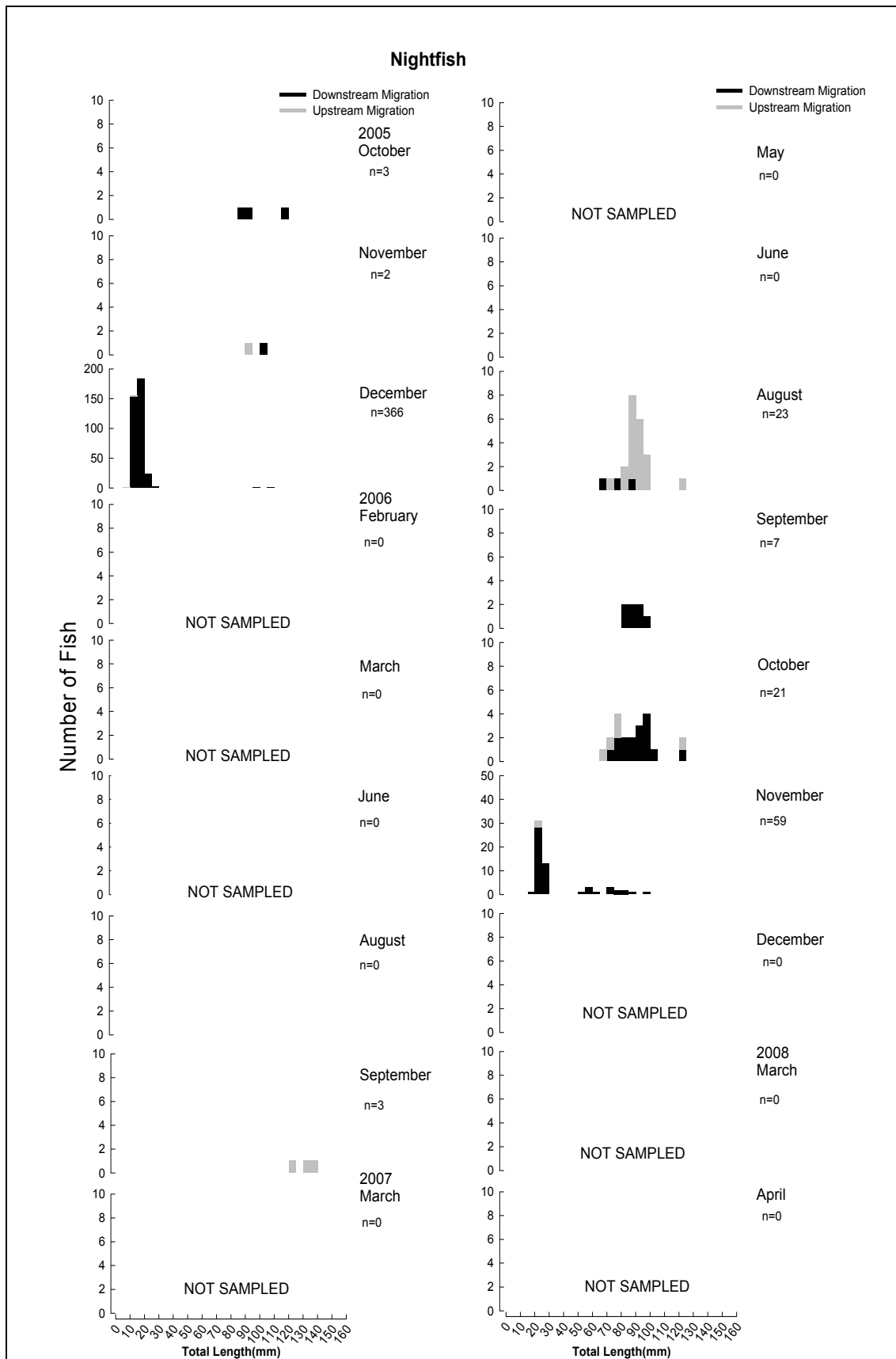




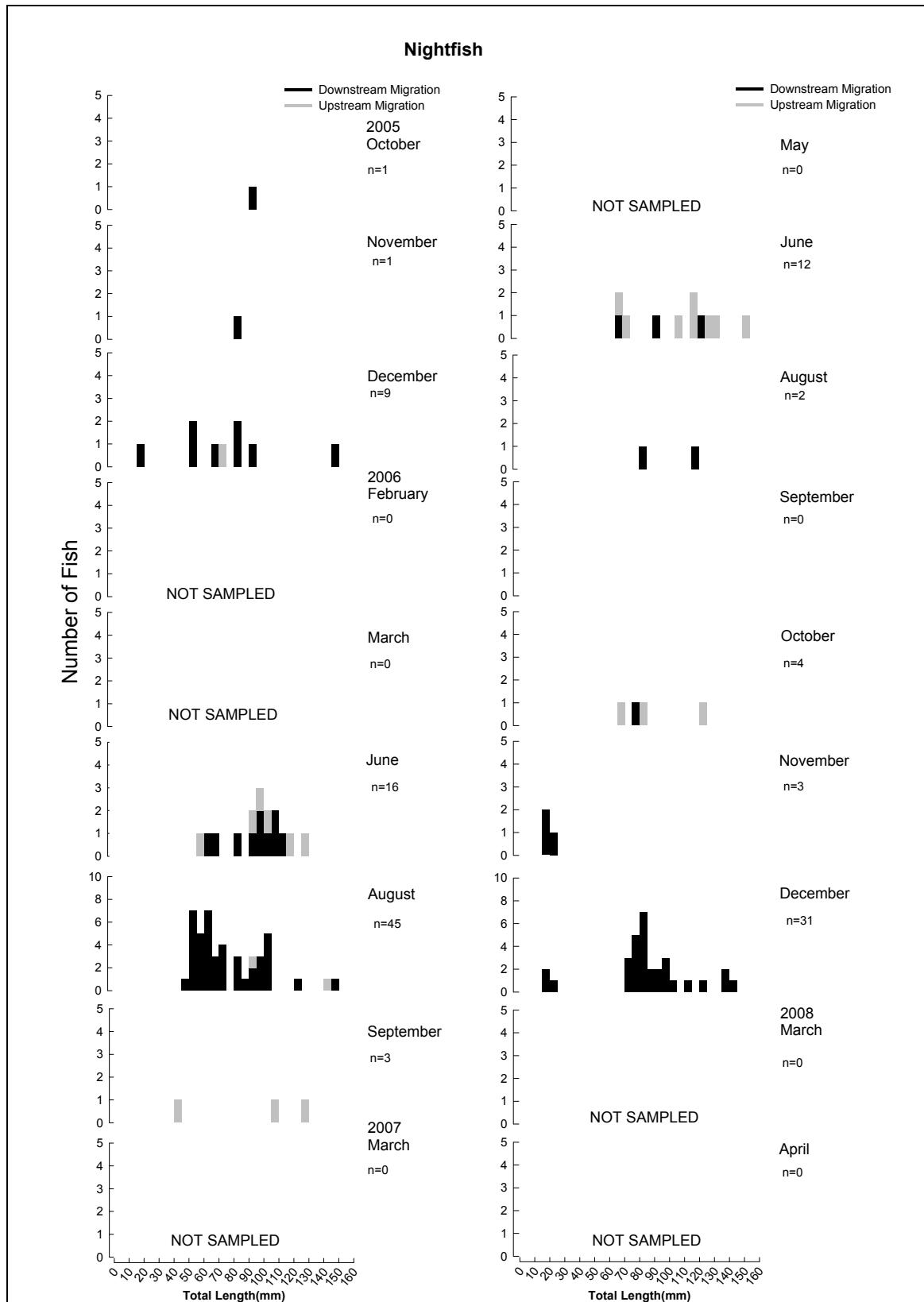
**Figure 46** Length-frequency histograms, differentiated by upstream and downstream movement, of Nightfish in Rosa Brook.



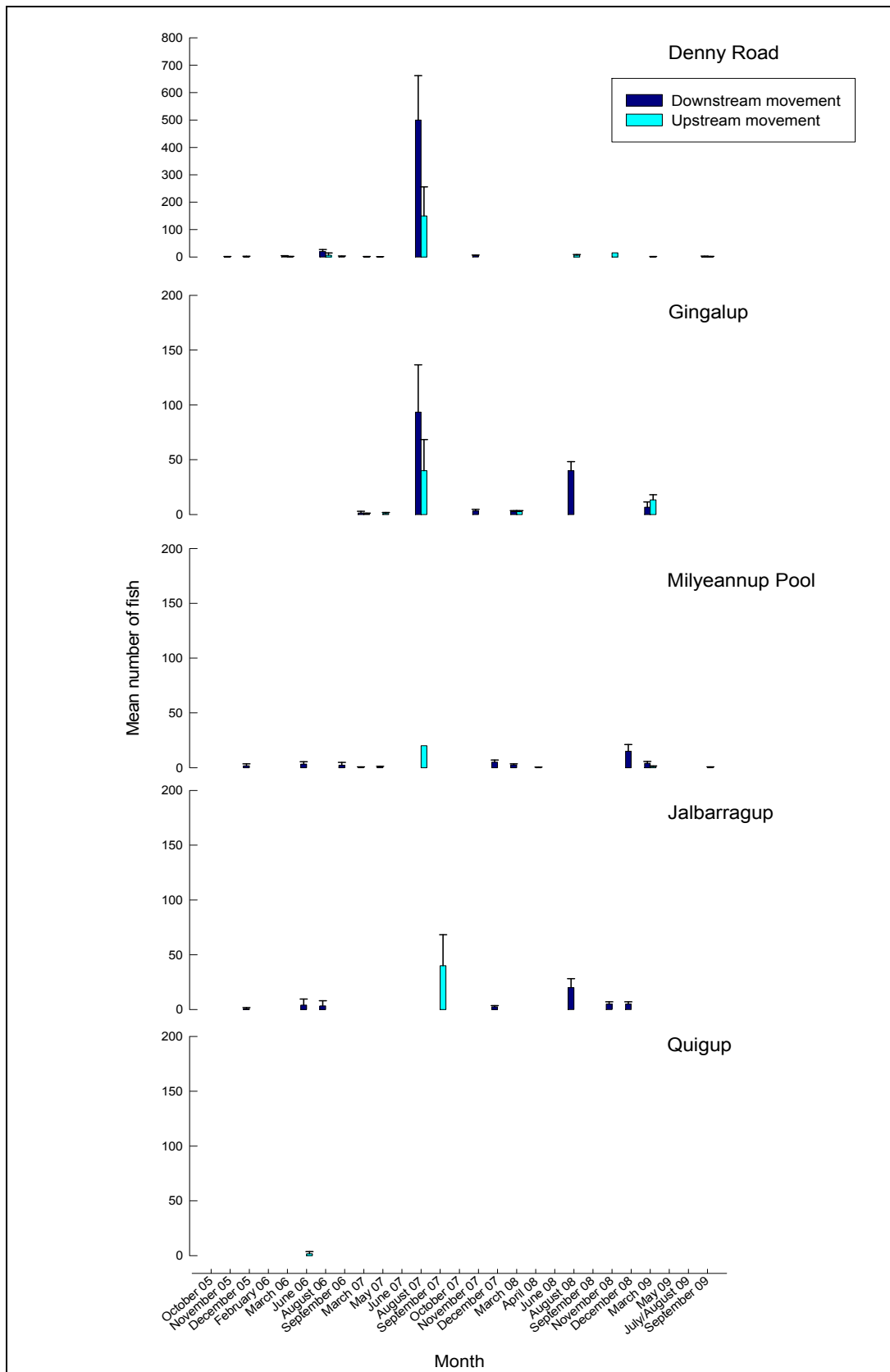
**Figure 47** Length-frequency histograms, differentiated by upstream and downstream movement, of Nightfish in Milyeannup Brook



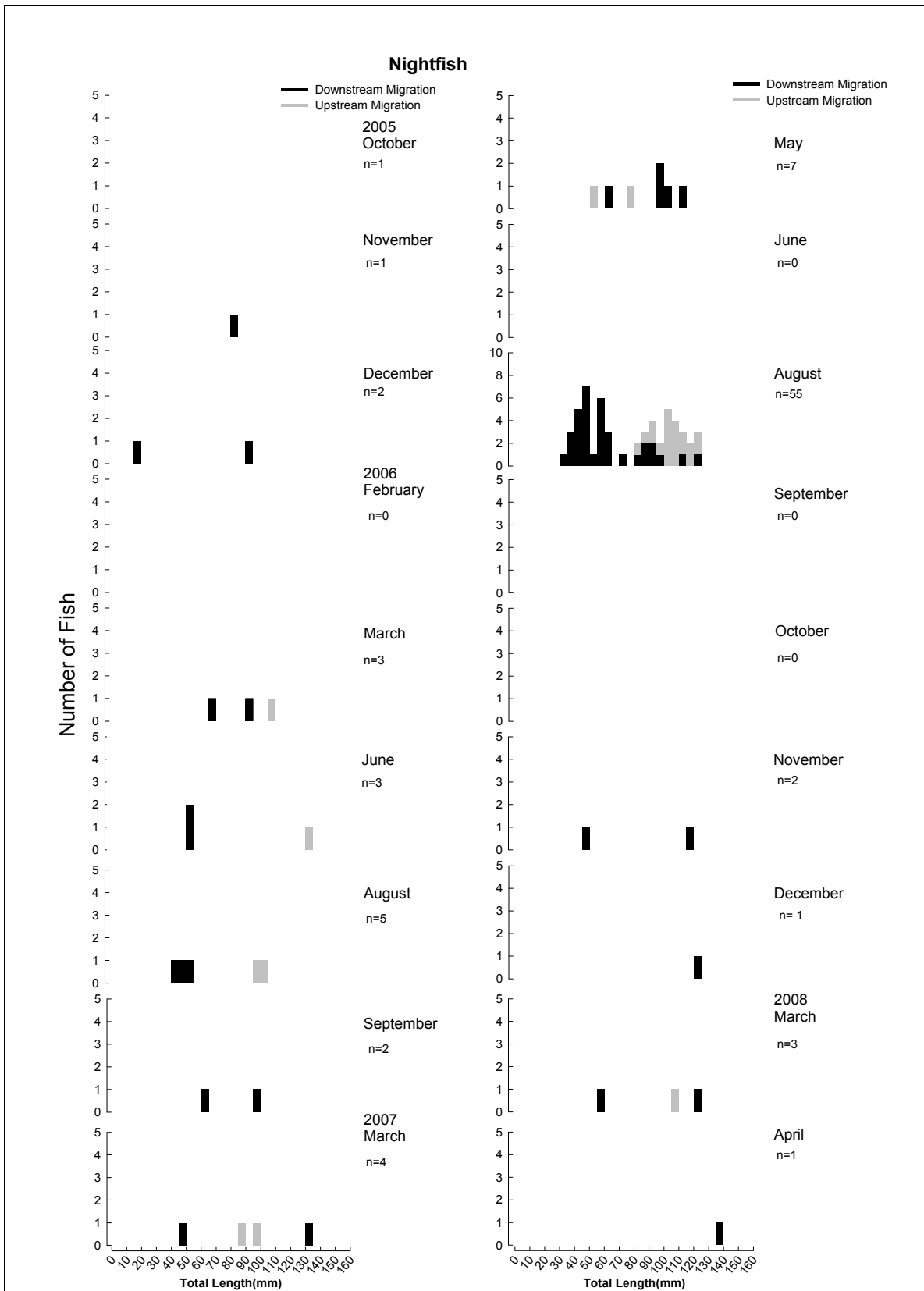
**Figure 48** Length-frequency histograms, separated by upstream and downstream migration, of Nightfish in Layman Brook.



**Figure 49** Length-frequency histograms, separated by upstream and downstream migration, of Nightfish in McAtee Brook.



**Figure 50** Upstream and downstream movement of Nightfish in the Blackwood River main channel.



**Figure 51** Length-frequency histograms, differentiated by upstream and downstream movement, of Nightfish in the Blackwood River main channel.



**Table 6** Correlations between upstream and downstream movement of Nightfish in the tributaries of the Blackwood River and prevailing environmental variables during the major migration periods. N.B. Data were log10 transformed, \* denotes correlation is significant at the 0.05 level \*\* at 0.01 level (2-tailed).

		Log temperature	Log conductivity	Log pH	Log O2	Log discharge	Log NTU	Downstream movement
Log conductivity	Pearson Correlation	.560						
	Sig. (2-tailed)	.149						
Log pH	Pearson Correlation	.127	-.311					
	Sig. (2-tailed)	.764	.453					
Log O2	Pearson Correlation	.022	.301	-.580				
	Sig. (2-tailed)	.958	.469	.132				
Log discharge	Pearson Correlation	.140	.731	.022	-.174			

Log NTU	Sig. (2-tailed)	.765	.062	.963	.709			
	Pearson Correlation	.292	.951(*)	-.311	-.033	.999(*)		
Downstream movement	Sig. (2-tailed)	.708	.049	.689	.967	.023		
	Pearson Correlation	-.660	-.608	.161	.182	-.589	-.388	
Upstream movement	Sig. (2-tailed)	.075	.110	.703	.667	.164	.612	
	Pearson Correlation	.191	.225	.678	-.228	.430	.264	-.187
	Sig. (2-tailed)	.650	.591	.064	.588	.335	.736	.657

\* Correlation is significant at the 0.05 level (2-tailed).

\*\* Correlation is significant at the 0.01 level (2-tailed)



## Milyeannup Brook baseflow fish habitat associations

Permanent flow in Milyeannup Brook was found to decrease from a distance of ~2500m from the Blackwood River in March 2006 (i.e. between Mil 6 and Mil 7) to between 1600m-1800 in 2007 and 2008. Site Mil 6 contained segmented, shallow pool habitats in March 2007 and 2008 and those sites upstream of that site were dry on both occasions (Table 7). Based on discharge estimates at Mil 3 in March 2007 and 2008, baseflow discharge was greater in 2007 (3.6 l/sec) compared to 2008 (2.0 l/sec). Negligible flow was observed at Mil 5 in both March 2007 and 2008 however; clearly deeper pool habitats existed in the former year presumably as a result of higher groundwater level that increased level of baseflow discharge downstream at Mil 3 (Table 7, Figure 52).

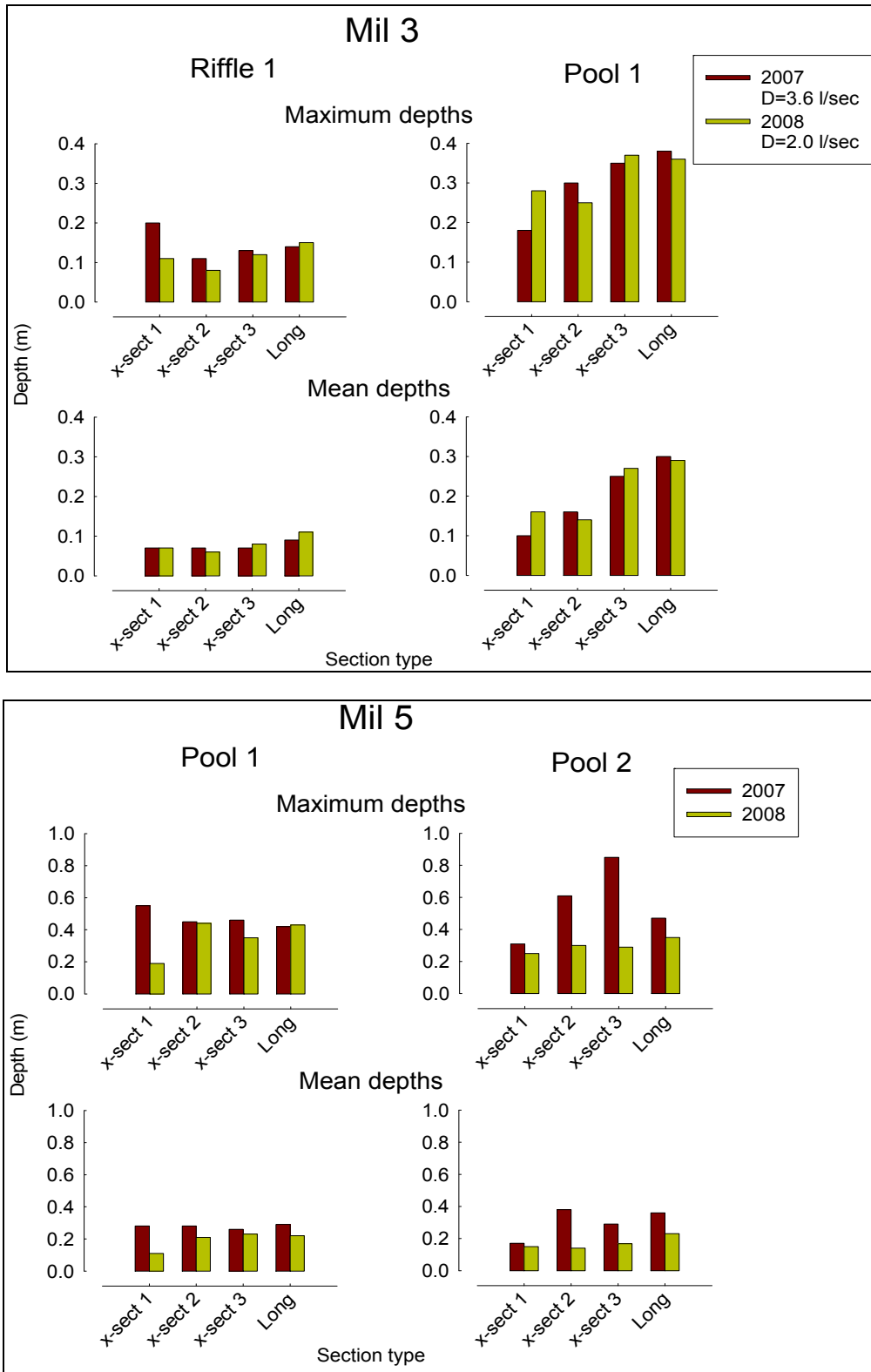
Densities of fishes in the habitats at the various sites are presented in Table 8. Balston's Pygmy Perch were consistently recorded at sites Mil 3 (2007, 2008) and Mil 4 (in 2007: not sampled in 2006, 2008). This species was not recorded at Mil 5 suggesting its upstream distribution (despite inter-annual variations in discharge) to be consistently between Mil 4 and Mil 5 (i.e. <1600m from the Blackwood River). However, as with other species it appears that it may have a preference for deeper pool habitats as suggested by the fact that the species was only recorded in pools and an (albeit relatively weak at this stage i.e.,  $r^2 = 0.35$ ) positive relationship existed between habitat depth and density of the species in Mil 3 and Mil 4 (Figures 53-55). Balston's Pygmy Perch were found in pool habitats with an average depth of >~19cm.

Other native fishes were consistently recorded as far upstream as Mil 5; further than the Balston's Pygmy Perch (Figure 53-55). Although the Western Minnow, Western Pygmy Perch and Nightfish were generally more prevalent and concentrated in pool habitats, each was recorded in riffle zones on at least one occasion during the sampling in the three sites in March 2007 and 2008. This suggests that these species may occupy a wider range of aquatic habitats in Milyeannup Brook during baseflow conditions than Balston's Pygmy Perch.

Freshwater crayfishes were shown to occupy both pool and riffle habitats with no clear preference being evident (Figures 53-55). The Gilgie, Restricted Gilgie and Koonac are known to occupy a wide range of permanent and temporary aquatic systems in the south-west region (Austin and Knott, 1996) and therefore the occupation of both riffle and pool habitats within Milyeannup Brook is not unexpected. Furthermore, their ability to tolerate drought by burrowing into the watertable also allows them to permanently inhabit the upstream, seasonally inundated, sections of Milyeannup Brook (and other tributaries of the Blackwood River).

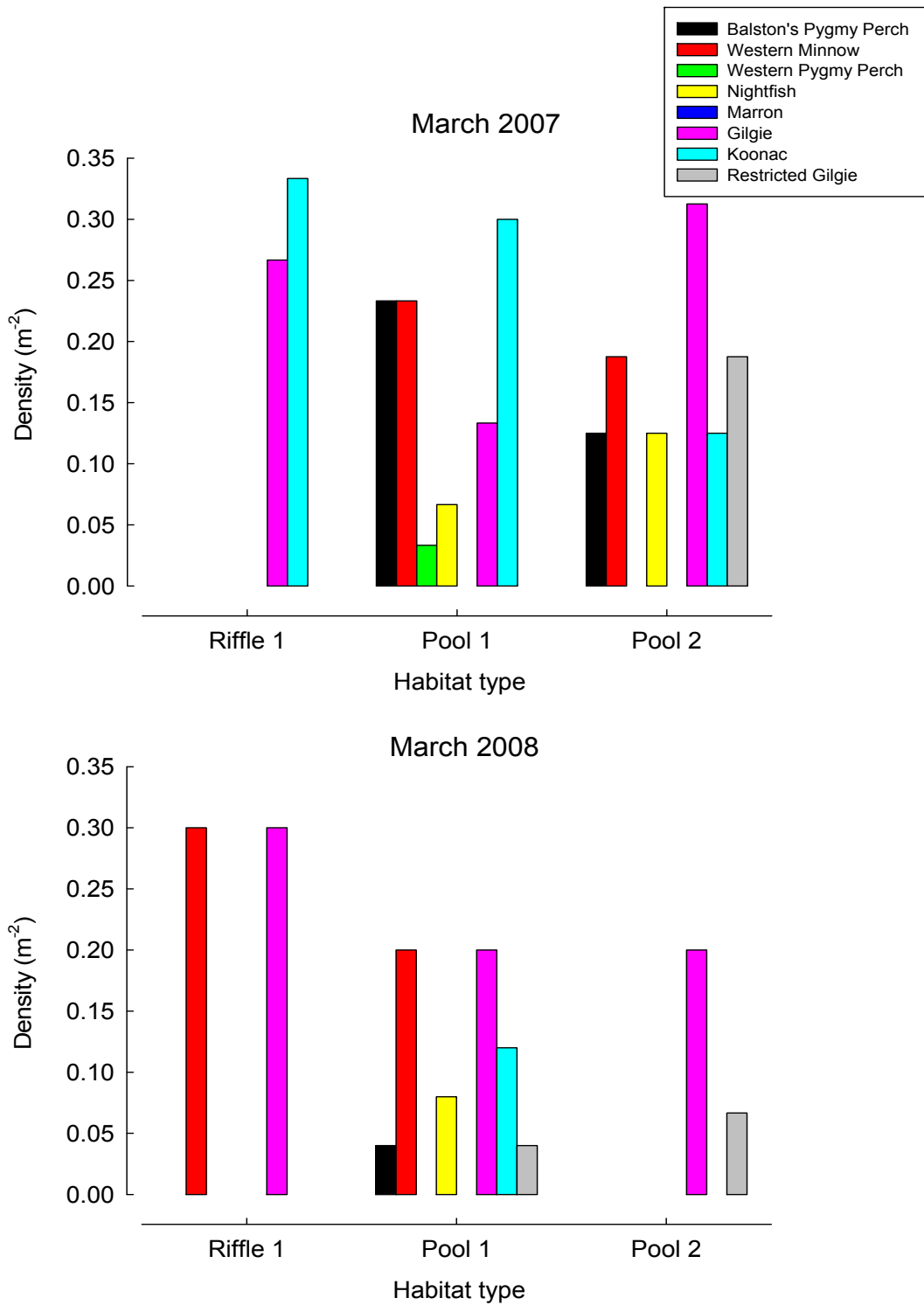
### Milyeannup Brook summary

- Inter-annual variation in baseflow in Milyeannup Brook is considerable as evidenced by a length of flow decrease from a distance of ~2500m from the Blackwood River in March 2006 down to between 1600-1800m in 2007 and 2008.
- The upstream extent of the baseflow distribution of Balston's Pygmy Perch in Milyeannup Brook is upstream distribution was consistently between Mil 4 and Mil 5 (i.e. <~1600m from the Blackwood River).
- Balston's Pygmy Perch was not recorded in riffle zones on any occasion and were only found in habitats with an average depth of >~19cm suggesting that it may have a preference for pool habitats.
- Western Minnow, Western Pygmy Perch and Nightfish were generally more prevalent and concentrated in pool habitats; however, each species was recorded in riffle zones suggesting they may have the ability to occupy a wider range of aquatic habitats in Milyeannup Brook during baseflow conditions than Balston's Pygmy Perch.
- Freshwater crayfishes were shown to occupy both pool and riffle habitats with no clear preference being evident.

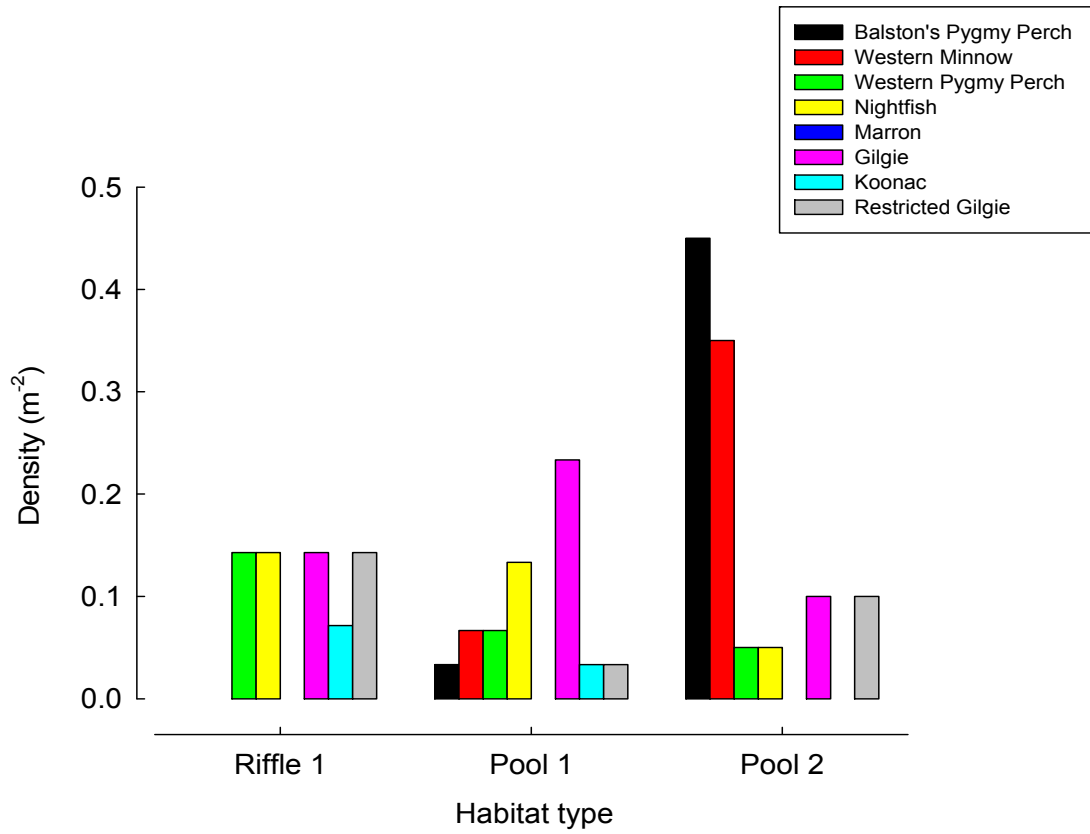


**Figure 52**

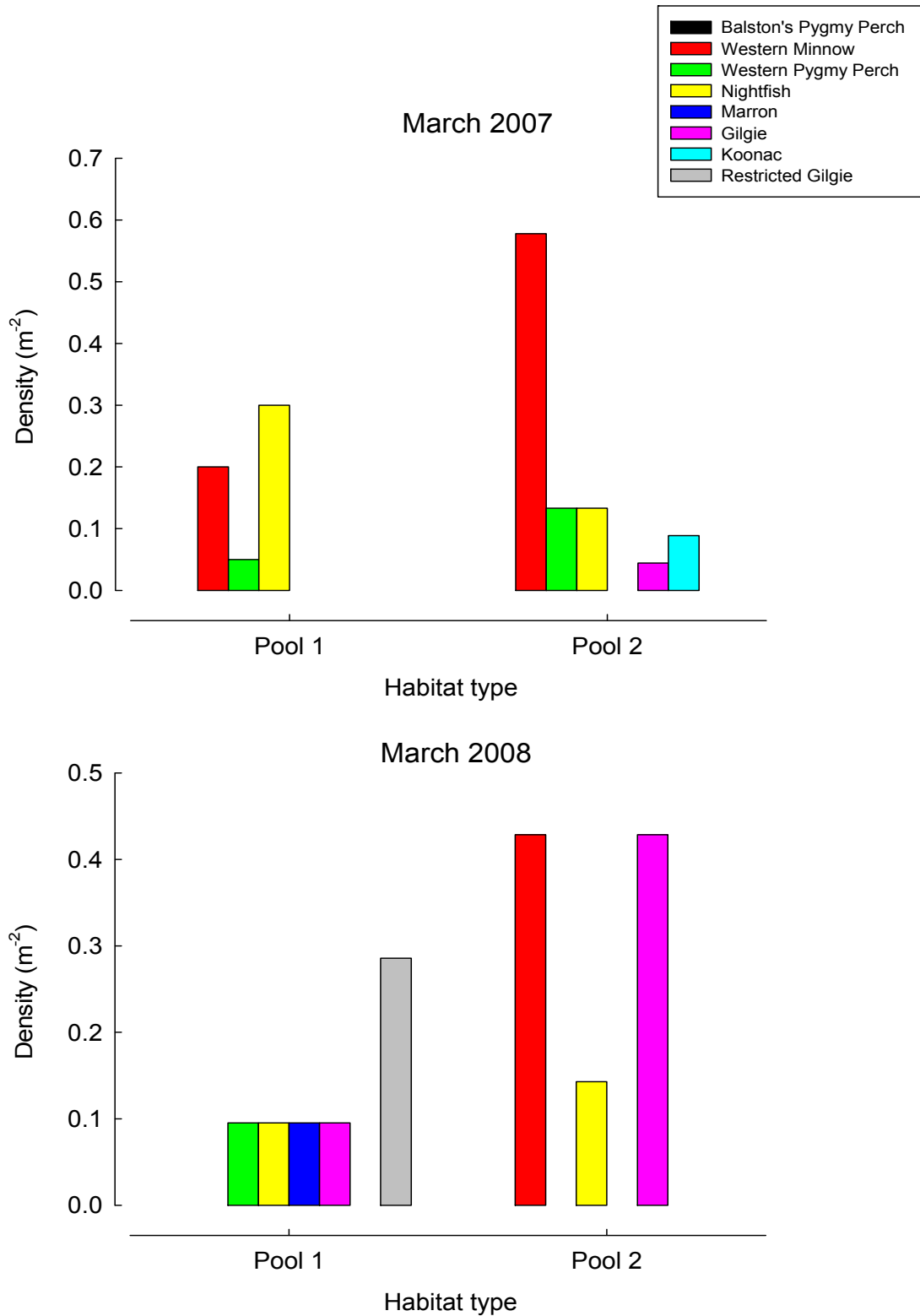
Maximum and mean depths of the cross (x-sect) and longitudinal (long) sections of dominant habitat types (either pool or riffle zones) at Mil 3 and 5. N.B. the greater depths in the pools in Mil 5 in the 2007 during greater discharge (measured at Mil 3) compared with 2008. Note also the lack of clear depth difference between those times in Mil 3 despite the increased discharge measured at that site.



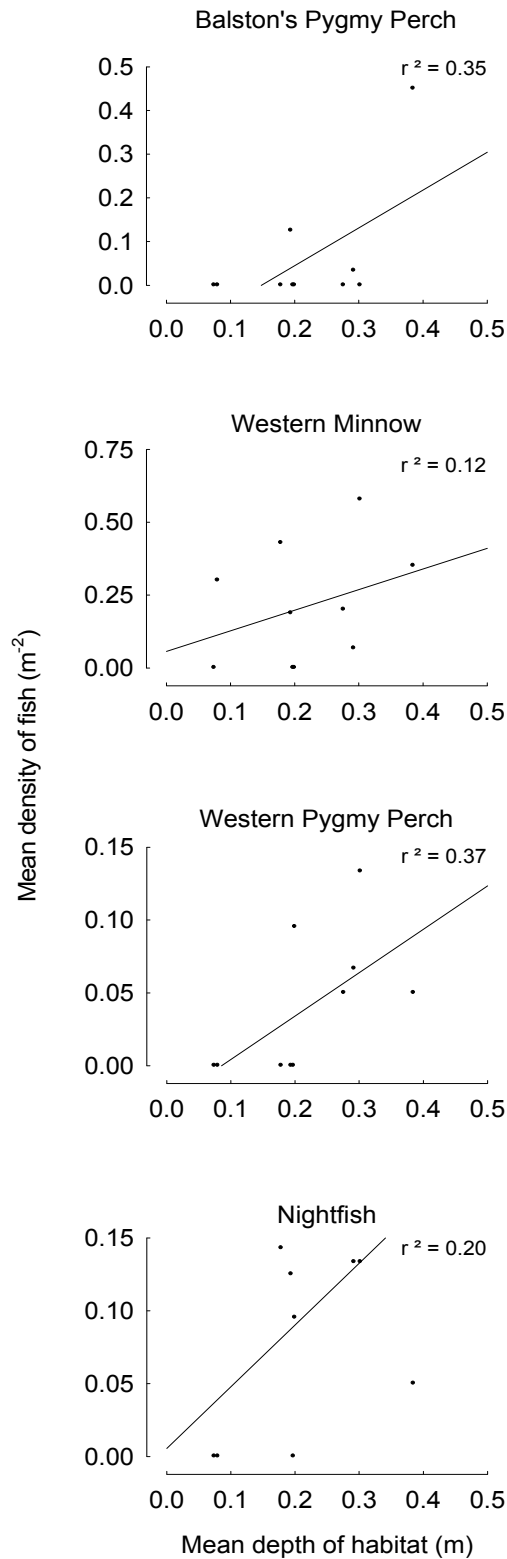
**Figure 53** Densities of fish and freshwater crayfish in the three habitat types at Mil 3 during baseflow in 2007 and 2008. N.B. the lower diversity of fish in the riffle habitat compared with the pool habitats.



**Figure 54** Densities of fish and freshwater crayfish in the three habitat types at Mil 4 during baseflow in 2007. N.B. the relatively high abundance of the Balston's Pygmy Perch in Pool 2.



**Figure 55** Densities of fish and freshwater crayfish in the three habitat types at Mil 5 during baseflow in 2007 and 2008. N.B. the lack of Riffle habitat in this section and it approximated the upstream point of permanent aquatic habitat with negligible flow being recorded in both years.



**Figure 56**

Preliminary relationships between the mean depths of habitats sampled within Mil 3, 4 and 5 and the mean density of fish within each of those habitats. N.B. the weak positive relationship between the depth and fish density of Balston's Pygmy Perch and Western Pygmy Perch.

**Table 7** Depths of pool and riffle habitats at sites sampled during baseflow period (March) in 2007 and 2008 in Milyeannup Brook. N.B. Included is the mean temperature, conductivity, pH and dissolved oxygen at each site. Note the reduced flow velocity and discharge during 2008 relative to 2007 and the greater depths at pools at Mil 5 in 2007 relative to 2008 whereas no clear differences existed at Mil 3 between those years. \*disconnected pool habitats at Mil 6 in March 2007 and 2008.

Site	Habitat type	Section taken	2007						2008								
			Max depth (m)	Av depth (m)	Av vel (m/sec) and disch (l/sec)	Temp (°C)	Cond (µS/cm)	pH	DO (ppm)	Max depth (m)	Av depth (m)	Av vel (m/sec) and disch (l/sec)	Temp (°C)	Cond (µS/cm)	pH	DO (ppm)	Turb (NTU)
Mil 3	Riffle 1	x sect 1	0.2	0.07					0.11	0.07							
		x sect 2	0.11	0.07					0.08	0.06							
		x sect 3	0.13	0.07					0.12	0.08							
		Longitudinal	0.14	0.09					0.15	0.11							
	Pool 2	x sect 1	0.18	0.10	V = 0.06 D = 3.6	16.5 (0)	483 (11.1)	5.69 (0.01)	5.30 (0.06)	0.28	0.16	V = 0.04 D = 2.0	15.1 (0.11)	481 (0.71)	5.26 (0.01)	5.36 (0.01)	1.52 (0.13)
		x sect 2	0.30	0.16					0.25	0.14							
		x sect 3	0.35	0.25					0.37	0.27							
		Longitudinal	0.38	0.30					0.36	0.29							
Mil 4	Pool 1	x sect 1	0.45	0.28		15.4 (0.04)	514 (7.79)	5.37 (0.04)	4.44 (0.02)								
		x sect 2	0.45	0.29													

		<b>x sect 3</b>	0.35	0.24														
		<b>Longitudinal</b>	0.48	0.41														
<b>Pool 2</b>		<b>x sect 1</b>	0.5	0.28														
		<b>x sect 2</b>	0.75	0.53														
		<b>x sect 3</b>	0.32	0.21														
		<b>Longitudinal</b>	0.89	0.57														
<b>Mil 5</b>	<b>Pool 1</b>	<b>x sect 1</b>	0.55	0.28						0.19	0.11							
		<b>x sect 2</b>	0.45	0.28						0.44	0.21							
		<b>x sect 3</b>	0.46	0.26						0.35	0.23							
		<b>Longitudinal</b>	0.42	0.29						0.43	0.22							
	<b>Pool 2</b>	<b>x sect 1</b>	0.31	0.17	<b>V = ~0</b>	17.6	556	5.38	2.68	0.25	0.15	<b>V = ~0</b>	15.1	591	4.98	4.45	1.51	
					<b>D = ~0</b>	(0.19)	(10.82)	(0.05)	(0.04)			<b>D = ~0</b>	(0.14)	(4.30)	(0.03)	(0.11)	(0.24)	
		<b>x sect 2</b>	0.61	0.38						0.3	0.14							
		<b>x sect 3</b>	0.85	0.29						0.29	0.168							
		<b>Longitudinal</b>	0.47	0.36						0.35	0.23							
<b>Mil 6</b>	<b>Pool 1*</b>	<b>Length (m)</b>	6							4								
		<b>Width (m)</b>	1							1.2								
		<b>Max depth (m)</b>	0.08							0.06								



<b>Pool 2*</b>	<b>Length (m)</b>	4	1
	<b>Width (m)</b>	0.8	1.2
	<b>Max depth (m)</b>	0.1	0.05
<b>Pool 3*</b>	<b>Length (m)</b>	5	4
	<b>Width (m)</b>	1	1
	<b>Max depth (m)</b>	0.1	0.08

<b>Mil 7A</b>	<b>DRY</b>	<b>DRY</b>
<b>Mil 7B</b>	<b>DRY</b>	<b>DRY</b>
<b>Mil 7</b>	<b>DRY</b>	<b>DRY</b>
<b>Mil 8</b>	<b>DRY</b>	<b>DRY</b>
<b>Mil 9</b>	<b>DRY</b>	<b>DRY</b>
<b>Mil 10</b>	<b>DRY</b>	<b>DRY</b>
<b>Mil 11</b>	<b>DRY</b>	<b>DRY</b>



Autumn 2008									
<b>Mil 5</b>	<b>34.1040</b>	<b>115.5708</b>							
	<b>3</b>	<b>5</b>							
Summer 2006			30	0.83	0.03	0.03	0.03		0.30
				(25)	(1)	(1)	(1)	0.03 (1)	(9)
Autumn 2007			44.5	0.39	0.09	0.22			0.02
				(0.27)	(0.06)	(0.12)			0.04 (0.06)
Autumn 2008			37.5						
<b>Mil 6</b>	<b>34.1068</b>	<b>115.5699</b>							
	<b>2</b>	<b>6</b>							
Summer 2006			20			0.600	0.300		0.100
						(12)	(6)		(2)
Autumn 2007	<b>Disconnected pools</b>		14						
Autumn 2008	<b>Disconnected pools</b>		10						

**Table 9** Mean densities of fish and freshwater crayfish during baseflow sampling in sites in Milyeannup Brook in 2006, 2007, 2008.

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