Effects of flooding on recruitment and dispersal of the Southern Pygmy Perch (*Nannoperca australis*) at a Murray River floodplain wetland

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Zeb Tonkin and Dr Alison King are freshwater ecologists, and Jobn Maboney is a senior technical officer with the Arthur Rylah Institute for Environmental Research, (Department of Sustainability and Environment, 123 Brown St., Heidelberg, Vic. 3089, Australia; Tel.: +61 3 9450 8600; Fax: +61 3 9450 8799; Email: zeb.tonkin@dse.vic.gov.au, alison.king@dse. vic.gov.au, john.maboney@dse. vic.gov.au). This work is part of a project funded by the Murray-Darling Basin Commission which looks at assessing fish breeding and recruitment in response to environmental flow management. **Summary** With limited evidence linking Australia's Murray-Darling Basin fish species and flooding, this study assessed annual variation in abundance and recruitment levels of a small-bodied, threatened floodplain species, the Southern Pygmy Perch (*Nannoperca australis*), in floodplain habitats (creeks, lakes and wetlands) in the Barmah-Millewa Forest, Murray River, Australia. Spring and summer sampling over a 5-year period encompassed large hydrological variation, including 1 year of extended floodplain inundation which was largely driven by an environmental water release, and 2 years of severe regional drought. Recruitment and dispersal of Southern Pygmy Perch significantly increased during the floodplain inundation event compared with the other examined years. This study provides valuable support for an environmental water allocation benefiting a native species, and explores the link between flooding and its advantages to native fish. This suggests that the reduced flooding frequency and magnitude as a result of river regulation may well be a major contributing factor in the species' decline in the Murray-Darling Basin.

Key words: *environmental water allocation, floodplain inundation, freshwater fish, Murray-Darling Basin.*

Introduction

owland river fish communities are thought to rely on regular floodplain inundations which provide a range of benefits such as spawning cues and an abundance of food and habitat (Junk et al. 1989; Bayley 1991). The flood pulse concept is often extrapolated to Australia's Murray-Darling Basin (MDB) fish community (e.g. the 'flood recruitment model', Harris & Gehrke 1994) despite limited evidence of any benefits floodplain inundation has had on any MDB species (Humphries et al. 1999; King et al. 2003; Mallen-Cooper & Stuart 2003; Graham & Harris 2005). With most rivers in the MDB now regulated, there has been a significant loss of low to medium level floods, which is believed to be a major contributing factor to the decline in the abundance and distribution of the Basin's native fish fauna (MDBC 2004). As a consequence, much attention has been given to the recovery of a more natural flow regime as a restoration strategy, particularly through environmental water allocations (EWA; Poff et al. 1997; Arthington et al. 2006). Indeed, the

provision of environmental water to currently over-allocated or overused systems to environmentally sustainable levels and improve environmental management practices is a key objective of the National Water Initiative (NWI 2005). Although these EWAs are being increasingly targeted at improving native fish populations, there are unfortunately only a few examples of a positive response by fish to an environmental flow (Poff *et al.* 2003; Arthington *et al.* 2006; King *et al.* in press).

The Barmah-Millewa Forest is a 70 000ha highly complex floodplain wetland system located on the Murray River in south-eastern Australia's MDB. The forest is one of the six recognized icon sites under the Murray-Darling Basin Commission's 'Living Murray Initiative' (COAG 2004), and is internationally recognized as an important wetland under the Ramsar Convention (Ward 2005). The forest's hydrology has been severely altered as a result of the regulation of the Murray River, and poses a major threat to the ecological values of the forest (Ward 2005). In an attempt to return a more natural hydrological regime to the forest, the Barmah-Millewa Forest receives an annual EWA which has been used three times since its inception in 1993 (King et al. 2007). It was not until the latest of these events, the 2005 EWA, that an intense fish monitoring programme was undertaken to assess the response of the forest's fish community to such an event. King et al. (in press) documented increases in spawning and/or recruitment of larger, native riverine fish species Golden Perch (Macquaria ambigua), Silver Perch (Bidyanus bidyanus), Murray Cod (Maccullochella peelii peelii) and Trout Cod (Maccullochella macquariensis) during this EWA-driven flood year. As part of this programme, this paper assesses annual variation in abundance and recruitment of a small bodied, cryptic floodplain species, the Southern Pygmy Perch (Nannoperca australis).

The Southern Pygmy Perch is the most widespread of the six nannopercid species, occurring from coastal south-west Western Australia to southern Queensland (Humphries 1995). Although relatively common in coastal Victoria, the Southern Pygmy Perch is patchily distributed in northern Victoria and is now listed as Endangered in

New South Wales (established under part 7a of the NSW Fisheries Management Act 1994) and South Australia (SA Fisheries Act 1982; Action Plan for South Australian Freshwater Fishes 2007). The species reaches a maximum size of 85 mm (although rarely >65 mm) and prefers slow flowing or still waters containing dense habitat such as aquatic vegetation or complex woody debris and is therefore commonly associated with floodplain environments such as wetlands and billabongs (see also Kuiter et al. 1996; Lintermans 2007). The species is able to tolerate a broad range of temperatures and extremely low dissolved oxygen levels (McNeil & Closs 2007), which is typical of such environments. Submerged and aquatic vegetation are also the preferred spawning site of the species where females randomly disperse their small demersal eggs (Llewellyn 1974). Unsurprisingly, habitat alteration such as loss of aquatic vegetation and seasonal flow changes/reductions are listed as possible reasons for the species' decline (Lintermans 2007). The aim of this paper was to assess annual variation in abundance and recruitment levels of this species in floodplain habitats of the Barmah-Millewa Forest over a 5-year period encompassing large variation in hydrological conditions including an environmental watering event.

Methods

As part of a larger fish monitoring programme (see King *et al.* 2007), sampling was conducted at 11 floodplain sites which encompassed four forest creeks, two large lakes and five smaller wetlands. Sampling was performed on a monthly basis from mid-September to the end of February over a 5-year period (2003/2004, 2004/2005, 2005/2006, 2006/2007 and 2007/2008).

The 5-year study period encompassed a range of hydrological conditions resulting in contrasting differences in floodplain inundation between years. This included brief minor floodplain inundation during early spring for the first two years, major extended spring/summer floodplain inundation encompassing the EWA event during the middle year (2005/2006) and no flooding in the final two years. The latter conditions resulted in the complete drying of many of the floodplain habitat sites towards the end of the study.

Sampling was conducted using modified quatrefoil light traps (Floyd et al. 1984; Secor et al. 1992), which are a useful passive technique for collecting fish from a range of meso-habitat types and depths. Light traps were set on dusk with a yellow 12-h light stick, and were retrieved as early as possible the next day. Active sampling was also conducted in all habitat types using the Sweep Net Electrofishing (SNE) method (King & Crook 2002). The SNE method is a modified standard backpack electrofishing unit (Smith-Root Model 12, NE Salmon Creek Ave., Vancouver, WA, USA), with a 15-cm diameter anode ring, and fitted with a moulded plastic rectangular frame $(25 \times 30 \times 2 \text{ cm})$ with an attached 250 µm mesh sampling net. A replicate SNE sample involved approaching the selected habitat, activating the anode and moving at a constant speed in a forward zigzag motion for 20 s of electrofishing time (see King et al. 2007). Five replicate samples of both light trap and SNE were taken at each site (only three replicates were taken in smaller wetland habitats), throughout the duration of the study. The 2006/2007 and 2007/2008 seasons consisted of extremely low water levels, resulting in a number of the wetland sites drying.

After collection, fish were preserved in 95% ethanol and later measured for standard length (L_s - nearest 0.1 mm) and weight (nearest 0.001 g). These measurements were performed after a minimum of 10 days to allow shrinkage associated with preservation to have stabilized (Fey & Hare 2005). As no larvae were collected for the species over the entire 5-year period, limited conclusions could be drawn on spawning of the species. However, by using a combination of length frequency distributions and otolith microstructure analysis of the smaller size classes of fish $<30 \text{ mm } L_s$ (assumed to be juveniles according to Lintermans 2007), estimates of daily age (and therefore spawning time) and mean growth rates was available.

Unlike difficulties of annuli interpretation reported for the species (Humphries 1995), the microstructure of sagittal otoliths of early juvenile Southern Pygmy Perch possessed clear banding. Although daily increment deposition has not been validated for the species, daily deposition has been confirmed for another member of the genus, Nannoperca oxleyana (J. Knight, unpubl. data 2008). Furthermore, back calculation of increment counts did correspond to the length frequency distributions and approximate birthdates of the species presented by other authors (e.g. Humphries 1995), suggesting that increments are deposited on a daily basis for the species. Sagittal otoliths were removed with the aid of a stereo microscope and mounted in thermoplastic cement (proximal face down). Otoliths were then polished to the level of the primordium across the sagittal plain using 3 μ m lapping film and 0.5 μ m alumina slurry. Using a compound microscope $(400-1000 \times magnification)$ fitted with a digital camera and the image analysis system imagepro express (version 5.0.1.26, Media Cybernetics Inc., Bethesda, MD, USA) the number of increments from the first major increment outside the primordium to the edge of the otolith was counted three times, after which the mean ring count was determined (see Sweatman & Kohler 1991). The mean ring count for each fish was then used as the estimate for daily age; however, given the timing of deposition for the first increment is unknown, ages have been presented as 'estimated age' throughout. No attempt was made to assess the age structure of older fish given the difficulties of annuli interpretation reported by Humphries (1995).

Data analysis

To assess annual variation in Southern Pygmy Perch abundance, the total number of fish collected each month from all sites using both gear types was divided by the total number of replicates to give a standard catch per-unit effort (CPUE). This was used to correct for minor differences in sampling effort over the 5-year period due to factors such as gear malfunction or loss of sites due to drying. Monthly CPUE data were log-transformed to achieve normal distribution and tested for annual differences in abundance using a one-way analysis of variance (ANOVA). To assess annual variation in distribution, monthly presence/absence data for each site were log-transformed to



Figure 1. Annual variation in (a) the number of days of floodplain inundation; (b) numbers of Southern Pygmy Perch adults (black) and juveniles (grey) with the associated number of sites (triangles) in which the species was present and; (c) mean monthly catch-per-unit-effort (CPUE) of Southern Pygmy Perch during each year of the study.

achieve normal distribution and tested for annual differences using a one-way ANOVA. Although small sample sizes and restricted size ranges did not permit the development of a growth model for the population (see Campana & Jones 1992), length at estimated age comparisons was used to estimate mean individual growth rates for early juvenile fish using the following formula:

 $G_c = (L_s - 3.45)/A_c$

where G_c is the mean daily growth rate at capture (millimetres/day), L_s is the standard length at capture, 3.45 mm is the length at hatch as determined by Llewellyn (1974) and A_c is the estimated age at capture (days). Mean juvenile growth rates were log transformed and tested for annual differences using a one-way ANOVA.

Results

Over the 5-year period, 96 individual Southern Pygmy Perch were collected from creeks and wetlands in the BMF. Over half of these fish (n = 50) were collected in 2005/2006, when the maximum number of days of floodplain inundation occurred (Fig. 1a,b). There was a significant difference in the monthly CPUE of fish between sampling years (one-way ANOVA, F = 3, 3.97, P < 0.05; Fig. 1c), with a greater number of Southern Pygmy Perch collected in 2005/ 2006 and 2006/2007 than 2003/2004, 2004/2005 and 2007/2008, respectively. Southern Pygmy Perch were also recorded at significantly more sites in 2005/2006 and 2006/2007 (five sites), than the other 3 years (one site in 2003/2004 and 2004/ 2005) of the study (F = 3, 4.33, P < 0.05; Fig. 1b). No fish were collected in 2007/ 2008, primarily due to the majority of wetland sites being completely dry for the entire sample period after a prolonged period of no floodplain inundation (Fig. 1a).

In three of the 5 years, there were two distinct size classes of Southern Pygmy Perch captured, adults and early juveniles (Fig. 2). Using a combination of daily age estimation and length frequency distributions, we were able to determine that the smaller size class of fish (<30 mm) captured from November until February were young-of-year. More of these juvenile fish were collected in 2005/2006 (n = 21) compared to the other years (Fig. 1b). Daily age estimation indicated that regardless of year, all fish were spawned in October. Length frequency distributions also suggest this spawning time (Fig. 2). Early juvenile fish collected in 2005/2006 were larger than similar-aged fish collected from 2004/2005 (Fig. 3). A comparison of mean growth rates indicated early juvenile fish collected in 2005/2006 had significantly higher mean growth rates $(0.352 \pm$ 0.010 mm/day, n = 21) than those collected 2004/2005 (0.209 ± 0.029 mm/day, in n = 9) (F = 2, 51.79, P < 0.001). The single early juvenile fish captured in 2006/2007 had a mean growth rate of 0.207 mm/day.

Discussion

With limited evidence linking MDB fish species and floodplain inundation, the broad applicability of the flood-pulse concept and subsequent models (e.g. Harris & Gehrke 1994) has recently come under question for many species (Humphries et al. 1999; King et al. 2003; Graham & Harris 2005). The results of this study show the highest numbers of Southern Pygmy Perch were collected in 2005/2006 (the year of prolonged spring/summer floodplain inundation), with a high proportion of fish being juveniles. Many fish were also collected in the following year (2006/2007), however, catches were almost exclusively made up of adult fish, most likely as a result of the increased recruitment from the 2005/2006 flood season. Additionally, the only other year where a high number of juvenile fish were captured was 2004/2005, which



Figure 2. Southern Pygmy Perch length frequency histograms for each month sampled in 2004/2005 (n = 17), 2005/06 (n = 50) and 2006/2007 (n = 28). Note: 2003/2004, 2007/2008 not included, as very few fish were collected.

encompassed the next longest period of floodplain inundation for the study. Taken together, these results show that recruitment of Southern Pygmy Perch was greatest during years where prolonged spring/summer floodplain inundation periods occur, as predicted by the floodpulse concept and flood-recruitment models.

It must be noted that strong conclusions are limited given the restricted number of years for comparison and the lack of independent control sites which make it difficult to ascribe causation from correlative data. We can however, be confident that fish were recruited within the floodplain habitats of the BMF and not sourced from upstream given King *et al.* (2007) reported no captures of the species in the main river channel after 3 years of sampling.

Harris and Gehrke (1994) proposed that floods could enhance recruitment of Australian native freshwater fish either by spawning in direct response to floods or by indirectly increasing the survival of young by providing suitable food and habitat resources on the inundated floodplain. Although we are unable to comment on whether there was an increase in spawning activity of Southern Pygmy Perch, as no eggs or larvae were captured, the increased number of recruits during years of greater floodplain inundation suggests the second mechanism described by Harris and Gehrke (1994) and Junk *et al.* (1989), may have occurred. Furthermore, estimated mean growth rates of juvenile fish (derived from length at age data) suggest the extended floodplain inundation during 2005/2006 provided better conditions to facilitate early growth in this species. Although we have not been able to comment on the amount of spawning of Southern Pygmy Perch, it is possible that the higher water levels which inundated



Figure 3. Juvenile Southern Pygmy Perch estimated age (days) at length (L_s) data for 2004/2005 (crosses), 2005/2006 (black diamonds) and 2006/2007 (square). n = 21, 9 and 1, respectively.

vast areas of vegetation during the 2005/ 2006 years, may have benefited the spawning mechanism of the species. Llewellyn (1974) reported that while flooding was not necessary as a spawning stimulus, heavily weeded slow flowing areas are the preferred spawning areas of the species. Another member of the genus, *Nannoperca oxleyana*, has also been reported to preferentially spawn on submerged vegetation in aquaria (Leggett 1990; Knight *et al.* 2007).

Results of this study indicated that regardless of year, all of the juvenile fish collected in the BMF were spawned around October. Humphries (1995) also reported October spawning for a Tasmanian population of Southern Pygmy Perch, documenting female gonadosomatic indices peaking in October and, like the present study, most juveniles occurred in December. Llewellyn (1974) also reported the species breeding during September and October during a pond study in southern New South Wales. This suggests that spawning times are similar between populations; despite hydrological, latitudinal and habitat differences. Interestingly, this consistently restricted spawning period for Southern Pygmy Perch contrasts markedly with the known protracted spawning period of the northern coastal species N. oxleyana, which has recently been documented spawning from September until April in Northern New South Wales and Queensland (Knight et al. 2007).

In addition to the increased recruitment levels recorded during the 2005/2006 years, Southern Pygmy Perch were recorded from the greatest number of sites during the high flow year and the following year, which suggests that this flood event also facilitated the dispersal of Southern Pygmy Perch throughout the Forest. This result highlights the importance of flooding as a mechanism of dispersal and recolonization for this floodplain species, which is a common strategy employed by species inhabiting these floodplain environments (e.g. Winemiller et al. 2000; Granado-Lorencio et al. 2005). Of course, the increased number of sites fish were recorded from may also have been a result of increased recruitment by sparse local populations not detected by earlier sampling, rather than dispersal from other sites. Although flooding may provide new potential habitats, high productivity and an input of new recruits, extreme dry periods can result in catastrophic mortality (Winemiller et al. 2000). The last two years of this study were some of the area's driest periods in history, and resulted in two consecutive years of no floodplain inundation and a large proportion of aquatic floodplain habitats completely dried out. This ultimately led to the absence of the species from samples in the final year of sampling at all of the previously sampled sites. Additional surveys of the area have recorded low numbers of large adults (no juveniles) in some of the few remnant pools in the forest (Tonkin & Rourke 2008). Indeed, emergency management measures including targeted watering and relocation from drying pools to more permanent refuge areas have already occurred during the 2007/2008 season. This is by no means a long-term solution since Humphries (1995) reported the age structure of a Tasmanian population of Southern Pygmy Perch to be dominated by 0+ fish, with very few individuals living more than 1 year. Given that after 3 years of sampling, King et al. (2007) reported no captures of the species in the main river channel, the local BMF population of Southern Pygmy Perch is under serious threat of extinction, unless appropriately timed flooding can trigger successful recruitment and dispersal in the coming seasons.

Such a flood-reliant life-history strategy appears to be quite risky for a short-lived, MDB species, particularly given the huge hydrological variations that characterize Australian floodplain rivers. Additionally, river regulation in the MDB has resulted in a reduction in the frequency and magnitude of flooding events (MDBC 2004), and hence, is likely to have contributed to the species' decline and current conservation status. Although the Barmah-Millewa Forest currently receives a substantially reduced frequency and magnitude of flooding events than pre-river regulation, the forest still receives floods that cover 70% of its floodplain in 37% of years, and small localized flooding events occur more often (although they are mostly outside the natural seasonal inundation period for the period) (MDBC 2006). This fairly regular flooding frequency may help to explain why the Barmah-Millewa Forest has maintained a population of the species, and further highlights the fragility and importance of this complex wetland system as a key area for the conservation of Southern Pygmy Perch in the MDB.

The extended floodplain inundation during 2005/2006 included the largest EWA used to date in Australia (513 GL). Although the EWA resulted in other positive ecological outcomes, this research demonstrates that flooding, and the use of the EWA, increased recruitment and dispersal of a species of conservation significance, the Southern Pygmy Perch. This response from a small-bodied floodplain species adds further evidence to the response of fish to the Barmah-Millewa EWA, as presented by King et al. (in press), which documented increases in spawning and/or recruitment of four larger, native riverine species during this environmental watering event. With recent questioning of the relationship between flooding and Australian freshwater fish (e.g. Humphries et al. 1999; King et al. 2003; Mallen-Cooper & Stuart 2003; Graham & Harris 2005), the use of EWA's as a means to restore and enhance its freshwater fisheries has received much attention, particularly when the MDB is experiencing one of its worst recorded droughts. Therefore, although strong conclusions are limited, this work has suggested a link between flooding and a Murray-Darling native floodplain species, and has provided invaluable evidence of a native fish species benefiting from the use of environmental water. This research has provided much needed support for the use of managed flows for the environment as a method of fish rehabilitation not only in Australia's MDB, but in regulated floodplain rivers around the world.

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