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# RESPONSE BY FISH ASSEMBLAGES TO AN ENVIRONMENTAL FLOW RELEASE IN A TEMPERATE COASTAL AUSTRALIAN RIVER: A PAIRED CATCHMENT ANALYSIS

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## ABSTRACT

Defining appropriate environmental flow regimes and criteria for the use of environmental water allocations requires experimental data on the ecological impacts of flow regime change and responses to environmental water allocation. Fish assemblages in one regulated and one unregulated tributary paired in each of two sub-catchments of the Hunter River, coastal New South Wales, Australia, were sampled monthly between August 2006 and June 2007. It was predicted that altered flow regime due to flow regulation would reduce species richness and abundance of native fish, and assemblage composition would differ between paired regulated and unregulated tributaries. Despite significant changes in richness, abundance and assemblage composition through time, differences between regulated and unregulated tributaries were not consistent. In February 2007, an environmental flow release ('artificial flood') of 1400 ML was experimentally released down the regulated tributary of one of the two catchments over 6 days. The flow release resulted in no significant changes in fish species abundances or assemblage composition when compared to nearby unregulated and regulated tributaries. Flow regulation in this region has reduced flow variability and eliminated natural low-flow periods, although large floods occurred at similar frequencies between regulated–unregulated tributaries prior to and during 2006–2007, resulting in only moderate changes to regulated flow regimes. Barriers to dispersal within catchments also compound the effects of flow regulation, and findings from this study indicate that the location of migratory barriers potentially confounded detection of the effects of flow regime change. Further experimental comparisons of fish assemblages in regulated rivers will refine river-specific response thresholds to flow regime change and facilitate the sustainable use of water in coastal rivers. 

KEY WORDS: Australian smelt; Cox's gudgeon; environmental flow; fish assemblages; *Gobiomorphus coxii*; hydrology; *Retropinna semoni*;

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## INTRODUCTION

The timing and magnitude of river flow events, such as floods and droughts, affect the composition and abundance of fish assemblages (Jowett *et al.,* 2005). Variability within and between fish assemblages has been associated with gradients of flow variability in many rivers throughout the world (Gehrke *et al.,* 1995; Gehrke *et al.,* 1999; Cattanéo, 2005; Taylor *et al.,* 2006). Maintenance of natural flow characteristics is critical to conserve the natural variability of the aquatic ecosystems to maintain important ecological processes (Bunn and Arthington, 2002).

Water extraction and dams alter natural flow regimes (Singer, 2007). Regulated flow regimes are often characterized by reduced variability of flow therefore influencing aquatic habitat diversity (Mérigoux and Ponton, 1999), and in turn, affecting aquatic biota and the ecosystem processes that support fish assemblages. Although the influences of flow regulation on fish assemblages are sometimes detected immediately after the flow regime has been manipulated (e.g. Xie *et al.,* 2007), often the effects are not evident for years or even decades after flow regime change (e.g. Humphries *et al.,* 2002). Fish assemblages in rivers with a long history of flow regulation and catchment degradation have been found to have reduced species richness (Gehrke *et al.,* 1995), altered population abundances (Gehrke *et al.,* 1999) and assemblage composition and variability in both space and time (Humphries *et al.,* 2002).

Restoring natural variability to the flow regime of regulated rivers is often proposed as a method of rehabilitating their aquatic ecosystems (Marchetti and Moyle, 2001; Arthington and Pusey, 2003). The restoration of the natural flow regime aims to recover important characteristics of the natural flow regime required to maintain ecological processes of water-dependant environments, such as flood frequency (Arthington and Pusey, 2003; Welcomme, 2006). For example, the delivery of 'artificial floods' to maintain hydrological variability is frequently considered a key aspect in environmental flow plans (e.g. Valdez *et al.,* 2001; Ortlepp and Mürle, 2003). Maintaining natural hydrological variability in regulated rivers is also recommended as a tool

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to control alien fish species and to restore native fish populations (Valdez *et al.,* 2001). For example, in the regulated San Juan River (USA), densities of several native fish species increased in years of elevated spring flow releases from the Navajo Reservoir that mimicked natural high inflows over the same period (Propst and Gido, 2004). However, the responses of native and alien fish to environmental flow programs are likely to vary due to the various ecological needs of different species. The success of past and current environmental flow programs is likely limited by an inadequate understanding of these relationships between flow regime and biological interactions, which is essential if environmental flow programs are to be successful (Jowett *et al.,* 2005).

The Hunter River catchment, coastal New South Wales, is experiencing steep increases in population growth and water usage, and already has a number of regulated rivers with dams of different ages and designs. The fish fauna of the Hunter River catchment supports a variety of native and alien species with diverse life histories (Gehrke and Harris, 2000; Brooks *et al.,* 2004), but there is a need to understand how these assemblages respond to changes in flow regime. If there is an impact of regulation, can this be alleviated by environmental releases down regulated tributaries? In this study, the reach-scale associations between flow regime and fish assemblages at three sites along paired regulated and unregulated tributaries in two catchments of the Hunter River catchment were compared during 2006–2007. The responses by downstream fish assemblages to a single 6-day 'artificial flood' (hereafter referred to as an environmental flow release, EFR) in February 2007 in a regulated tributary were experimentally assessed to determine the potential benefits of an annual Environmental Contingency Allowance set by the local Water Sharing Plan (NSW Department of Water and Energy, 2007). We hypothesized that fish assemblages and populations in regulated tributaries would differ in composition and abundance compared to unregulated tributaries, with species richness and abundance of native fish being higher in unregulated tributaries than in their corresponding regulated partner. We also predicted that the EFR in a regulated tributary would restore fish assemblages to a more natural composition, judged as equivalent to that of the assemblage in the unregulated adjacent tributary.

## METHODS

### *Study region and sampling design*

The Hunter River catchment, coastal New South Wales, Australia (Figure 1), drains 22 000 $km^2$, and has a mean annual discharge of 1680 GL (Chessman *et al.,* 1997). Upland tributaries of the Hunter River catchment are characterized by cobble-pebble substrate of Devonian-Carboniferous sediment and basalt origin. Riparian vegetation is predominantly dry sclerophyll forest comprising native species including *Eucalyptus* spp., river oaks (*Casuarina cunning-*

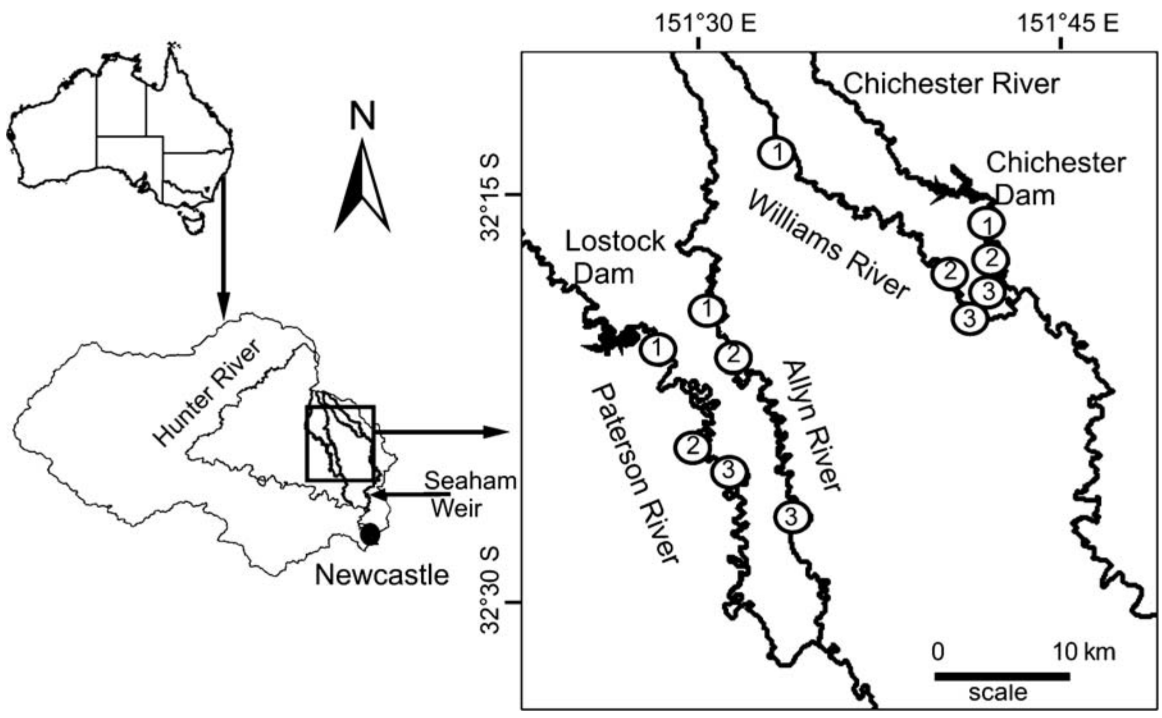


Figure 1. Map indicating study sites (numbered 1–3 in each river) in the Paterson and Williams catchments, each with a regulated and unregulated tributary



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*hamii*) and exotic willows (*Salix* spp.), and giant reed (*Arundo donax*). Catchment land use is predominantly grazing agriculture, timber harvesting or preserved as national park.

Median annual rainfall is between 1061 and 1278 mm within the eastern Hunter River catchment (Bureau of Meterology 2009), however, high inter-annual variability in rainfall occurs within the catchment (Chessman and Growns, 1994). Around 40% of rainfall occurs between January and March (austral summer) (Bureau of Meteorology 2009) and the region has a warm temperate climate (Chessman *et al.,* 1997). Annual rainfall between 2001 and 2006 was ~75% of median long-term records, and resulted in reduced frequency and magnitudes and floods.

Eight major water storages exist within the Hunter River catchment to mitigate floods and provide water for agriculture, industry and domestic use. The flow regime in reaches downstream of these water storages has reduced variability in the magnitude and frequency of high flows within the river channel (Erskine, 1985) and maintains steady low flows in dry periods when flows would normally cease (Chessman *et al.,* 1997). Despite these impacts, the total annual discharge volume continues to be largely unaffected when compared with pre-regulation conditions.

To compare fish assemblages in tributaries with and without flow regulation, a pair of similar-sized tributaries was chosen in each of the Paterson and Williams subcatchments of the Hunter River (Figure 1). One tributary was dammed whereas its neighbouring reference tributary retained a natural flow regime. Three sites nested in each tributary paired within two subcatchments enabled a spatially hierarchical sampling design to compare spatial and temporal patterns in species population abundance and assemblage composition between a regulated tributary and its unregulated 'reference' tributary (a multiple Before-After-Control-Impact-Paired, BACIP, design for the EFR, sensu Downes *et al.,* 2002).

The flow regime of the Paterson River is regulated by Lostock Dam, a 38-m high dam built in 1971 with a reservoir capacity of 20 GL. From 17th February 2007, an EFR was conducted (total volume 1400 ML with 100, 600, 300, 200, 100 and 100 ML delivered on days 1–6, respectively) from Lostock Dam, simulating a small spate (<1500 ML day$^{-1}$) that occur relatively infrequently in the regulated tributaries when compared to unregulated tributaries in January–April each year (Figure 2). The Allyn River (Figure 1) was used as the reference unregulated tributary of the Paterson catchment. In the Williams catchment, the Chichester River downstream of Chichester Dam experiences a regulated flow regime before its confluence with the Williams River, used here as the reference tributary (Figure 1). Chichester Dam is a 43-m high dam built in 1921 with a capacity of 21 GL to supply domestic water to the central coast region of NSW. No EFR occurred in the Chichester River during the study period. Three sites were sampled in each of the four rivers,

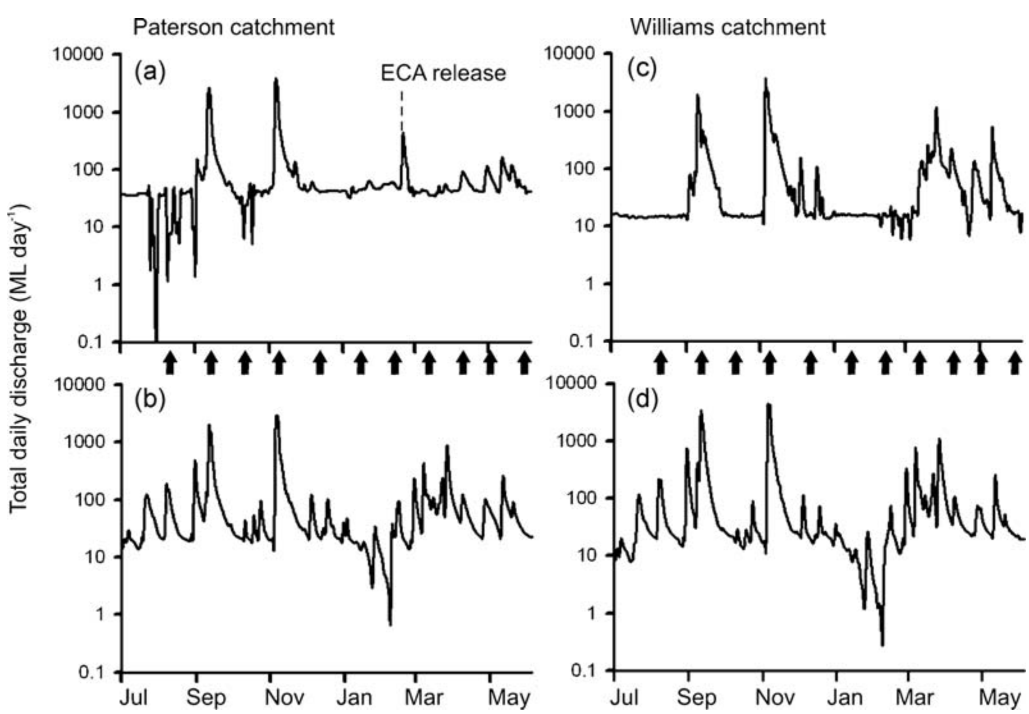


Figure 2. Hydrographs of total daily flow from July 2006 to June 2007 recorded in the regulated Paterson River (a), unregulated Allyn River (b), regulated Chichester River (c) and unregulated Williams River (d). The environmental flow release (EFR) and sampling dates are indicated by the dashed line and arrows, respectively



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yielding a total of 12 sites that each included run-riffle-pool habitats over approximately 100 m length with an average width of 12.5–14.8 m.

Both Lostock and Chichester Dams were consistently >80% full, due to generally consistent rainfall. These two dams have rarely dropped below 80% capacity since their construction, and during flooding water flows uncontrolled over the dam wall. Controlled releases of water from the dams are delivered via outlet valves at the bottom of the dam wall. Due to the depths of the two dams, combined with their consistently high levels, temperature and oxygen stratification probably occurs so that during periods of controlled releases (when dams are not spilling), unnaturally cold hypoxic water is discharged.

### *Sampling methods*

Fish were sampled monthly at each site between August 2006 and June 2007 to test if differences in fish assemblages between regulated and unregulated tributaries, and effects of the EFR, were consistent through time or associated with seasonal events such as spawning. Sweep net electrofishing (SNE), an effective method of sampling small-bodied fish (King and Crook, 2002), was used to semi-quantitatively sample (based on catch-per-unit-effort, CPUE) all habitats at each time. SNE was operated at 400–500 volts DC pulsed at 30 Hz and 12% duty cycle using a Smith-Root LR-24 backpack electrofishing machine (Vancouver, Washington, USA) with a 15-cm anode ring used to increase the voltage gradient necessary to effectively collect small fish. Attached to the anode pole was a 25 cm × 30 cm frame fitted with a tapered mesh net (250 μm).

Four samples were collected to sample the fish assemblage at each site. Each replicate was a composite sample of 18 sequential 10 s periods of electrofishing, totalling 180 s shock time per sample. Each composite sample was taken from either a riffle, run or pool habitat while the operator moved upstream in a single pass. Samples were preserved in 70% ethanol and returned to the laboratory. All fish were identified to species and standard length was measured to 0.1 mm. Data from a pilot study in May 2006 confirmed that four composited within-site replicates were adequate to detect the presence of all species at each site, and differences of 50% species richness and 80% abundance of all fish and individual species populations with 80% power between each pair of regulated and unregulated tributaries.

### *Data analysis*

*Hydrological analysis.* Seventeen flow metrics were calculated from daily flow data from each of the four study rivers for July 2002–June 2007 using the Time Series Module of the River Analysis Package (RAP) (Marsh, 2004). These metrics included mean and median daily flows, 10th and 90th percentile flows, flow variability (coefficient of variation, index of flow variability and standard deviation), rate of rise, duration and number of rises, base flow index and flood flow index. Principal components analysis (PCA) was used to portray variation in flow regimes between 2002 and 2007 of the two regulated and two unregulated rivers in terms of these 17 characteristics. Data were normalized prior to PCA, and analysis was done using PRIMER v6.1.11.

*Fish assemblage analysis.* Prior to analysis, residuals were examined for heterogeneous variances and normality was tested using Shapiro-Wilk tests in Statistix v.8 (Analytical Software, USA). Fourth-root transformation was applied when necessary to stabilize variances. All data were analysed using a series of mixed-model analyses of variance (ANOVA) using Systat v.12 (Systat Corporation, Evanston, Illinois, USA). Two ANOVA models were used to test for the differences in species richness, total fish abundance and abundances of dominant native and alien species (Australian smelt (*Retropinna semoni*), Cox's gudgeon (*Gobiomorphus coxii*) and eastern gambusia (*Gambusia holbrooki*)) between catchments, regulated-unregulated rivers, sites and time on dependant variables. Model 1 (Table I) compared the dependent variables between Catchments, Regulation (nested within Catchment), Sites (nested with Regulation) and Time before the EFR in February 2007. Catchment had two levels (Paterson and Williams catchments) as a fixed factor. Although the factor 'Regulation' was nested in Catchment, it was treated as a fixed factor rather than random due to differences in the nature of regulation and management of each regulated flow regime. Nested factors are usually treated as random factors, although exceptions can be made in experimental designs (e.g. Downes *et al.,* 2006). In this study, the regulated Paterson River supplies water for irrigation (using run-of-river transfers), whereas water is diverted from the Chichester River at Chichester Dam to supply water for domestic uses in the central coast region of NSW. Therefore, the effects of flow regulation on the flow regime are unique to each regulated–unregulated river pair (see Results), necessitating nesting rather than crossing Regulation and Catchment factors. The three Sites were tested as a random factor to generalize the effects of flow regime on each tributary as longitudinal differences in fish assemblages between sites in each river were minimal. Sample times (7 levels; monthly samples from August 2006 to February 2007) were designated as a fixed factor.

Model 2 (Table I) compared the dependent variables between Catchments, Regulation and Sites before and after the environmental flow release that occurred in the Paterson River. The design was similar to Model 1, but only used data from the three sampling times immediately before (December 2006, January 2007, February 2007) and after (March, April and May 2007) the EFR to create a



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Table I. Statistical model used for ANOVA and PERMANOVA

| Model 1 (Differences between flow regime) | | | | Model 2 (Before-After EFR) | | | |
|---|---|---|---|---|---|---|---|
| Source of variation | Type | d.f. | Denominator for F test and VC | Source of variation | Type | d.f. | Denominator for F test and VC |
| Catchment | F | 1 | S(R(C)) | Catchment | F | 1 | S(R(C)) |
| **Regulation (C)** | F | 2 | S(R(C)) | **Regulation (C)** | F | 2 | S(R(C)) |
| Site (Re(C)) | Ra | 8 | Residual | Site (Re(C)) | Ra | 8 | Residual |
| Time | F | 6 | S(R(C)) × T | Period | F | 1 | S(R(C)) × P |
| C × T | F | 6 | S(R(C)) × T | Time (P) | F | 4 | S(R(C)) × T(P) |
| **Re(C) × T** | F | 12 | S(R(C)) × T | C × P | F | 1 | S(R(C)) × P |
| S(Re(C)) × T | M | 48 | Residual | C × T(P) | F | 4 | S(R(C)) × T(P) |
| Residual | Ra | 252 | | **Re(C) × P** | F | 2 | S(R(C)) × P |
| Total | | 335 | | **Re(C) × T(P)** | F | 8 | S(R(C)) × T(P) |
| | | | | S(Re(C)) × P | M | 8 | Residual |
| | | | | S(Re(C)) × T(P) | M | 32 | Residual |
| | | | | Residual | Ra | 216 | |
| | | | | Total | | 287 | |

Type, F: fixed factor, Ra: random factor, M: mixed (F × Ra). VC: variance component. Terms presented in **bold** are of key interest in this study.

balanced design to increase statistical power (see Underwood 1997). Model 2 included the additional factor of Period as a fixed factor representing 'before' and 'after' sampling, with the Time (3 levels) as a fixed factor nested within each Period.

Null hypotheses of no differences in fish assemblage composition between catchments, river types, sites, period and time were tested using the experimental models (Table I) by Permutational Multivariate Analysis of Variance (PERMANOVA) (Anderson, 2001). Samples were standardized by total abundance (to give relative abundance of each species), fourth-root transformed and then compared based on the Bray-Curtis dissimilarity measure using 9999 permutations. Samples were ordinated using multi-dimensional scaling (MDS) and data for each river within each catchment were presented separately from a single MDS ordination to enable direct comparison and clarify presentation. All multivariate analyses were done using PRIMER v6.1.11 with the PERMANOVA+ 1.0.1 add-on package.

All fish assemblage data were analysed using second-stage community analyses (Clarke *et al.,* 2006) to test for interactions between the assemblage composition trajectories in different rivers over time. On second-stage ordination plots, points that are close together indicate similarities in time trajectories (Clarke *et al.,* 2006). One-way second-stage ANOSIMs for each of the Paterson and Williams catchments were used to examine the temporal differences between regulated and unregulated rivers in each catchment. As only three replicate sites were sampled in each river over time, ANOSIM can only detect differences at $P = 0.1$, therefore second-stage ANOSIM results were considered significant at $P \leq 0.1$.

## RESULTS

### *Hydrological analysis*

The first two principal components (PCs) of the PCA of flow regimes of the regulated Chichester and Paterson Rivers and unregulated Allyn and Williams Rivers from 2002 to 2007 explained 72.3% of the total variation described by the 17 flow metrics (Figure 3). PC1 indicates a gradient of increasing magnitude and rate of falling flows and mean daily flow whereas along PC2, there is a gradient of increasing flow variability and flood flow index and reduced lower base flow index (Figure 3). The two regulated rivers differed considerably in hydrology. The Paterson River was characterized by lower flow variability and flood flow index values and higher base flow index values, whereas the Chichester River had higher flow variability, flood flow index scores and lower base flow index scores. The two unregulated rivers had similar flow regimes. All rivers had similar annual patterns in flow regimes (Figure 3).

### *Fish fauna*

A total of 6260 fish was collected, representing 11 native species and a single alien species, gambusia (Table II). Australian smelt and Cox's gudgeon were the only species collected at all sites in all four study tributaries, accounting for 88.8% of the total catch. Freshwater mullet (*Myxus petardi*), bullrout (*Notesthes robusta*) and southern-blue eye (*Pseudomugil signifer*) were found at low abundances only in the Paterson River catchment. Shortfinned eel (*Anguilla australis*) and striped gudgeon (*Gobiomorphus australis*) were each only sampled in three of the four tributaries (Table II). Flathead gudgeon (*Philypnodon grandiceps*) were sampled in all rivers,



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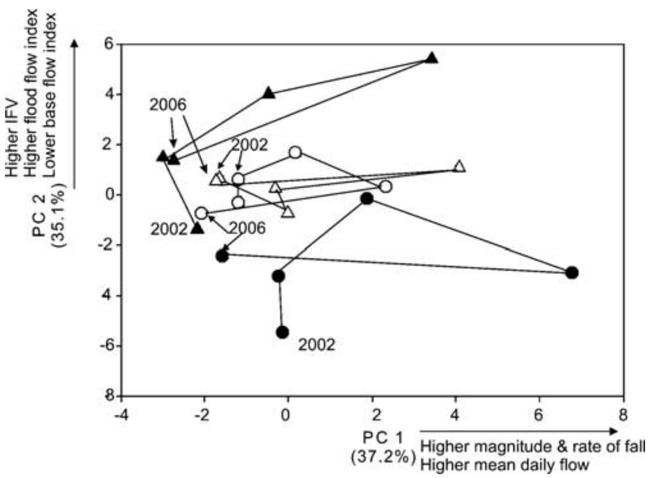


Figure 3. PCA plot of the first two principal component axes (PC1 vs. PC2) for annual flow metrics of the unregulated Allyn (clear circles) and Williams (clear triangles) rivers and regulated Chichester (solid triangles) and Paterson (solid circles) rivers from 2002 to 2006 calendar years. Each symbol (e.g. 2002) represents a single year from July–June

although in greater abundances in the Williams catchment. Australian bass (*Macquaria novemaculeata*) were collected in all rivers, although in low numbers.

*Associations between fish assemblages and flow regime*

Fish species richness did not differ between regulated–unregulated flow regimes, whereas sampling time explained the most (22.5%) variation (Table III). A significant Catchment × Time interaction indicated that differences between catchments were not consistent through time, with greater species richness found in the Paterson catchment when compared to the Williams catchment in November 2006 and January 2007. Temporal differences in total fish abundance between regulated–unregulated tributaries were not consistent and explained 13.5% variation (Table III), with greater abundances of fish sampled in the unregulated river of the Williams catchment compared to the regulated tributary in November 2006 (Figure 4). Sampling immediately following flooding across all four study rivers in September 2006 collected very few fish, explaining the reduced species richness and abundance of fish (Figure 4).

Differences in the abundance of Australian smelt between regulated and unregulated tributaries were only detected in the Williams catchment in September and November 2006 (Table III; Figure 5). Changes in the abundance of Australian smelt across all rivers through time explained the greatest proportion of total variation (48%, Table III). Abundances of Cox's gudgeon were significantly higher in the unregulated tributary of the Williams catchment compared with the regulated tributary in October and November 2006 (Figure 5). Cox's gudgeon were consistently more abundant in the Paterson catchment than the Williams catchment. Gambusia did not differ in abundance between catchments or regulated–unregulated rivers (Table III), and changes through time explained only small proportions of the total variance (Table III).

*Effects of the environmental flow release on fish populations and assemblages*

Species richness did not differ between regulated and unregulated tributaries, and no effect of the EFR was detected with non-significant Regulation (Catchment) × Period and Regulation (Catchment) × Time (Period) interactions

Table II. Summary of fish sampled in the Paterson and Williams catchments between August 2006-June 2007

| Species name | Common name[a] | Paterson catchment | | Williams catchments | | Total |
|---|---|---|---|---|---|---|
| | | Allyn (UR) | Paterson (R) | Williams (UR) | Chichester (R) | |
| *Anguilla australis* | Shortfinned eel[D] | | 3 | 2 | 1 | 6 |
| *Anguilla reinhardtii* | Longfinned eel[D] | 23 | 73 | 11 | 5 | 112 |
| *Gambusia holbrooki*[A] | Eastern gambusia[ND] | 124 | 41 | 13 | 13 | 191 |
| *Gobiomorphus australis* | Striped gudgeon[D] | 7 | 12 | | 1 | 20 |
| *Gobiomorphus coxii* | Cox's gudgeon[D] | 471 | 582 | 184 | 128 | 1365 |
| *Macquaria novemaculeata* | Australian bass[D] | 1 | 1 | 2 | 4 | 8 |
| *Myxus petardi* | Freshwater mullet[D] | 33 | 5 | | | 38 |
| *Notesthes robusta* | Bullrout[D] | 1 | | | | 1 |
| *Philypnodon grandiceps* | Flathead gudgeon[ND] | 1 | 2 | 78 | 80 | 161 |
| *Pseudomugil signifer* | Southern blue-eye[U] | | 4 | | | 4 |
| *Retropinna semoni* | Australian smelt[ND] | 427 | 429 | 2530 | 807 | 4193 |
| *Tandanus tandanus* | Freshwater catfish[ND] | 56 | 49 | 28 | 28 | 161 |
| Total abundance | | 1144 | 1201 | 2848 | 1067 | 6260 |
| Total no. species | | 10 | 11 | 8 | 9 | 12 |

[a]common name according to McDowall (1996), [A] denotes alien species, [D, ND] and [U] indicates known diadromous, non-diadromous and unknown/ undetermined migrations (according to McDowall 1996, Gehrke *et al.,* 2002, Pusey *et al.,* 2004 and Miles *et al.*, 2009), UR: unregulated, R; regulated.



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Table III. Mean square, F-ratio and probability results of 4-way analysis of variance (Model 1) testing differences in species richness, total fish abundance, and the abundance of Australian smelt, Cox's gudgeon and gambusia. Significant P values ($\leq$0.05) are presented in bold and variance components (VC) are indicated

| Source of variation | Species richness | | | | Total abundance | | | | Australian smelt | | | | Cox's gudgeon | | | | Gambusia | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | MS | *F* | *P* | VC | MS | *F* | *P* | VC | MS | *F* | *P* | VC | MS | *F* | *P* | VC | MS | *F* | *P* | VC |
| Catchment | 9.0 | 7.9 | **0.023** | 3.5 | 0.5 | 0.6 | 0.467 | 0 | 12.7 | 27.7 | **0.001** | 7.0 | 18.1 | 11.9 | **0.009** | 19.2 | 1.2 | 2.5 | 0.15 | 2.8 |
| Regul (C) | 0.7 | 0.6 | 0.577 | 0 | 1.0 | 1.3 | 0.330 | 0.5 | 2.2 | 4.8 | **0.042** | 2.0 | 0.5 | 0.3 | 0.719 | 0 | 0.7 | 1.5 | 0.289 | 1.7 |
| Si (R(C)) | 1.1 | 1.6 | 0.135 | 1.1 | 0.8 | 4.8 | **<0.001** | 4.1 | 0.5 | 1.7 | 0.100 | 0.6 | 1.5 | 6.7 | **<0.001** | 9.0 | 0.5 | 5.0 | **<0.001** | 8.9 |
| Time | 15.4 | 14.9 | **<0.001** | 22.5 | 11.5 | 29.6 | **<0.001** | 40.7 | 24.6 | 53.1 | **<0.001** | 48.0 | 3.6 | 9.6 | **<0.001** | 13.1 | 0.5 | 2.8 | **0.020** | 3.9 |
| C × T | 3.6 | 3.4 | **0.007** | 7.9 | 0.6 | 1.5 | 0.189 | 1.5 | 0.7 | 1.5 | 0.191 | 1.0 | 0.5 | 1.4 | 0.252 | 1.1 | 0.4 | 2.4 | **0.038** | 6.3 |
| R(C) × T | 1.8 | 1.7 | 0.086 | 4.8 | 1.3 | 3.4 | **0.001** | 13.5 | 1.8 | 4.0 | **<0.001** | 11.0 | 0.7 | 2.0 | **0.045** | 6.1 | 0.2 | 1.4 | 0.177 | 3.9 |
| S(R(C)) × T | 1.0 | 1.4 | **0.043** | 5.8 | 0.4 | 2.3 | **<0.001** | 9.6 | 0.5 | 1.7 | **0.005** | 4.6 | 0.4 | 1.7 | **0.007** | 7.2 | 0.2 | 1.7 | **0.006** | 10.6 |
| Residual | 0.7 | | | 54.3 | 0.2 | | | 30.1 | 0.3 | | | 25.9 | 0.2 | | | 44.2 | 0.1 | | | 61.9 |

(Table IV). Significantly greater numbers of species were collected in the Paterson catchment than in Williams before and after the EFR (Table IV; Figure 4). Mean total fish abundance per sample was not significantly different between catchments or flow regime, and a non-significant interaction between Regulation (Catchment) × Time factors indicated that total fish abundance was not altered by the EFR. In all rivers, fish abundances were consistently higher before the EFR (indicated by the significant Period term explaining 26.9% variation, Table IV), indicating that fish abundances had similar patterns between rivers.

Australian smelt were consistently more abundant in the Williams catchment than in the Paterson catchment but abundances did not differ between regulated–unregulated tributaries in either catchment or in response to the EFR (Table IV; Figure 5). Differences in the abundance of Cox's gudgeon were only significant between the Paterson and Williams catchments and in all rivers through time (Table IV), but showed not significant differences between regulated–unregulated tributaries or after the EFR. Abundances of gambusia were greater in the Paterson catchment than the Williams catchment (Table IV), and were significantly higher in the unregulated Allyn River than in the regulated Paterson River (Table IV; Figure 5).

*Differences between flow regime and effects of the EFR on fish assemblage composition*

Fish assemblage composition differed significantly between catchments, particularly for the Catchment × Time interaction term (Table V). Diadromous species, particularly longfinned eel, Cox's gudgeon and freshwater mullet were more abundant in the Paterson catchment than in the Williams catchment. Differences in fish assemblage composition between regulated–unregulated rivers changed significantly over time, as indicated by the significant Regulation (Catchment) × Time interaction, although this contributed little to the total variation (4.5%, Table V).

The EFR had no significant effect on fish assemblage composition (non-significant Regulation (Catchment) × Period and Regulation (Catchment) × Time (Period) interactions, Table V; Figure 6). Differences between fish assemblage composition between the Paterson and Williams catchments were consistent through time and contributed to a large proportion of total variance. Differences between sites in rivers indicated that within-tributary assemblage variability was maintained throughout the sampling period, however this variability was consistent among all river before and after the EFR indicated by the non-significant interaction terms (Table V).

Differences in temporal trajectories of fish assemblage between regulated and unregulated tributaries in the Paterson and Williams catchments were inconsistent. In the





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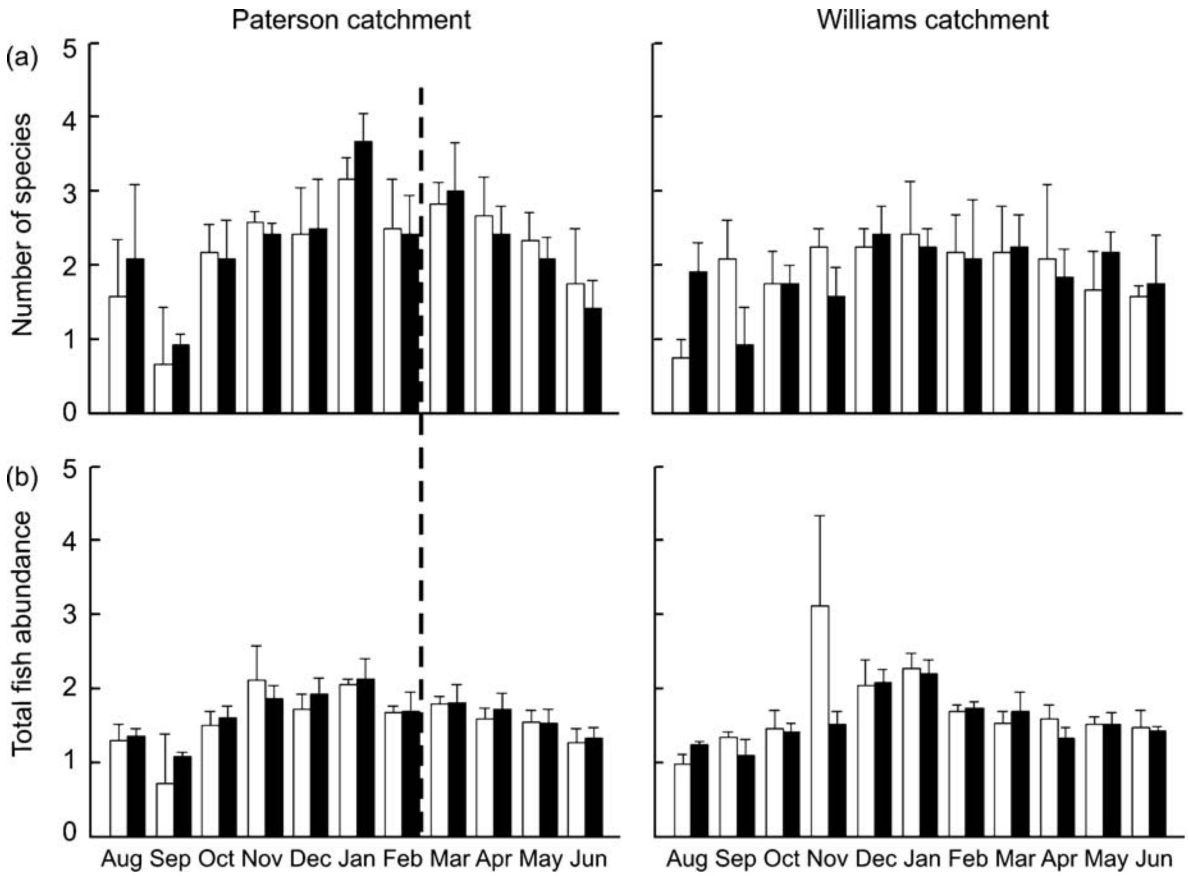


Figure 4. Mean (+1 s.d.) of (a) number of species and (b) total fish abundance (fourth-root transformed) per sample in the unregulated (open bars) and regulated (solid bars) tributaries of the Paterson and Williams catchments between August 2006–June 2007. The EFR is indicated by the dashed line

Paterson catchment, second-stage ANOSIM tests found significantly different trajectories between the regulated and unregulated tributary (ANOSIM $R = 0.37$, $P = 0.1$) indicating that temporal changes in composition were inconsistent between rivers, particularly between sampling events 5–9 (Figure 6). In the Williams catchment, temporal trajectories were similar between both regulated and unregulated tributaries (ANOSIM $R = 0.185$, $P = 0.2$).

## DISCUSSION

### *Patterns in fish assemblages associated with flow regulation*

There was little difference in fish species richness and total abundance between regulated and unregulated tributaries of the Paterson and Williams catchments whereas species richness between the two study catchments differed significantly. The two dams regulating river flow in this study are known to spill frequently, thereby maintaining frequency of large floods and some level of variability in the flow regime likely to result in only moderate changes to flow regime when compared to regulated rivers in other studies (e.g. Humphries *et al*., 2008). Given the effect of flow regulation on the flow regime is not as marked as other regulated rivers where fish assemblages have been considerably altered (e.g. Gehrke *et al.,* 1995; Humphries *et al.,* 2008), it seems likely that the impact of flow regulation on fish assemblages is minimal in the coastal rivers assessed in the present study.

Neither fish species populations nor assemblages showed consistent differences in regulated tributaries when compared with unregulated tributaries in either of the two catchments. Anthropogenic impacts on riverine fish assemblages are considered best analysed by young-of-the-year (YOY) fish rather than adults as densities of YOY fish are often strongly associated with hydrology (e.g. Mérigoux and Ponton, 1999). Almost all of the fish species considered abundant (>1% of the total catch) in this study are short-lived species rarely exceeding 1–2 years of age, such as Australian smelt (maximum length 60 mm, Rolls unpublished data). Humphries *et al.,* (2008) studied fish assemblages in the Campaspe River (heavily regulated) and the Broken River (mildly regulated) in the southern Murray-Darling



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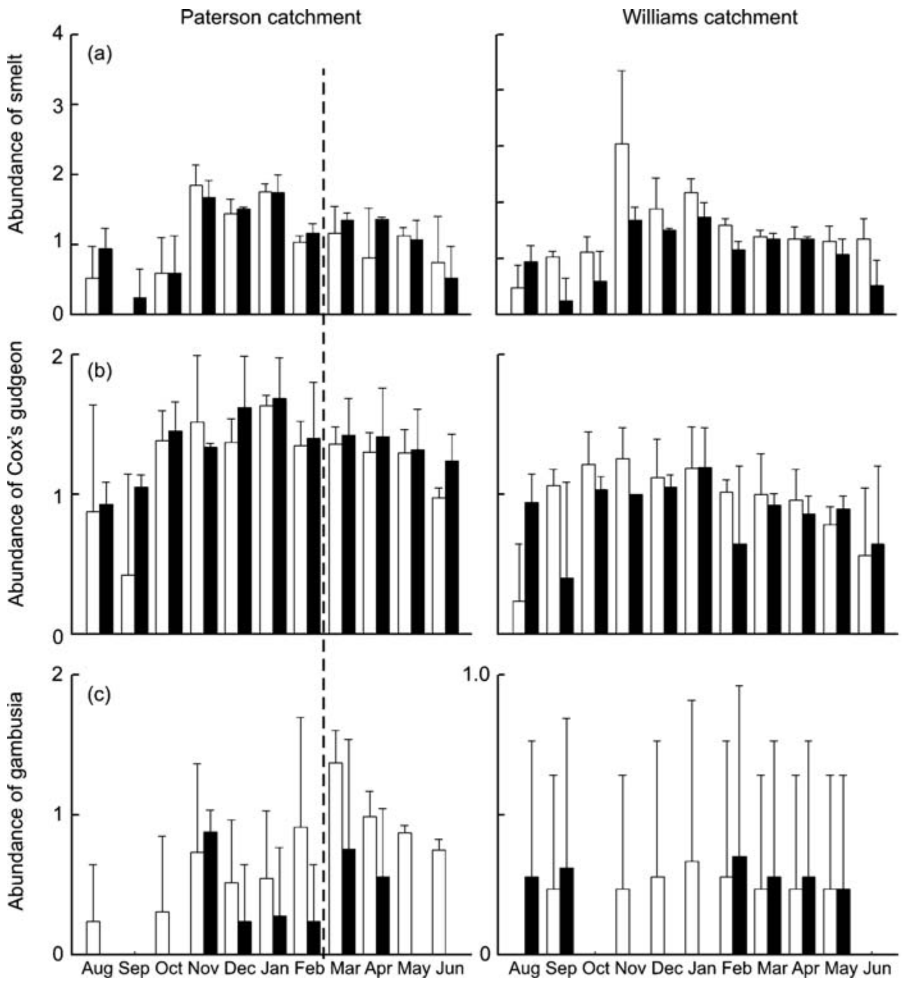


Figure 5. Mean (+1 s.d.) of (a) Australian smelt, (b) Cox's gudgeon and (c) gambusia (all fourth-root transformed) per sample in the unregulated (open bars) and regulated (solid bars) tributaries of the Paterson and Williams catchments between August 2006–June 2007. The EFR is indicated by the dashed line

Basin, Australia, over 7 years, and found that fish composition did not appear to be influenced by hydrology, although abundances of YOY golden perch (*Macquaria ambigua*), European perch (*Perca fluviatilis*) and common carp (*Cyprinus carpio*) were significantly influenced by hydrological variables such as the variability in flow events. Differences in fish populations of assemblages between regulated and unregulated tributaries would have therefore been expected to be apparent in this study. The effect of flow regulation on fish assemblages in the Paterson and Williams catchments is likely to be small, probably as the effect of flow regulation on the flow regime in regulated tributaries is minimized by frequent dam spilling events maintaining hydrological variability.

The significant differences in fish assemblages between the two study catchments and significantly lower abundances of common species such as Cox's gudgeon in the Williams catchment than in the Paterson catchment are likely to be due to barriers to connectivity between the two catchments and the lower Hunter River estuary. Seaham Weir in the lower Williams catchment has structures to facilitate fish passage, but these probably ineffective for temperate Australian freshwater fish species as they were historically designed for non-native salmonids (Stuart and Mallen-Cooper, 1999; Thorncraft and Harris, 2000). In this case, it is difficult to separate the effects of flow regime change on fish assemblages from other confounding factors such as the physical structures that control flow and also impact on fish movements and migration.

Differences in the abundance of Cox's gudgeon and Australian smelt between regulated–unregulated rivers were inconsistent with studies in other temperate coastal catchments. In this study, Cox's gudgeon were more abundant in the unregulated tributary of the Williams catchment only during October–November 2006, the peak period of juvenile migration to upland reaches (Pusey *et al.,* 2004). In the Hawkesbury-Nepean catchment, Cox's gudgeon were recorded in greater abundances in unregulated



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Table IV. Mean square, F-ratio and probability results of five-way analysis of variance (Model 2) testing differences in species richness, total fish abundance, and the abundance of Australian smelt, Cox's gudgeon and gambusia

| Source of variation | Species richness | | | | Total abundance | | | | Australian smelt | | | | Cox's gudgeon | | | | Gambusia | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | MS | *F* | *P* | VC | MS | *F* | *P* | VC | MS | *F* | *P* | VC | MS | *F* | *P* | VC | MS | *F* | *P* | VC |
| Catchment | 19.5 | 7.1 | **0.029** | 10.7 | 0.1 | 0.3 | 0.583 | 0 | 10.6 | 47.0 | **<0.001** | 11.2 | 30.4 | 22.4 | **0.001** | 37.7 | 4.3 | 11.3 | **0.010** | 9.8 |
| Regul (C) | <0.1 | <0.1 | 0.984 | 0 | 0.1 | 0.4 | 0.702 | 0 | 1.0 | 4.3 | 0.054 | 1.6 | 0.2 | 0.1 | 0.867 | 0 | 3.3 | 8.7 | **0.010** | 14.8 |
| Si (R(C)) | 2.8 | 3.7 | **<0.001** | 7.8 | 0.3 | 3.6 | **0.001** | 4.2 | 0.2 | 0.9 | 0.493 | 0 | 1.4 | 5.6 | **<0.001** | 8.7 | 0.4 | 2.2 | **0.028** | 3.2 |
| Period | 3.8 | 4.5 | 0.067 | 1.9 | 10.0 | 54.8 | **<0.001** | 26.9 | 20.5 | 27.0 | **0.001** | 21.2 | 3.0 | 18.4 | **0.003** | 3.7 | 0.4 | 2.2 | 0.176 | 0.6 |
| Time (P) | 3.8 | 4.9 | **0.003** | 5.9 | 1.5 | 9.3 | **<0.001** | 11.0 | 4.1 | 10.9 | **<0.001** | 12.0 | 1.2 | 5.0 | **0.003** | 3.7 | 0.5 | 2.5 | 0.060 | 2.4 |
| C × P | <0.1 | <0.1 | 0.950 | 0 | 1.8 | 9.9 | **0.014** | 8.9 | 2.8 | 3.6 | 0.092 | 4.3 | <0.1 | 0.4 | 0.564 | 0 | 0.3 | 1.5 | 0.257 | 0.5 |
| C × T(P) | 1.8 | 2.3 | 0.082 | 3.9 | 0.2 | 1.5 | 0.212 | 1.5 | 0.4 | 1.1 | 0.375 | 0.2 | <0.1 | 0.4 | 0.798 | 0 | 0.3 | 1.4 | 0.244 | 1.4 |
| R(C) × P | 0.4 | 0.5 | 0.615 | 0 | <0.1 | <0.1 | 0.907 | 0 | 0.2 | 0.3 | 0.754 | 0 | <0.1 | 0.2 | 0.797 | 0 | 0.1 | 0.6 | 0.561 | 0 |
| R(C) × T(P) | 0.5 | 0.6 | 0.778 | 0 | 0.1 | 0.6 | 0.768 | 0 | 0.1 | 0.2 | 0.985 | 0 | 0.3 | 1.7 | 0.223 | 0.7 | 0.2 | 1.2 | 0.397 | 0.8 |
| S(R(C)) × P | 0.8 | 1.1 | 0.335 | 0.8 | 0.2 | 1.9 | 0.065 | 2.8 | 0.8 | 3.1 | **0.002** | 6.6 | 0.2 | 0.7 | 0.724 | 0 | 0.2 | 1.1 | 0.352 | 0.6 |
| S(R(C)) × T(P) | 0.8 | 1.1 | 0.371 | 1.2 | 0.2 | 1.7 | **0.018** | 6.4 | 0.4 | 1.5 | **0.039** | 5.1 | 0.2 | 1.0 | 0.525 | 0 | 0.2 | 1.2 | 0.219 | 3.2 |
| Residual | 0.7 | | | 67.8 | 0.1 | | | 38.4 | 0.2 | | | 37.7 | 0.2 | | | 45.5 | 0.2 | | | 62.7 |

tributaries when compared to regulated tributaries (Gehrke *et al.,* 1999). These patterns may be a consequence of higher abundances of Cox's gudgeon in flowing riffle habitat that is susceptible to changes in flow (Pusey *et al.,* 2004). Abundances of Australian smelt were greater in regulated reaches of the Hawkesbury-Nepean catchment (Gehrke *et al.*, 1999), although in the southern Murray-Darling Basin the proportion of Australian smelt larvae was higher in the Broken River when compared to the more heavily regulated Campaspe River (Humphries *et al.,* 2002). However in the present study, greater abundances of Australian smelt were only found in the unregulated tributary of the Williams catchment in September and November 2006. These inconsistencies between separate studies suggest that these species display thresholds to flow regime alteration and that recruitment success strongly determines the population-level effects of flow regulation.

Flow regulation has been suggested to reduce the resilience of fish assemblages to invasion by alien species (Gehrke and Harris, 2001), yet this hypothesis was not supported in this study. Gambusia, the only alien species collected in this study, were rare (3% of all fish). The very high residual variance component calculated during ANOVA for gambusia (61.9–62.7%) possibly masked responses to flow regime differences by this species uncommon in the rivers in this study. The lack of other alien species sampled in this study also fails to support the prediction that fish assemblages stressed by flow regulation are dominated by alien species. Anthropogenic hydrological disturbance benefits the native roundhead galaxias (*Galaxias anomalus*) to the detriment of the alien brown trout (*Salmo trutta*) in southern New Zealand (Leprieur *et al.,* 2006). Restoring elements of the flow regime influenced by flow regulation has been suggested to be an effective tool for controlling alien species (e.g. Marchetti and Moyle, 2001). The lack of abundant populations of alien species in the rivers sampled in this study suggests that the current extent of flow regulation does not facilitate the establishment of significant populations of alien species in this region, assuming that alien fish are able to access these rivers, habitats are suitable and that the sweep-net electrofishing used here did not bias against them. However, using SNE, King (2004) sampled ~6.8 times more gambusia than Australian smelt juveniles and adults, indicating that the method used in this study is adequate to detect small-bodied and juvenile alien species.

### *Responses of fish assemblages and populations to an environmental flow release*

The EFR during this study was delivered to experimentally mimic a small flood event that now occur with reduced frequency during the summer–autumn period in the two




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Table V. Mean square, F-ratio and probability results of Permutational Multivariate Analysis of Variance (PERMANOVA) testing differences in fish assemblage composition between regulated and unregulated tributaries (Model 1) and responses to the environmental flow release (Model 2). Significant differences ($P \leq 0.05$) are in bold and VC indicates variance component

| Source | Associations with flow regime (Model 1) | | | | Source | Effect of flow release (Model 2) | | | |
|---|---|---|---|---|---|---|---|---|---|
| | MS | *F* | *P* | VC | | MS | *F* | *P* | VC |
| C | 61 977 | 9.7 | **0.003** | 12.1 | C | 61 054 | 11.2 | **0.003** | 20.3 |
| R(C) | 3303 | 0.5 | 0.794 | 0 | R(C) | 4435 | 0.8 | 0.613 | 0 |
| S(R(C)) | 6397 | 4.5 | **<0.001** | 6.5 | S(R(C)) | 5461 | 4.9 | **<0.001** | 9.5 |
| T | 21 165 | 9.2 | **<0.001** | 14.4 | P | 10 508 | 8.2 | **0.004** | 3.4 |
| C × T | 3858 | 1.7 | **0.025** | 2.4 | T(P) | 4289 | 4.1 | **<0.001** | 3.6 |
| R(C) × T | 3775 | 1.6 | **0.014** | 4.5 | C × P | 3520 | 2.8 | 0.051 | 1.6 |
| S(R(C)) × T | 2312 | 1.6 | **<0.001** | 8.1 | C × T(P) | 1961 | 1.9 | 0.067 | 2.0 |
| Residual | 1429 | | | 52.2 | R(C) × P | 1564 | 1.2 | 0.324 | 0.4 |
| | | | | | R(C) × T(P) | 1052 | 1.0 | 0.483 | 0 |
| | | | | | S(R(C)) × P | 1274 | 1.1 | 0.291 | 0.7 |
| | | | | | S(R(C)) × T(P) | 1049 | 0.9 | 0.633 | 0 |
| | | | | | Residual | 1111 | | | 58.4 |

regulated tributaries when compared with the unregulated tributaries. Responses to the EFR were not detected at either the population or assemblage level. This may be due to the fact that river assemblages, including fish, do not respond to small floods in the short term, and show greater changes after long periods of sequential small floods (Robinson and Uehlinger, 2008). Pires *et al.,* (2008) suggest that large floods in Mediterranean climates may influence population dynamics for fish species sensitive to flow, whereas the responses at the assemblage level may not be apparent. Similarly, brown trout abundances did not show any increase until 2 years following a 3-year period of a series of small flushing flows along the River Spöl in Switzerland (Ortlepp and Mürle, 2003). This suggests that the response by fish assemblages and populations following small environmental flows may not be detectable until well after (e.g. years) restoration attempts.

Flow down the two regulated tributaries, particularly the Paterson River, is now permanent as the river channel is used to transport irrigation water from Lostock Dam during dry periods. Low flows or cease-to-flow events occur in the unregulated tributaries, such as the Allyn River, and were associated with consistent peaks in fish abundance as a possible consequence of limited habitat. In the Waipara River, New Zealand, Jowett *et al.,* (2005) found no correlations between abundances of adult fish and the frequency and magnitude of high flows, whereas changes in fish abundances were consistent with the frequency and duration of low flow events. In temperate Australia, one of the major problems associated with flow regulation is probably

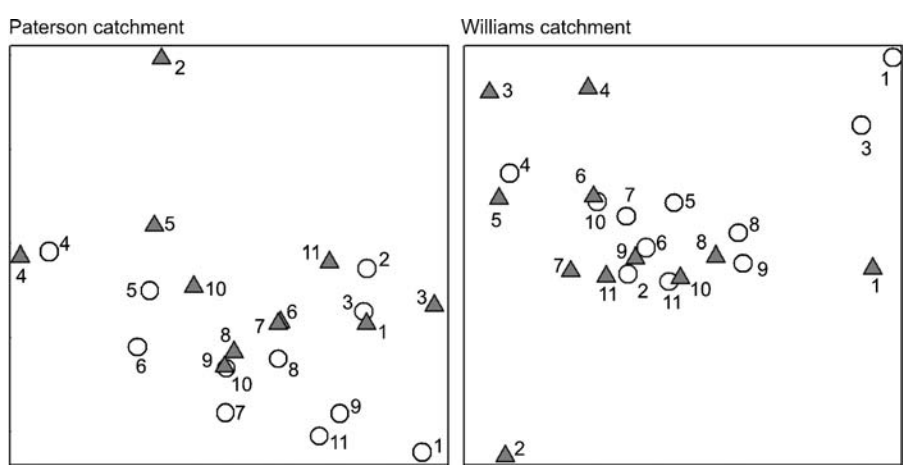


Figure 6. Two-dimensional MDS ordination (stress = 0.22) indicating similarities in fish assemblages between monthly sample times (August 2006–June 2007, 1–11, respectively). Regulated and unregulated sites in each catchment are indicated by grey triangles and open circles, respectively. Points presented are the average composition of all three sites in each river reach. The EFR occurred between sampling occasions 7 and 8



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'anti-drought', the removal of the important and natural low-flow events from the flow regime to which indigenous aquatic biota (including fish) are adapted (McMahon and Finlayson, 2003). Low flows are a natural phenomenon in temperate Australian rivers, and although often perceived to be a problem by public (Thoms and Sheldon, 2002), restoration of natural low flows in regulated rivers is recognized as providing the natural flow regime for regulated rivers (Reich *et al.,* 2010). If possible, experimental assessment of low flow periods in regulated rivers that are affected by constant flows, such as the Paterson and Chichester Rivers, will improve our knowledge of the most appropriate management strategies that are focused on delivery of environmental water allocations.

## *Conclusions*

At least four possible reasons exist to explain why the EFR tested in this study did not alter fish assemblages. Firstly, high within-river (site-site) variability in fish assemblages makes it difficult to detect more subtle differences between rivers (i.e. between regulated and unregulated), therefore requiring larger numbers of sites in each river to quantify this variability with a repeated measures design such as that used in this study. However, increasing the number of sites in each river would be a trade off by reducing sampling frequency, therefore resulting in possible short-term patterns being missed by less frequent sampling. Secondly, the timing of the EFR was followed by a series of natural spates in all rivers, and may also have occurred during a period where fish assemblages were not affected by flood events that may have improved abundances. Thirdly, the duration of sampling following the EFR may have been too short, and the variable hydrology pre- and post-EFR would make it unrealistic to detect responses to a single flow release. For example, ecological responses to a series of 15 artificial floods in a regulated river in Switzerland over an 8 year period were only detected after the first 3 years (Robinson and Uehlinger, 2008), suggesting that single artificial floods have little long-term consequences and that a series of EFRs are more likely to contribute to sustained ecological changes. Finally, disturbances acting at larger scales throughout the Hunter River catchment, for example river training work and desnagging (e.g. Erskine, 1990; Erskine, 2001; Brooks *et al.,* 2004), may have an overriding primary influence on fish assemblages, rather than flow regime change that affects relatively fewer river reaches. In this study, flow regime change has not been associated with loss of fish species biodiversity, and is similar to that reported from rivers from the wider coastal NSW region (Gehrke and Harris, 2000).

The use of large restoration projects as ecological experiments is probably the best way to understand ecosystems at the scales of interest for environmental managers (Poff *et al.,* 2003). This study found no significance in fish assemblages or populations associated with flow regime change, although anthropogenic impacts on rivers (flow regulation, riparian clearing, habitat loss etc) almost always occur simultaneously (Stewart-Koster *et al.,* 2010). The impacts of flow regime change on fish populations and assemblages often do not become apparent until long after flow regulation has occurred (possibly decades-centuries) as the impacts are likely to become more evident over longer times scales and in conjunction with other disturbances occurring simultaneously. However, by sampling small-bodied short-lived species, it would be expected that if there were any effects of flow regulation, that this study would have been able to detect them. Accordingly, as single EFRs are unlikely to contribute to sustained ecological change, more frequent and repeated uses of environmental water allocations may be needed in this region under the climatic conditions that were experienced in this study to promote a shift in fish assemblage composition.

This study used a multiple Before-After-Control-Impact (MBACIP) design to test if differences between regulated–unregulated tributaries were consistent in each catchment. MBACI(P) designs are advocated for determining if the effects of human activity are consistent between different rivers (Downes *et al.,* 2002), however such designs are limited in large scale assessments of the effects of river regulation and EFRs. Despite high statistical power, this study found few differences in fish assemblages between regulated and unregulated tributaries in either of the two catchments, and no consequences of a single EFR. This indicates that fish assemblages have not been impacted by river regulation in the eastern Hunter River catchment, and that current flow management rules in the Paterson and Chichester Rivers are not exceeding ecological thresholds that would result in poor fish assemblage condition.

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University of New England Animal Ethics Committee (AEC permit 06/003) and NSW Department of Primary Industries scientific collection permit (P06/0014).

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