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2016–17 Basin-scale evaluation of Commonwealth environmental water — Fish

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Final Report

2016–17 Basin-scale evaluation of Commonwealth environmental water – Fish

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Contents

List of tables

List of figures

Executive summary

This report summarises findings from year three of a five-year program to assess Basin scale outcomes for fish from environmental water delivery under the Commonwealth Environmental Water Office (CEWO) Long Term Intervention Monitoring (LTIM) Program. The analyses presented integrates data collected from multiple sites across the Basin, in particular from seven Selected Areas where standardised monitoring protocols have been adopted to support an integrated analysis (see Stoffels & Bond 2016).

The multi-site analyses presented here are designed to complement the reporting on outcomes from each of these seven Selected Areas. As well as integrating data from multiple rivers, this 2016–17 report involves an analysis of data collected over the first three years of the program, with the focus on demonstrating the use of statistical modelling approaches to report on and assess outcomes from environmental watering, and in particular to assist in separating out the effects associated with environmental watering actions from the effects of the considerable background variation in hydrology that occurs in the Basin.

Environmental context

While hydrology during the first two years of LTIM was characterised by below average discharge, the third year of LTIM (2016–17) was defined by significant natural flooding in the southern Basin. Large areas of the inundated floodplain in the Murray and Murrumbidgee catchments had not been flooded since the Millennium Drought, resulting in the liberation of very large quantities of accumulated terrestrial carbon as dissolved organic carbon. This liberation of organic material provided an accessible carbon source for microorganisms and microinvertebrates. As a result of this large increase in ecosystem respiration, the 2016 flood resulted in hypoxic flows in the Lachlan and Edward-Wakool (severe hypoxia), and the Murrumbidgee, Goulburn and Lower Murray (mild hypoxia).

Synthesis of Selected Area fish outcomes

During 2016–17 a total of 58 Commonwealth environmental water actions were delivered throughout the Basin, with expected outcomes for fish. While these covered a range of objectives the most common were fish habitat, movement and spawning. There were also a small number of watering actions with primary expected outcomes aimed at ameliorating the effects of hypoxia on fish. The majority of watering actions with primary expected outcomes for fish were delivered as freshes (including weir pool raising and lowering in the Lower Murray) with a smaller number of base flows delivered mainly for fish habitat and condition, and contributions to several bankfull flows aimed mostly at fish movement.

Commonwealth environmental water contributed to fish spawning in several rivers across the Basin in 2016–17, with strong evidence of Commonwealth environmental water supporting spawning and recruitment in Gunbower Creek and the lower Darling River. Commonwealth environmental water was also successful in facilitating fish movement within and between a number of river systems including between the Murray and Goulburn Rivers, the Barwon and Macquarie Rivers and the lower Darling and Murray Rivers. The effectiveness of Commonwealth environmental water in ameliorating hypoxic conditions was varied across Selected Areas where this was monitored.

Results of Basin-scale evaluation

Data collected in the first three years of LTIM was analysed using statistical models to evaluate two aspects of fish in the Basin:

1. the state of fish populations focussed on age structure (recruitment and survival) and condition of three large bodied native fish: bony bream, golden perch and Murray cod; and
2. fish community dynamics of all small and large bodied native fish.

It was originally planned that the 2018 analysis would also include models of fish spawning (Stoffels *et al.* 2016). Data collection and storage in the LTIM database has been hampered by a range of issues related to both the implementation of different methods in several locations as well as data quality assurance and control. This has prevented an analysis and development of the spawning model in this third LTIM year. Larval fish data issues have been resolved and it is anticipated that development of spawning models will be possible in the subsequent LTIM fish reports.

It should also be noted that due to data limitations, the models developed and presented here for fish populations and communities have not been cross validated (Hyndman & Athanasopoulos 2018). Therefore, all results should be considered low confidence inferences. Cross-validation of models will become possible as the program evolves, and longer time-series of data are available from each selected area.

Population dynamics

Models examining the individual condition of the three target large-bodied native fish, found that bony herring condition was not related to river flows and that recruitment and survival of this species was variable across the Basin, with no obvious patterns in relation to flow delivery. Results relating golden perch and Murray cod condition to patterns of hydrologic variability, however, yielded several new ecological insights, most notably:

- The impact of flows on body condition is season-specific;
  - High spring (in particular) and summer flows were associated with positive trends in fish condition, as measured at the end of the flow delivery season (autumn).
  - High autumn flows were associated with minor increases in the golden perch condition, but also appeared to be related to reduced Murray cod condition. At present there is no explanation for this association, and further data are required to establish the veracity of this finding.
  - High winter flows have subtle, but significant, positive effects on golden perch condition but no significant effects on Murray cod condition.
- Golden perch condition was positively related to both seasonal patterns of flow during a flow-delivery year, as well as the observed discharge in the previous water year. Thus, appropriately timed higher flows across multiple years may have cumulative benefits for golden perch condition.
- Body condition is also negatively correlated with the abundance of conspecifics, which suggests populations of golden perch and Murray cod may be food or habitat limited, with body condition decreasing as a result of competition.

In terms of the effects of environmental water (including Commonwealth environmental water) on fish populations, there is some evidence to suggest that:

- Commonwealth environmental water delivery during 2014–15 increased Murray cod condition in the Edward-Wakool and in the Gwydir.
- The delivery of Commonwealth environmental water in the Goulburn in autumn 2014–15 may have resulted in a decrease in Murray cod condition, although further data are required to confirm this possible effect.
- Environmental water delivery (Commonwealth environmental water and other environmental water sources) during 2014–15 and 2015–16 increased golden perch condition within the Goulburn, Murrumbidgee, Lachlan and Lower Murray Selected Areas.

Community dynamics

There was little change observed in the Basin's fish communities during the first two years of LTIM. The large-scale natural floods in 2016–17, however, had an observable effect on some species through reduced dissolved oxygen (hypoxia). While the analysis of data across multiple Selected Areas could not detect a consistent response to hypoxia in most fish species, this may reflect issues in detection of hypoxic conditions at some sites due to the timing of dissolve oxygen logger deployment, rather than a true inconsistent response. There were however consistent responses to hypoxia detected in two species; common carp and Murray cod:

- Carp abundances increased due to successful spawning and recruitment. This is the expected response of this species to flooding.
- Carp were also better able to cope with hypoxic conditions than native species such as Murray cod. As a result, there were lower rates of mortality of carp relative to Murray cod.
- Murray cod abundances declined at sites where flooding and hypoxia occurred. The magnitude of the response in Murray cod, however, varied considerably across Selected Areas.

We were unable to determine how specific water actions aimed at ameliorating hypoxia affected fish populations, including Murray cod and common carp. Monitoring of fish movement detected no movement of individual fish towards localised hypoxia refuges within the Edward-Wakool, but there were few data points, and thus we cannot conclude whether this strategy was effective. Irrespective of our ability to evaluate exactly how water actions ameliorated impacts of hypoxia, we argue that the decisions underpinning those water actions were sound for at least two reasons:

- Given the magnitude of natural flooding, Commonwealth environmental water was insufficient to alter water quality except at a very local scale.
- Given the high rates of mortality of Murray cod, enhancing the survival of even very small numbers, could help increase the resilience of local populations, and hence the capacity for the population to increase again in subsequent years.

Adaptive management

There are a number of key adaptive management recommendations that have arisen for the evaluation of data across the first three years of the LTIM project:

- Flows delivered in spring and summer have the greatest positive impact on golden perch and Murray cod condition. Spring-summer flow delivery may result in synergistic impacts on fish populations—promoting spawning while at the same time increasing energy acquisition/storage, which in turn may feed into enhancing other population processes in subsequent years.
- At this stage, the models indicate that autumn flow pulses may decrease Murray cod condition, although no mechanism has yet been identified, and this observation requires further investigation. It may, however, have implications for environmental flow releases delivered for other purposes, as well as for autumn irrigation flows.
- Planning of flow delivery each year should give thought to flows delivered in the previous year, which may affect the condition response of golden perch. In particular, priority should be given to delivering water actions for golden perch in the year immediately following a high flow year.
- It is important to note that when impacts of watering actions are additive across years:
  - observable impacts will take years to occur, and to detect reliably
  - there may be greater ecological returns in the long-term, without necessarily allocating more water

- o a great challenge will be the optimisation of multi-year flow regimes within catchments.
- Commonwealth, state and other environmental water use has synergistic effects on fish condition, so should be managed in a coordinated way. It is noted this is already occurring.
- Hypoxic blackwater events have the potential to reduce the longer-term benefits from environmental watering actions. There is a need for innovative thinking and further modelling to understand how the dynamics of (a) river-floodplain hydrological connectivity; and (b) floodplain organic carbon loads, can be actively adaptively managed, at scales that go beyond Selected Areas.
- The models suggest that shorter periods of hypoxia (e.g. a reduction in the duration of events from 14 to 7 days) can have a significant benefit in reducing mortality rates. Given that hypoxic blackwater events are likely to be unavoidable, a worthwhile focus for environmental watering actions could be to try and reduce the number of days over which hypoxic conditions persist. In addition, prioritising outcomes for Murray cod at locations not impacted by blackwater would be an effective use of environmental water.

1   Introduction

1.1   Context

The Long-Term Intervention Monitoring (LTIM) Project aims to understand the impacts of Commonwealth environmental water in the Murray-Darling Basin (the Basin) and seeks to improve capacity to support flow allocation decisions. It does this through the monitoring and evaluation of the response of six ecological indicators to managed flows in the Basin. While this document focuses on outcomes for fish, the other Basin Matters are: hydrology, vegetation diversity, stream metabolism, ecosystem diversity and biodiversity. These Basin Matter indicators are being monitored across seven LTIM 'Selected Areas' throughout the Basin. Fish are a target in all but one of these Selected Areas (Junction of the Warrego and Darling rivers), and so this document commonly refers to fish data collected across six Selected Areas. The Selected Areas are monitoring sites which can be used to infer outcomes of environmental watering more broadly across the Basin.

Fish are an important component of the Basin and can also provide a valuable indicator for understanding flow response. Evaluation of native fish diversity, condition, reproduction and recruitment contribute to understanding the benefit of environmental water and to the biodiversity outcomes sought by the Murray–Darling Basin Plan (Commonwealth of Australia, Basin Plan 2012). Fish have substantial socioeconomic value and often play important roles in food web and ecosystem processes (Holmlund & Hammer 1999). Here we present the 2016–17 Basin Matter report for fish monitoring and evaluation within the LTIM Project. This is the third of five Fish Basin Matter reports to be delivered within the project. Further details on methods are provided in Stoffels & Bond (2016).

1.2   Objectives

1.2.1   Overarching objectives of the Fish Basin Matter

LTIM evaluation questions can be divided into those that concern short- and long-term outcomes to flows. These short- and long-term evaluation questions reflect the fact that certain ecological variables can respond rapidly to environmental change, while others are slower to respond (Levin 2000). The LTIM evaluation questions for fish are:

- long-term
    - What did Commonwealth environmental water contribute to sustaining native fish populations?
- short-term
    - What did Commonwealth environmental water contribute to sustaining native fish reproduction?
    - What did Commonwealth environmental water contribute to sustaining native fish survival?

A key objective of LTIM is to improve our capacity to predict ecological response to flow events and regimes (Gawne *et al.* 2013, 2014).

1.2.2   Specific objectives for 2016–17 evaluation

The general objectives for the 2016–17 LTIM Fish Basin Matter report are to present analyses of the impacts of Commonwealth environmental water and, more broadly, river flows, on fish responses. This is undertaken in part by developing predictive models informed by observed responses to all sources of flow variability, and then using that capability to make inferences about the contributed effects of environmental watering using flow scenarios that include/exclude Commonwealth environmental water. The motivation for this work is the long-term goal of providing scientifically

defensible reporting of the outcomes of Basin Watering Strategy. Developing predictive models facilitates the separation of the effects of Commonwealth watering actions from the effects of the background (non-environmental water) hydrological variability. Selected Area reports present several analyses of localised fish responses to natural and managed flows. Our Basin Matter report complements and builds upon those reports by (see Appendix A for more details on data collection and model development):

1. **Review technical reports for short-term (1 year or less) outcomes from Commonwealth environmental watering actions during 2016–17 and present a brief synthesis of those outcomes**. This brief synthesis is structured by three groups of expected outcomes of Commonwealth environmental water in 2016–17: reproduction, movement and ameliorating hypoxia.

2. **Undertaking a Basin-scale analysis of the response of fish *condition* to flows.** This work provides an evaluation of how environmental water delivery within and across years has affected fish condition, which provides a simple metric reflective of the overall health of individual fish. The specific objectives of this component were to:
   a. Develop statistical models using data collected from Selected Areas to determine how river flows affect condition of three target fishes: bony herring (*Nematalosa erebi*), golden perch (*Macquaria ambigua*) and Murray cod (*Maccullochella peelii*), which were among the only large-bodied species caught at most sites. We designed the models to answer the following questions:
       i. Does the timing (season) of flow delivery affect fish condition?
       ii. Does the response of fish condition in a given year depend on flows delivered in years previous?
       iii. Does the response of fish condition to flows depend on the relative density of the species in the river?
   b. Use these models to estimate how environmental water delivery has contributed to fish condition.
   c. Identify possible options for changes in managed flow delivery that could improve fish condition.

3. **Undertake a Basin-scale analysis of the impact of the 2016 flooding events in the southern Basin, and the associated environmental hypoxia**. The specific objectives of this component were:
   a. Determine how the 2016 Basin-scale flooding event affect the composition of the Basin's fish community – which species were most affected?
   b. Focusing on the species most affected, draw upon data from multiple Selected Areas to parameterise statistical models that enable us to answer the following questions:
       i. To what extent were changes in abundance due to flooding and hypoxia? We aim to separate the effects of floods and hypoxia to isolate the relative impact of these two major drivers of change that unfolded during spring-summer 2016.
       ii. Precisely how does the magnitude of environmental hypoxia (number of days within a river segment where minimum daily DO drops below 4 mg L^{-1}) affect the magnitude of change in the abundance of fishes?
   c. Evaluate the water delivery decisions and discuss challenges of managing the risks that hypoxic blackwater poses to Basin Plan objectives.

It was originally planned that the 2018 analysis would also include models of fish spawning (Stoffels *et al.* 2016). Data collection and storage in the LTIM database has been hampered by a range of issues related to both the implementation of different methods in several locations as well as data quality assurance and control. This has prevented an analysis and development of the spawning model in this third LTIM year. Larval fish data issues have been resolved and it is anticipated that development of spawning models will be possible in the subsequent LTIM fish reports.

2 Environmental context

2.1 Summary

Hydrology, dissolved oxygen and river metabolism

- While hydrology during the first two years of LTIM was characterised by below average discharge, the third year of LTIM 2016–17) was characterised by significant natural flooding in the southern Basin.
- Large areas of the inundated floodplain in the Murray and Murrumbidgee catchments had not been flooded prior to the millennium drought, resulting in the liberation of very large quantities of accumulated terrestrial carbon as dissolved organic carbon (DOC). This DOC provided an accessible carbon source for microorgansims and microinvertebrates.
- As a result of this large increase in ecosystem respiration, the 2016 flood resulted in hypoxic flows in the Lachlan and Edward-Wakool (severe hypoxia), and the Goulburn and Lower Murray (mild hypoxia).

Water actions targeting fish outcomes during 2016–17

- During 2016–17 a total of 58 Commonwealth environmental water actions were delivered throughout the Basin, with expected outcomes for fish. While these covered a range of objectives the most common were fish habitat, movement and spawning. There were also a small number of watering actions with primary expected outcomes aimed at ameliorating the effects of hypoxia on fish.
- The majority of watering actions with primary expected outcomes for fish were delivered as freshes (including weir pool raising and lowering in the Lower Murray) with a smaller number of base flows delivered mainly for fish habitat and condition, and contributions to several bankfull flows aimed mostly at fish movement.
- Commonwealth environmental water contributed to fish spawning in several rivers across the Basin in 2016–17, with strong evidence of supporting spawning and recruitment in Gunbower Creek and the Lower Darling River.
- Commonwealth environmental water was also successful in facilitating fish movement in a number of river systems including between the Murray and Goulburn Rivers, the Barwon and Macquarie Rivers and the Lower Darling and Murray Rivers.
- The effectiveness of Commonwealth environmental water in ameliorating hypoxic conditions was varied across Selected Areas where this was monitored.

2.2 Hydrology, dissolved oxygen and river metabolism

Environmental conditions of the Basin during 2016–17 contrasted very strongly with those of the first two years of LTIM (2014–15 and 2015–16). While the first two years of LTIM were characterised by lower than average rainfall and low river discharges, a major flooding event that spanned much of the Basin was a salient feature of 2016. Flood magnitudes were highest in the southern catchments of the Basin; the Murrumbidgee, Edward-Wakool, Lachlan, Murray, and Goulburn Rivers (Figure 1). The Gwydir River, in the northern Basin, also experienced a relatively short natural flood, when peak discharge was well above the average (Figure 1c).

Dissolved oxygen (DO) and temperature data were recorded under the LTIM Project with continuous loggers for the river metabolism monitoring within the Murrumbidgee, Goulburn, Lower Murray, and Edward-Wakool Selected Areas. Some of these loggers were affected by flooding resulting in gaps in measurements. Data availability for the Lachlan Selected Area in particular is sparse, limited to one river metabolism monitoring site that operated through the flood event, and with no permanent continuous DO data at any stream gauge for this Selected Area. Looking across sites, decreases in the mean daily DO levels (red dots in Figure 2) demonstrate that hypoxia was widespread throughout the Basin during the spring-summer of 2016. However, the magnitude of the environmental hypoxia event varied among Selected Areas. In certain areas, such as the Lachlan, decreases in DO resulted in high magnitudes of hypoxic flows, with DO being reduced to below 4 mg L^{-1} for months (Figure 2e). In other sites such as the Murrumbidgee River system, the timing of the gaps in the available DO data limit our ability to accurately interpret the impact that flooding had on DO levels, hence the occurrence of hypoxic conditions associated with the flood event (Figure

2a**)**. These data issues will be addressed over time as more data become available but must be acknowledged as a potential source of uncertainty and error in the models.
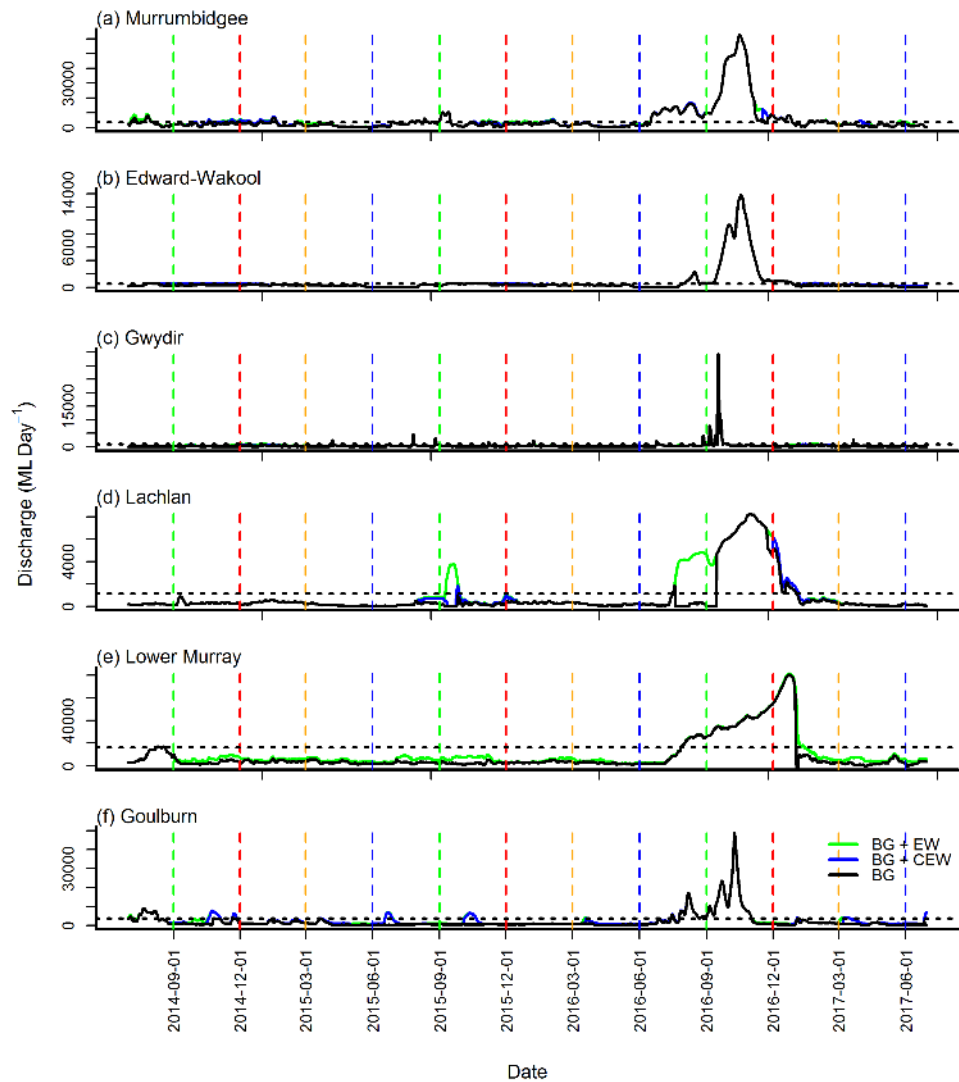
Figure 1. Hydrological dynamics of the fish focal zones within the six LTIM Selected Areas with monitoring programs targeting fish. Discharge within any given day has been decomposed into background flows (BG; black), background flows plus Commonwealth environmental water (BG + CEW; blue) and background flows plus all environmental water (BG + EW; green). In the Lower Murray, the contribution of Commonwealth environmental water has been absorbed into EW, due to difficulties isolating the Commonwealth environmental water component. Horizontal dashed lines indicate mean discharge at the gauge used for that area during the period. Extent of y-axis differs between plots.
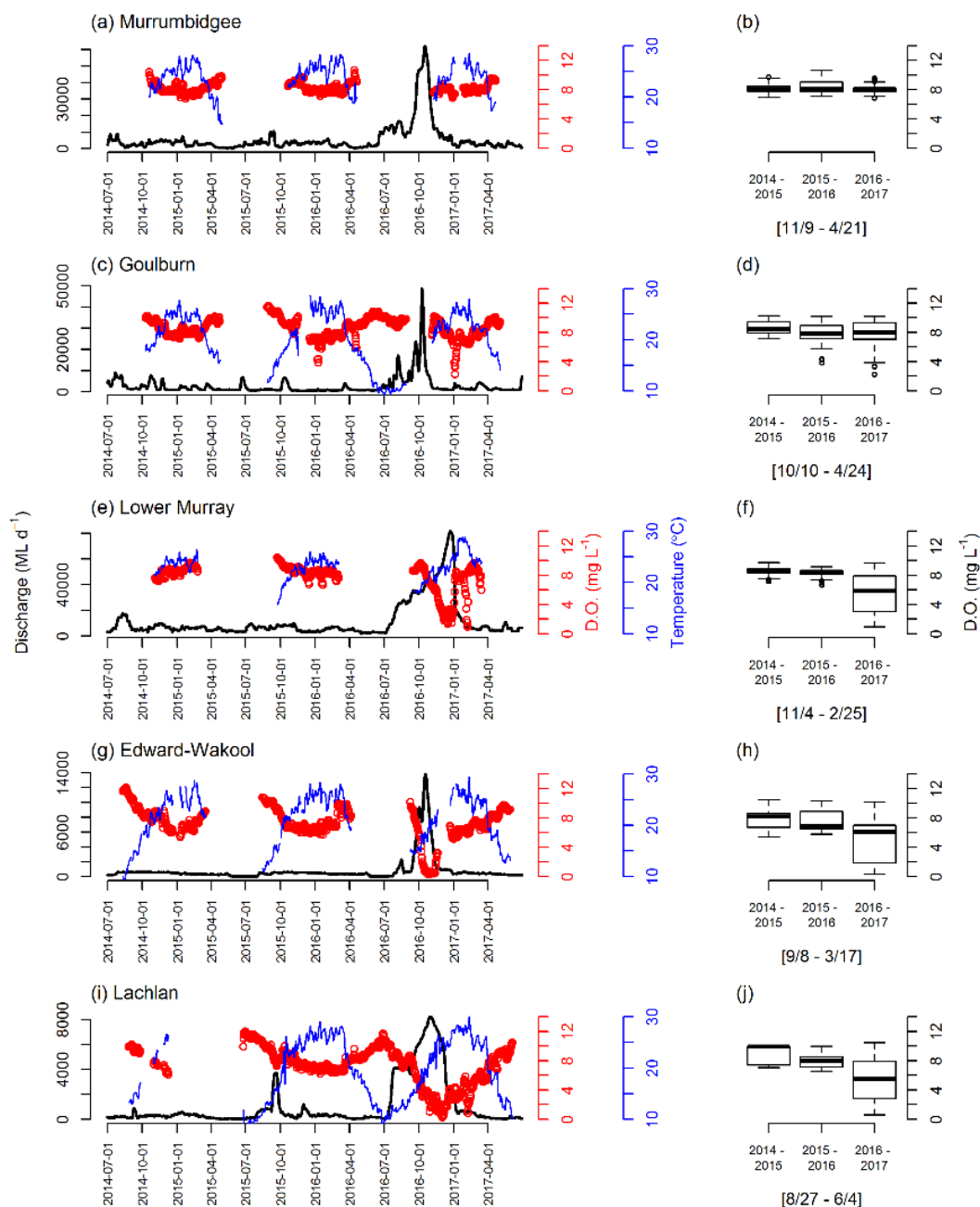
Figure 2. Time series of mean daily discharge (black), mean daily dissolved oxygen (red) and mean daily water temperature (blue) within each of the five study rivers over the three years of LTIM.

2.3   Water actions targeting fish outcomes, 2016–17

During 2016–17 a total of 58 Commonwealth environmental water actions were delivered throughout the Basin, with expected outcomes for fish. Together, these water actions comprised about 1 658 745 ML of Commonwealth environmental water[1]. Of these actions, over half targeted fish habitat and / or movement, and one third targeting spawning and / or recruitment noting that most watering actions have multiple expected outcomes (Figure 3).

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[1] Note that this figure includes watering actions where water was reused and so accounted for more than once. Many watering actions also included other sources of environmental water.

There were 38 Commonwealth environmental watering actions delivered in 2016–17 with primary expected outcomes for fish. Of these, the most common expected outcomes related to movement, dispersal or connectivity for native fish, followed by habitat and recruitment. There were four watering actions (three in the Edward Wakool and one on the Murrumbidgee) that were specifically aimed at ameliorating low dissolved oxygen for fish (Figure 3).

In terms of the flow component for each fish primary expected outcome, the vast majority were delivered as freshes (noting that this figure includes eight watering actions that were weir pool raising and lowering and not true freshes) (Figure 4). Base flows were delivered across several rivers including the Goulburn, Broken and Namoi rivers for improved condition, movement and habitat. Commonwealth environmental water contributed to bankfull flows in the Border Rivers, Lower Balonne and Murrumbidgee with primary fish expected outcomes for movement.
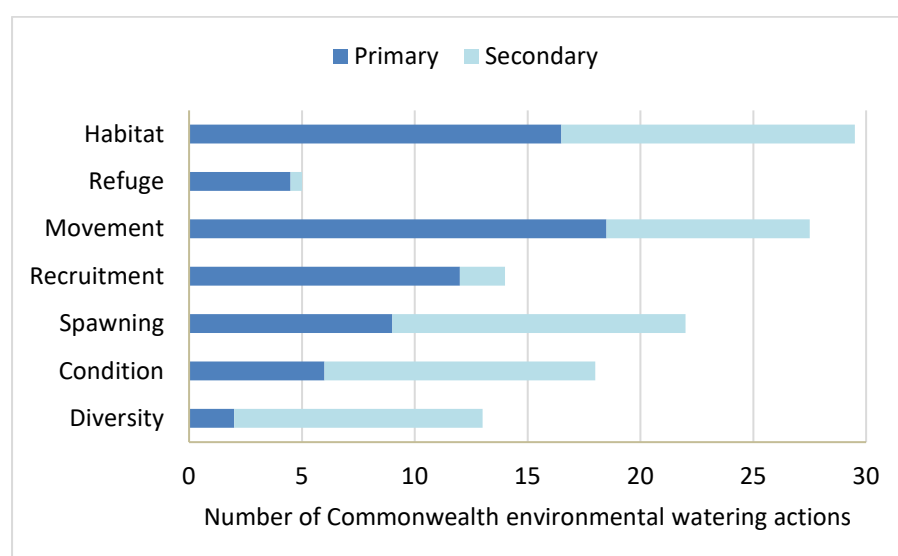
Figure 3. Commonwealth environmental watering actions in 2016–17 with expected outcomes for fish.
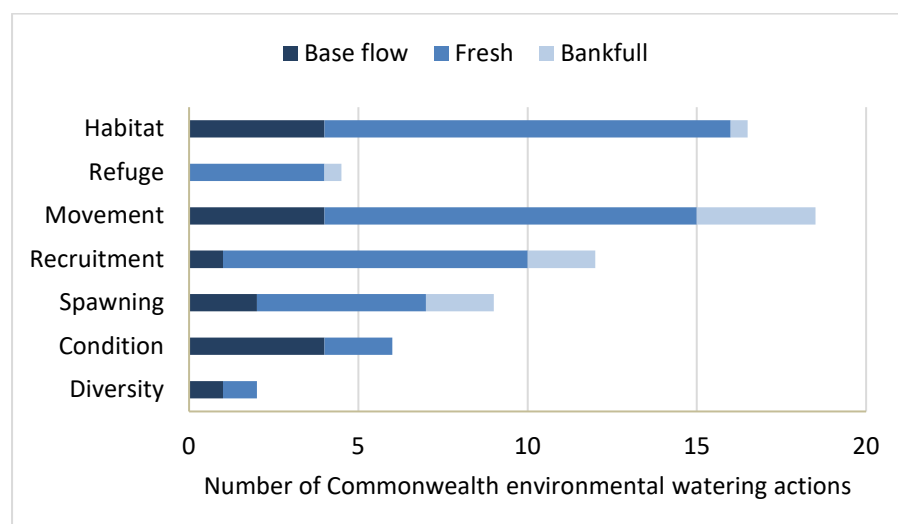
Figure 4. Flow components of Commonwealth environmental watering actions in 2016–17 with primary expected outcomes for fish.

Of the 58 watering actions that were delivered with expected outcomes for fish, 23 were monitored as part of LTIM and other CEWO commissioned monitoring programs (Table 1). This monitoring included assessments of reproduction (spawning and recruitment), movement and the effects of hypoxia.

Table 1. Summary of observations and other information from monitored watering actions with expected outcomes for fish in 2016–17. Note that many of these actions involved multiple water sources (in addition to Commonwealth environmental water). Additional information on the portfolio of environmental water can be found in the Basin Matter Hydrology report (Stewardson & Guarino 2018). Also note, that this table does not include monitoring of fish in LTIM Selected Areas that was not directly linked to a Commonwealth environmental watering action.

Surface water region/asset	Commonwealth environmental water volume (ML)[1]	Dates[1]	Flow component	Expected ecological outcome[1]	Observed ecological outcome	Source of information
Border Rivers – Severn River	823.53	01/07/16 - 30/06/17	Bankfull	Support movement/migration of large bodied fish including Murray cod, golden and silver perch, eel-tailed catfish.	Five native species of fish recorded, including three threatened species: eel-tailed catfish, Murray cod and silver perch. Flows considered too low to adequately inundate Murray cod spawning habitat.	NSW Department of Primary Industries & Queensland Department of Agriculture and Fisheries (2017).
Border Rivers - Dumaresq-Macintyre River and fringing wetlands	914.3 14376.8 6492.1	07/07/16 - 13/07/16	Fresh, Bankfull, Fresh	Support dispersal, reproduction and recruitment of native including Murray cod and eel-tailed catfish.	Ten native species of fish recorded including four threatened species: eel-tailed catfish, Murray cod, purple-spotted gudgeon and olive perchlet. Some evidence of spawning and recruitment of carp gudgeon and Murray cod.	
Central Murray – Gunbower Creek	23563	01/07/16 - 30/06/17	Base flow	Maintain the diversity and condition of small and large-bodied native fish populations in Gunbower Creek through the provision of habitat and opportunities for breeding and recruitment.	There was evidence of recruitment in five native species: Australian smelt, carp gudgeon, Murray cod, Murray-Darling rainbow fish and unspecked hardy-head. The average abundance of Murray cod and silver perch in Gunbower Creek in 2016 was higher than in any previous year. There are now juvenile and sub-adult size classes of Murray cod clearly represented in Gunbower Creek.	Bloink & Robinson (2016)
Macquarie – Lower Macquarie River	27583	16/04/17 - 15/05/17	Fresh	Connect the mid-Macquarie River to the Barwon River via the lower Macquarie Marshes and lower Macquarie River, to facilitate native fish movement between the river systems. Provide opportunities for juvenile native fish such as golden perch, silver perch, and spangled perch to emigrate into the Macquarie catchment.	Golden perch juveniles were detected in small numbers in the lower Macquarie River prior to the water delivery and increased in numbers following the full connection with the Barwon River. There was evidence of juvenile golden perch and other channel specialists such as spangled perch migrating in an upstream direction. In stream barriers are likely impacting on the movement of fish.	Davis *et al.* (2017)
EdwardWakool: Wakool River	29306.63	31/10/16 - 31/12/16	Fresh	To provide hypoxic water refuge for fish and other aquatic biota.	Only a small proportion of the tagged sample of fish survived the hypoxic blackwater event. Timing of	Watts *et al.* (2017)

Surface water region/asset	Commonwealth environmental water volume (ML)[1]	Dates[1]	Flow component	Expected ecological outcome[1]	Observed ecological outcome	Source of information
Edward Wakool: Edward River	74822.7	24/10/16 - 08/12/16	Fresh		flow delivery was generally too late to protect the majority of individuals.	
Edward Wakool: Colligen-Neimur	3240.67	17/10/16 - 16/12/17	Fresh			
Edward Wakool: Yallakool Creek	27581	01/01/17 - 30/03/17	Fresh	To influence different changes in river rises that may contribute to silver perch spawning. To provide a number of peaks at a time when water temperature is suitable for silver perch spawning. To determine if river level rises timed with periods of 23°C water temperature and no moon (dark night sky conditions) has any influence on silver perch spawning response.	No evidence of golden perch or silver perch spawning was found during the sampling period. No eggs or larvae of golden perch or silver perch were detected with targeted sampling from Sept to Dec 2016. Monitoring, however, occurred prior to the delivery of environmental water and so could not be assessed. It is likely that the hypoxic conditions that occurred in November 2016 affected spawning and recruitment.	Watts *et al.* (2017)
Goulburn - Lower Goulburn River	9250	01/07/16 - 05/08/16	Base flow	Provide suitable habitat to support native fish survival and condition (through submergence of snags, increasing availability of slack and deep water habitat and encouraging aquatic vegetation and planktonic production for food).	Recommended base flows provided for adults. It is not possible to associate fish population makeup or diversity to the provision of base flows at this stage. The provision of base flows and freshes in the 2016–17 season contributed to variation in the type and distribution of hydraulic habitat known to be of value to fish.	Webb *et al.* (2017)
Goulburn - Lower Goulburn River	8200	02/11/16 - 09/01/17	Base flow			
Goulburn - Lower Goulburn River	64290	01/03/17 - 03/04/17	Fresh	Promote fish migration	Attraction flows saw the movement of some tagged sub-adult silver perch into the lower Goulburn River. Silver perch tagged in the Murray River moved into the Goulburn River in March-April 2017 coinciding with a within-channel environmental flow fresh in the Goulburn River	Webb *et al.* (2017)
Goulburn - Lower Goulburn River	39585	04/04/17 - 25/06/17	Base flow			
Lower Darling-Lower Darling River	71248.6	02/10/16 - 08/01/17 24/04/17 - 30/06/17	Fresh, Base flow	Support Murray cod spawning, recruitment and larval dispersal in the Lower Darling main channel. Support golden and silver perch spawning and dispersal.	Strong spawning results observed for Murray cod, with 885 Murray cod larvae collected and follow-up monitoring indicated good recruitment. Also spawning of silver perch and golden perch. Recruitment and large-scale (500 – 1000 km) dispersal of golden perch from nursery grounds in the Menindee Lakes to the Lower Darling River.	Sharpe & Stuart (2018)

Surface water region/asset	Commonwealth environmental water volume (ML)[1]	Dates[1]	Flow component	Expected ecological outcome[1]	Observed ecological outcome	Source of information
Lower Murray - Berri Evaporation Basin	707	01/01/17 - 30/06/17	Wetland	Support nationally threatened Murray hardyhead populations	No Murray hardyhead were caught in monitoring; but individuals were observed. Likely that the species dispersed due to the extensive flood event.	CEWO acquittal report (unpublished)
Lower Murray - Rufus River	29570	17/12/16 - 01/01/17	Fresh	Maintaining current species diversity, extending distributions and improving breeding success and numbers of short, moderate and long-lived native fish species by: Managing water quality risks to vulnerable populations and species; Providing in-stream habitat for fish; Increasing the presence of fast flowing fish habitat along the River Murray.	Natural flooding reduced dissolved oxygen levels to below 50% saturation (~4.5 mg / L). Commonwealth environmental water that supplemented releases from Lake Victoria maintained oxygen levels above 4 mg / L in the Rufus River, which potentially provided refuge areas for aquatic organisms.	Ye *et al.* (2017)
Lower Murray - Lock 9	0	15/07/16 - 30/12/16	Fresh	Maintaining current species diversity, extending distributions and improving breeding success and numbers of short, moderate and long-lived native fish species by: • Increasing the presence of fast flowing fish habitat along the River Murray and, where feasible, increased lateral connectivity with anabranches and low elevation floodplain wetlands. • Providing in-stream habitat for fish and thereby supporting recruitment of fish (including golden and silver perch spawned in spring 2015), particularly by increasing the availability of food resources and habitat during periods where flows would be unnaturally low. • Improving the body condition of mature fish during winter/spring ('pre-spawning conditioning') and providing opportunities for spawning during spring (subject to appropriate seasonal conditions).	Commonwealth environmental water did not contribute to the presence of any new cohorts of golden perch (age 0+, 1+) in the Lower Murray River, despite spawning during spring/ summer 2016. It is possible that hypoxic blackwater from natural flooding impacted directly on egg and larval development, or indirectly via the effect of reduced food resources. Commonwealth environmental water did not contribute to the presence of any new cohorts of silver perch (age 0+, 1+) in the Lower Murray River. No silver perch spawning was detected.	Ye *et al.* (2017)
Lower Murray - Lock 9	0	30/04/17 - 30/06/17	Fresh			
Murrumbidgee - Murrumbidgee River	150978	28/10/16 - 05/01/17	Fresh, bankfull	Provide in-channel refuge habitat and in-channel movement opportunities for native fish from areas of low dissolved oxygen levels in the lower reaches to refuge habitat	Commonwealth environmental water contributed to improving water quality and ameliorating the effects of hypoxia in the Murrumbidgee. Dissolved oxygen concentrations recovered to > 4 mg/L five	Wassens *et al.* (2017)

Surface water region/asset	Commonwealth environmental water volume (ML)[1]	Dates[1]	Flow component	Expected ecological outcome[1]	Observed ecological outcome	Source of information
				upstream Support the movement of native fish and other aquatic animals from the floodplain to the river channel, thereby minimising or preventing stranding of these animals on the floodplain	days sooner with environmental water than without.	
Murrumbidgee - Eulimbah	2320	28/11/16 - 03/03/17	Wetland	Supporting the habitat requirements of native fish and other aquatic animals.	These actions were successful in prolonging the period of fish occupation in target wetlands allowing the development of more mature fish and repeated opportunities for spawning. We note, however, that overall wetland fish communities remain in poor condition and are dominated by highly abundant opportunistic generalist species with floodplain specialist species, those that still survive in parts of the Murrumbidgee Catchment (such as Murray hardyhead), generally absent from wetland data.	Wassens *et al.* 2017
Murrumbidgee - Telephone Bank	5425	24/11/16 - 20/03/17	Wetland			
Murrumbidgee - Yanga NP	2155	29/10/16 - 13/02/17	Wetland			
Murrumbidgee - Nap Nap	630	03/01/17 - 07/01/17	Wetland			
Warrego: Lower Warrego River and fringing wetlands.	5865	23/09/16 - 10/10/16	Bankfull	Support fish migration and spawning opportunities especially large bodied species including golden perch.	Evidence of breeding in bony herring, spangled perch and Murray-Darling rainbowfish in response to delivered flows. Also, evidence of improved population structure in Hyrtl's catfish. Commonwealth environmental water contributed to connectivity through the Warrego channel for 20 days. Abundance and richness declined as waterholes dried.	Southwell *et al.* (2017a)
	7762.5	08/10/16 - 28/10/16	Fresh	Provide opportunities for native fish (e.g. fish spawning, recruitment and movement) (Warrego River).		

Reproduction

Monitoring of fish in 2016–17 provided some evidence of Commonwealth environmental water benefiting native fish either by stimulating spawning or providing conditions to improve survival of larval fish. Fish spawning and / or recruitment was reported coinciding with the delivery of environmental water in the Dumaresq-Macintyre River (NSW DPI & Qld Department of Agriculture and Fisheries 2017) and the Lower Warrego River (Southwell *et al.* 2017a). CEWO commissioned monitoring (outside of LTIM) provided strong evidence linking Commonwealth environmental water to native fish reproductive outcomes in two locations:

- Gunbower Forest – water was delivered in 2015–16 and 2016–17 as part of a three year Environmental Water Agreement with the Commonwealth Environmental Water Office (CEWO) to provide the fish hydrograph from 2015–2018 in Gunbower Creek. Prior to the implementation of environmental water in Gunbower Creek, the system dried to a series of residual pools in the off-irrigation season. This was recognised as having a deleterious effect on fish recruitment and survival, with no Murray cod in size classes that represent fish less than three years of age (Sharpe *et al.* 2014). Following the implementation of Commonwealth environmental watering there was evidence of recruitment in five native species: Australian smelt, carp gudgeon, Murray cod, Murray-Darling rainbow fish and unspecked hardy-head There are now juvenile and sub-adult size classes of Murray cod clearly represented in Gunbower Creek (Bloink & Robinson 2016).
- Lower Darling River – Commonwealth environmental water was delivered to the lower Darling between October and December 2016 to support Murray cod breeding ecology, supporting courtship, nest selection, spawning and nest retention by avoiding rapid drops and rises in river levels throughout the breeding period. Monitoring across a 500 km stretch of the river from the Menindee Lakes to Wentworth indicated success of the environmental watering action with strong spawning responses from Murray cod (Sharpe & Stuart 2018).

Movement

In 2016–17 supporting movement and dispersal of native fish was the primary expected outcome of around one third of the Commonwealth environmental watering actions targeting fish (19 of 58 watering actions). There are several examples that demonstrate the effect of Commonwealth environmental water on biological connectivity in 2016–17:

- Movement of eel-tailed catfish and Murray cod in response to natural flows and environmental water in the Gwydir and Mehi Rivers (Southwell *et al.* 2017b).
- Increases in diversity and abundance of native fish in the connected waterholes of the Lower Warrego River in response to Commonwealth environmental water (Southwell *et al.* 2017a).
- Positive effects of "attraction flows" provided by environmental water in the Goulburn River system. Silver perch tagged in the Murray River moved into the Goulburn River in March-April 2017 coinciding with a within-channel environmental flow fresh in the Goulburn River (Webb *et al.* 2017).
- Managed environmental water in autumn 2017 connected the Lower Macquarie to the Barwon River facilitating the upstream movement of golden perch and spangled perch. Monitoring indicated that although the managed flow resulted in movement of native fish, in-channel infrastructure acts as barriers to migration in the lower Macquarie River downstream of Marebone Weir, limiting the benefits to native fish populations (Davis *et al.* 2017).
- In spring and summer of 2016–17 environmental water, including Commonwealth environmental water, was delivered to complete the nursery function of the Menindee Lakes by dispersing early juvenile golden perch into the lower Darling River, Great Darling Anabranch and Murray River populations. Golden perch spawned in the Border Rivers in

October 2016, moving downstream into nursery grounds in the Menindee Lakes in late spring. Environmental flows then enabled dispersal of the young golden perch into the lower Darling River and Great Darling anabranch where they made up 50% of the population. It is expected that these young golden perch would then have dispersed into the Murray River demonstrating the effectiveness of environmental water outcomes over large scales and multiple river systems (Sharpe & Stuart 2018).

Ameliorating hypoxia

Monitoring in the Selected Areas reported variable success of Commonwealth environmental water in ameliorating the effects of hypoxic blackwater events and increasing dissolved oxygen. In the Edward-Wakool, the Selected Area report concluded that the three environmental watering actions aimed at providing hypoxic water refuges for fish were unsuccessful, with only a small portion of large bodied native fish that had been tagged surviving the blackwater event. It was also suggested that the hypoxic conditions that occurred in November 2016 affected spawning and recruitment. They concluded that the timing of the flow delivery was too late to protect the majority of individuals (Watts *et al.* 2017). In the Lower Murray, it was suggested that hypoxic blackwater from natural flooding impacted directly on golden perch egg and larval development, or indirectly via the effect of reduced food resources. In the Rufus River natural flooding reduced dissolved oxygen levels to below 50% saturation (~4.5 mg / L). Commonwealth environmental water that supplemented releases from Lake Victoria maintained oxygen levels above 4 mg / L in the Rufus River, which potentially provided refuge areas for aquatic organisms (Yen *et al.* 2013). Commonwealth environmental water also contributed to improving water quality and ameliorating the effects of hypoxia in the Murrumbidgee. Dissolved oxygen concentrations recovered to > 4 mg/L five days sooner with environmental water than without (Wassens *et al.* 2017).

The effect of hypoxia on fish communities in the Basin was also explored using community dynamics models (see section 4).

3 Population dynamics

3.1 Summary

Effects of hydrology and other environmental variables

Models relating golden perch and Murray cod condition to patterns of hydrologic variability indicated that the impact of flows on body condition is season-specific;

- High spring (in particular) and summer flows were associated with positive trends in fish condition, as measured at the end of the flow delivery season (autumn).
- High autumn flows were associated with minor increases in the golden perch condition, but also appeared to coincide with reduced Murray cod condition. At present there is no explanation for this association, and further data are required to establish the veracity of this finding.
- High winter flows have subtle, but significant, positive effects on golden perch condition but no significant effects on Murray cod condition.
- Golden perch condition was positively related to both seasonal patterns of flow during a flow-delivery year, as well as the observed discharge in the previous water year.
- Thus, appropriately timed higher flows across multiple years may have cumulative benefits for golden perch condition.
- Body condition is also negatively correlated with the abundance of conspecifics, which suggests populations of golden perch and Murray cod may be food or habitat limited, with body condition decreasing as a result of competition.

Impacts of environmental water

With respect to Murray cod:

- Commonwealth environmental water delivery during 2014–15 increased Murray cod condition in the Edward-Wakool and in the Gwydir.
- Environmental water delivered during 2014–15 in the Gwydir, additional to Commonwealth environmental water, increased Murray cod condition.
- There was delivery of Commonwealth environmental water within the Goulburn during autumn 2014–15, which from the current data may be predicted to have decreased Murray cod condition. At present there is no clear explanation for this association, and further data are required to establish the potential cause. One possibility is higher autumn flows leading to lower water temperatures, which may affect fish condition.

With respect to golden perch:

- Environmental water, but not Commonwealth environmental water alone, delivered during 2014–15 and 2015–16 increased golden perch condition within the Goulburn and Murrumbidgee;
- Environmental water delivery during 2014–15 and 2015–16 increased golden perch condition within the Lachlan and Lower Murray (noting that the Commonwealth environmental water was not differentiated from all environmental water in both those systems for 2015–16).
- Both above inferences concerning environmental water impacts inherently incorporate effects of environmental water delivery during both 2014–15 and 2015–16, because the best fitting statistical model of golden perch condition incorporates 2-year flow impacts on condition.

Adaptive management

- Flows delivered in spring and summer have the greatest positive impact on golden perch and Murray cod condition. Spring-summer flow delivery may result in synergistic impacts on fish populations—promoting spawning while at the same time increasing energy acquisition/storage, which in turn may feed into enhancing other population processes in subsequent years.
- At this stage, the models indicate that autumn flow pulses may decrease Murray cod condition, although no mechanism has yet been identified, and this observation requires further investigation, but may have implications for environmental flow releases delivered for other purposes, as well as for autumn irrigation flows.
- Planning of flow delivery each year should give thought to flows delivered in the previous year, which may affect the condition response of golden perch. In particular, priority should be given to delivering water actions for golden perch in the year immediately following a high flow year.
- It is important to note that when impacts of watering actions are additive across years:
  - observable impacts will take years;
  - there may be greater ecological returns in the long-term, without necessarily allocating more water;
  - a great challenge will be the optimisation of multi-year flow regimes within catchments.
- Commonwealth, state and other environmental water use has synergistic effects on fish condition, so should not be managed in isolation.

3.2   Recruitment and survival

3.2.1   Effects of hydrology and other environmental variables

Variation in the rates of survival from one age class to another (e.g. from year 0 to year 1) within a population is ultimately an important determinant of population trends. However, estimating survival rates in natural populations is challenging because it is difficult to track the fate of individual organisms. However, changes in survival rates from one year to the next can be estimated by examining changes in the age structure from one year to the next. Years with higher survival will tend to have a relative increase in the number of individuals reaching the next age class and vice versa. This variation can then be linked to hydrologic variables using statistical models, to identify the influence that flow variability (including that from environmental watering actions) has on recruitment and survival. This section reports on the preliminary efforts to infer the influence of CEW on recruitment and survival rates.

Initial analysis of the age structure of golden perch, bony herring and Murray Cod demonstrated that given the uncertainty in the current data (representing just three survey years), recruitment and survival could be explored for only bony herring and Murray cod. At the Basin-scale, across multiple Selected Areas, bony herring recruitment (measured as the abundance of the young-of-year cohort in autumn) —varies significantly through years (Figure 5). At this stage, however, there are no obvious systematic patterns in bony herring recruitment across Selected Areas. Likewise, for bony herring survival, with the exception of the Lower Murray, Basin-scale patterns are not obvious given the data currently available. Within the Lower Murray there was a weak trend for reduced survival of bony herring from 2016 to 2017, across all age-classes (Figure 5). The causes of this are uncertain, however this may be due to a negative effect of the 2016 flood, and/or the associated environmental hypoxia that followed.
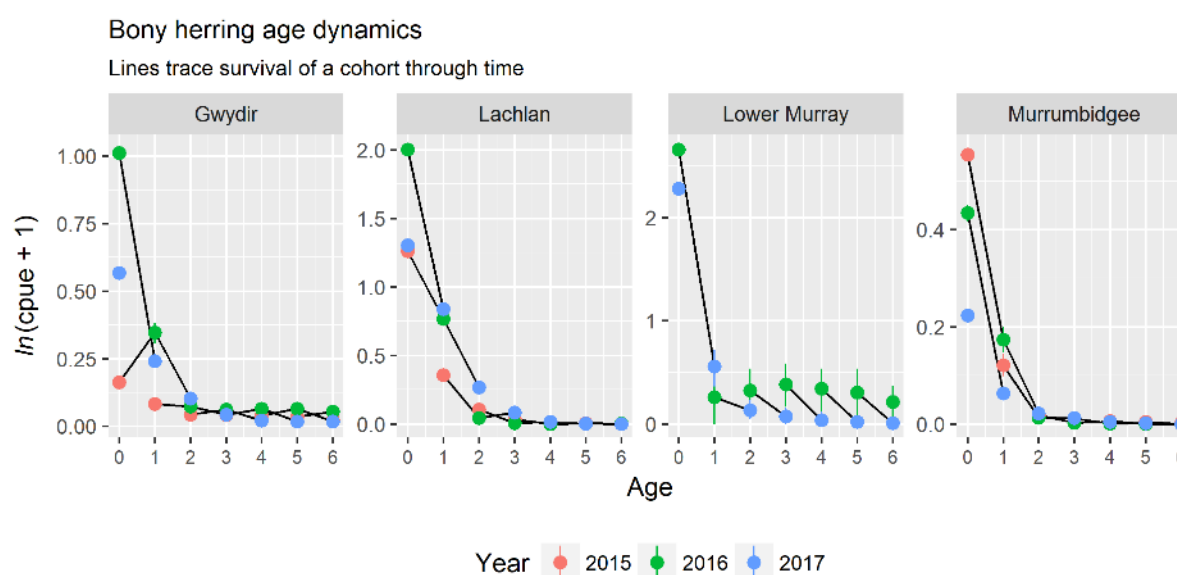
Figure 5. Changes in the abundance of age-classes of bony herring within Selected Areas, across the first three years of LTIM. Mean abundance +/- 95% confidence intervals are presented, where the confidence intervals present uncertainty in the mapping of lengths to age. Bony herring were not a target species of the Lower Murray during 2014–15.

Murray cod recruitment showed strong and consistent patterns at the Basin-scale with the most obvious trend being the negative effect of the 2016 floods on recruitment (Figure 6). In the Edward-Wakool and Lachlan Selected Areas, recruitment in 2016–17 was particularly poor where environmental hypoxia was recorded as extreme (see section 4 on Community dynamics). These

results are concordant with the hypothesis that Murray cod are a low-flow recruiter (Humphries *et al.* 1999; Humphries 2005), or not influenced by high flows in terms of recruitment.
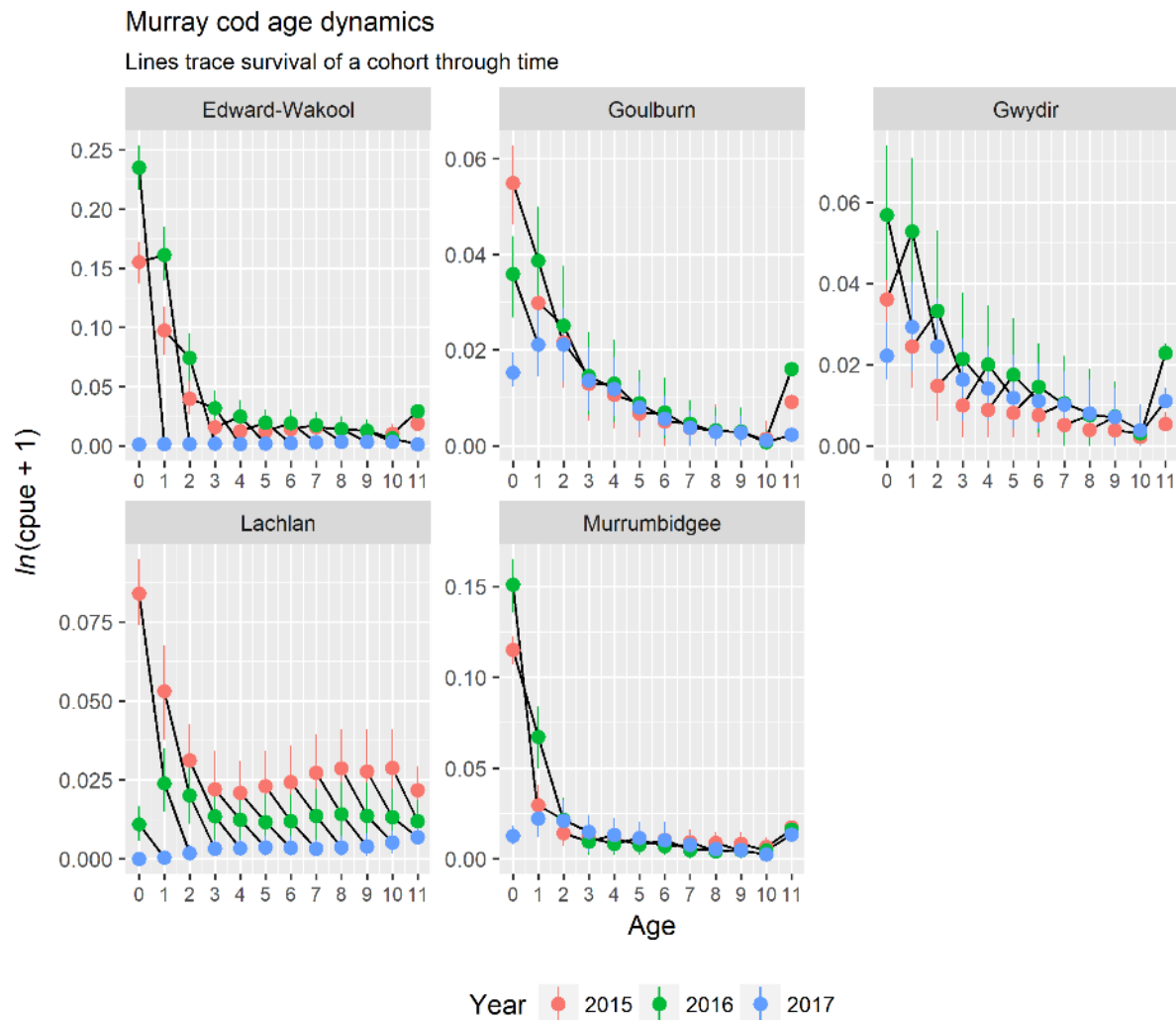
Figure 6. Changes in the abundance of age-classes of Murray cod within Selected Areas, across the first three years of LTIM. Mean abundance +/- 95% confidence intervals are presented, where the confidence intervals present uncertainty in the mapping of lengths to age. Insufficient data for the Lower Murray.

3.2.2 Effects of environmental water and adaptive management

Although good progress is being made towards being able to provide scientifically defensible statements concerning the impacts of environmental watering, at present the models demonstrate that large hypoxic floods negatively affect rates of survival, which is consistent with the widely observed fish kills that occurred in 2016 where flood waters were hypoxic. Note, that the observed mortality is most likely caused by hypoxia, rather than by floods. However, flooding is the primary cause of large-scale hypoxic blackwater events in the basin. The 2016 event represents an extreme case of how flow variability can affect survival, and the detection of more subtle changes in rates of survival such as those that are expected to occur from environmental watering are likely to take significantly longer to be detectable given underlying uncertainty in the data associated with sampling uncertainty. We discuss the significance of the observed effects of hypoxic conditions for adaptive management in Section 4 (Community dynamics).

3.3 Mean individual condition

3.3.1 Effects of hydrology and other environmental variables

Condition of golden perch and Murray cod was found to vary among Selected Areas and years (Figure 7). Golden perch of the Edward-Wakool and Goulburn generally exhibit average to above-average condition during the period, with other Selected Areas exhibiting average to below-average condition. Following the 2016 floods golden perch exhibited strong increases in median condition in all Selected Areas with the exception of the Gwydir Selected Area. Prior to the 2016 floods, golden perch exhibited declining condition from 2015 to 2016 in the Goulburn, Lachlan, Lower Murray and Murrumbidgee (Figure 7).
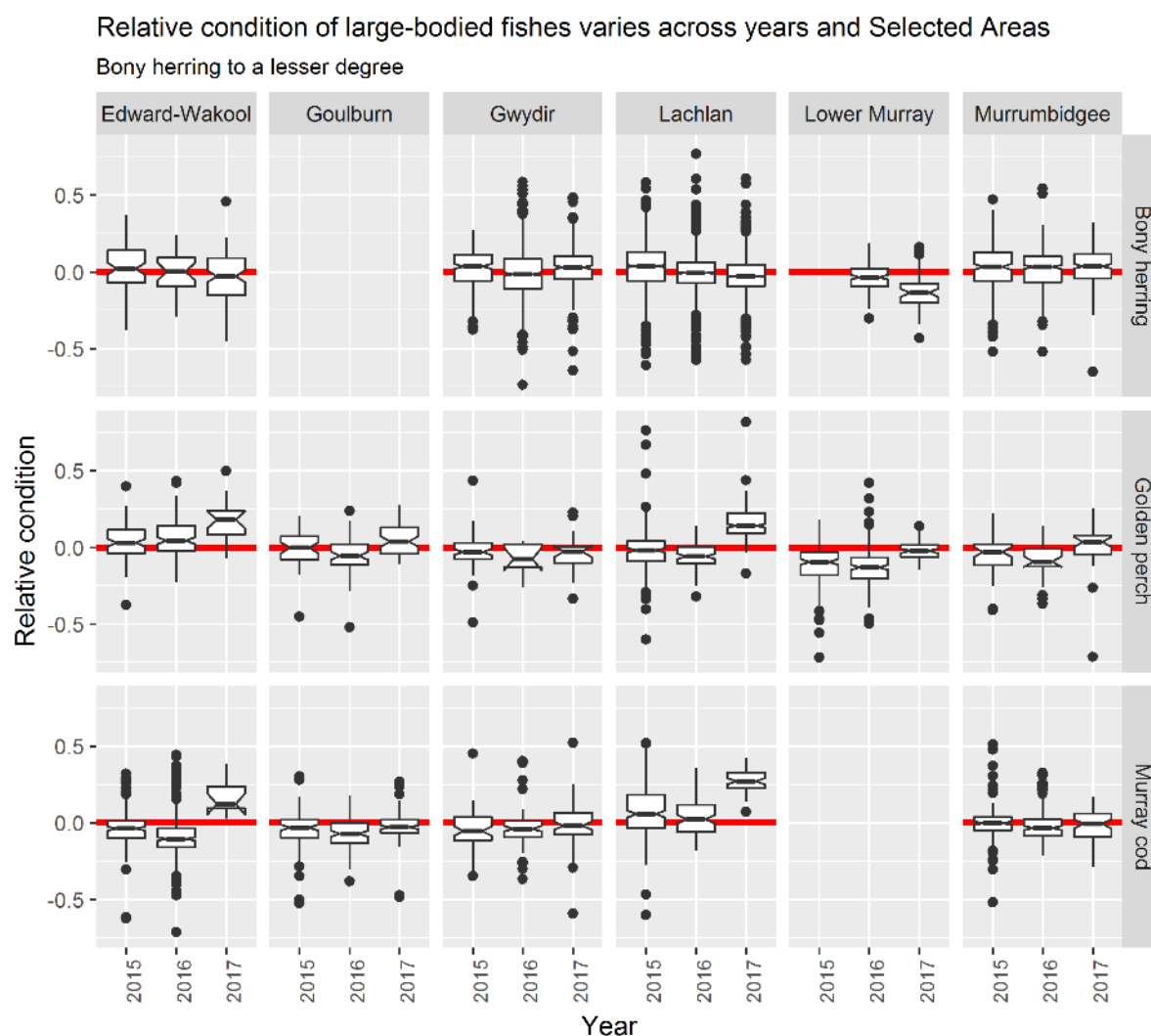
Figure 7. Boxplots of relative body condition of the three target species across years and Selected Areas. Thick horizontal lines are medians; the box is defined by the 25th and 75th percentiles (lower and upper quartile, respectively); dashed lines have lengths of 1.5 times the spread (spread = difference between quartiles). Points outside this range are outliers. Notches can be viewed as 95% confidence intervals for the median. Red horizontal line is basin-wide mean condition for the species. Blank panels indicate insufficient data.

Murray cod populations of the Lachlan and Murrumbidgee Selected Areas generally had better condition than individuals within the other Selected Areas during the period (Figure 7). With respect to variation through time, Murray cod, like golden perch, exhibited strong variation in median condition among years. The strongest differences were observed in the Edward-Wakool and the Lachlan, where the entire distribution of individual condition increased following the 2016 flood

(Figure 7). No increase in the condition of Murray cod was observed in the Gwydir, Goulburn and Murrumbidgee despite the fact these Selected Areas also experienced large floods in 2016. This pattern contrasts with golden perch, where an increase in condition was more widespread following the 2016 flood. With the exception of the Lower Murray, bony herring exhibit little variation in condition (Figure 7). At this stage, the reasons for this are not clear. It is possible that condition is not a good Basin-scale indicator of flow response for bony herring, and analysis of growth responses to flow variables may provide greater insights. Work to examine variation in growth rates is currently underway.

Modelling has showed there were strong and significant associations between river flows and the condition of golden perch and Murray cod. The optimal models provide a good fit to the data considering the variability in individual body condition, and, therefore, may provide a useful tool for evaluating the impacts of environmental water. However, because of the relatively small number of years of data currently available, the model predictions should be treated as preliminary. As the LTIM program continues the confidence in the observed relationships will increase.

Season-specific impacts of flows

The magnitude and directionality of the effect of flow on golden perch and Murray cod condition depended on seasonality for both species (Figure 8; Figure 9). The condition of both species increased with median daily discharge in both spring and summer (Figure 8; Figure 9). Spring flows had stronger positive effects on golden perch condition than summer flows (Figure 8). The condition of Murray cod, however, appeared to increase with equal magnitude in response to both spring and summer flows (Figure 9). Throughout much of the Basin, high flows during spring and early summer are expected under the natural flow regime. It is possible that invertebrate consumers (which are key to energy processing and transfer to fishes) may respond particularly strongly to spring and summer flows, in turn leading to increased golden perch and Murray cod condition, but the influence of this is uncertain.

Autumn median daily discharge had a negative effect on golden perch and Murray cod condition (Figure 8; Figure 9). This effect was particularly strong for Murray cod; indeed, median autumn flow was the variable with the greatest overall effect size in the model of Murray cod condition. With respect to golden perch, median autumn flow had relatively minor negative effects on condition in comparison to the strong positive effects of spring and summer flows. Further data will be required to validate this observation and begin to develop an understanding of any potential causal links.

Figure 8. Predictions (+/- 80% CI) of how the environment affects golden perch condition. The variable with the largest individual effect size (median daily discharge in spring) is on the x-axes. Individual panels present effects of spring discharge on condition at different values of autumn and summer median daily discharge. All predictions made within domain of data used for model fitting. Different panels defined by quantiles of observed flow data; the 10th, 25th, 50th (median), 75th and 90th percentiles. 'High density' and 'Low density' defined by 90th and 10th percentiles of golden perch CPUE (catch per unit effort). 'High flow LY' and 'Low flow LY' defined by 90th and 10th percentiles of median daily discharge in the previous flow delivery year.

Murray cod, *Maccullochella peelii*

Condition increases with Summer and Spring flows, but decreases with high Autumn flows
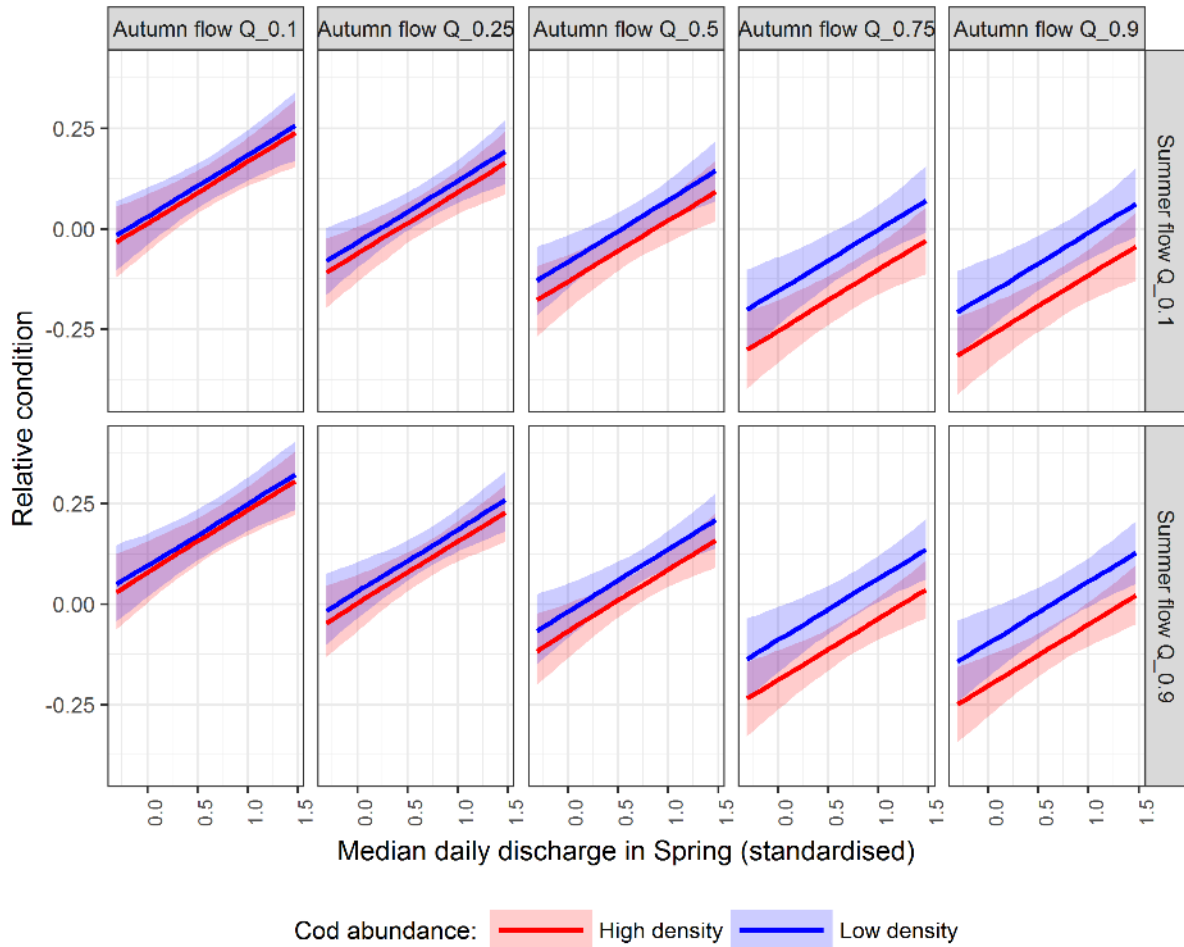
Figure 9. Predictions (+/- 80% CI) of how the environment affects Murray cod condition. The variable with the largest individual effect size (median daily discharge in autumn) is on the x-axes. Individual panels present effects of autumn discharge on condition at different values of summer and spring median daily discharge. All predictions made within domain of data used for model fitting. Different panels defined by quantiles of observed flow data; the 10th, 25th, 50th (median), 75th and 90th percentiles. 'High density' and 'Low density' defined by 90th and 10th percentiles of Murray cod CPUE. 'High flow LY' and 'Low flow LY' defined by 90th and 10th percentiles of median daily discharge in the previous flow delivery year.

Multi-year flow regimes affect golden perch condition

Exploring the influence of multiple year flows on golden perch condition indicated that condition in a year is not just affected by seasonal flows in that year, but also by flows in the previous year. Adding the previous year median discharge to the model improved model likelihood thus indicating that golden perch condition is influenced by antecedent flow conditions. Indeed, the results indicate that it is possible that if the right flow conditions exist, golden perch may accumulate condition over several years. However, in contrast to golden perch, median discharge of the preceding year was not a significant predictor of Murray cod condition.

Golden perch and Murray cod condition is affected by abundance of conspecifics

Body condition of both golden perch and Murray cod declined as population densities of conspecifics (other members of the same species) increased. Two possible hypotheses can be used to explain this result: First, the relationship may be an artefact of sampling where conditions

promoting an increase in condition and those causing a reduction in abundance just happened to occur at the same time but are not causally related. Second, it is possible that energy acquisition in golden perch and Murray cod is density-dependent. That is, as density of conspecifics increases, competition for food increases, resulting in a decline in condition. Further interrogation of the data indicates this first hypothesis is unlikely (see Section 4) and while demonstrating density-dependent condition would require much more data, the results highlight uncertainties concerning the dynamics of Basin fish populations.

3.3.2   Effects of environmental water

Note: Due to insufficient data, the intervention analysis presented in this section is based on models that have not been cross-validated (Hyndman & Athanasopoulos 2018). As such, all inferences presented in this section should be viewed as 'low confidence' inferences. Cross-validation of condition models will be possible with additional years of data collection.

The effects of environmental water on golden perch (Figure 10) and Murray cod condition (Figure 11) were examined under three hydrological scenarios to predict mean fish condition. The purpose of the three scenarios was to explore the additive contribution of environmental water compared to background (non-environmental water) flows. The scenarios explored were:

1. background river flows in the absence of any environmental water (black line);
2. background flows plus flows due to Commonwealth environmental water, but not all environmental water (blue line);
3. background flows plus all environmental water (green line).

From the models, a significant effect of environmental watering is inferred to have occurred when confidence intervals from the different scenarios do not overlap.

Golden perch

With respect to golden perch, we can state with *low confidence* that:

*   Environmental water, but not Commonwealth environmental water alone, delivered during 2014–15 and 2015–16 was attributed to increases in golden perch condition within the Goulburn and Murrumbidgee Selected Areas over the evaluation period (Figure 10);
*   Environmental water delivery during 2014–15 and 2015–16 was attributed to increases in golden perch condition within the Lachlan and Lower Murray over the evaluation period (Figure 10; noting that the Commonwealth environmental water was not differentiated from all environmental water in both those systems for 2015–16).
*   High flows in 2016/17 had a positive effect on fish condition, although those high flows occurred independently of Commonwealth environmental water delivery.

Both of the above inferences concerning environmental water impacts inherently incorporate and consider cumulative effects of environmental water delivery during 2014–15 and 2015–16 years, due to the fact that the optimal model of golden perch condition incorporates effects of antecedent flow on condition. We cannot evaluate the effects of environmental water on golden perch condition at the time step of first year of LTIM (2014–15), due to the optimal model incorporating multi-year flow impacts on condition; and the fact that we do not have counterfactual hydrological data for 2013–14, the year prior to the first year of LTIM.

There was a strong influence of the positive effect of 'background' flows (mostly natural flows in this case) on golden perch condition in all Selected Areas between 2016 and 2017. This increase in condition occurred in association with, and for those fish that survived, the 2016 floods. Given the background flow conditions during 2016 and 2017 the model was unable to determine impacts on

golden perch condition resulting from environmental water delivered specifically during 2016–17 within any of the Selected Areas (Figure 10).

There was no evidence of water delivery within the Edward-Wakool improving golden perch condition in any year (Figure 10), however this is not surprising given golden perch condition were not an objective of water delivery within the Edward-Wakool.
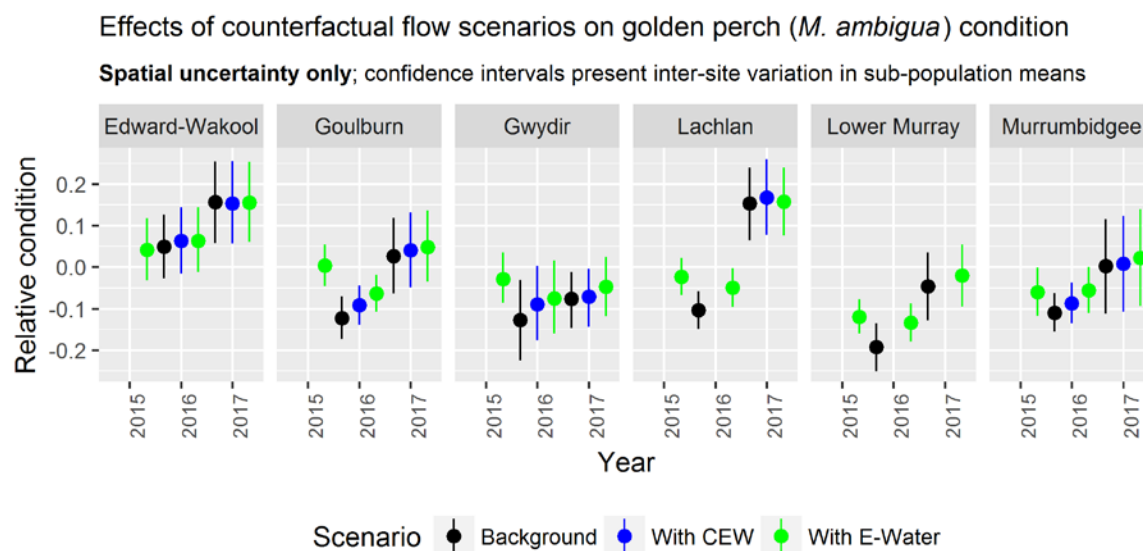
Figure 10. Predicted outcomes for golden perch condition under different hydrological scenarios considering the impact of background and environmental water flows.

Murray cod

With respect to Murray cod, we can state with *low confidence* that:

- Commonwealth environmental water delivery during 2014–15 increased Murray cod condition in the Edward-Wakool and in the Gwydir, as measured during 2015 (Figure 11).
- Environmental water delivered during 2014–15 in the Gwydir, additional to Commonwealth environmental water, increased Murray cod condition (Figure 11).
- Delivery of Commonwealth environmental water within the Goulburn during 2014–15 decreased Murray cod condition. Indeed, there was a general trend for environmental water delivery within the Goulburn to have a negative impact on Murray cod condition (Figure 11).
- High flows in 2016/17 had a positive effect on Murray cod condition in the Edward-Wakool and Lachlan rivers, although those high flows occurred independently of Commonwealth environmental water delivery.

We cannot evaluate the effects of environmental water delivery on Murray cod within the Lower Murray as there was insufficient condition data in this Selected Area. As more condition data is collected and as model confidence increases, we will be able to predict flow impacts beyond the areas that contributed data for parameter estimates and with a higher level of confidence.

The natural floods within the Edward-Wakool and Lachlan Selected Areas during 2016 resulted in a positive effect on the condition of those Murray cod that managed to survive the floods (Figure 11). This observation points to the potential benefits of floods for fish growth, but that those positive effects can be offset by higher rates of mortality when floods are associated with hypoxia. There was no evidence that discerned that environmental water delivered in the 2016–17 flood year impacted Murray cod condition (positively or negatively) within any of the Selected Areas (Figure 11).
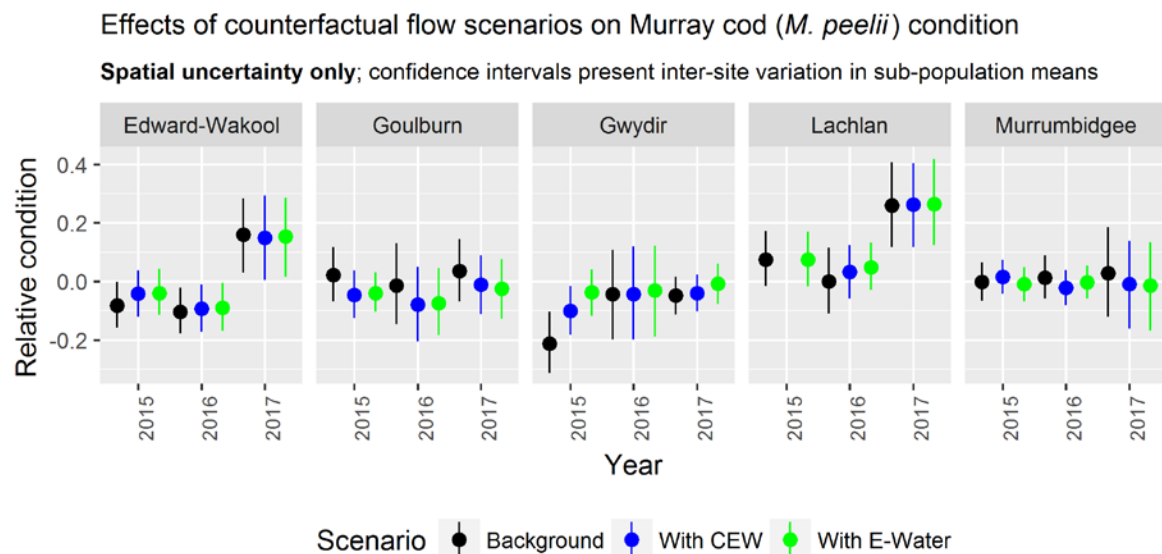
Figure 11. Predicted outcomes for Murray cod condition under different hydrological scenarios considering background and environmental water flows.

3.4 Adaptive management

Flows delivered in spring and summer may achieve the greatest positive impacts on golden perch and Murray cod condition. Flows in autumn should be delivered with caution.

For both golden perch and Murray cod, the effect of flow seasonality on fish condition was important, with spring discharge having the greatest positive effect. With respect to flow management within the Basin, this result implies that delivery of environmental water in spring may not just be good for spawning of species cued by spring-summer pulses (like golden perch and silver perch; King et al. 2009), but also good for condition of large-bodied fishes more generally (e.g. Tonkin et al. 2017). Even the condition of species like Murray cod, whose spawning is not dependent on spring flow pulses (Humphries 2005), responds positively to spring pulses. It follows that delivery of spring flows may be a 'win-win', since it may enhance several population processes of native fishes, as well as certain food web processes that fish are dependent on.

The condition of both golden perch and Murray cod appeared to exhibit a strong negative response to higher autumn flows where these occurred (e.g. in the Goulburn River). The mechanisms underlying this response are unknown, and there is a need to better understand how natural and managed flows affect food web processes within the Basin. Research to better understand these relationships is currently underway within the fish and food web themes of the Environmental Water Knowledge and Research (EWKR) Project.

The Goulburn was the only Selected Area where environmental flows elevated autumn discharge, and in accordance with model predictions, in areas where Murray cod condition was modelled, it is the only Selected Area were higher autumn flows were associated with declines in Murray cod condition. There may however be benefits in autumn environmental watering beyond those considered in these results, including providing movement and dispersal opportunities, enhancing breeding of small bodied generalists, and increasing survival of silver perch fingerlings.

When making flow decisions in any given year, flows delivered in the current year may affect how golden perch condition responds to flows in the subsequent year

Our analysis indicates that golden perch condition is affected by river flows across multiple years, meaning that flows in preceding years have an influence on condition. Consideration of the additive benefits of watering across years may provide opportunity to see greater ecological return, without necessarily allocating more water overall. There would be benefit in future work in undertaking a sensitivity analysis to separate the relative contribution to golden perch condition of environmental flows from previous years from those in the present year. Additionally, approaches exist to explore the optimisation of multi-year flow regimes (e.g. Horne *et al.* 2016) that could provide benefit to support multi-year decision making within LTIM.

Commonwealth and state environmental water use has synergistic effects, so should not be managed in isolation

The statistical modelling showed that, while Commonwealth environmental water on its own may have marginal effects on fish condition, combining that contribution with other water sources can result in significant additional improvements to fish condition. Therefore, continued collaboration amongst water managers from multiple jurisdictions is considered advantageous in achieving best possible outcomes.

4 Community dynamics

4.1 Summary

Effects of hydrology and water quality

- While little change in the Basin's fish community was observed during the first two years of LTIM, the large-scale natural floods of 2016 had significant and strong mixed effects on the Basin's fish community.
- With respect to environmental hypoxia, the analysis of data across multiple Selected Areas found only common carp and Murray cod to show consistent responses. No consistent response to environmental hypoxia was observed for other fish species.
- This may reflect issues with hypoxic conditions going undetected at some sites due to the timing of dissolved oxygen (DO) logger deployment, an issue that will be addressed over time as more data become available.
- With respect to the impacts of hypoxic blackwater floods on carp and Murray cod:
  - Carp abundances increased due to successful spawning and recruitment. This is the expected response of this species to flooding.
  - Carp were also better able to cope with hypoxic conditions than native species such as Murray cod. As a result, there were lower rates of mortality of carp relative to Murray cod.
  - Murray cod abundances declined at sites where flooding and hypoxia occurred.
- The magnitude of hypoxia, and hence impacts on the carp and Murray cod populations, varied considerably across the Selected Areas.

Impacts of environmental water

- We were unable to determine how specific water actions aimed at ameliorating hypoxia affected fish populations, including Murray cod and common carp.
- Monitoring of fish movement detected no movement of individual fish towards localised hypoxia refuges within the Edward-Wakool, but there were few data points, and thus we cannot conclude whether this strategy was effective.
- Irrespective of our ability to evaluate exactly how water actions ameliorated impacts of hypoxia, we argue that the decisions underpinning those water actions were sound for at least two reasons:
  - Given the magnitude of natural flooding, Commonwealth environmental water was insufficient to alter water quality except at a very local scale.
  - Given the high rates of mortality of Murray cod, enhancing the survival of even very small numbers, could help increase the resilience of local populations, and hence the capacity for the population to increase again in subsequent years.

Adaptive management

- Hypoxic blackwater events have the potential to reduce the longer-term benefits from environmental water actions. There is a need for innovative thinking and further modelling to understand how the dynamics of (a) river-floodplain hydrological connectivity; and (b) floodplain organic carbon can be actively adaptively managed, at scales that go beyond Selected Areas.
- The models suggest that shorter periods of hypoxia (e.g. a reduction in the duration of events from 14 to 7 days) can have a significant benefit in reducing mortality rates.
- Given that hypoxic blackwater events are likely to be unavoidable, a worthwhile focus for environmental watering actions could be to try and reduce the number of days over which hypoxic conditions persist.
- Prioritising Murray cod recruitment outcomes at locations unaffected by blackwater will contribute to improved populations at a Basin scale.

4.2 Effects of hydrology and water quality

Community dynamics

Assessment of LTIM survey data demonstrated that after three years of sampling, the dominant feature of community dynamics at the Basin-scale was the impact of the 2016 floods and environmental hypoxia event. That is, the fish community changes that were observed between 2015 and 2016 were minor compared with the changes that occurred between 2016 and 2017, the two surveys that occurred on either side of the Basin-wide floods and the hypoxic events in some Selected Areas. Within the Murrumbidgee, the magnitude of change following the flood was 1.65 times that of the change between 2015 and 2016 (Figure 12). In the Lachlan, where the hypoxia event was significant, the magnitude of change was 6.50 times that of the change between 2015 and

2016 (Figure 12). Generally, the shifts in community composition were high within Selected Areas where hypoxia was observed as significant.
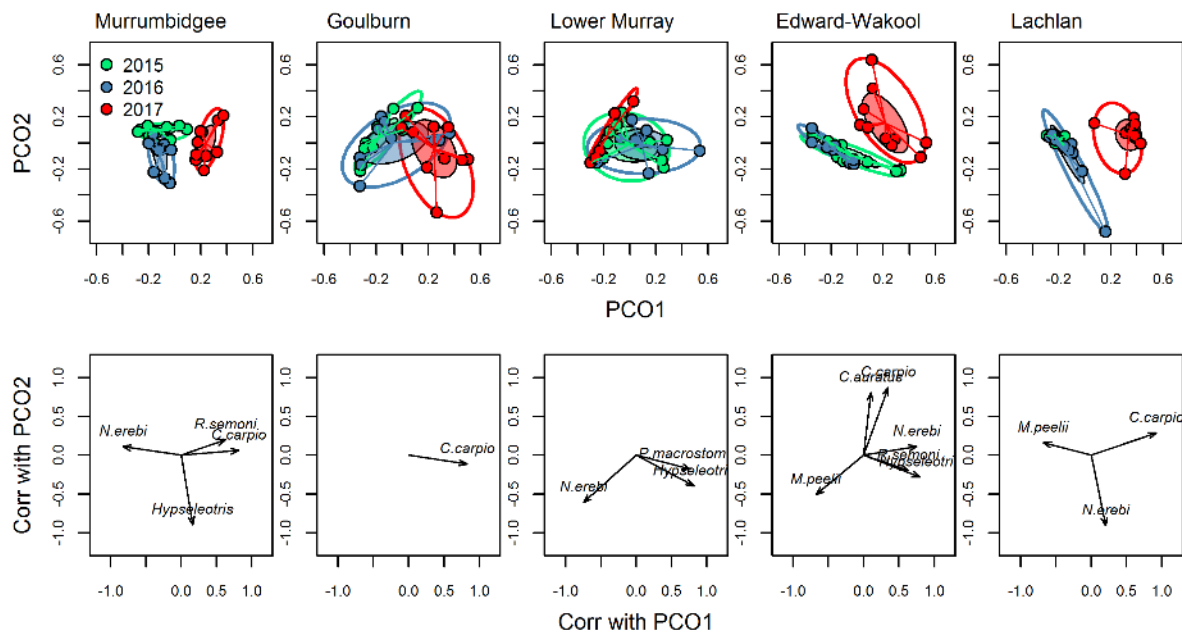
Figure 12. Top row: Ordinations (principal coordinate analysis: PCO) of changes in fish community structure among years for five Selected Areas. Flooding occurred between the 2016 and 2017 samples. Presented are points corresponding to individual sites within rivers; lines from points group (year) centroids; filled ellipses represent standard deviations around centroids; and ellipsoid hulls enclose all points within groups. Bottom row: Vector plots showing the correlation species abundances have with PCO axes. Only correlations > 0.6 are plotted.

Abundance

Although there was clearly a strong change in the Basin's fish community following the 2016 flood, not all fishes showed consistent and/or significant responses. For example, within the Lachlan, carp (*Cyprinus carpio*) abundance following hypoxic flows *increased* by more than 20-fold, and in the Edward-Wakool carp abundance *increased* four-fold. All increases in carp abundance were attributed to strong spawning and recruitment events associated with the 2016 flooding (Figure 13). Whether this strong cohort persists across multiple years will be revealed by future surveys.

Murray cod abundance *declined* by 77% within the Lachlan, and by 96% within the Edward-Wakool. In the Murrumbidgee carp increased by five-fold and Murray cod declined by 56%. Declines in Murray cod abundance were associated with reduced survivorship of all ages, but strong declines in recruitment and juvenile survival made a disproportionately large contribution to overall decline (Figure 6; Figure 13).

Abundances of all other fish species (both small and large-bodied species) changed within the Selected Areas following the 2016 flooding, but the direction of these changes (increasing or decreasing abundance) showed no consistent response across Selected Areas.
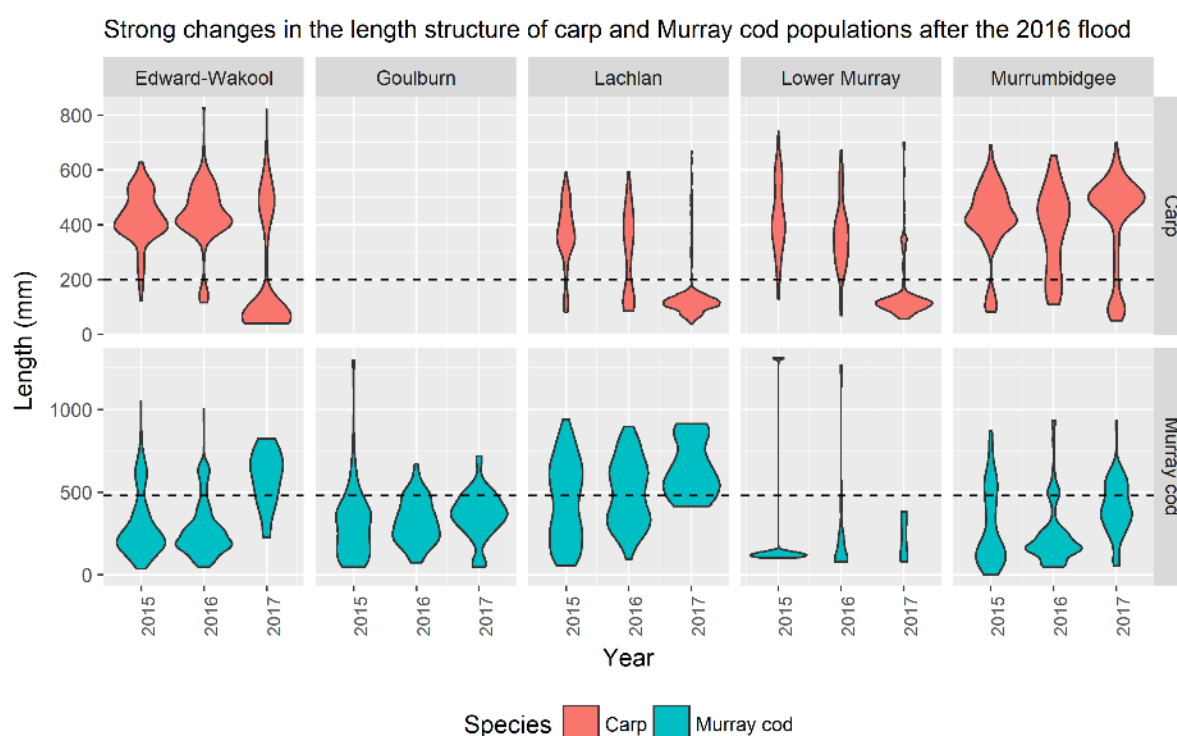
Figure 13. Relative distributions of fish lengths for carp and Murray cod, the two species impacted by the hypoxic blackwater flows. Dashed line indicates approximate maximum length of young-of-year carp, and length at maturity for cod.

To evaluate how hypoxia and high flows interacted to affect the abundance of carp and Murray cod we developed models to bring data together across five Selected Areas (Figure 2; Figure 12). The two predictor variables used in these models were the number of high-flow days and the number of hypoxic days. Model results for carp suggest the number of high-flow and hypoxic days had simple, *positive* and additive effect on carp abundance. In strong contrast with carp, both the number of high-flow days and, in particular, the number of hypoxic days had significant *negative* effects on Murray cod abundance. The interaction between these two predictor variables was significant, so that, as the number of high-flow days increases, the negative impact of hypoxia appears to weaken. Possible mechanisms underlying the modelled interaction between number of high-flow and hypoxic days are not obvious. One explanation is that extended periods of high flow provide a dilution effect relative to events that are long enough only to inundate the floodplain.

Murray cod are known to be susceptible to relatively mild environmental hypoxia (King *et al.* 2012; Small *et al.* 2014), and the reduction in Murray cod abundance we observed following the 2016 floods is largely a consequence of mortality. This result is corroborated by the many observations of Murray cod deaths throughout the Basin during the hypoxic floods. In contrast, carp are known to be very resistant to environmental hypoxia, having several traits that allow them to survive hypoxia (Lomholt & Johansen 1979; Zhou *et al.* 2000; Stecyk & Farrell 2006). In addition, they are also known to spawn on floodplains during floods (Koehn 2004). Therefore, flooding alone—irrespective of hypoxia—can provide enhanced spawning and recruitment opportunities for carp.

4.3   Effects of environmental water in ameliorating hypoxia

Several watering actions in 2016–17 had primary goal of ameliorating the effects of hypoxic water on fish. Development of models linking hydrology and dissolved oxygen has not yet been explored, and so while we have thus far been able to show that the number of hypoxic days has strong effects

on carp and Murray cod abundances, we do not, in turn, yet know how water delivery affects the number of hypoxic days. Such interactions will require future investigation.

While the benefits of environmental watering in providing local refuges from hypoxia remain unclear, we suggest that given the magnitude of reductions in Murray cod abundance (up to 96% loss in some regions), even small increases in survival of Murray cod could help to increase the resilience of the Murray cod population within the Basin.

4.4 Adaptive management

The primary outcome in terms of adaptive management to support fish community outcomes, is that while flood events may increase food availability and increase the condition of native fish, where those floods are associated with hypoxic blackwater, there clearly are significant and strong detrimental effects on survival of species such as Murray cod. Therefore, it follows that it is important to increase our understanding of how to either reduce these risks or to better mitigate the impact of hypoxic conditions when they do occur. This could include increasing our understanding of the spatial and temporal dynamics of hypoxia based on different hydrographs, as well as the history of flooding. For example, it will be important to ascertain whether the inundation of some low lying areas during years when minor flooding ay occur can help to reduce the risks of hypoxia by reducing the loads of dissolved organic carbon liberated during larger floods. Another important question is the scales over which fish can move to access localised refuges of higher water quality, and what role such interventions might play in enhancing survival rates and population resilience.

Results from the modelling thus far suggest that relatively small changes in the number of hypoxic days can have strong and significant effects on the fish community outcomes. Thus, even if we cannot avoid hypoxic events altogether, if such events occur small reductions in the number of hypoxic days can have very large positive biodiversity outcomes for the Basin. In addition, the outcomes of monitoring of environmental watering actions in the lower Darling, indicated that targeting Murray cod spawning and recruitment outcomes at locations that are not likely to experience hypoxic conditions can be an effective use of environmental water.

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Appendix A: Methods

All sampling methods, including the logic and rationale underlying their design, are presented in detail within Hale *et al.* (2013). Throughout this document — and LTIM documentation in general — Category 1, 2 and 3 methods are mentioned. Hale *et al.* (2013) explains the reasoning underlying the implementation of categorised methods, and explains in detail all Category 1 and 2 methods. Here we very briefly define Category 1–3 methods:

- Category 1 methods are standardised methods implemented across all six Selected Areas with a fish-monitoring focus. Data generated from Category 1 methods are used for Basin-scale analyses of fish response to flow and environmental change.
- Category 2 methods are standardised methods implemented across a subset of the six Selected Areas with a fish-monitoring focus. They may also be used for Basin-scale analyses, but are primarily used to inform area-scale evaluation of flow impacts.
- Category 3 methods are not standardised, but are area-specific methods primarily aimed at informing area-specific evaluation questions. In many instances, however, Category 3 methods are sufficiently similar—in some cases identical, but with greater effort—to Category 1 methods to be used for basin-scale analyses.

Study sites – LTIM Selected Areas

LTIM monitoring takes place at seven Selected Areas (Figure 1A). Not all seven areas implement the same methods, nor are they monitoring the same responses to flows. Monitoring and evaluation of fish response to flows at the basin-scale focuses on spawning, movement within Selected Areas, condition of individuals, and the dynamics of population and community composition. Table 1 presents an overview of where various monitoring activities are taking place.

Table 1A. Table highlighting data collected within each area. 'Cat' refer to category of sampling methodology (Hale et al. 2013). Filled cells indicate the sampling methods of the far left column are being implemented within that Selected Area.

	Goulburn	Edward-Wakool	Murrumbidgee	Lachlan	Gwydir	Lower Murray	Warrego-Darling
Cat 1 – Annual population / community censuses	■	■	■	■	■	■	
Cat 1 & 3 – Spawning response	■	■	■	■		■	
Cat 2 – Movement response	■	■			■		
Cat 3 – Various indicators	■	■				■	■

Hydrology

Time series of daily mean discharge for each Selected Area are supplied by the LTIM hydrology team (Michael Stewardson and Enzo Guarino) each year. Table 2 presents the gauges used for discharge data supporting fish analyses. The statistics in Table 2 associated with log-normalisation of discharge series will be explained later.
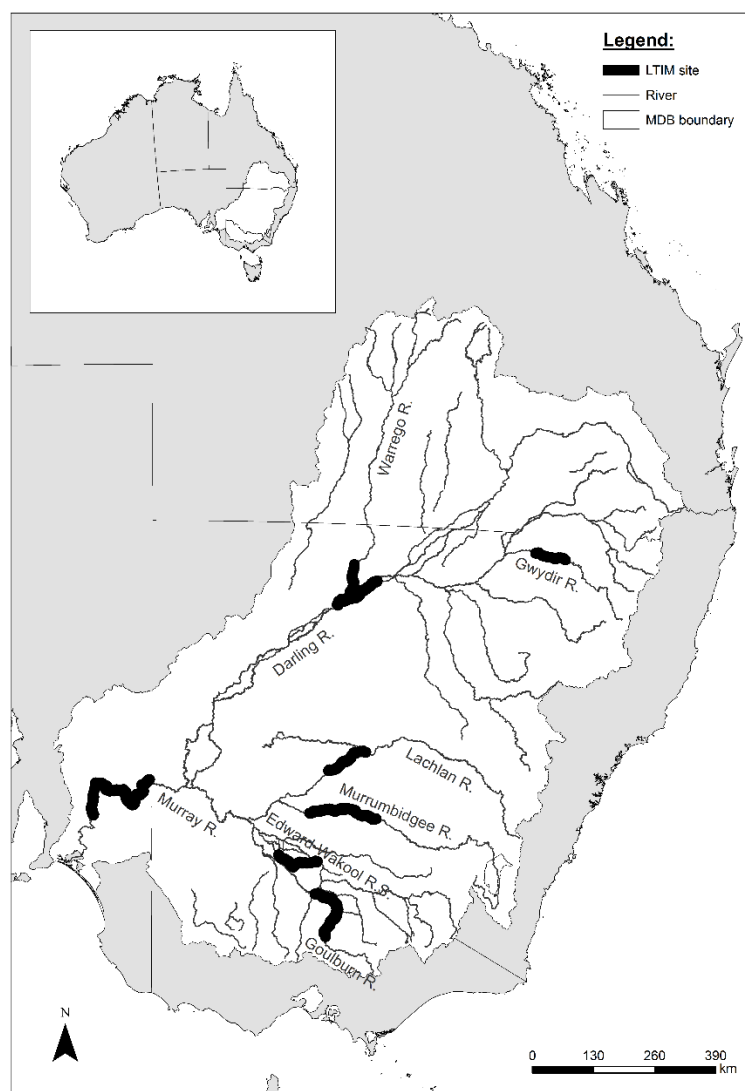
Figure 1A. Map of the Murray-Darling Basin and the locations and spatial extents of the seven Selected Areas of LTIM. Map prepared by Kyle Weatherman.

Logging of dissolved oxygen and temperature

Dissolved oxygen (DO) and water temperature were monitored within Selected Areas using multi-parameter water quality probes. Probes were deployed to monitor DO and temperature dynamics during the environmental flow delivery season within each year, which was generally between August and March. However, the exact timing and duration of deployment varied among rivers and years. Probes were calibrated following the manufacturers' specifications prior to deployment and every 4-6 weeks thereafter, as required. Individual measurements of DO and temperature occurred every 10 minutes. Data presented in this report come from one logging station, containing a single probe, established amongst the ten fish monitoring sites (see Fish Sampling) within each area (Table 2A). Dynamics of DO and temperature shown in this report were corroborated with those generated by other logging stations up/downstream of the study segments of the five rivers.

Table 2A. Gauges yielding hydrological time series used for fish basin-scale analyses. 'Zone' refers to the name given to LTIM zones. 'Gauged' indicates whether the raw discharge time series are obtained from a specific gauge (TRUE) or modelled/inferred based on data from those gauges (FALSE). Columns 'Census' and 'Larvae' indicate whether, respectively, population/community censuses and/or larval data are collected from that LTIM site. 'WQ' indicates whether dissolved oxygen and temperature loggers were deployed at that site. Remaining columns present statistics used for transforming daily mean discharge series into standard log-normal deviates (Mean and StDev) and the percentiles are the corresponding threshold values for log-normal deviates of flow series. 'N' and 'Start' are, respectively, the number of daily mean discharge values and the first year of discharge data that have contributed to the standardisation.

| | | | | | | | | Mean, StDev of ln(discharge + 1) | | Percentiles used as thresholds | | | | | | | | |
| | | | | | | | | Mean | StDev | 5th | 10th | 25th | 50th | 75th | 90th | 95th | | |
Selected Area	Zone	Gauge name	Gauge #	Gauged	Census	Larvae	WQ	Mean	StDev	5th	10th	25th	50th	75th	90th	95th	N	Start
Edward-Wakool	Zone1	YallakoolOfftake	409020	TRUE	FALSE	FALSE	TRUE	4.400	2.168	-2.029	-2.029	-0.337	0.237	0.598	0.988	1.476	22908	1954
Edward-Wakool	Zone2	WakoolOfftake	409019	TRUE	FALSE	FALSE	FALSE	3.198	2.115	-1.512	-1.512	-0.649	0.146	0.554	1.242	1.777	15559	1974
Edward-Wakool	Zone3	EWZone3_Model	NA	FALSE	TRUE	TRUE	FALSE	4.349	2.402	-1.810	-1.810	-0.394	0.275	0.606	1.017	1.466	24236	1951
Goulburn	Zone2	McCoys	405232	TRUE	FALSE	TRUE	TRUE	7.194	1.268	-1.050	-0.963	-0.758	-0.362	0.511	1.682	2.092	18831	1965
Goulburn	Zone1	Murchison	405200	TRUE	FALSE	TRUE	TRUE	6.677	1.508	-1.040	-0.871	-0.723	-0.468	0.669	1.684	2.064	23421	1951
Gwydir	GinghamGwydir	Pallamallawa	418001	TRUE	FALSE	FALSE	FALSE	6.445	1.354	-1.538	-1.183	-0.690	-0.075	0.690	1.321	1.635	16259	1972
Gwydir	GinghamGwydir	Tyreel	418063	TRUE	TRUE	FALSE	FALSE	4.231	1.539	-1.932	-1.210	-0.647	-0.035	0.771	1.273	1.516	11503	1985
Gwydir	GinghamGwydir	Millewa	418066	TRUE	TRUE	FALSE	FALSE	3.414	1.629	-2.037	-1.686	-0.586	0.102	0.701	1.262	1.528	10028	1988
Gwydir	GinghamGwydir	Yarraman	418004	TRUE	TRUE	FALSE	FALSE	5.250	1.666	-1.557	-1.072	-0.555	0.004	0.600	1.035	1.617	16750	1964
Lachlan	Zone1	Hillston	412039	TRUE	TRUE	TRUE	FALSE	6.071	1.425	-1.563	-1.214	-0.567	-0.053	0.428	1.595	1.787	22502	1955
Lachlan	Zone1	Whealbah	412078	TRUE	FALSE	FALSE	TRUE	5.281	1.444	-1.407	-1.138	-0.584	-0.079	0.396	1.677	2.115	9273	1968
Lachlan	Zone1	Booligal	412005	TRUE	FALSE	FALSE	FALSE	5.489	1.596	-1.437	-1.144	-0.605	-0.110	0.638	1.545	1.682	24032	1951
Lower Murray	Floodplain	Lock6	4260511	TRUE	FALSE	TRUE	TRUE	8.122	1.160	-1.854	-1.500	-0.599	0.118	0.617	1.321	1.665	7189	1994
Lower Murray	Floodplain	Lock5	4260513	TRUE	FALSE	FALSE	FALSE	8.690	0.898	-1.643	-1.204	-0.609	-0.058	0.578	1.393	1.743	10265	1981
Lower Murray	Floodplain	Lock4	4260515	TRUE	FALSE	FALSE	FALSE	8.610	0.798	-1.729	-1.197	-0.633	-0.004	0.575	1.372	1.872	6772	1994
Lower Murray	Gorge	Lock1	4260903	TRUE	TRUE	TRUE	TRUE	8.899	1.303	-1.420	-1.130	-0.665	-0.147	0.755	1.390	1.652	18953	1951
Lower Murray	Gorge	Lock3	4260517	TRUE	FALSE	FALSE	FALSE	8.564	0.863	-1.589	-1.181	-0.601	-0.092	0.548	1.430	1.852	7043	1994
Lower Murray	Gorge	Lock2	4260519	TRUE	FALSE	FALSE	FALSE	8.502	0.879	-1.683	-1.299	-0.545	-0.072	0.578	1.369	1.854	7056	1994
Murrumbidgee	Carrathool	Carrathool	410078	TRUE	TRUE	TRUE	TRUE	7.618	1.018	-1.593	-1.349	-0.665	-0.006	0.607	1.317	1.765	7783	1995
Murrumbidgee	Narrandera	Narrandera	410005	TRUE	FALSE	TRUE	FALSE	8.650	0.896	-1.682	-1.338	-0.662	0.093	0.550	1.231	1.794	16012	1973

Fish sampling

Spatial configuration of fish sampling

LTIM samples are collected within a hierarchy of spatial sampling units, following Gawne et al. (2013): 'Zones' are nested within Selected Areas, and 'sites' are nested within zones. Zones and sites for fish sampling have the following characteristics:

A 'zone' was a subset of a Selected Area that represents a spatially, geomorphologically and/or hydrologically distinct unit at reach-segment scales (Fausch *et al.* 2002). A Selected Area may comprise multiple zones, but each Selected Area contained a 'focal zone', from which samples for basin-scale analyses were collected. The focal zone of each Selected Area was likely to receive Commonwealth environmental water at least once in the next five years and was associated with specific expected ecological outcomes within that same time frame. Aquatic habitat within focal zones were representative of the Selected Area as a whole.

Within zones, ten channel sites were established for sampling. A site was defined as an 800 m reach of channel (Figure 2A). Sites were selected to be representative of the zone as a whole, were randomly located, and their locations are fixed throughout the LTIM program. Site locations have been fixed such that we do not conflate spatial and temporal sources of variation.

Within sites, a sampling grid was established to ensure individual samples can be sampled randomly with respect to spatial environmental heterogeneity, such that samples were representative of that site as a whole (Figure 2A). Each 800 m site is subdivided by fixed transects spaced 50 m apart. Points of intersection between the transects and the river bank defined the sampling grid (Figure 2A).

The sample design specified in Figure 2A defines two key sampling locations: electrofishing (EF) units (16 in total), and passive-gear sample (PS) waypoints (34 in total). Use of these EF units and PS waypoints for larval sampling and annual censuses of population and community structure will be explained below.
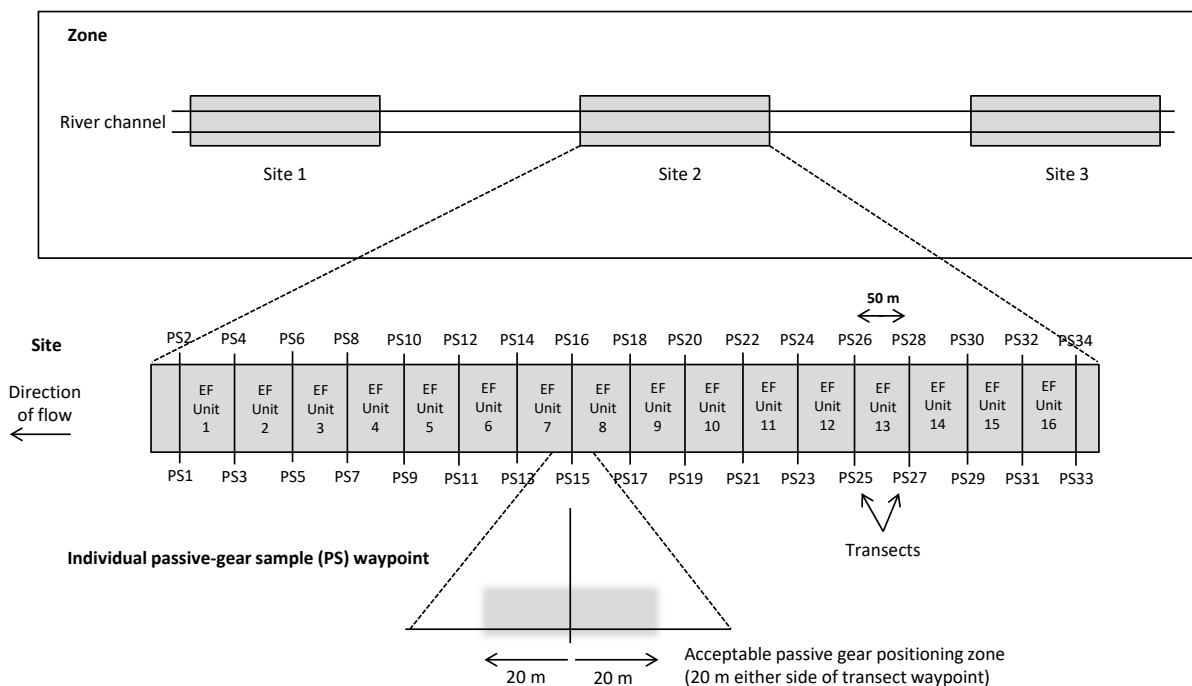
Figure 2A. Diagram of the spatial configuration of sampling, showing sites nested within a zone, and individual sampling locations/units within sites.

Larval fishes

Larval sampling methods differ among Selected Areas and are a mix of Category 1 and 3 methods. The Category 1 larval sampling method currently in use is not the method presented in Hale et al. (2013). Instead, larval sampling under the current Category 1 larval method had the following specifications:

Sampling took place at three of the ten sites within the focal zone of each Selected Area. Each site was sampled for larvae five times (five sampling 'events'; [Event 1, Event 2, …, Event 5]) within each flow delivery year (corresponds with a financial year; July 1 – June 30) and, as much as practicable, all three sites were sampled on the same day within each event. Within each unique site-event combination, the standard method called for ten light-trap and three drift samples.

The same modified quatrefoil light-traps (Humphries et al. 2002) were used at each Selected Area, to eliminate spatial bias. Mesh was fitted around the light traps to eliminate larger fish from entering the trap, and eating the sample (3 mm knot-to-knot). Each light trap was 'baited' with a yellow Cyalume® 12 h light stick (or equivalent manufacturer, but yellow in colour). The ten light traps set within each of the three sites were set in the afternoon and retrieved the following morning. Within each site, during each event, the light traps were set at a random subset of 10 PS waypoints from the total of 34 (Figure 2A). Abundances from light-trap samples were uploaded to the LTIM database as catches-per-unit-effort (CPUE), with units of number of individuals, per trap, per hour within a site-event combination (i.e. variance across traps within sites was 'averaged away' prior to uploading).

The three drift samples within a site-event combination were one of two types:

A. If there was sufficient flow velocity within the focal zone, drift samplers were established to passively filter larvae from a moving water column.
B. If there was insufficient water velocity within the focal zone, three larval tows were obtained, whereby the drift net is pushed through the water column by boat, and larvae are actively filtered from the water column.

Irrespective of the whether the sample was active or passive, drift nets were constructed of 500 µm mesh, and had an opening diameter of 50 cm, tapering over 1.5 m to an opening of 9 cm, where a 'reducing bottle' was fitted. Volume through the net was estimated so that larval abundances in drift nets could be expressed as a density: number of individuals per m³. Volume sampled by the net is estimated as $\pi r^2 \cdot v \cdot t$, where r is radius in metres, v is mean velocity in m s^{-1}, and t is time set in seconds. Thus for both active and passive drift samples, data uploaded to the LTIM database had CPUE units of mean number of individuals per m³. Again, each row in the LTIM database is a mean CPUE and disregards variation across the three drift samples within a site-event combination.

Population and community censuses

Category 1 annual censuses took place once each year during autumn and, unlike larval methods, follow Hale et al. (2013). Within the six Selected Areas, annual censuses consisted of intensive electrofishing for large-bodied species and fyke netting for small-bodied species.

Within each of the ten sites the entire 800 m site was electrofished. Within each electrofishing unit of a site (EF unit; Figure 2A) two 'shots' of 90 s 'on-time' was carried out. This resulted in a total of 2880 s (48 min on-time) for each site. No more than 180 s of shocking was allocated to each EF unit, such that electrofishing effort was spread out across the entire site, giving a random sample with respect to the (site's) environment. Within EF units the location of shots was left to the discretion of the sampler. Abundances from electrofishing were calculated as number of individuals per second 'on-time' within each site and year.

Ten fyke net (fish trap) samples were also taken from within each site. Fyke nets were randomly positioned at PS waypoints and set overnight. Abundances from fyke nets were uploaded to the LTIM database as mean numbers of individuals per net, per hour within each site and year (variation across nets within a site-year combination is disregarded).

Data analysis

All analyses and visualisations were carried out using R version 3.4.0 (R Core Team 2017).

Preparation of hydrology, temperature and dissolved oxygen data

Series for three variables were calculated for each of the gauges presented in Table 1A: (1) 'background' (BG) daily discharges; the estimated discharge in the absence of any environmental water; (2) background discharges plus the contribution of Commonwealth environmental water (BG + CEW); (3) background daily discharges plus the contribution of all environmental water (BG + EW), where environmental water includes, Commonwealth environmental water, state environmental water (e.g. VEWH allocations), The Living Murray flows, and certain inter-valley transfers.

We required a hydrological index that can be standardised across rivers having mean daily discharges that differ by orders of magnitude. Towards this end, we transformed all daily discharge variables from each gauge into *standard log-normal random variables* (Wackerly et al. 2002):

$$\widetilde{D}_{ij} = (D_{ij} - \overline{D}_j)/\delta_j$$

where D_{ij} is the ln(x+1)-transformed discharge on day *i* at gauge *j*; $\overline{D}_j$ and δ_j are, respectively, the mean and standard deviation of those log-transformed values at that gauge; and $\widetilde{D}_{ij}$ is the log-normal deviate. By transforming all discharge values have placed all discharge values from different rivers on a common scale that should (approximately) quantitatively map to ecological effects in a similar manner across selected areas. Discharge values at each gauge were standardised for all three series (BG; BG + CEW; BG + EW). For clarity, we hereafter refer to $\widetilde{D}_{ij}$ as 'standardised discharge.'

If one wishes to determine any of the threshold values of Table 2A in units of ML d^{-1}, then the formula for back-transformation is:

$$D_{ij} = \exp(\widetilde{D}_{ij}\delta_j + \overline{D}_j) - 1$$

Time series of daily mean water temperature and dissolved oxygen for focal zones (Table 2A) were obtained from the LTIM DO logger database. Water temperature and DO data were only available for certain subsets of each year.

Once all hydrology, temperature and DO data were sourced and tidied they were then ready for generation of various predictor variables; variables that would serve as covariates in subsequent modelling of fish response to flows. Certain hydrology predictor variables were defined by the number of days where standardised discharge was above or below some threshold. Hydrology thresholds were defined by specific percentiles of the standard log-normal variables; the 5th, 10th, 25th, 50th, 75th, 90th and 95th percentiles. The values of these thresholds, as well as the quantity and span of data that went into their calculation, are presented in Table 2A.

The specific predictor variables generated were dependent on the temporal grain of analysis they were intended for. Samples of the structure of fish populations (including condition) and communities are collected once annually, so we require predictor variables describing the statistical properties of the environment over the year preceding the population/community sample. Analyses pertaining to effects of flow on the structure of populations and communities have an annual temporal grain. Each year a set of base predictors with an annual temporal grain are generated; they are described in Table 3A. For each of the flow-related variables in Table 3A we were able to

estimate the counterfactual values—the values of that predictor variable expected under the three hydrological scenarios BG, BG + CEW, BG + EW.

Table 3A. Definition of base predictors with an annual temporal grain. R scripts that collate and tidy the raw data, then estimate these predictors are available from R. Stoffels.

Predictors	Counterfactual scenarios?	Definition
flow_Q1; flow_Q2; flow_Q3	Yes	The first, second (median) and third quartiles of mean daily standardised discharge (ML/day) within each of the four seasons (3 x 4 = 12 values per annum, per gauge), and the year as a whole (3 x 1 values per annum, per gauge).
meanDO_Q1; meanDO; Q2; meanDO_Q3	No	The first, second (median) and third quartiles of mean daily dissolved oxygen concentration (mg/L) within each of the four seasons (3 x 4 = 12 values per annum, per gauge), and the year as a whole (3 x 1 values per annum, per gauge).
minDO_Q1; minDO_Q2; minDO_Q3	No	The first, second (median) and third quartiles of minimum daily dissolved oxygen concentration (mg/L) within each of the four seasons (3 x 4 = 12 values per annum, per gauge) and the year as a whole (3 x 1 values per annum, per gauge).
meanTemp_Q1; meanTemp_Q2; meanTemp_Q3	No	The first, second (median) and third quartiles of mean daily water temperature (°C) within each of the four seasons (3 x 4 = 12 values per annum, per gauge), and the year as a whole (3 x 1 values per annum, per gauge).
maxTemp_Q1; maxTemp_Q2; maxTemp_Q3	No	The first, second (median) and third quartiles of maximum daily water temperature (°C) within each of the four seasons (3 x 4 = 12 values per annum, per gauge), and the year as a whole (3 x 1 values per annum, per gauge).
GPP_Q1; GPP_Q2; GPP_Q3	No	The first, second (median) and third quartiles of daily Gross Primary Production (mg O_2 L^{-1} Day^{-1}) within each of the four seasons (3 x 4 = 12 values per annum, per gauge), and the year as a whole (3 x 1 values per annum, per gauge).
ER_Q1; ER_Q2; ER_Q3	No	The first, second (median) and third quartiles of daily Ecosystem Respiration (mg O_2 L^{-1} Day^{-1}) within each of the four seasons (3 x 4 = 12 values per annum, per gauge), and the year as a whole (3 x 1 values per annum, per gauge).
vLowDays	Yes	The number of days within the season where the mean daily standardised discharge was less than or equal to the 5th quantile of long-term standardised daily discharge levels (4 values per annum, per gauge), and the year as a whole (1 value per annum, per gauge).
lowDays	Yes	The number of days within the season where the mean daily standardised discharge was less than or equal to the 10th quantile of long-term standardised daily discharge levels (4 values per annum, per gauge), and the year as a whole (1 value per annum, per gauge).
highDays	Yes	The number of days within the season where the mean daily standardised discharge was greater than or equal to the 90th quantile of long-term standardised daily discharge levels (4 values per annum, per gauge), and the year as a whole (1 value per annum, per gauge).
vHighDays	Yes	The number of days within the season where the mean daily standardised discharge was greater than or equal to the 95th quantile of long-term standardised daily discharge levels (4 values per annum, per gauge), and the year as a whole (1 value per annum, per gauge).

meanDODays6; meanDODays4; meanDODays2	No	The number of days within the season where the mean daily dissolved oxygen concentration was less than or equal to 6, 4 and 2 mg L^{-1} (3 x 4 values per annum, per gauge), and the year as a whole (3 values per annum, per gauge).
minDODays4; minDODays2	No	The number of days within the season where the minimum daily dissolved oxygen concentration was less than or equal to 4 and 2 mg L^{-1} (2 x 4 values per annum, per gauge), and the year as a whole (2 values per annum, per gauge).
meanTempDays20; meanTempDays25	No	The number of days within the season where the mean water temperature exceeded 20 or 25 °C (2 x 4 values per annum, per gauge), and the year as a whole (2 values per annum, per gauge).
maxTempDays25; maxTempDays30	No	The number of days within the season where the maximum water temperature exceeded 25 or 30 °C (2 x 4 values per annum, per gauge), and the year as a whole (2 values per annum, per gauge).

Fish population dynamics

To determine how the states of fish populations varied within and among Selected Areas, we compared and contrasted the age structure and fish condition among years. LTIM's three large-bodied target fishes were the focus of our population analysis: bony herring, golden perch (both in the periodic guild) and Murray cod (equilibrium guild).

Age structure

Comparisons of age and length structure among Selected Areas involved two steps:

1. Develop age–length keys (ALKs) for the three target species, using LTIM age–length data — sourced from the LTIM otolith collections — from each Selected Area.
2. Use fish length data obtained from Category 1 censuses (Hale et al. 2013) which serve as input to an algorithm that estimates the proportionate age composition of the length sample.

Step 1: Development of ALKs

Development of the ALK for each species follows standard techniques in fisheries stock assessment (Quinn and Deriso 1999). An ALK is a matrix whose *i,j*th entry is the probability that a fish of length-class *i* is of age *j* [$P(j|i)$]. When constructing ALKs, the width of length classes (in cm) varied among species, and reflected the range in length for each species. That is, the larger the species gets, the wider the length class used for ALK construction. For bony herring, golden perch and Murray cod, the length class widths were 1, 2 and 3 cm, respectively.

A single ALK was developed for each of the three target species using age–length pairs, obtained from otoliths collected across the Selected Areas. Within species, age–length data from Selected Areas were pooled to obtain large samples for estimates of the $P(j|i)$ values. In pooling data, we made the assumption that age–length functions of fish populations do not vary across Selected Areas. If this assumption is false, our current ALKs will inflate errors around our estimates of the proportionate age composition of each population. In the long term, we aim to relax this assumption, by collecting more otoliths from target species, towards development of Selected Area–specific ALKs. The total number of age–length pairs that were used to estimate ALKs for each species were as follows:

- Murray cod: 1420 individuals.
- Golden perch: 951 individuals.
- Bony herring: 1456 individuals.

Each ALK went through three stages of development. First, the raw age–length pairs of a species were used to populate the matrix, such that the i,jth entry represents the number of individuals within length class i of age j. Second, we made the assumption that lengths within an age cohort were normally distributed. A normal curve was fitted to the data within each column of the ALK using maximum likelihood estimation. Thus, we now have an ALK whose i,jth entry is the expected frequency of individuals in length class i of age j, assuming a normal length distribution within each age cohort. At this stage, column totals — but not row totals — sum to one. The final stage of ALK development was to transform the matrix such that row totals sum to one, as is required when row entries represent the probabilities $P(j|i)$.

Step 2: Inferring age structure from ALKs

ALKs were used in a Monte Carlo (MC) algorithm to determine the mean proportionate age composition for each species within each Selected Area. This algorithm also estimates 95% confidence intervals (CIs) about the proportionate age composition, where the CIs represent uncertainty due to variation in length at age.

Once a vector of fish lengths is passed to this algorithm, an individual run consists of assigning an age to each individual fish length with probability $P(j|i)$. Because ages are assigned to lengths probabilistically, an individual length will not necessarily be assigned to the same age between runs. By running this MC algorithm many times, we estimate mean proportionate age composition of a length sample and 95% CIs around each mean. The width of CIs is, therefore, determined by the distribution of $P(j|i)$ values within each row of the ALK and the size of the length sample passed to the MC algorithm. Mean proportionate age compositions, and their corresponding CIs, were multiplied by catches per unit effort (CPUEs) to yield the CPUE of each cohort, within each Selected Area, and the corresponding uncertainty.

R scripts for the above analyses are available from the lead author of this document.

Condition

Relative condition of target species bony herring, golden perch and Murray cod were was modelled as a function of (a)biotic variables. We determined how *relative* condition of individuals within a species varies through space and time as follows: After pooling all length-mass pairs of a species (across all years and Selected Areas), a simple linear regression between *ln* mass (g) and *ln* length (mm) was executed and the residuals were extracted as indicies of relative condition, *Con* (Ogle 2016). This step of the condition analysis removes any effect of body mass on condition. *Con* is centred on zero with a normal distribution. Values less than and greater than zero indicate, respectively, below- and above-average condition, relative to all other individuals sampled throughout the Basin, over all years. Prior to modelling, outliers were defined as those residuals with absolute values exceeding four times the mean of Cook's distance for all residuals (Cook 1977).

Following the removal of body length effects on *Con*, linear mixed effects regression was used to determine how the (a)biotic environment affects *Con* (Pinheiro and Bates 2000). The fixed terms of the model include flow variables and catch per unit effort (CPUE) of the species. CPUE was included to account for possible impacts density may have on intraspecific competition for food resources, hence fish condition. To determine how timing of flows affects fish condition we selected four flow predictors: the median daily discharge in the Winter (*flowWin*), Spring (*flowSpr*), Summer (*flowSum*) and Autumn (*flowAut*) of the flow delivery year within which the Autumn population census occurred. However, it is possible that the impact of flows on *Con* may be cumulative across more than a single year, so we also included the median daily discharge for the entire year *prior* to the flow delivery year within which the fish population was censused (*flowLastY*). To be clear, if Con_{it} is the relative condition of individual i in flow delivery year t, then $flowLastY_t$ is defined by the median daily discharge in flow delivery year t-1.

Our model selection strategy follows that of Diggle (2002), and recommended by Zuur et al (2009). We start with the full fixed component of the model (the 'beyond optimal' model) and alter the random component of the model towards finding the optimal random structure first. The fixed component of the beyond optimal *Con* model was not an exhaustive parameterisation of all combinations of predictors, but a parameterisation that reflected our primary questions of the data:

$$Con_{it} = \alpha + \beta_1 flowWin_t + \beta_2 flowSpr_t + \beta_3 flowSum_t + \beta_4 flowAut_t + \beta_5 cpue_t +$$
$$\beta_6 flowSum_t \cdot cpue_t + \beta_7 flowAut_t \cdot cpue_t + \beta_8 flowLastY_t + \varepsilon_{it} \qquad \varepsilon_{it} \sim N(0, \sigma^2)$$
$$(1)$$

Where *Con*$_{it}$ is the condition of individual fish *i* (1,2,…) in year *t* (2015, 2016, 2017), predictors are as described earlier, α is the intercept, the β are slope parameters, and ε_{it} is the residual error. We restricted any interaction cpue has with flow to seasons when feeding activity is likely to be highest to match metabolic demands, and when flows may be lowest (hence competition highest); Summer and Autumn. Exploratory plots indicated that Murray cod Con may exhibit quadratic relationships with *flowSum* and *flowAut*, so the beyond optimal model for that species included four additional parameters—one for each individual quadratic term, and one each for their interaction with cpue.

We wished to answer two broad questions with respect to the random component of our model: First, does explicitly accounting for the nested structure of our data (with sites nested in Selected Areas) improve the fit of models, or is allowing for intercepts to vary over Selected Areas sufficient? Second, do we need to allow the slopes of key linear relationships to vary spatially, or is allowing for spatial variation in intercepts alone sufficient? Eighteen different random structures were tested to answer these questions. To save space we do not list those 18 equations here, but instead state the different random structures verbally:

1. Random intercept for Selected Area;
2. Random intercept and slope (*flowLastY*) for Selected Area;
3. Random intercept and slope (*flowAut*) for Selected Area;
4. Random intercept and slope (*flowSum*) for Selected Area;
5. Random intercept and slope (*flowSpr*) for Selected Area;
6. Random intercept and slopes (*flowAut* and *flowSum*) for Selected Area;
7. Random intercept and slopes (*flowAut* and *flowSpr*) for Selected Area*;*
8. Random intercept and slopes (*flowSum* and *flowSpr*) for Selected Area*;*
9. Random intercept and slopes (*flowAut*, *flowSum* and *flowSpr*) for Selected Area*;*
10. An additional nine models the same as above but with intercepts and slopes also varying between sites within Selected Areas.

As was the case for the fixed component of the model, it will be obvious from the above that we did not implement an exhaustive search across all possible random terms (given the fixed terms of Equation 1). Instead, we included random terms that we thought would lead to less biased, more robust inferences concerning the fixed terms of primary interest; namely, the impacts of flows. The random structure that yielded the lowest Akaike Information Criterion (AIC) was selected as the most parsimonious model after the first step of the selection process. Potential bias was checked by examining plots of standardised residuals against fitted values and predictor variables. During this first step of the model selection process all models were fit using restricted maximum likelihood.

The aim of the second step was to simplify the fixed component of the model. This involved determining which single-term deletions could be made that increased AIC of the model without significantly reducing the likelihood. The models were nested so the log-likelihood ratio test was used to determine whether single-term deletions significantly reduced likelihood. Use of the log-likelihood ratio test required parameter estimation using maximum likelihood (Zuur et al. 2009).

The final model selected for each species was that model for which any further single-term deletion significantly decreased likelihood. The final step of the model selection process was to refit the final model using restricted maximum likelihood, undertake a visual check of plots of standardised residuals against fitted values and covariates, then graphically examine the direction and magnitude of effects.

Linear mixed-effects modelling was carried out using the lme4 package (Bates et al. 2015), with additional model analysis facilitated by the sjstats package (Lüdecke 2018), the merTools package (Knowles and Frederick 2016), and lmerTest (Kuznetsova et al. 2017).

Predictions from the fitted models were used to determine whether environmental watering actions had an impact on golden perch and Murray cod condition. That is, we predicted mean condition of each species under each of the three hydrological scenarios within each Selected Area:

4. 'background' river flows in the absence of any environmental water (BG);
5. background flows plus flows due to Commonwealth environmental water, but not all environmental water (BG + CEW);
6. background flows plus all environmental water (BG + EW).

Inferences concerning environmental water effects are presented under three confidence levels. If we infer an effect of environmental water with:

- 'low confidence', then that means the 55% confidence intervals corresponding to two hydrological scenarios do not overlap, but 75% and 95% confidence intervals do;
- 'medium confidence', then the 75% confidence intervals do not overlap, but the 85% confidence intervals do;
- 'high confidence', then the 95% confidence intervals do not overlap.

Fish community dynamics

For all analyses pertaining to community structure, data were first range standardised by re-scaling all abundance estimates to between 0 and 1. Range standardisation was carried out across all small-bodied species, and across all large-bodied species separately, to account for differences in absolute abundance estimates from fyke net and electrofishing sampling techniques, which were used to estimate abundances of these two groups groups respectively. As stated in the Standard Methods (Hale et al. 2013), small-bodied species comprised any species belonging to the families Retropinnidae, Eleotridae, Galaxiidae, Melanotaeniidae, Atherinidae, Ambassidae and Poeciliidae. Large-bodied fishes are classified as belonging to the other families of the Basin. The range-standardised CPUE from site k, species j and year i is defined:

$$CPUE_{ijk}^{RS} = \frac{CPUE_{ijk} - \min(CPUE_{jk})}{\max(CPUE_{jk}) - \min(CPUE_{jk})}$$

where the js belong to one of two sets defining the domains of the minimum and maximum functions; the set of small-bodied fishes, or the set of large-bodied fishes. Range standardisations were carried out to put the very different CPUE units of these two sets of species (ind net^{-1} h^{-1} *versus* ind sec^{-1} on-time) on the same range [0,1]. In doing so, both large-bodied and small-bodied species exert equal influence on all community analyses, be they univariate (e.g. species evenness) or multivariate (e.g. Bray-Curtis similarity) analyses.

Ordination was used to determine the magnitude and direction of change in species composition among years within rivers. In order to make meaningful visual contrasts of the magnitudes of compositional changes across years, among rivers, we used Principal Coordinates Analysis (PCO), which plots samples in a 2D space defined by absolute (dis)similarities. Permutation tests following Canonical Correspondence Analysis (CCA) enabled tests for significant change across years. Vector plots and SIMPER analysis (Clarke 1993) were used to identify species making the greatest

contribution to interannual change. Ordination and SIMPER were carried out on Bray-Curtis similarities. Packages vegan and labdsv were required for multivariate analyses (Roberts 2016, Oksanen et al. 2018).

Analysis of fish community dynamics for the present report focused on the impact of the 2016 flood and associated environmental hypoxia. Once the above multivariate analysis had facilitated identification of which species were most affected by the hypoxic flows, we proceeded to univariate analysis, whereby we modelled changes in catch-per-unit-effort (CPUE) of a species as a function of dissolved oxygen and discharge using linear mixed effects models (aka hierarchical or multilevel models). In these models the random factors were River and Site, and our minimal random term was to have Site nested within River.

Two fixed variables were included, one of which was the number of days each year (1 July – 31 June) when mean daily discharge exceeded the 75th percentile of the long-term distribution of daily discharges for that river. Mean daily discharge data from different rivers were all placed on a comparable scale by transforming discharge values of each river into random log-normal deviates. This flow variable was selected from a larger set of flow variables following graphical analysis of the relationships between fish cpue and flow variables. Henceforth, we refer to this variable as the number of high-flow days, *F*.

The second fixed variable was designed as an indicator of the magnitude of the hypoxic event within each river. Three variables were developed: the number of days when (1) mean daily DO was below 6 mg L^{-1}; (2) mean daily DO was below 4 mg L^{-1}; (3) minimum daily DO was below 4 mg L^{-1}.

Our model selection strategy broadly followed that of Zuur et al. (2009) and consisted of four steps. The criteria by which models were selected at each step included Akaike Information Criteria (AIC) and, more importantly, visual examination of model fits through plotting of normalised residuals against fitted values and environmental variables (Pinheiro and Bates 2000). First we wished to determine which of the three DO variables best explained variance in fish cpue. To do this we fit the following model to the data using restricted maximum likelihood:

$$C_{ijk} = (\alpha + \alpha_{jk}) + \beta_1 O + \beta_2 F + \beta_3 O \cdot F + \varepsilon_{ijk}; \qquad \varepsilon_{ijk} \sim N(0, \sigma^2) \qquad (1)$$

where C_{ijk} is the cpue of a species at site k (k = 1, 2,…,10) within river j (j = 1, 2,…,5); α is the fixed intercept and α_{jk} is the random intercept with zero mean and standard deviation to be estimated as a parameter. Variables O and F are, respectively, one of the three DO variables described above and the number of high-flow days. β_1 to β_3 are slope parameters, and ε_{ijk} are within-group errors.

Following the selection of the DO variable that best explained species' cpue, the second step was to determine whether random slopes for the DO variable improved the fit of the models. This was done comparing Models 1 and 2:

$$C_{ijk} = (\alpha + \alpha_{jk}) + (\beta_1 + \beta_{1,jk})O + \beta_2 F + \beta_3 O \cdot F + \varepsilon_{ijk}; \qquad \varepsilon_{ijk} \sim N(0, \sigma^2) \qquad (2)$$

where $\beta_{1,jk}$ is the random slope of the DO effect within each site, within each river, with zero mean and standard deviation to be estimated as a parameter. After determining the best model from Steps 1 and 2, we then the determined whether heterogeneity was present and whether model fit could be improved by adding variance functions to either Model 1 or 2. Assuming Model 2 provides the better fit, we then added the following variance functions:

$$C_{ijk} = (\alpha + \alpha_{jk}) + (\beta_1 + \beta_{1,jk})O + \beta_2 F + \beta_3 O \cdot F + \varepsilon_{ijk}; \qquad \varepsilon_{ijk} \sim N(0, \sigma_j^2 \exp(2\delta O_{ijk})) \quad (3a)$$

$$C_{ijk} = (\alpha + \alpha_{jk}) + (\beta_1 + \beta_{1,jk})O + \beta_2 F + \beta_3 O \cdot F + \varepsilon_{ijk}; \qquad \varepsilon_{ijk} \sim N(0, \sigma_j^2 \exp(2\delta F_{ijk})) \quad (3b)$$

$$C_{ijk} = (\alpha + \alpha_{jk}) + (\beta_1 + \beta_{1,jk})O + \beta_2 F + \beta_3 O \cdot F + \varepsilon_{ijk} ; \quad \varepsilon_{ijk} \sim N(0, \sigma_j^2 \exp(2\delta[O_{ijk} + F_{ijk}]))$$
(3c)

Models 3 all allow for heterogeneity among rivers while, in addition, Models 3a to 3c allow for, respectively, residual heterogeneity to be an exponential function of (a) DO; (b) number of high-flow days; and (c) the sum of both variables.

Following specification of the random structure of the models, the fourth and final step was to test whether model fits could be improved by removing fixed terms. Mixed-effects modelling was carried out using the nlme package (Pinheiro et al. 2017), with supporting functions provided by the sjstats package (Lüdecke 2018).

Further graphical univariate analyses were carried out to compare and contrast mean species richness, mean Pielou's evenness, and mean nativeness among years within Selected Areas, and among Selected Areas. Mean (and SEs, from which CIs are obtained) values of these statistics were calculated treating sites within years (n = 10) as replicates. Species richness refers to the number of species within an area. Pielou's evenness has range [0,1] and equals 1 when all species' abundances of an assemblage are equal (1/n if there are n species) and so, in conjunction with richness, helps provide a picture of how well represented each species is, within the local assemblage of a Selected Area. Nativeness is defined within the fish Basin Matter as the proportion of total range standardised CPUEs comprised of native species. For these three variables we infer a 'significant' difference between two groups when 95% confidence intervals of those groups do not overlap. Note that LTIM nativeness and SRA nativeness are not quantitatively comparable as they are based on different sampling protocols.

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