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Lower Murray
2019-20 Technical Report

A report prepared for the Commonwealth Environmental Water Office by the South Australian Research and Development Institute, Aquatic Sciences

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Cover photos: microinvertebrate (Cladocera) – the University of Adelaide; Coorong aerial shot – DEW; others – SARDI.

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EXECUTIVE SUMMARY

From 2019-20 to 2021-22, the Commonwealth Environmental Water Office (CEWO) Monitoring, Evaluation and Research (MER) Project will monitor and evaluate ecological outcomes of environmental water delivery in the Lower Murray, along with six other Selected Areas in the Murray–Darling Basin (MDB). This project, with an integrated research component, extends the monitoring activities of the Long-term Intervention Monitoring Project (2014-15 to 2018-19) and aims to demonstrate the ecological outcomes of Commonwealth environmental water delivery and support adaptive management.

During 2019-20 (a hydrologically dry year), ~750 GL[a] of Commonwealth environmental water was delivered to the main channel of the Lower Murray, in conjunction with other environmental flows (e.g. the Murray–Darling Basin Authority's The Living Murray Initiative), coordinated through a series watering events across the southern MDB to achieve multi-site environmental outcomes. Environmental water contributed to 39% of the total annual flow volume to the Lower Murray River (LMR) (at South Australian border, QSA), of which 81% was Commonwealth environmental water. Environmental water delivery largely consisted of return flows from upstream watering events (e.g. in the Murray and Goulburn rivers) and promoted winter (up to ~11,000 ML/d) and spring (up to ~15,600 ML/d) flow pulses in the LMR. Environmental water was also delivered to the LMR via direct trades from summer–late autumn, mainly to support continuous flows to the Lakes and Coorong. Barrage flows in 2019-20 were comprised of 100% Commonwealth environmental water.

Nine indicators were used to evaluate the ecological response to Commonwealth environmental water in the Lower Murray. Three indicators (*Hydrology (channel)*, *Stream Metabolism and Water Quality* and *Fish (channel)*) primarily aimed to evaluate Basin-scale objectives and outcomes, and in some instances, also local (Selected Area) objectives, following basin-wide standard protocols. Six indicators (*Hydraulic Regime*, *Matter Transport and Coorong Habitat, Littoral Vegetation Diversity and Productivity, Microinvertebrate Assemblage, Murray Cod Recruitment* and *Flow-cued Spawning Fish Recruitment*) aimed to address local evaluation questions, using area-specific methods. Additional contingency monitoring of lamprey migration was conducted during winter–summer 2019.

Key findings and ecological outcomes

Environmental water delivery contributed to some ecological improvements in the Lower Murray Selected Area in 2019-20:

- **Connectivity**: Commonwealth environmental water improved longitudinal hydrological connectivity via increasing annual flow by 32% in the LMR, meeting the Basin-wide environmental watering target of >30% increase in flows in the Murray River (calculated at the SA border). Commonwealth environmental water contributed to lateral connectivity throughout the water year, increasing the maximum inundation area, occurring in October 2019, by 517 ha.
- **Hydraulic diversity**: Commonwealth environmental water increased the duration and extent of 'flowing water' (lotic) habitat, with an extra 34 km (10%) of the LMR

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[a] Environmental water volumes and percentages provided here exclude wetland pumping as this investigation focuses on the main channel of the LMR.

characterised by mean water velocities >0.3 m/s for at least 30 days. This hydraulic variability may have benefited native animals (e.g. Murray cod) adapted to lotic riverine environments.

- **Water level variability**: Environmental flows, in combination with weir pool manipulations, increased water level variability (interquartile range) increased by 0.13 m in the tailwaters (i.e. just downstream of each weir) of the LMR.
- **Littoral vegetation**: Native plant species diversity increased (38–100%) at multiple spatial scales following the inundation of littoral zones by spring flows, and river red gum seedling gemination was supported in the LMR. Variable water levels also produced conditions suitable for the recruitment of specialised riparian species, therefore increasing plant functional diversity.
- **Water quality**: Commonwealth environmental water increased water mixing (velocities >~0.2 m/s) and oxygen exchange at the water surface, reducing the risk of low dissolved oxygen across an extra 104km for 30 days. This was particularly the case during spring–summer – the period that corresponds with highest ecosystem respiration rates and the primary reproductive season of many species that generally favour DO >5 mg/L.
- **River productivity**: Primary production, which supports aquatic food webs (e.g. invertebrates and fish), increased slightly (1%) in response to the physical changes generated by the addition of environmental water as the influence on channel volume was constrained by generally stable weir pool levels in the regulated LMR.
- **Matter transport**: Environmental water moderately increased the transport of nutrients, which would likely have stimulated primary and secondary productivity in downstream ecosystems, providing potential benefits to food webs of the LMR, Lower Lakes, Coorong and Southern Ocean, adjacent to the Murray Mouth.
- **Microinvertebrates**: Despite slight increases in primary production in the LMR main channel due to environmental water, the diversity and density of microinvertebrates increased during spring–summer by 11% and 23%, respectively, and the density of preferred prey species of large-bodied native fish larvae increased by 37% during spring 2019 due to environmental water.
- **Murray cod recruitment**: There was strong recruitment of Murray cod suggested by high abundance of young-of-year (YOY, age 0+). The increased extent of favourable (lotic) habitat by the spring flow pulse during the spawning/early larval period, and increased larval food resources, may have supported spawning and recruitment in this species.
- **Barrage flows**: Continuous barrage flows (including for fishway operations) were maintained by Commonwealth environmental water (100%) in this dry year.
- **Lamprey migration**: Barrage flows facilitated connectivity between freshwater, estuarine and marine environments, and promoted lamprey migration. During winter–spring 2019, moderate–high abundances of pouched and short-headed lamprey, relative to previous years, were found passing the Murray barrages with migrations that continued for 100's of kilometres upstream.
- **Salt export and reducing import**: Commonwealth environmental water substantially increased salt export out of the Basin (modelled 624,000 tonnes in 2019-20), reduced salt import into the Coorong (2 million tonnes), and reduced salinity levels in the Coorong, which was considered to be crucial for maintaining ecosystem health and species diversity.

- ***Ruppia* and fish habitats**: Environmental water substantially increased favourable fish habitat for estuarine species (e.g. 40% increase in the area of suitable habitat for mulloway in 2019-20 due to environmental water deliveries from 2017-18 to 2019-20). Over the three years, environmental flows also led to some improvements in habitat suitability for *Ruppia* seed production and life-cycle completion in the southern Coorong.
- **Flow-cued spawning fish**: No YOY of golden perch were detected in the LMR during autumn 2020, suggesting localised recruitment failure and a lack of immigration from spatially distinct spawning sources (e.g. mid Murray or Darling rivers).
- **Fish assemblage**: With low in-channel flows (<18,000 ML/d) in the LMR since the 2016-17 flood, the current (2020) fish assemblage in the main channel represents one typical of low flows, with high abundances of small-bodied species, and a lack of recruitment of native, large-bodied flow-cued spawners.

Key learnings and management implications

- In the highly regulated LMR, environmental water can be used to help reinstate key features of the natural hydrograph to support hydrodynamic and ecosystem restoration; for example, to reinstate winter freshes and in-channel spring–early summer flow pulses <20,000 ML/d.
- It is increasingly evident that reaching and sustaining flows >20,000 ML/d in the LMR is challenging with existing volumes of environmental water and delivery constraints, even when coordinating flow deliveries across the southern MDB. This has not been achieved under regulated conditions during the past six years. Under wetter scenarios, flows >20,000 ML/d may be achieved by delivering environmental water in conjunction with unregulated flows.
- In the LMR, increasing flows to 20,000–45,000 ML/d significantly improves hydraulic conditions (e.g. increased velocity and water level). Weir management, particularly lowering, could also be considered to complement flows to achieve hydraulic rehabilitation and promote lotic conditions. To inform flow management and maximise ecological outcomes, however, we need to better understand the effect of specific aspects of flow (e.g. timing, magnitude and duration) on ecological processes and the hydraulic requirements of flow-dependant species.
- Evidence from this and allied investigations suggest that under current constraints and weir operating regimes, flows >20,000 ML/d are likely required to significantly influence golden perch spawning and recruitment in the LMR.
- Environmental water delivery that promotes longitudinal and lateral connectivity will enhance productivity in the LMR. Lateral connectivity may increase carbon and nutrient inputs while longitudinal connectivity will help facilitate the transport and dispersal of matter and aquatic biota (e.g. aquatic plant propagules, microinvertebrates, fish larvae).
- In the regulated LMR, the influence of environmental flows on riverine production is largely restricted by stable water levels set by weirs. To improve riverine productivity, water deliveries in conjunction with more natural water level changes are desirable.
- Environmental flows are pivotal in maintaining barrage flows and end-of-system connectivity in the MDB, particularly during low flow periods, when there would

otherwise be negligible water and matter exchange between the Lower Lakes and Coorong. This is critical for a functioning river system and supporting species' life history processes (e.g. migration of diadromous fish).

- Barrage flows play a key role in salt export from and reducing salt import to the MDB, maintaining estuarine habitat (e.g. for *Ruppia* and fish), ecosystem functions and biodiversity in the Coorong and reducing the risk of Murray Mouth closure.
- The timing of environmental flow delivery should continue to align with ecological objectives and consider biological processes and life history requirements (e.g. reproductive season of flow-dependent species in spring/summer, spawning migration of diadromous fishes in winter/spring, or reducing salinities and maintaining water levels in the Coorong during summer/autumn).
- Flow management should consider the source of water (i.e. origin), when possible, which can influence water quality (e.g. turbidity, dissolved organic carbon, the amount and form of nutrients), ecological processes (e.g. primary/secondary productivity) and subsequent biological responses.
- Furthermore, maintaining flow integrity from its source (e.g. Darling River, Murray upstream or major tributaries) to the end of the Murray River system is important to support broad-scale ecological processes and outcomes (e.g. improved productivity, migration of diadromous species, enhanced spawning, larval fish dispersal and recruitment of flow-dependent species).

More specific management considerations are provided in Sections 2 and 3, based on ecological outcomes and findings from indicators.

1 INTRODUCTION

1.1 Flow regimes and riverine ecology

River regulation and flow modification have severely impacted riverine ecosystems throughout the world, including the Murray–Darling Basin (MDB) (Maheshwari *et al.* 1995; Kingsford 2000; Grill *et al.* 2019). The southern MDB is highly regulated, where natural flow regimes have been substantially altered, leading to decreased hydrological (e.g. discharge) and hydraulic (e.g. water level and velocity) variability, and reduced floodplain inundation (Maheshwari *et al.* 1995; Bice *et al.* 2017). The Murray River downstream of the Darling River junction is modified by a series of low-level (<3 m) weirs (Figure 1), changing a connected flowing river to a series of weir pools (Walker 2006). The flow regime has been further exacerbated by upstream diversions and increased extraction. These have had profound impacts on riverine processes and ecosystems (Walker 1985; Walker and Thoms 1993; Wallace *et al.* 2014).

Flow regimes play a critical role in determining the distribution and abundance of native aquatic biota (Koehn *et al.* 2020a; 2020b), and the ecological integrity of floodplain rivers (Junk *et al.* 1989; Poff *et al.* 1997; Bunn and Arthington 2002). Lotic (flowing water) habitats, characteristic of the Murray River before weir construction, are integral to the ecological and life history processes of many native biota that are adapted to flowing riverine environments. For example, they provide stimuli for the spawning of flow-cued species (e.g. silver perch *Bidyanus bidyanus*) (Tonkin *et al.* 2019), facilitate downstream drift and transportation of plankton, macroinvertebrates and fish larvae, and provide diverse hydraulic habitats that are suitable for a range of species (e.g. Murray cod, *Maccullochella peelii* and Murray crayfish, *Euastacus armatus*) (Mallen-Cooper and Zampatti 2018). Increased variability in water levels improve lateral connectivity and increase transport of material from off-channel habitats to enhance productivity and support food webs (Baldwin *et al.* 2016), benefit fringing and floodplain vegetation (e.g. Cooling *et al.* 2010), and assist in the regular “re-setting” of biofilms (Steinman and McIntire 1990), which are key components of riverine food webs.

In the MDB, environmental flows have been used to re-establish key features of the natural flow regime (MDBA 2012; Koehn *et al.* 2014; Webb *et al.* 2017). In South Australia, the main channel of the Lower Murray River (LMR) represents a significant ecological asset to be targeted for environmental watering (MDBC 2006; DEWNR 2015). To achieve the greatest ecological benefits from available environmental water, it is important to understand biological and ecological responses to flow regimes. This provides critical knowledge to underpin environmental flow management in the LMR.

1.2 CEWO Monitoring, Evaluation and Research Project

From 2014-15 to 2018-19, the five-year Commonwealth Environmental Water Office (CEWO) Long-Term Intervention Monitoring (LTIM) Project was conducted to monitor and evaluate ecological outcomes of Commonwealth environmental water delivery in the MDB. The project was implemented across seven Selected Areas throughout the MDB, including the Lower Murray, to assess and evaluate both Basin-scale and Selected Area (local) responses to environmental flows. The overall aim of this project was to demonstrate the ecological outcomes of Commonwealth environmental water delivery and support adaptive management. The current CEWO Monitoring, Evaluation and Research (MER) Project extends the LTIM Project activities to June 2022.

In the Lower Murray, the CEWO MER Project (2019-20 to 2021-22) focuses on the main river channel between the South Australian border and Wellington (LMR), with one indicator (i.e. Matter Transport and Coorong Habitat) extending to the Lower Lakes and Coorong (Figure 1). The riverine monitoring sites (for indicators) cover three geomorphic zones (floodplain, gorge and swamplands) (Figure 1).

A total of nine indicators were established to assess ecological responses to environmental water delivery in the Lower Murray. Three indicators (***Hydrology (channel)***[b], ***Stream Metabolism and Water Quality*** and ***Fish (channel)***) followed standard protocols to support quantitative Basin-wide and Selected Area evaluation, where applicable (Hale *et al.* 2014). Six indicators (***Hydraulic Regime***, ***Matter Transport and Coorong Habitat***, ***Littoral Vegetation Diversity and Productivity, Microinvertebrate Assemblage, Murray Cod Recruitment*** and ***Flow-cued Spawning Fish Recruitment***) were developed to address Selected Area-specific objectives and test a series of hypotheses with respect to biological/ecological response to environmental flows. Under the MER Project, an integrated research project explores the links between key indicators to improve our understanding of how flow influences the ecological processes that drive recruitment of key fish species (Murray cod). Contingency monitoring activities (e.g. monitoring lamprey migration in 2019-20) are also being undertaken in response to opportunities as they arise to support current monitoring and/or to inform environmental water use planning and management.

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[b] Hydrology (Channel) does not directly address any specific CEWO evaluation question, but provides fundamental information for analysis and evaluation of monitoring outcomes against hydrological conditions and environmental water delivery for all other indicators. Results for this indicator are presented in Section 1.4.

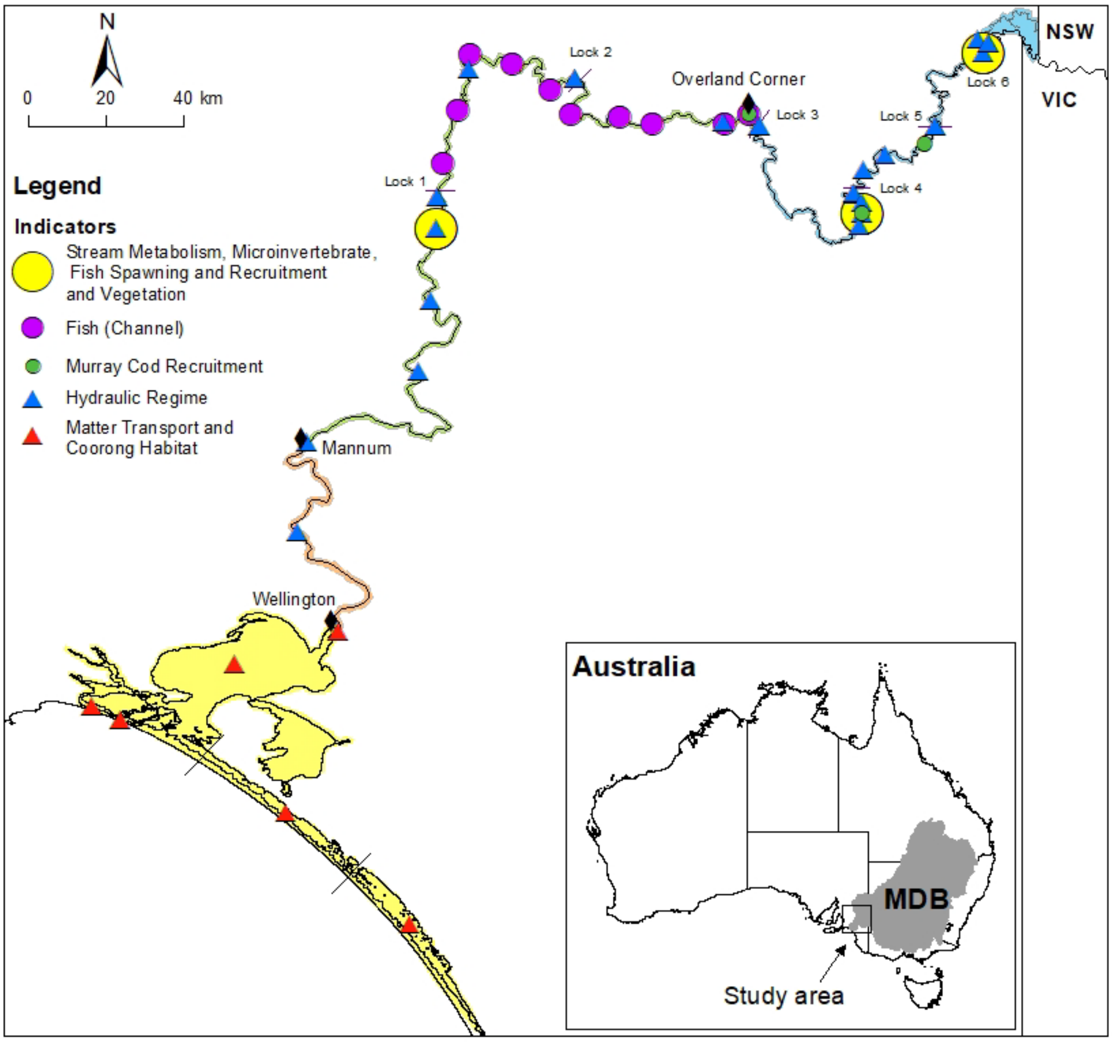

Figure 1. Map of the Lower Murray Selected Area showing the Lower Murray River floodplain (blue), gorge (green) and swamplands (orange) geomorphic zones, and the Lower Lakes and Coorong (yellow). Sampling sites are indicated by coloured circles (field monitoring) and triangles (modelling). Larval fish sampling for Fish Spawning and Recruitment was not undertaken in 2019-20.

1.3 Expected outcomes in the Lower Murray

For the period of the MER Project (2019-20 to 2021-22), it is expected that the majority of the Commonwealth environmental water deliveries to the Lower Murray will contribute to base flows and freshes in the LMR channel (Figure 2), and maintain river flows to the Lower Lakes and Coorong. These particular flows aim to achieve a variety of environmental outcomes including those relating to fish, vegetation, birds, water quality and river function, Lower Lakes water levels, salt export and connectivity between freshwater, estuarine and marine environments (Appendix A), although only some of these are monitored through this project.

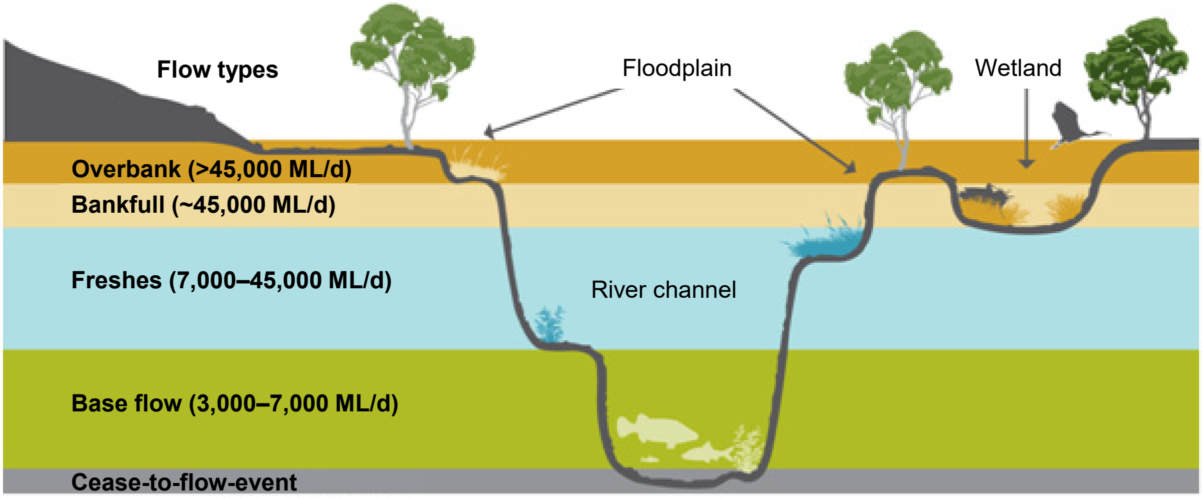

Figure 2. The various flow types of the Lower Murray River as described in the Murray–Darling Basin Plan (MDBA 2011). This diagram represents an idealised, unconstrained river reach.

In the Lower Murray, environmental water delivery that contributes to base flows and freshes increases stream velocity, mixing and dilution; increases variability in water levels; increases the inundated area of littoral zone of channels, low-lying wetlands and floodplains; and improves connectivity between freshwater, estuarine and marine environments (Ye *et al.* 2020). These changes to hydrological/hydraulic conditions in the LMR are expected to lead to:

- Maintained dissolved oxygen and water quality due to increased mixing and discharge;
- Increased productivity due to lateral transport of organic material;
- Increased transport of dissolved and particulate matter (salt and nutrients) downstream due to mobilisation and increased discharge;
- Increased littoral understorey vegetation diversity, productivity and community resilience due to increased water levels;
- Increased microinvertebrates (and egg-bank) diversity and abundance due to increased inundated area in littoral and off-channel habitats from increased water levels and discharge;
- Increased larval abundance of flow-cued spawning fish species (golden perch *Macquaria ambigua* and silver perch) due to the provision of flow-cues for spawning and increased larval drift and dispersion;
- Increased recruitment of flow-cued spawning fish species due to increased spawning and larval drift, and enhanced survival rate due to increased productivity;
- Improved recruitment and population resilience of main channel specialist fish species (Murray cod) due to increase in lotic habitat and productivity;
- Increased salt export out of the MDB; reduced salt import into the Coorong, and reduced salinities; and
- Improved fish habitats in the Coorong, and also *Ruppia tuberosa* habitats at higher flow.

Over the long-term (decades), environmental water delivery is expected to make a significant contribution to achieving ecosystem outcomes in the Lower Murray, through restoring ecological processes and improving habitat for biota in the main channel and floodplain/wetlands. A consolidated view of the expected outcomes driven by flow for the Lower Murray is presented in Figure 3 below, which includes core monitoring indicators of the MER Project. The conceptual diagram demonstrates the inter-relationships between the changes of hydrological/hydraulic regime and riverine productivity (stream metabolism, vegetation, microinvertebrates) and matter transport, and how these may influence fish spawning and recruitment and the overall fish assemblage in the Lower Murray.

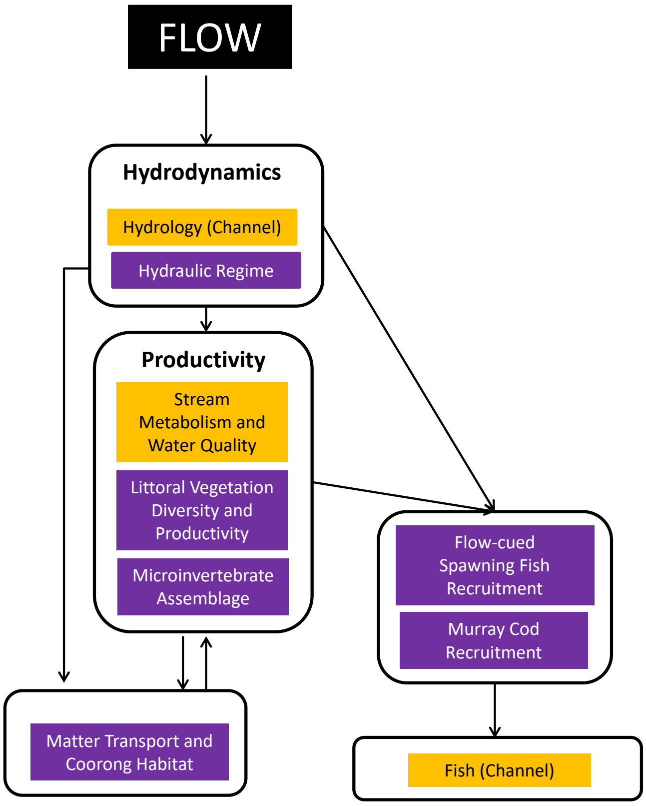

Figure 3. Cause and effect diagram of flow for the main channel of the Lower Murray with respect to the proposed indicators. Magnitude, timing and duration are factors of flow (in black). Yellow indicators followed standard protocols to support quantitative Basin-wide and Selected Area evaluation, where applicable. Purple indicators were developed to address objectives and test Selected Area-specific hypotheses with respect to biological/ecological response to environmental flows.

1.4 Environmental water delivery

Since 2011-12, environmental water has been delivered to the Lower Murray (Figure 4), to restore ecological health (www.environment.gov.au/water/cewo). During the LTIM and MER projects (2014-15 to 2019-20), an average of ~700 GL/year of Commonwealth environmental water has been delivered to the LMR, in conjunction with other environmental flows (i.e. water from The Living Murray (TLM) Initiative, Victorian Environmental Water Holder, River Murray Increased Flows, and New South Wales Department of Planning, Industry and Environment) (Table 1; Ye *et al.* 2020), with an average of 602 GL/year of Commonwealth environmental water flowing through the barrages into the Coorong (Table 2). Deliveries to the LMR largely occurred as return flows, during winter and spring–early summer, through coordinated watering events across the southern connected Basin to achieve multi-site environmental outcomes. Direct orders of environmental water to the South Australian border also occurred for specific purposes, often during summer–autumn, to provide flow for the Lakes and Coorong.

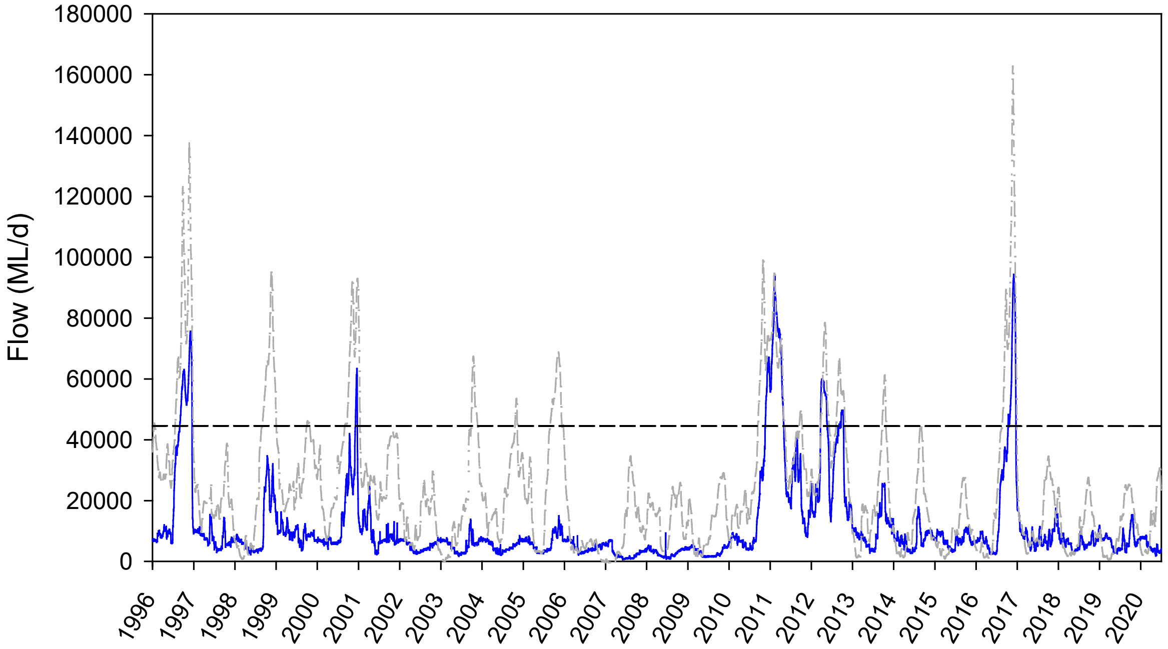

Figure 4. Daily flow (ML/d) in the Lower Murray River (LMR) at the South Australian border (blue solid line) from January 1996 to July 2020, compared to modelled flow under natural conditions (grey dashed line). Approximate bankfull flow in the main channel of the LMR is shown (black dashed line).

Table 1. Total annual volumes (gigalitres, GL) of environmental water (eWater), including Commonwealth environmental water (CEW), delivered to the Lower Murray River (LMR) channel (excludes wetland use*) and the proportion contribution towards total flow to the LMR (QSA). Volumes are provided by the CEWO, include the environmental components of the South Australian entitlement and exclude environmental water delivered from the SA Minister for Environment and Water and Accolade Wines (predominantly for wetlands). TLM = The Living Murray, VEWH = Victorian Environmental Water Holder, RMIF = River Murray Increased Flows, NSW DPIE = New South Wales Department of Planning, Industry and Environment.

Water year	2014-15	2015-16	2016-17	2017-18	2018-19	2019-20
CEW	581	798	618	898	549	750
TLM	107	101	234	176	96	68
VEWH	26	15	43	30	35	60
RMIF			100	53	111	52
NSW DPIE				9		
Total eWater	714 (25%)	914 (37%)	996 (11%)	1167 (43%)	791 (32%)	931 (39%)

* A total of 9.7 GL of CEW was also delivered for wetland watering in 2019-20.

Table 2. Annual flow over the Murray barrages (total volume, GL) from 2014-15 to 2019-2020, showing contribution by Commonwealth environmental water (CEW). CEW and total flow volumes are based on South Australian barrage dashboard accounting data. Matter transport results in Section 2.3.1 are based on different modelled data, and may not necessarily reflect the accounted data presented here.

Water year	2014-15	2015-16	2016-17	2017-18	2018-19	2019-20
Total	987	561	6484	854	377	685
CEW	454	561	802	757	377	685

In 2019-20, flow remained in-channel and was similar to four of the previous five years, which were hydrologically dry (i.e. flow remained <18,000 ML/d at the South Australian border, Figure 4). During this year, ~931 GL of environmental water (excluding wetland use, 39% of the total flow), including ~750 GL of Commonwealth environmental water, was delivered to the LMR (Table 1).

Environmental water delivered to South Australia from mid-July to mid-August 2019, largely supported by flow from the Goulburn River, contributed to a winter flow pulse of 11,000 ML/d in the LMR (Figure 5). Similarly, environmental water delivered between late September and early November 2019, supported by return flows from the Murray and Goulburn rivers and Barmah–Millewa Forest, promoted an in-channel spring flow pulse peaking around 15,600 ML/d in mid-October 2019 (Figure 5).

Environmental water delivery to the LMR from summer–late autumn comprised of direct trades[c] at the South Australian border (Figure 5a). During this period, Commonwealth environmental water played a critical role in maintaining barrage releases (Table 2). Barrage flows in 2019-20 were entirely comprised of (100%) Commonwealth environmental water.

The physical source of flows to the LMR during 2019-20 are presented in Figure 5b. Flow to South Australia mainly comprised flow from the upper Murray River, with greater proportional flow from Lake Victoria, resulting from direct trades, after December 2019.

Key watering events in the LMR during 2019-20 and the targeted expected outcomes of these deliveries are presented in Appendix A. Environmental water also supported other complementary management actions to achieve ecological outcomes in the Murray River; key activities from downstream of the Darling River junction to Wellington are summarised in Appendix B. These included manipulations of Weir Pools 2, 6, 7, 8 and 15, and wetland watering by pumping.

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[c] "Direct trade" refers to an order for a specified volume of environmental water to be delivered at the South Australian border. Typically a timing and profile for the delivery is specified and river operators can meet the order by providing water from any available source.

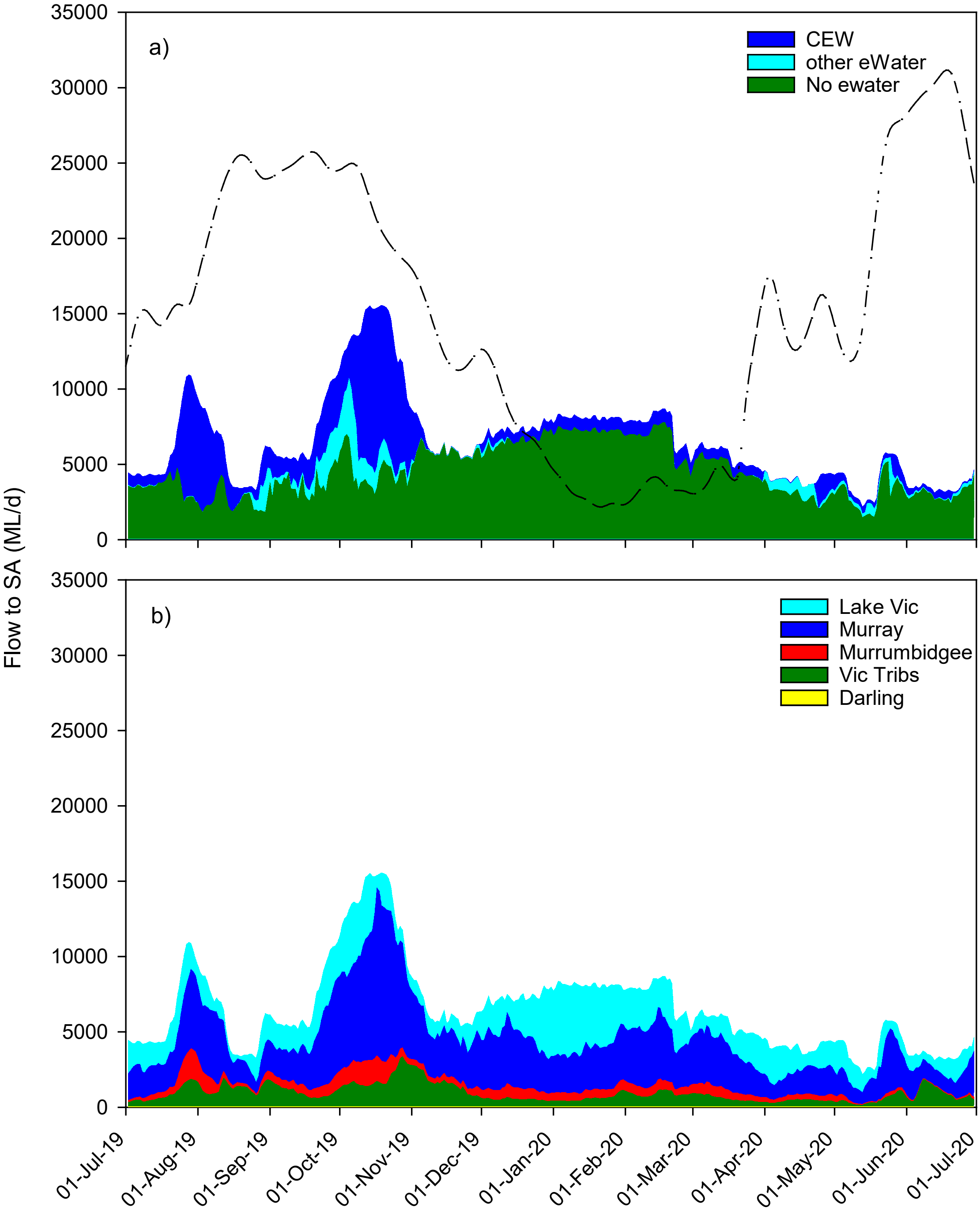

Figure 5. Flow to South Australia from July 2019 to July 2020 showing the (a) contribution of environmental water (eWater) and (b) source of all (environmental and consumptive) water (MDBA). CEW = Commonwealth environmental water. Modelled flow under natural conditions is shown by the dotted black line. 'Bigmod salinity routines' was used as a proxy for transport of biological matter, to estimate the proportion of the flow that originated at different upstream tributaries[d].

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[d] Molecules of water, nutrients, and the biological matter transported downstream often move slower than the wave front that is recorded as the change in flow discharge (Chow *et al.* 1988). To account for this, the MDBA has used Bigmod salinity routines as a proxy for transport of biological matter, to estimate the proportion of the flow at the South Australian border that originated at different upstream tributaries. While acknowledging potential difference in travel time between salt and other matter, this approach is preferred over estimating travel times based on observed changes in flow along the main channel.

1.5 Purpose of the CEWO MER report for 2019-20

This report presents the key findings from monitoring and research in the Lower Murray during 2019-20, and answers CEWO evaluation questions about ecological responses to Commonwealth environmental water deliveries (Sections 2, 3 and 4). Refer to previous annual reports (Ye *et al.* 2016b; 2017; 2018; 2019; 2020) for Lower Murray monitoring findings from 2014-15 to 2018-19, and the *Monitoring, Evaluation and Research Plan for the Lower Murray* (SARDI *et al.* 2019) for a detailed description of methods for current activities. Specific management recommendations for environmental flows in the Lower Murray are provided in Section 2, with general management implications summarised in Section 5, based on monitoring and evaluation outcomes, and expert knowledge. Findings from the integrated research project will be presented in a separate interim report and be incorporated into the final MER technical report (2021-22). Monitoring and evaluation of Commonwealth environmental water delivery in the Lower Murray focusses on the main watering period of spring–summer; therefore, our findings and recommendations on environmental water management are most relevant to this period.

2 INDICATORS

2.1 Hydraulic Regime

Background

The discharge, or *hydrology*, in the Lower Murray Selected Area was determined through routine monitoring. The hydrology expected to have occurred without environmental water components was determined by the Murray–Darling Basin Authority (MDBA) using a counterfactual modelling approach (Ye *et al.* 2020).

The *hydraulic* characteristics (e.g. depth/water level or flow velocity) of fluvial ecosystems result from the interaction of discharge and physical features (e.g. channel morphology, woody debris, man-made structures, etc.), and have a profound influence on river ecosystem structure and function (Statzner and Higler 1986; Biggs *et al.* 2005; Bice *et al.* 2017) (also see Section 1.1). It is these hydraulic characteristics that biota can sense and respond to, i.e. a change in velocity or water level, rather than a change in discharge.

The purpose of this indicator was to quantify the changes in hydraulics due to the delivery of environmental water using hydraulic models, to provide a basis to infer ecological changes caused by environmental water. This approach is particularly important in the LMR where a given discharge may not produce the same hydraulic response, as downstream structures (weirs) will also influence the hydraulics that are occurring.

Hypothesis

Commonwealth environmental water will promote a greater extent of lotic habitat as evidenced by increased water velocities and variability in water levels.

Methods

A steady-state modelling approach was adopted, similar to that used in the Goulburn (Webb *et al.* 2015) and Edward-Wakool (Watts *et al.* 2015) Selected Areas. For each weir pool within the Lower Murray Selected Area, i.e. Weir Pools 1 to 5, as well as the river between Lock 1 and Wellington, a range of steady state flow scenarios were simulated in the hydraulic models (2,000–100,000 ML/d) and a range of weir pool levels required to cover the range of conditions experienced. Models used for this analysis are outlined in McCullough *et al.* (2017) and Montazeri and Gibbs (2019). For each steady state scenario, a range of hydraulic metrics were computed, including the 10th, 50th and 90th percentile velocities within the weir pool, the proportion of the weir pool exceeding 0.2 and 0.3 metres per second (m/s), and water levels at regular locations along the weir pool. The former water velocity (>0.2 m/s) represents favourable velocities that entrain and transport/disperse phytoplankton, zooplankton and fish larvae (Gibbs *et al.* 2020) and allow gas exchange at the water surface (see Section 2.2 Stream Metabolism), and the latter (>0.3 m/s) represents flowing water (lotic) conditions for riverine biota (Bice *et al.* 2017).

To enable a consistent comparison of in-channel velocity changes due to environmental water, the same area was used for all velocity analyses. The area used for velocity analysis for each weir pool comprised the inundated area at flows of 5,000 ML/d and normal pool level conditions. As changes to in-channel velocities were the focus of this analysis, this approach of eliminating additional inundated areas was considered reasonable. Additionally, where the full inundated area was used, the disproportionate increase in area of slow flowing backwaters compared to in-channel area as discharge increased had a large impact on the overall proportion of the weir pool with low velocities.

Post-processing correction to the modelled water levels downstream of each lock was undertaken. A linear regression relationship between flow and the difference in modelled and recorded water level was used to correct for any systematic bias introduced by errors in the hydraulic model. Following this correction, the remaining residual error between the modelled and recorded data each day was applied to all scenarios, which represents random error introduced by other factors, such as wind setup. This results in modelled water levels that are the same as the observed water level downstream of each lock for the scenario representing observed conditions (*All Water*) with consistent corrections applied to the without environmental water scenarios.

Environmental water scenarios

With the lookup information derived from the hydraulic models, the time series of discharge for each of the environmental water scenarios presented in Section 1.4 and the downstream water level each day for each weir pool, time series of hydraulic parameters were interpolated using linear bivariate interpolation (R version 3.6.0 and akima package 0.6-2). Four scenarios have been considered based on the discharge data available, i.e. with all environmental water ("***All Water***", representing observed conditions), without Commonwealth environmental water ("***No CEW***"), without any environmental water ("***No eWater***"), and a representation of natural conditions, Without Development ("***WoD***"). The discharge time series for these scenarios were provided by the MDBA, and the data accounts for changes in diversions expected within South Australia by assuming full utilisation of the entitlements recovered for the environment in the without environmental water (*No CEW* and *No eWater*) scenarios. *WoD* results were not simulated downstream of Lock 1, due to limited information on the natural downstream water level for this scenario.

The observed water levels at each lock, and at Wellington, were used as inputs for the *All Water* scenario. For the without environmental water scenarios, the weir pool manipulation at Lock 2 was removed, and instead the water level was assumed to be at normal pool level during these periods. For the Below Lock 1 reach the influence of environmental water on the water level in the Lower Lakes was incorporated, based on MDBA water balance modelling and the recorded water level at Wellington.

Results

A summary of the results at the Lower Murray Selected Area scale can be seen in Figure 6. For the velocity metrics, only results above Lock 1 were presented here to enable a comparison against the representation of natural conditions from the *WoD* scenario. Figure 6 includes the discharge at the South Australian border for the different scenarios, the resulting area inundated (from Lock 6 to Wellington), and length of the river between Lock 6 and Lock 1 experiencing lotic conditions, based on thresholds of velocity >0.2 m/s and >0.3 m/s.

Velocity

The modelling indicates that there were substantial short-term changes in the length of river with velocities exceeding 0.2 m/s in 2019-20 due to Commonwealth environmental water. An additional 176 km (52% of the reach) exceeded this threshold for 2 weeks, and 104 km of river (31% of the reach) for a duration of 30 days (Figure 6). Increases in the proportion of the river with a velocity exceeding the higher threshold of 0.3 m/s were also modelled, 74 km (22%) for 14 days and 34 km (10%) for 30 days due to Commonwealth environmental water. This can be compared to the without development hydraulic conditions, where even in this low flow year the full reach was expected to experience cross-section averaged velocities exceeding 0.3 m/s for approximately 2 months. The velocity magnitudes are presented at a weir pool scale in Figure 7, with the median velocity in the weir pool each day shown as a solid line, and the range in velocities (as the 10th and 90th percentiles) shown as the shaded band. The results suggest a relatively consistent response across the weir pools to the two flow pulses created by environmental water, however, below Lock 1 the modelling indicated lower velocities due to the deeper river in this reach.

Water level and area

The inundation area expected for the different scenarios can be seen in the second panel of Figure 6, where flows were below bankfull level (approximately 45,000 ML/d), and hence changes in inundation due to environmental water above Lock 1 were limited. Small increase in area occurred when environmental water increased the discharge above 10,000 ML/d, or due to the weir pool raising at Lock 2. The increase in surface area due to environmental water is also due to higher water levels in the Lower Lakes, and all of the increase in area in the latter half of 2019-20 occurs below Lock 1. The increased water level in the Lower Lakes due to environmental water can be seen in the below Lock 1 panel in Figure 8, which is at Wellington for this downstream end of the river reach below Lock 1.

The upstream end of the weir pool is the least influenced by the downstream weir and hence most responsive to changes in discharge when the weirs are controlling water levels (below 54,000–67,000 ML/d, depending on the weir). Environmental water created some variability in water levels at the upstream end of each weir pool that would not have occurred otherwise, and this was complemented by weir pool manipulation at Lock 2. Overall, the variability in water levels tended to follow the rise in August and fall in November in the without development river height scenario, albeit without the sustained levels throughout this period (Figure 9).

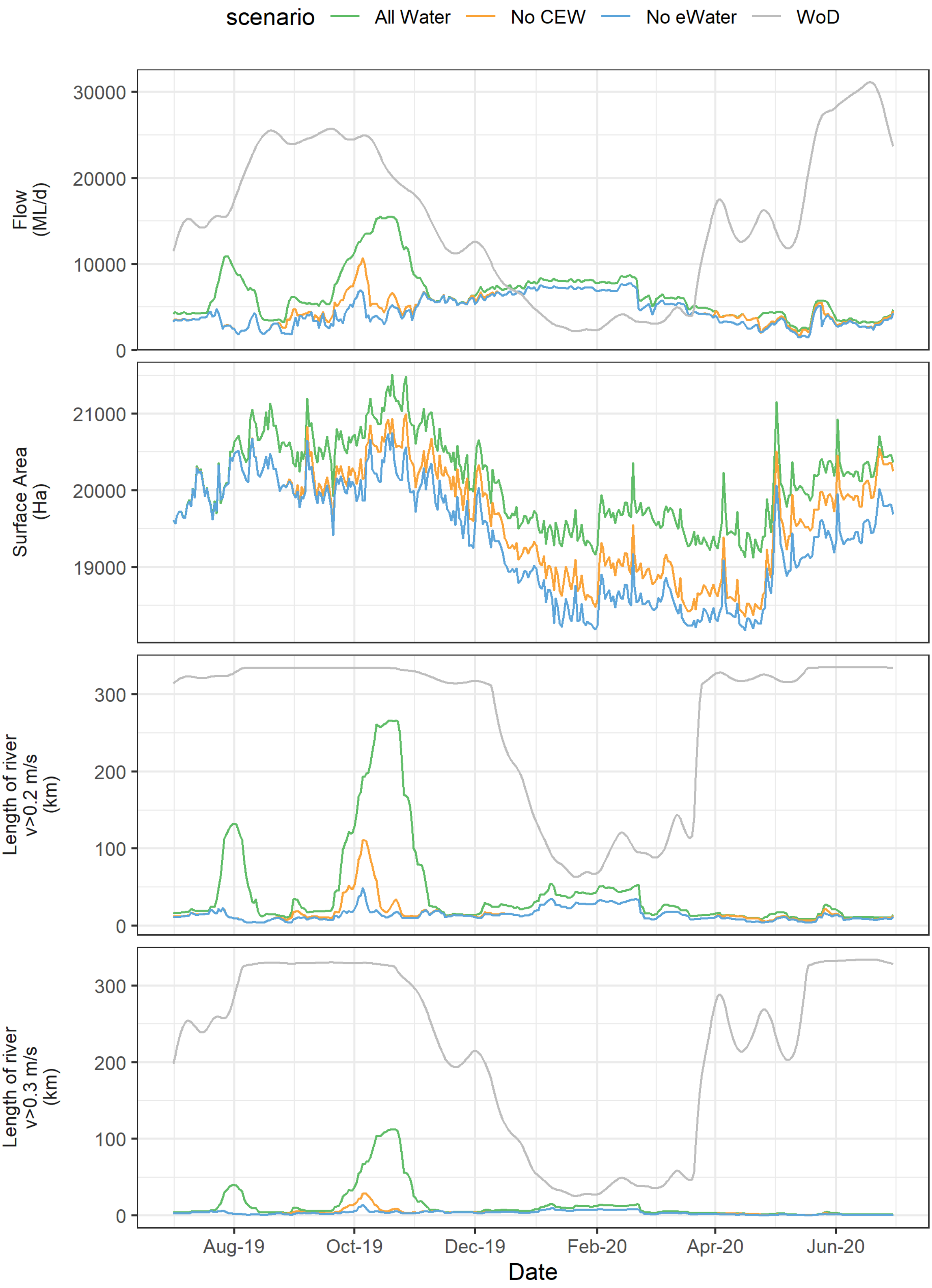


Figure 6. Discharge (flow to South Australia), inundated area between Wellington and Lock 6, and length of river with faster flowing velocities (v>0.2 m/s and v>0.3 m/s) for the Lower Murray River (LMR) between Locks 1 & 6 (excluding anabranches). Total length of river assessed for the length of river metrics in the LMR = 345 km.

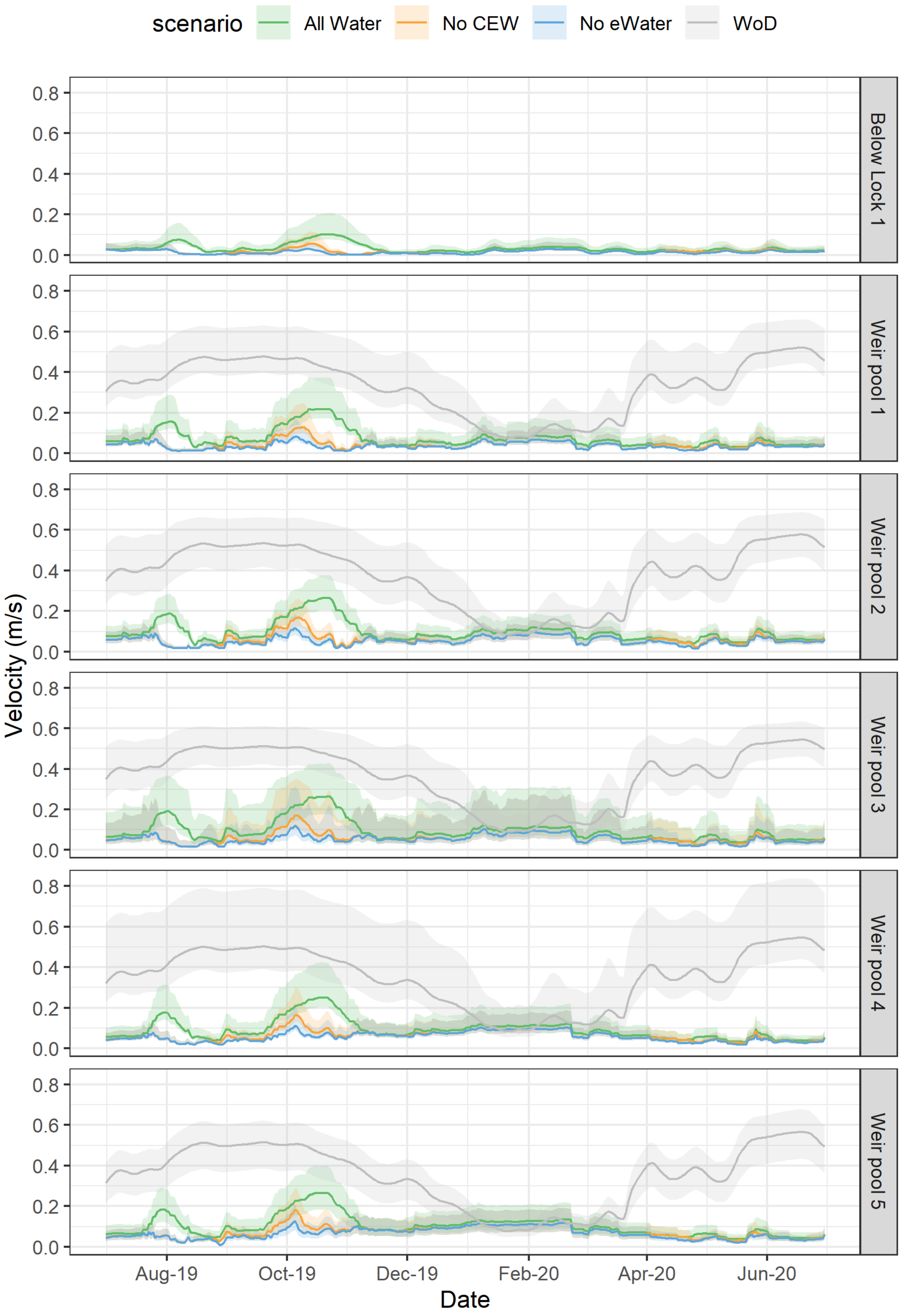


Figure 7. Median modelled velocity in each weir pool (line), with the range of velocities within the weir pool (the shaded area), defined by the 10th and 90th percentiles, in the Lower Murray River.

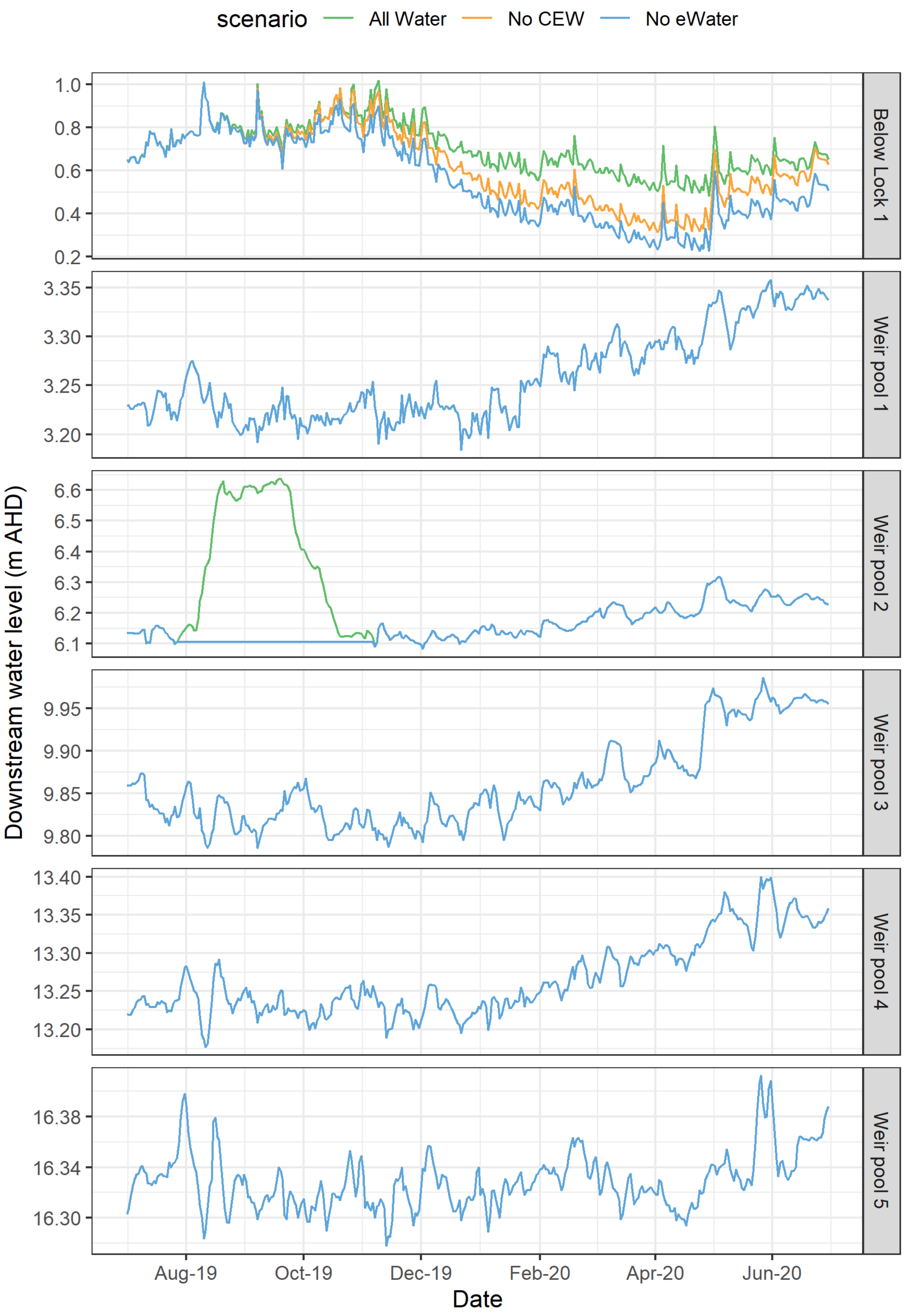


Figure 8. Modelled water level at the downstream end (i.e. at the lock and weir) of each weir pool in the Lower Murray River.

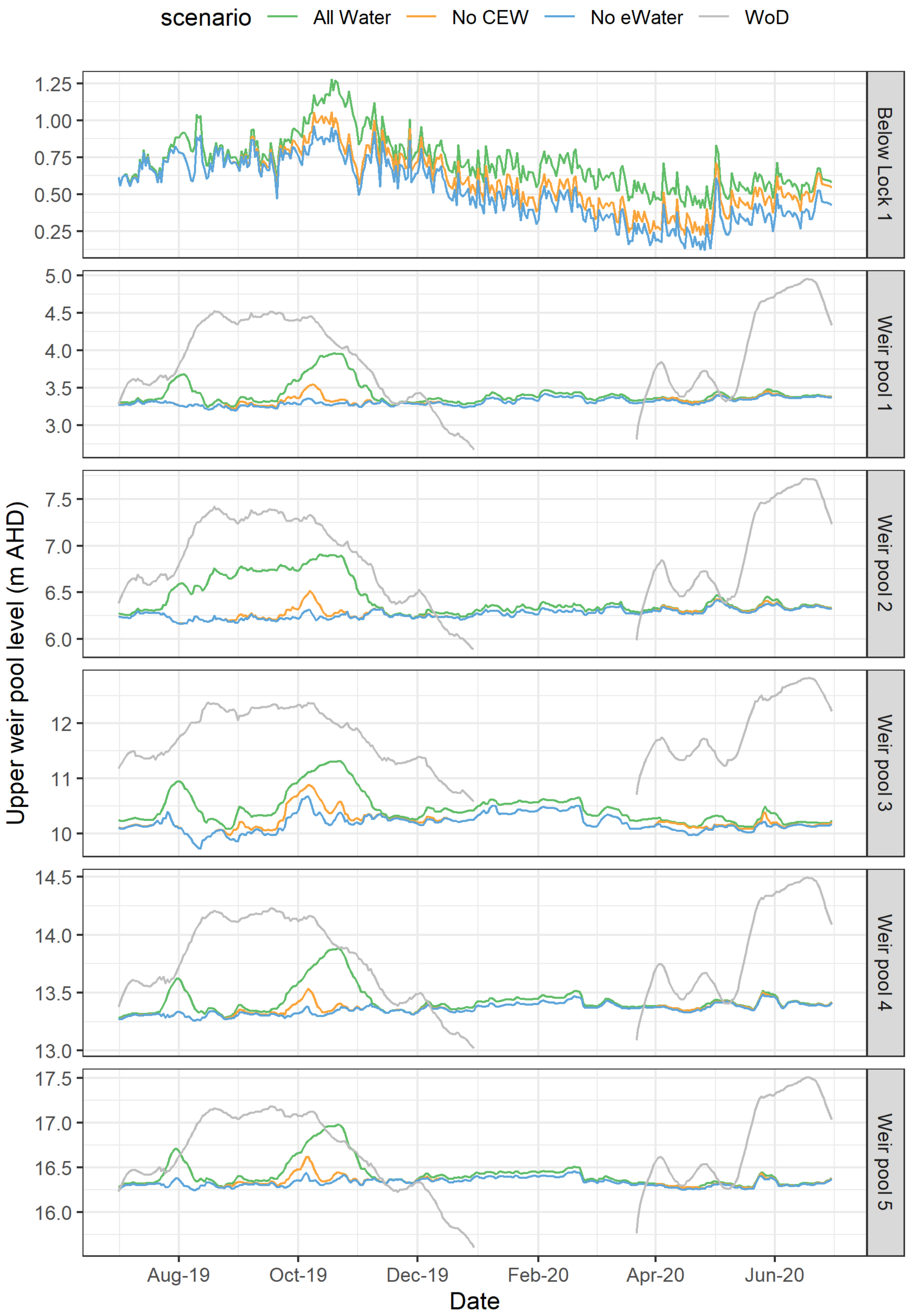


Figure 9. Modelled water level at the upstream end of each weir pool in the Lower Murray River.

Evaluation

To evaluate the contribution of Commonwealth environmental water towards a short- (annual) or long-term (multi-year) outcome, a contribution significance level was assigned to each evaluation question. The level was viewed as 'to what extent Commonwealth environmental water contributed towards that observed outcome, with the ecological significance of the outcome considered where possible'. For example, the level assigned may be similar for an ecologically significant outcome towards which Commonwealth environmental water made a minor contribution versus an outcome that was considered minor but towards which Commonwealth environmental water made a major contribution. The thresholds for assigning the significance vary among indicators and questions, ranging from using defined percentages or values of change, to qualitative assessment based on expert opinion.

Table 3. Hydraulic Regime evaluation questions and answers. CEW = Commonwealth environmental water, eWater = environmental water.

CEWO evaluation questions	Outcomes of CEW delivery					
	2014-15	2015-16	2016-17	2017-18	2018-19	2019-20
What did CEW contribute to hydraulic diversity within weir pools?	13 km, 4% (17 km, 5%)	18 km, 5% (22 km, 6%)	20 km, 6% (53 km, 15%)	36 km, 10% (49 km, 14%)	15 km, 4% (19 km, 6%)	An additional 34 km or 10% of lotic conditions created by CEW for at least 30 days (An additional 74 km or 22% for at least 14 days)
	CEW provided minor contributions towards increasing lotic habitat in the LMR over the last six years, except in 2017-18 and 2019-20, when CEW had a moderate contribution (an additional 34–36 km of river (10%) for 30 days). Length of river with lotic conditions (velocity >0.3 m/s) has been used to represent hydraulic diversity. If there is some flowing water, it is expected there will be greater hydraulic diversity due to changes in habitat complexity (bends, backwaters, benches, etc.). Length of river with lotic conditions exceeded for 30 days over the year is presented, and 14 days in brackets. The time periods represent a number of flow-related ecological or life-history processes that could occur over periods of ~2–4 weeks. Total length of river assessed in the LMR = 345 km.					
What did CEW contribute to variability in water levels within weir pools?	0.10 m	0.15 m	0.08 m	0.17 m	0.10 m	Interquartile range (IQR) in water level increased by 0.13 m due to CEW in the tailwaters (i.e. just downstream of each weir) across Weir Pools 1–5.

CEWO evaluation questions	**Outcomes of CEW delivery**					
	2014-15	**2015-16**	**2016-17**	**2017-18**	**2018-19**	**2019-20**
	CEW increased water level variability (IQR) in the tailwaters of weir pools during each year of the assessment, with the greatest increase in variability during 2017-18. IQR is a measure of variability, as the difference between the 75th and 25th percentile values for water level over the year. If the IQR increases, the variability must have increased.					
What did CEW contribute to hydrological connectivity?*	28 ha 20%	963 ha 37%	0 ha 5%	831 ha 39%	484 ha 20%	Maximum area inundated increased by 517 ha due to eWater. 32% increase in volume at the SA border.
	CEW contribution to connectivity has been assessed laterally, as the increase in maximum inundated area each year, and longitudinally, as the percentage increase in flow volume each year (as used for Basin Scale assessment). CEW contribution to lateral and longitudinal hydrological connectivity was variable over the last six years. In 2019-20, CEW increased lateral connectivity throughout the water year, and this was largely due to the increased water levels in the Lower Lakes, as well as the Lock 2 weir pool raising. Longitudinal hydrological connectivity was increased by CEW in 2019-20, where the Basin-wide eWater strategy target for a 30% increase in flows in the Murray River was met. Subsequently, a 'moderate' contribution has been assigned for hydrological connectivity.					

* Inundation results for 2014-15 to 2018-19 do not include the river below Lock 1.

Contribution (to what extent CEW contributed towards the outcome, with the significance of the outcome considered):
⬜ Unknown 🟥 Negative ⬜ None/negligible 🟩 Minor 🟩 Moderate 🟦 Substantial

Discussion

2019-20 was a dry year, where 1,568 GL of the full 1,850 GL of South Australia's entitlement flow was delivered, and there was no unregulated flow. In dry years like this, Commonwealth environmental water provides a substantial contribution to longitudinal connectivity, with a 32% increase in flow volume at the South Australian border.

The evaluation of Commonwealth environmental water contribution to hydraulic diversity within weir pools is in line with previous years (Ye *et al.* 2020). Figure 10 presents the flow to South Australia over the past six years to compare the events of flow delivery. It can be seen that events of similar magnitude and duration to 2019-20, despite different timing, also occurred in 2014-15 and 2017-18. Ye *et al*. (2020) demonstrated that the increase in discharge to this magnitude (e.g. 17,840 ML/d in 2017-18), compared to ~12,000 ML/d in the two dry years of 2015-16 and 2018-19, increased the length of the river with lotic velocities created by Commonwealth environmental water by a factor of 2.0–2.8. This result highlights the substantial increases in lotic habitat in the LMR by increasing flow to South Australia in the order of ~20,000 ML/d.

A range of metrics have been considered to assess the evaluation questions in Table 3. Relevant velocity thresholds, proportions of the river, time of year and duration required for different ecological processes to be promoted are the focus of further research, as the empirical evidence relating to the conditions occurring, and different ecological processes being promoted, continues to improve. It is expected that this hydraulic information, and the methodology developed to derive it for the future, will help to develop eco-hydraulic relationships.

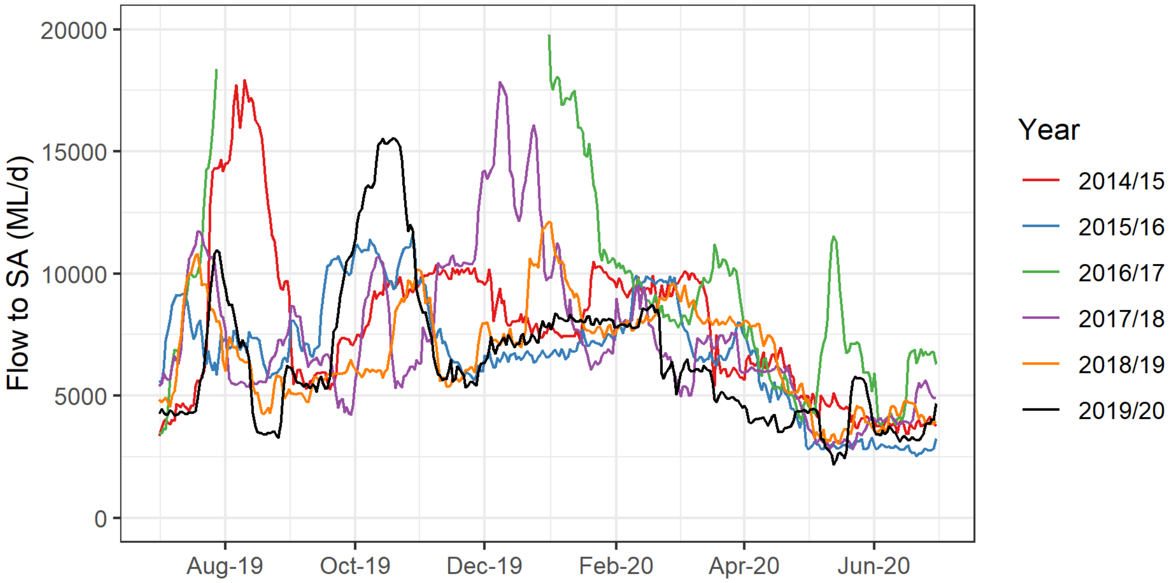


Figure 10. Flow to South Australia over the past six years, where events of similar magnitude and duration have occurred in 2014-15, 2017-18 and 2019-20. The high flow year of 2016-17 peaked at 94,350 ML/d.

Management implications

The restoration of lotic habitats is important for river ecosystem function and specifically to support key life history processes for many native biota that are adapted to flowing riverine environments. For example, they provide stimuli for spawning of flow-cued species (e.g. silver perch) (Tonkin *et al.* 2019), facilitate downstream drift and transportation of plankton, macroinvertebrates and fish larvae (Gibbs *et al.* 2020), and provide diverse hydraulic habitats that are suitable for a range of species (e.g. Murray cod) (Zampatti *et al.* 2014). The reduction in the abundance and distribution of riverine (lotic) biota (e.g. Macquarie perch *Macquaria australasica* and Murray crayfish *Euastacus armatus*) throughout the MDB (Lintermans 2007) highlights the importance of restoring hydraulic conditions (e.g. lotic habitats), which is particularly needed in the heavily regulated LMR.

In three of the past six years Commonwealth environmental water has contributed to flow events in the order of 15,000 ML/d at the South Australian border, demonstrating this is achievable within the current operating constraints and water availability. This has led to a substantial increase in the proportion of the river experiencing lotic velocities (>0.3 m/s), comparing the 10,000 ML/d to 15,000 ML/d results in Figure 11 and Figure 13. However, these figures indicate the lower end of each weir pool does not reach lotic velocities at 15,000 ML/d, and monitoring has indicated these flows have not been sufficient to reliably trigger some ecological responses, most notably recruitment of flow-cued species such as golden perch (see Section 2.6).

To investigate management options to further increase lotic conditions closer to the full reach beyond flow events in the order of 15,000 ML/d, scenarios representing a further increase to flow or including weir pool lowering have been considered. Figure 12 presents velocities along the LMR between Locks 1 and 6 for a 15,000 ML/d flow with a 1 m weir pool lowering at each weir compared to a flow of 20,000 ML/d with no lowering. While there are some differences due to the different mechanisms used to increase velocity, the two results are similar. The continuity equation indicates that velocity is equal to the flow divided by cross-sectional area, and to achieve the same velocity as an increase in flow of 33% (from 15,000 ML/d to 20,000 ML/d) a reduction in cross-sectional area of the same magnitude is required. Due to the relatively deep sections in the LMR a large weir pool lowering such as this (i.e. 1 m) is required to provide a similar increase in velocity as an increase in flow from 15,000 ML/d to 20,000 ML/d.

This comparison is further demonstrated in Figure 13, presenting the velocities against river distance instead of as a map. It should be noted that there is a difference in scale between Figures 11 and 12 compared to Figure 13, where Figures 11 and 12 presents the average velocity every 100 m along the river, compared to an average every 1 km in Figure 13. This increase in the averaging length reduces the variability in the results in Figure 13, however allows for a consistent comparison across multiple scenarios for the full length of river from Lock 1 to Lock 6. Figure 13 also includes smaller weir pool lowerings, of 0.3 and 0.6 m. The increase in the free-flowing section of the river with increasing size of the weir pool lowering can be seen, as the higher velocities occur further downstream within each weir pool. The corresponding water levels can be seen in Figure 14, where the water surface gradient (slope of the water level) increases with flow from 10,000 to 15,000, and to 20,000 ML/d, which also indicates sections of free flowing river with higher velocities. The 1 m weir pool lowering at 15,000 ML/d creates a water surface gradient similar (i.e. parallel) to the 20,000 ML/d flow, albeit at lower water levels.

Weir pool lowering events have value to increase velocities and also to increase variability of water levels. Variable water levels, and the coinciding periods of exposure and submergence of substrates beyond the euphotic zone, can result in the regular “re-setting” of biofilms (Steinman and McIntire 1990). The biofilm is a key component of riverine food webs, and this re-setting of the biofilm algal community produces higher quality food resources dominated by diatoms and unicellular algae (Wallace and Cummings 2016). These results have been presented to provide an indication of the scale of increase in flow or weir pool lowering required to take the next step in providing lotic conditions in the LMR.

Conclusion

2019-20 was a dry year, where 1,568 GL of the full 1,850 GL of South Australia’s entitlement flow was delivered, and there was no unregulated flow. Under these flow conditions and in the absence of environmental water, water levels would have been very stable throughout the year, with low hydraulic diversity and minimal lotic conditions in the LMR. In 2019-20, Commonwealth environmental water provided a substantial contribution to longitudinal connectivity, with a 32% increase in flow volume at the South Australian border, and 100% of the barrage flow that occurred (see Section 2.3).

The evaluation of Commonwealth environmental water contribution to hydraulic diversity within weir pools is in line with previous years, where in three of the past six years, Commonwealth environmental water created flow events similar to that occurring in 2019-20, exceeding 15,000 ML/d for a period of 7–14 days. These events lead to a substantial increase in the proportion of the river experiencing lotic velocities (>0.3 m/s) compared to flow below 10,000 ML/d. However, the results indicate the lower end of each weir pool does not reach lotic velocities at 15,000 ML/d. This can be compared to the without development hydraulic conditions, where even in such a low flow year the full reach was expected to experience cross-section averaged velocities exceeding 0.3 m/s for approximately 60 days.

In combination with weir pool raisings, environmental water also increased water level variability, as demonstrated by time series of modelled water levels with and without the environmental water, and the resulting interquartile range.

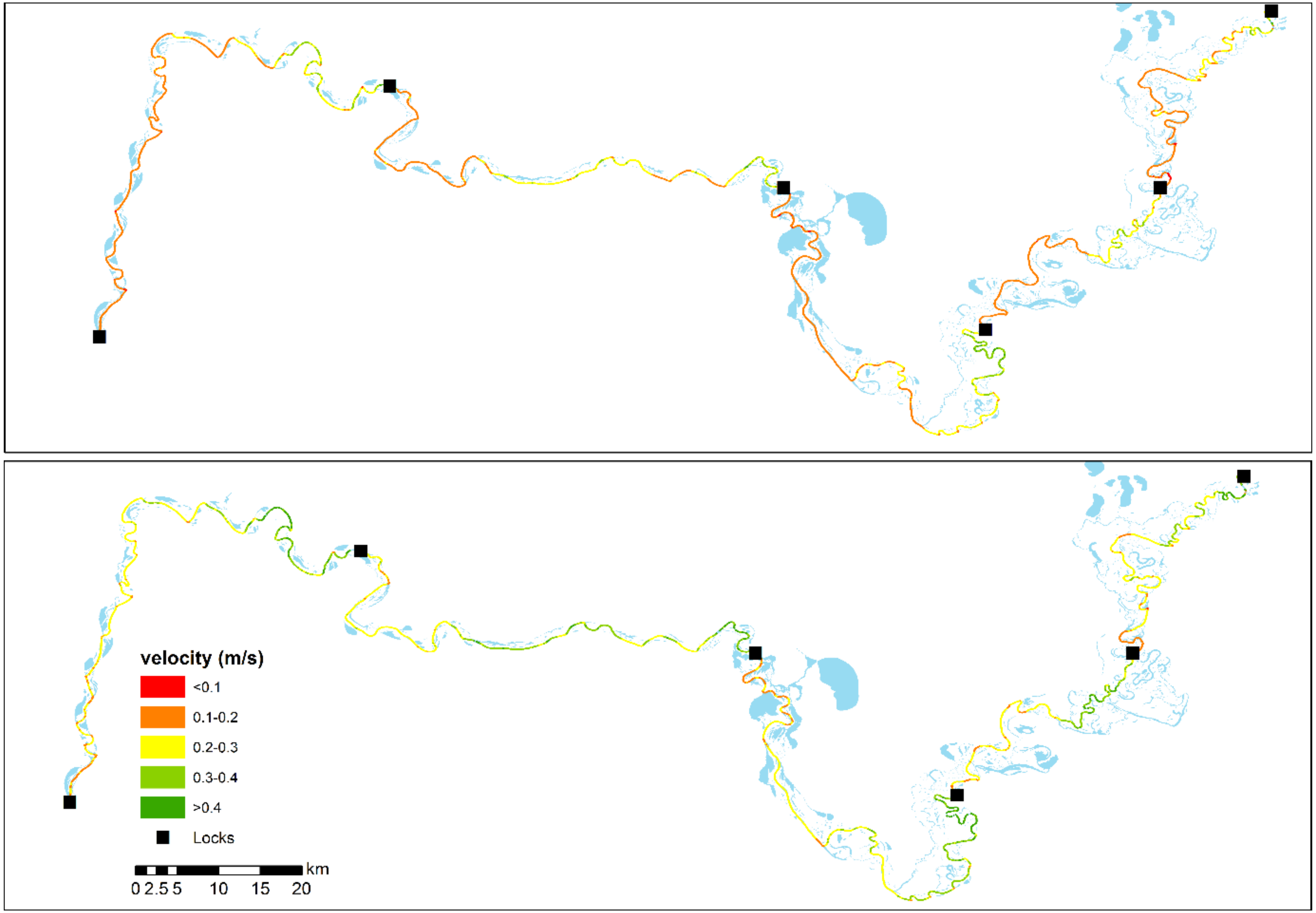


Figure 11. 100m average velocity for flow to South Australia of 10,000 ML/d (above) and 15,000 ML/d (below). Locks are indicated as black squares, from Lock 1 to Lock 6.

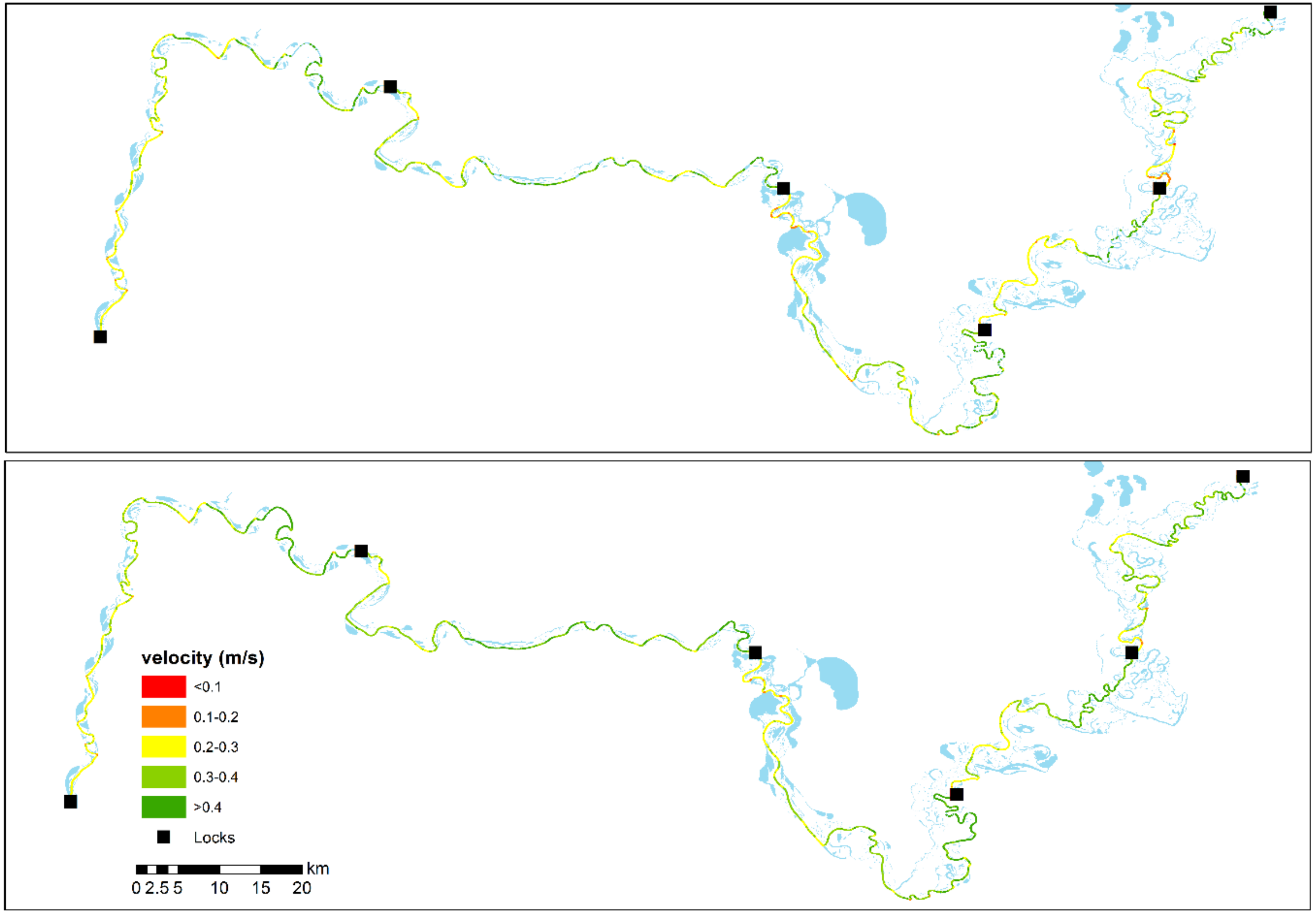


Figure 12. 100m average velocity for flow to South Australia of 15,000 ML/d and a 1 m weir pool lowering at each lock (above) and 20,000 ML/d with no lowering (below). Locks are indicated as black squares, from Lock 1 to Lock 6.

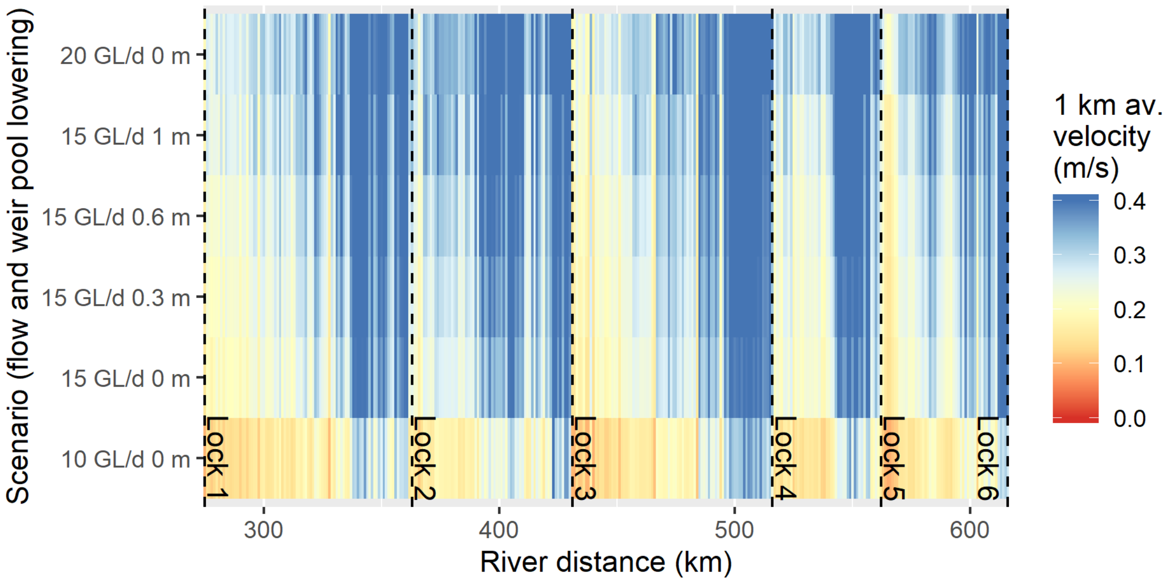


Figure 13. 1 km average velocity from Lock 1 to Lock 6 for scenarios of different flows to South Australia and weir pool lowering.

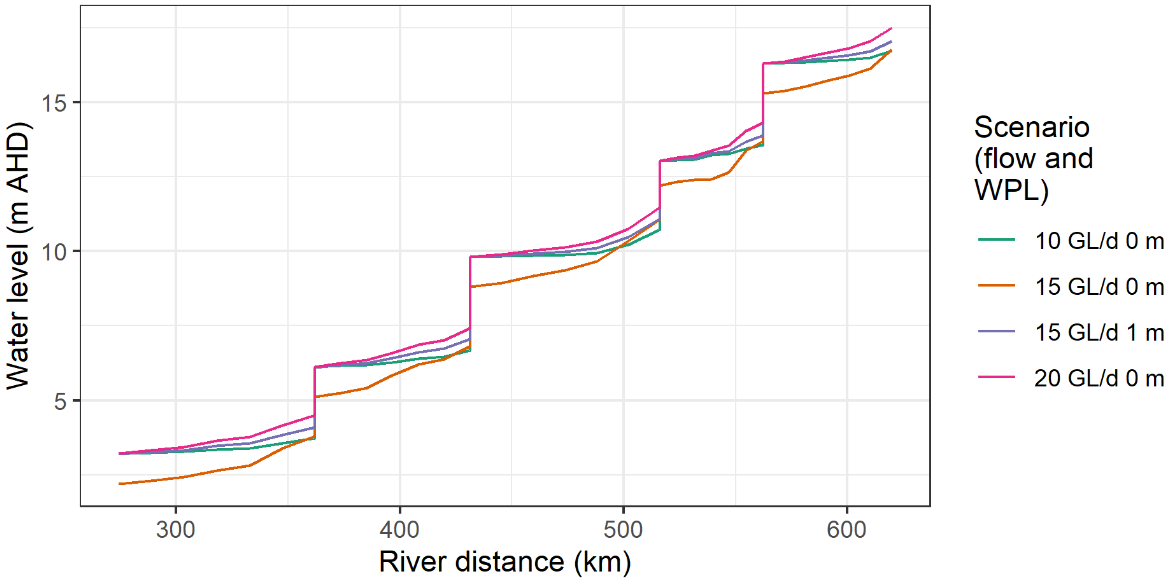


Figure 14. Modelled water level profile along the Lower Murray, from Lock 1 (274 km) to Lock 6 (620 km), for scenarios with flow increasing from 10,000 ML/d (10 GL/d) to 20,000 ML/d (20 GL/d).

2.2 Stream Metabolism and Water Quality

Background

River metabolism measurements estimate the in-stream rates of gross primary production (GPP) and ecosystem respiration (ER), providing information on the sources and utilisation of organic carbon by riverine food webs (Odum 1956; Young and Huryn 1996; Oliver and Merrick 2006). Comparing rates of photosynthesis and respiration helps describe the fundamental trophic energy connections that characterise different food web types. It can indicate whether production or decomposition processes predominate, and whether the organic food materials have come from within the river (autochthonous sources) or from the surrounding landscape (allochthonous sources). The magnitude and characteristics of the metabolic processes indicate the size of the food web and its capacity to support higher trophic levels, including fish, which are key targets for ecosystem management (Odum 1956; Sellars and Bukaveckas 2003; Oliver and Merrick 2006; Oliver and Lorenz 2010). As concentrations of dissolved oxygen are monitored to estimate rates of stream metabolism, these measurements provide ancillary information on the suitability of oxygen levels to support aquatic biota.

Photosynthetic microbes, comprised of microalgae and cyanobacteria, are autotrophs and fix dissolved carbon dioxide using the energy of sunlight to form organic materials for cell growth (Sellers and Bukaveckas 2003; Oliver and Merrick 2006; Várbíró *et al.* 2018). The availability of light for phytoplankton photosynthesis is influenced by the depth of light penetration and the intensity of water mixing that circulates phytoplankton through the upper illuminated surface layers. The availability and concentrations of nutrients also influence photosynthesis and the formation of organic materials (Reynolds 1984). So photosynthetic processes are affected by flow rates and water quality, and these are influenced by the catchment water sources, including environmental water contributions.

Net ecosystem production (NEP), the difference between GPP and ER, is considered a measure of the overall carbon balance, and frequently used as an estimate of the basal food resource supply (Odum 1956; Young and Huryn 1996; Oliver and Merrick 2006). If GPP>ER, carbon is accumulating, while if GPP<ER, carbon is being lost from the system. However, this interpretation implicitly assumes that fixation of carbon through photosynthesis is the source of organic material fuelling respiration. This is not usually the case, as respiration is also due to the decomposition of allochthonous organic carbon by heterotrophs which results in heterotrophic production, an additional food resource not accounted for in the NEP calculation. Terrestrial organic carbon enters rivers in particulate and dissolved forms, but dissolved organic carbon is most actively incorporated by heterotrophic microbes such as bacteria and is a major driver of heterotrophic respiration (Graeber *et al.* 2018). Flow patterns are important in determining the sources and supply of terrestrial organic carbon, and environmental flows and their management impact the carbon supply to the river food web (Oliver and Merrick 2006; Baldwin *et al.* 2016).

Major hypotheses

Increased flow (including the delivery of environmental water) into the LMR (peak and duration) in spring/summer will:

- Alter phytoplankton photosynthesis and the supply of autochthonous organic carbon to food webs if changes in-channel flow volumes and water quality modify light and nutrient availability;
- Enhance ecosystem respiration (ER) rates and heterotrophic production if flows better connect the channel with riparian, wetland or floodplain areas, increasing the supply of allochthonous organic carbon; and
- Reduce the likelihood of low dissolved oxygen (DO) concentrations by increasing water mixing in otherwise low flow zones except if flows carry excessive loads of organic carbon.

Methods

Field sampling

Rates of stream metabolism were estimated from daily changes in concentrations of dissolved oxygen (DO), which also provided information on the suitability of DO to support the aquatic biota. Monitoring consisted of *in situ*, continuous ten-minute interval logging of DO, water temperature, and incident light at three river sites. One site was downstream of Lock 6 ('Lock 6' herein) in the floodplain geomorphic zone, the second was downstream of Lock 4 ('Lock 4' herein), and the third downstream of Lock 1 ('Lock 1' herein) in the gorge geomorphic zone (Figure 1). Monitoring occurred from 3 September 2019 to 19 February 2020, with occasional interruptions (ca. one day) during probe maintenance.

Two metre, depth-integrated water samples were collected during the 10 probe maintenance field trips (ca. fortnightly), and analysed for chlorophyll *a*, total nitrogen, combined nitrate and nitrite, ammonium, total phosphorus, dissolved forms of phosphorus, and dissolved organic carbon. The detailed monitoring and analytical protocols described in Hale *et al.* (2014) were followed, but with some minor adjustments as detailed in Ye *et al.* (2018). In addition, the vertical light attenuation for Photosynthetically Active Radiation (PAR) was measured on each occasion using LiCOR underwater sensors.

Water quality measurements and vertical attenuation coefficients were considered relevant for three days before and three days after sampling, and this extended data set (61 points) was used to explore relationships with metabolic rates. Collected water quality data were supplemented with monitoring data from nearby sites provided by the Australian Water Quality Centre of South Australia Water.

Estimating metabolic rates

Daily volumetric rates for GPP and ER were estimated over 24-hour periods from midnight to midnight with the *BASE* program (Grace *et al.* 2015). This uses Bayesian regression routines to fit the measured changes in DO concentrations to a widely applied model that describes the daily fluctuations in water column DO concentrations (Odum 1956; Young and Huryn 1996; Oliver and Merrick 2006). Oxygen based metabolic rates were converted to carbon units by assuming that the photosynthetic and respiratory quotients were equal to one, each mole of oxygen transfer matched by a mole of carbon dioxide.

The measured volumetric rates of metabolism were integrated over river depth and width based on channel characteristics derived from a morphometric model linked to water level (Section 2.1). The morphometric data included the average cross-sectional areas, average depths, and average widths at different flow levels for the river reaches at each monitoring site. Daily flows measured during the monitoring periods were adjusted using hydrological modelling to estimate the flows and water levels that would have occurred without Commonwealth environmental water, and without any environmental flows.

Cross-sectional metabolic rates were calculated as the product of the volumetric rate and the average cross-sectional area. The influence of flow on the average depth and cross-sectional area was a function of the channel morphometry, and increased flows increased or decreased these depending on channel shape. Volumetric rates are reported as gm/m^3/day (numerically equal to mg/L/day), area as m^2 and so cross-sectional rates are for a 1 m length of river. Metabolic rates were integrated over time by summing daily rates.

The influence of light on photosynthesis was determined using the mean light intensity encountered by phytoplankton mixed through the water column. The mean intensity depends on the incident irradiance (*Io*), the vertical attenuation of light passing through the water column (*kd*), and the average depth (*zave*). If the average depth is greater than the depth of the illuminated surface layer, then the mean irradiance (*Im*) is given by (Oliver and Merrick 2006):

$$Im = \frac{Io}{kd * zave} \qquad (1)$$

The amount of phytoplankton present was estimated from the chlorophyll concentration. Rates of GPP were standardised to chlorophyll concentrations (GPP(b)) and compared with the corresponding mean irradiances *Im*. The relative changes in GPP(b) due to Commonwealth environmental water, and total environmental water were calculated from the changes in *Im* resulting from these flows. Conversion of modelled GPP(b) estimates to GPP requires knowledge of the chlorophyll concentration. It is assumed that the chlorophyll concentration on each day remains the same with and without environmental flows, and relative changes in GPP(b) and GPP are equivalent. This assumption is necessary as predicting the chlorophyll concentrations of the counterfactual flows is not possible, in part because the relative contributions of different water sources to these flows is unknown.

The contributions to ER by photosynthetic and heterotrophic microbiota was investigated using multiple regression of potential respiratory sources. GPP provided a measure of the organic carbon produced by photosynthesis, which is a major source of phytoplankton respiration (Beardall and Raven 1990), while dissolved organic carbon (DOC) concentrations provided a measure of the resource supply for respiration of heterotrophic microbes. Previous analyses identified these as major contributors to ER (Ye *et al.* 2020). Respiration rates were averaged for each three-day period before and after the field sampling dates to align with water quality data and compared with similarly averaged

GPP rates. Averaging helps to account for lagged metabolic interactions such as respiration rate being influenced by the GPP of the preceding day.

Overall NEP was calculated as the difference between volumetric GPP and ER but is not considered an informative parameter. The phytoplankton net production (PNP) was calculated as the difference between the measured GPP and the estimated phytoplankton community respiration (PCR). Metabolism measurements do not provide estimates of the gross production of bacteria, and bacterial net production rates (BNP) were determined from the relationship between BNP, BCR and bacterial growth efficiency (BGE):

$$BNP = \frac{BGE * BCR}{1 - BGE} \qquad (2)$$

The BGE is influenced by a range of environmental conditions including the chemical composition of DOC. However, an average value of 0.2 was considered reasonable for the Murray River within its typical temperature range and water quality attributes (Rivkin and Legendre 2001; Marra and Barber 2004; Berggren and del Giorgio 2015), and BNP was estimated as BCR/4.

Hydrological modelling was used to estimate the flows that might have occurred if Commonwealth environmental water and environmental water had not been delivered. However, identifying the water sources contributing to these modelled flows was not possible, and so forecasting water quality characteristics such as turbidity and dissolved organic carbon, or biological attributes such as chlorophyll concentrations could not be done, even though the analyses show these influence metabolism. To avoid this issue and provide illustrative examples of metabolic responses to flow changes, a simplifying scenario was adopted. It was considered that the mix of water sources remained the same without environmental flows and their removal did not have a major effect on water quality. This rephrases the question to assessing the influence on metabolism of changing flows of matching water quality.

Results

Dissolved oxygen

The time series of dissolved oxygen (DO) concentrations showed similar patterns across sites and were generally higher than saturation concentrations, indicating positive photosynthetic production (Figure 15). No consistent association was observed with flow, and the seasonal decline in DO concentration was attributed to the increasing water temperature. However, the high flow period in October appeared to slow the seasonal water temperature rise and stabilised the DO concentration over this period.

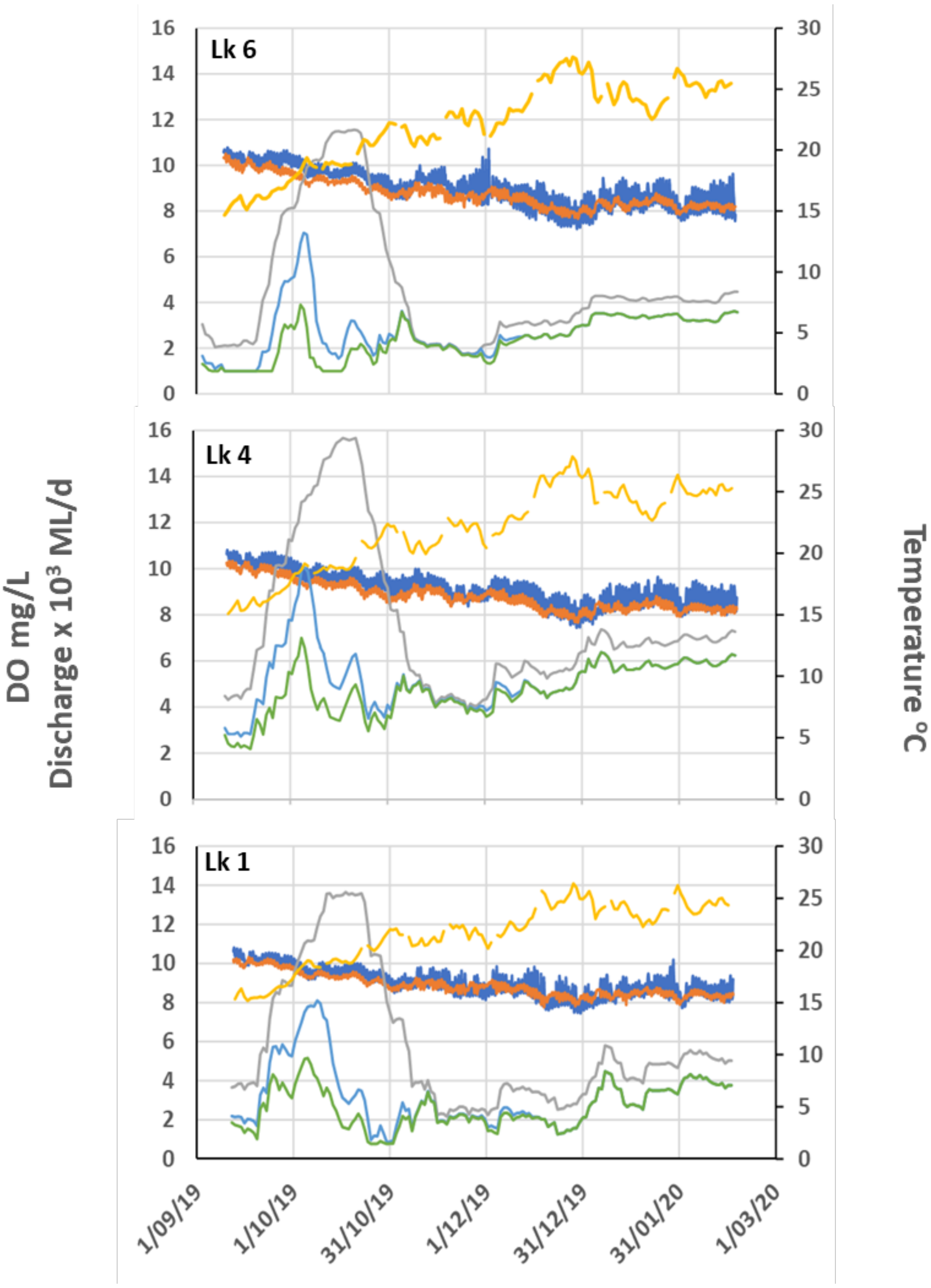


Figure 15. Dissolved (DO, ●) and saturated (●) oxygen concentrations, temperature (●) and the daily discharge observed (●) and modelled without CEW (●) and without environmental water (●) at Locks (Lk) 6, 4 and 1.

Water mixing, gas exchange and metabolism

Low flows that result in reduced water velocities influence the reliability of metabolism estimates because turbulent mixing declines and the DO measurements no longer represent activity throughout the water column. This is demonstrated by the change in the gas exchange coefficient (k) in response to average water velocity, calculated as the flow (m^3/s) divided by the cross-sectional area (Figure 16). Although following different curves, the gas exchange coefficient reduces systematically at each site as velocity declines within the channel, until below ca. 0.18 m/s it becomes increasingly variable. This reflects the reduced influence of low velocities on turbulent mixing and the increased importance of daily meteorological conditions in modifying thermal and chemical stratification. The relatively consistent relationship between k and velocities above 0.18 m/s breaks down once flows spread onto the floodplain, but this did not occur in the 2019-20 season.

The magnitude of the gas exchange coefficient has an important influence on the likelihood of oxygen depletion. If the consumption of oxygen is greater than its resupply from the atmosphere, then the oxygen concentration falls until the consumption and resupply rates are in balance. Consequently, as flow and water velocity decline the respiration rates that can be offset by oxygen resupply also decline due to reductions in the gas exchange coefficient and can lead to deleterious oxygen concentrations. At Lock 6, velocities of 0.18 m/s occur at flows of 4,800 ML/d and 0.1 m/s at flows of 2,500 ML/d.

During September and early October 2019 at Lock 6 modelled flows without environmental water were half the observed flows and low enough to increase the likelihood of oxygen depletion, especially as temperatures increased (Figure 15). It was considered that environmental water contributed to the maintenance of DO concentrations during this period. In late October and during November, flows were at similar levels to September and oxygen depletion was considered likely based on water velocities, but there was no problematic oxygen depletion, suggesting low respiration rates were offset by the low gas exchange rates.

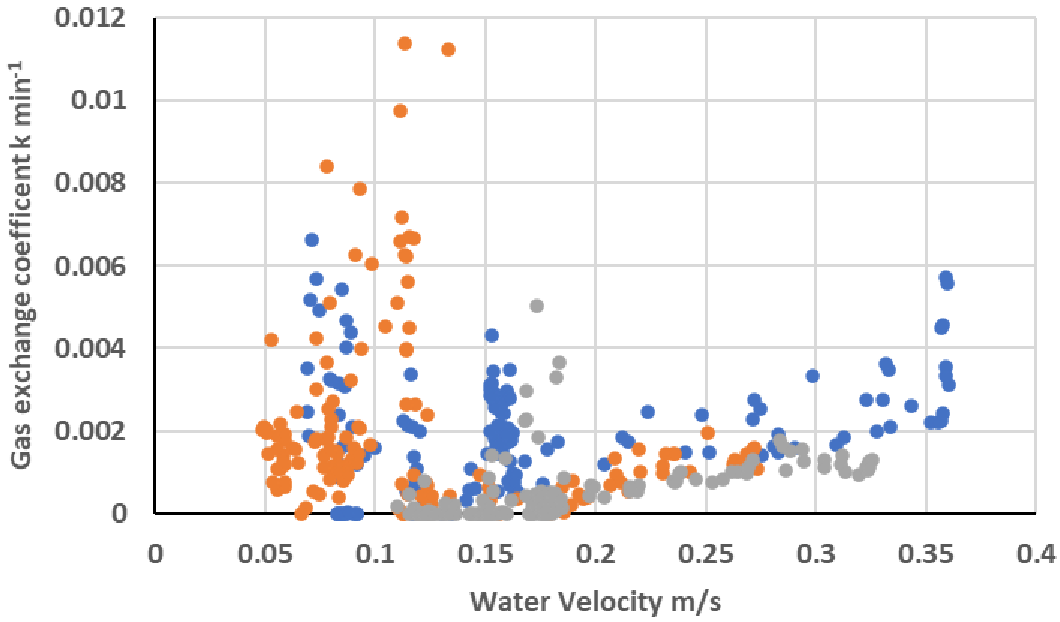


Figure 16. The relationship between the gas exchange coefficient (k) and water velocity at Lock 6 (●), Lock 4 (●) and Lock 1 (●) during 2019-20.

Daily metabolic rates

Patterns of daily GPP (photosynthesis) and ER (respiration) were similar across sites over the monitoring period (Figure 17) and comparable with rates measured in previous years

(Ye *et al.* 2020). A spike in metabolic activity in January at the Lock 1 site requires further investigation as it is suspected biofouling.

Despite large fluctuations in discharge (Figure 15), volumetric rates of metabolism over the monitoring period seemed largely unaffected by flows. GPP ranged from 1 to 5 mgO_2/L/d, gradually increasing over the monitoring season, and was mirrored by changes in ER. If it is assumed that ER was largely associated with phytoplankton, then the average daily NEP (mgO_2/L/d) calculated as the difference in GPP and ER, was 0.54 at Lock 6, 0.23 at Lock 4, and 0.47 at Lock 1. The cumulative NEP over the monitoring period was 83, 34 and 69 mgO_2/L, or approximately 31, 13 and 26 mgC/L respectively.

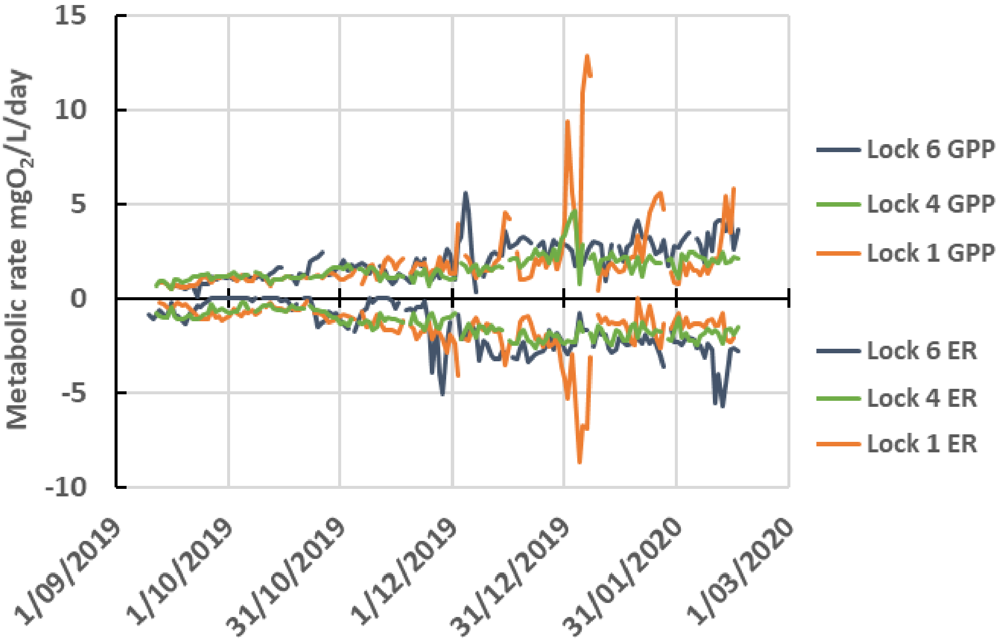


Figure 17. Daily gross photosynthesis (GPP) and ecosystem respiration (ER) rates at each of the three monitoring sites.

Determinants of gross primary production

Critical velocities of 0.18–0.20 m/s have previously been used to identify periods when in-channel mixing was sufficient to ensure metabolism estimates were reliable (Oliver and Lorenz 2010; Ye *et al.* 2020). The analysis of 2019-20 GPP measurements from times when water velocities at the sites exceeded 0.18 m/s supported previous findings of a significant linear correlation between the chlorophyll specific rate of gross photosynthesis (GPP(b)) and the mean irradiance in the mixed water column (*Im*) (Figure 18) with a linear regression slope equivalent to that calculated in previous analyses (Oliver and Merrick 2006; Ye *et al.* 2020).

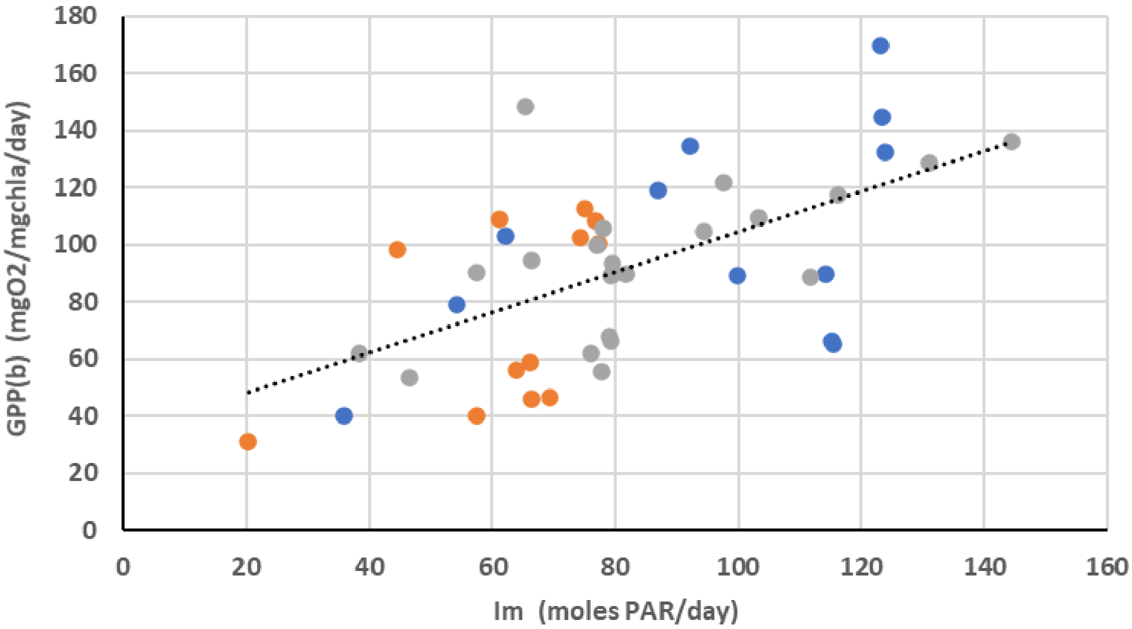


Figure 18. The response during 2019-20 of GPP per unit chlorophyll to the mean irradiance of the water column at Lock 6 (●), Lock 4 (●) and Lock 1 (●) when water velocities were greater than 0.18 m/s. Regression, y = 0.70x+34.2, r^2=0.35.

In naturally flowing rivers there is a relationship between flow and water depth, but at the study sites within the weir pools of the LMR, this connection is decoupled. At these sites there is minimal change in water depth unless flows are sufficient to overwhelm the functioning of the weirs, or alternatively weir operations are managed to influence depth. The high flows associated with environmental water delivery between mid-September 2019 and early November 2020 resulted in depth changes at each of the sites (Figure 19). At Lock 6 the average depth of 2.5 m was maintained for most of the season but increased by up to 0.5 m during the peak flows. At Lock 4 the water level was less stable, slowly increasing from 2.5 to 2.8 m over the season, and with a peak in the high flow period that increased the depth by a maximum of 1m. The monitoring site at Lock 1 is not in a weir pool but is influenced by water levels in Lake Alexandrina and by winds aligned with the long river reach that push water upstream. At this site water depth slowly declined from 3.2 to 3.0 m over the monitoring period but increased during the high flow period by a maximum of 0.3 m.

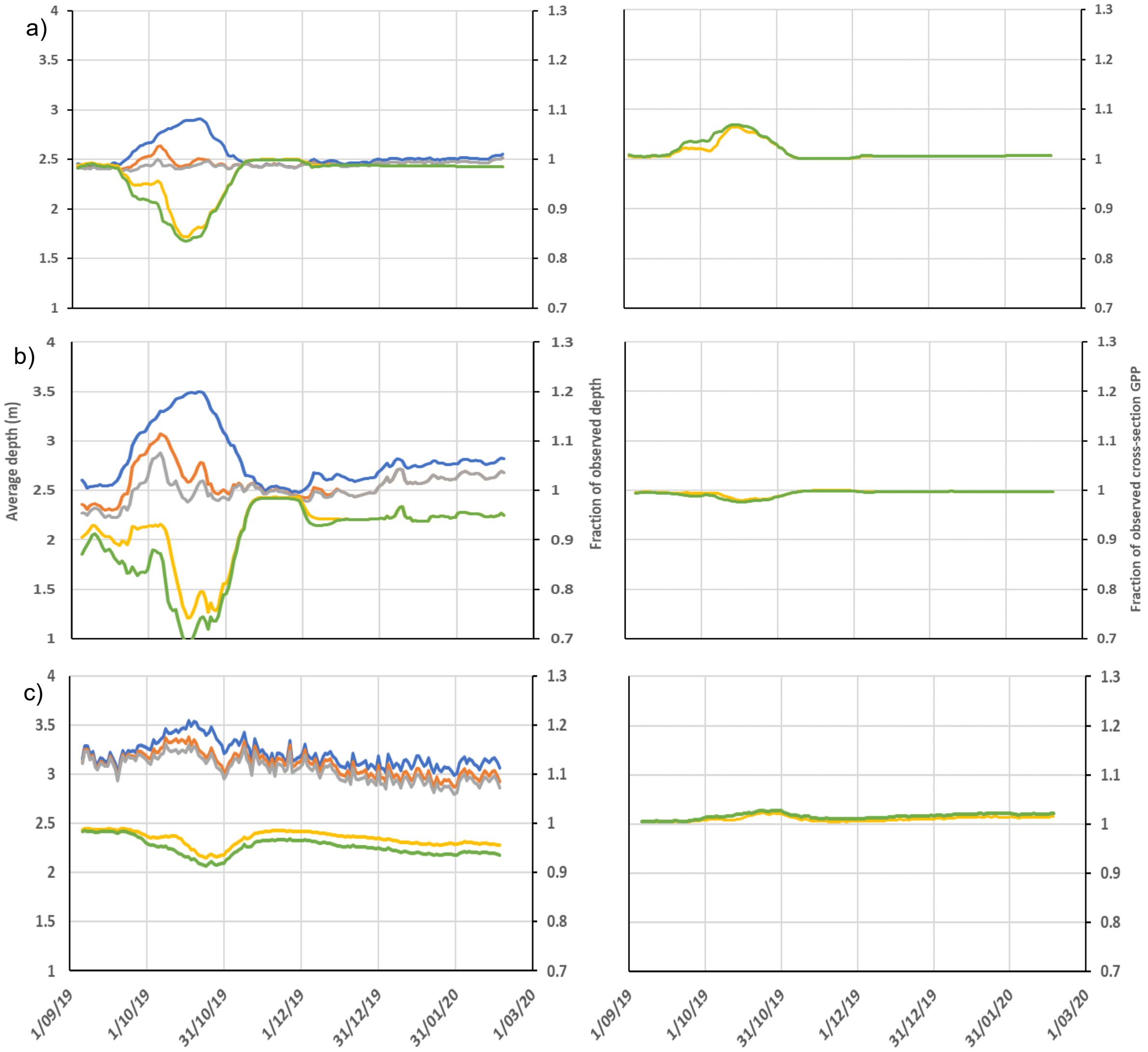


Figure 19. (Left) The average depths at (a) Lock 6, (b) Lock 4 and (c) Lock 1 observed (●), and for modelled flows without CEW (●), and without environmental water (EW) (●), and the fractional reduction in GPP(b) due to the increased depths associated with CEW (●) and without EW (●). (Right) The fractional change in cross-sectional GPP due to the combined change in depth and cross-sectional area for modelled flows without CEW (●) and without EW (●).

As *Im* is inversely related to the average depth of the water column (Equation 1), increases in flow that lead to increases in depth reduce the available light and reduce GPP(b) in inverse proportion to the relative change in depth, provided other conditions remain the same (Ye *et al.* 2020). Accepting that water quality remains unchanged between the flow conditions, the effect of environmental flows on the relative change in GPP(b) was determined from the ratio of the modelled water depth without environmental water to the observed depth with all flows, in effect assessing the influence of environmental water delivery on GPP(b) (Figure 19). At Lock 6, the increases in flow due to Commonwealth environmental water had a small effect on GPP(b) except during the high flows of mid- to late October 2019 when estimated rates were depressed by a maximum of 16% at the flow peak. At Lock 4, environmental water had a larger effect, reducing GPP(b) by about

6–10% and with a maximum reduction of around 30% at the flow peak. At Lock 1 GPP(b) was generally reduced by 1–4% with a maximum reduction of 7%.

If chlorophyll concentrations and *kd* remain unchanged for the daily comparisons of flows with and without environmental water, then fractional changes in volumetric GPP(b) and GPP are equivalent. The influence of the changing daily depth ratios (Figure 19) on the actual magnitude of GPP were generally small. The period of maximum change was during the second half of October when GPP was reduced by 0.5 $mgO_2/L/d$.

The cumulative effect of the depression in GPP due to additional flows can be assessed from the integrated production over the monitoring period, estimated as the sum of daily rates. Due to the increased flows associated with Commonwealth environmental water, the cumulative production over the monitoring period reduced at Lock 6 from 334 to 326 mgO_2/L (125 to 122 mgC/L), at Lock 4 from to 265 to 244 mgO_2/L (99 to 91 mgC/L), and at Lock 1 from 310 to 300 mgO_2/L (116 to 112 mgC/L). These small differences reflect the minimal changes in weir pool depths.

The volumetric rate of GPP measures the concentration change in the supply of primary production but does not describe the total supply in the river, which will depend on the cross-sectional area of the flow (Ye *et al.* 2020). The cross-sectional production, or GPP per linear metre of river, was calculated from the volumetric production rate and the cross-sectional area. To encompass the influence of depth changes associated with flows, the volumetric GPP rates adjusted for the relative changes in average depth were used with the corresponding cross-sectional areas to assess the changes in cross-sectional production. Despite reductions in the volumetric GPP(b) at all sites, especially during the high flow period (Figure 19), the production per metre of river increased at Lock 6 and Lock 1 (Figure 19). These changes were small, at Lock 6 a maximum increase of 7% and at Lock 1 a maximum increase of 3%. In contrast the cross-sectional production at Lock 4 decreased in response to the high flows by a maximum of 3% (Figure 19). These opposite effects are a function of the relationships between depth and cross-sectional area at the different sites.

The effects of flow on the cumulative cross-section production over the monitoring period were small within sites. At Lock 6 Commonwealth environmental water increased production from 99.7 to 100.8 gO_2/m (37.4 to 37.8 gC/m) and at Lock 1 from 153.5 to 155.0 gO_2/m (57.5 to 58.1 gC/m). Commonwealth environmental water had the opposite effect at Lock 4 with the cumulative cross-section production reducing from 111.6 to 111.1 gO^2/m (41.9 to 41.7 gC/m) over the monitoring period.

In contrast, the large differences in the cumulative cross-sectional production between sites, for example Lock 4 being 1.5 times higher than Lock 6, reflected the average cross-sections at the sites, 526 m^2 at Lock 1, 456 m^2 at Lock 4 and 311 m^2 at Lock 6. Volumetric rates of GPP were not substantially different between the sites (Figure 17) and consequently the cumulative cross-sectional production rates were proportional to the cross-sections. This demonstrates the large changes in GPP that will occur in response to flow at river sites where the naturally related changes in depth and cross-sectional area occur.

Determinants of ecosystem respiration

Previous analyses indicated that ER was largely associated with carbon sources from GPP and DOC (Ye *et al.* 2020). Rates of ER and GPP were converted to carbon units (mgC/L/day) and combined in a multiple linear regression assuming there were no other major sources of respiration. This yielded a relationship with regression coefficients that were similar to previous estimates (Ye *et al.* 2020):

ER = 0.68 (±0.06)*GPP + 0.05(±0.01)*DOC

Based on these an initial effort was made at evaluating separately the respiration rates due to phytoplankton (autotrophs) and bacteria (heterotrophs) across sites for 2019-20, but it is stressed that these are approximations only due to the limited data set and unexplained variation. Values reported (Figure 20) are for occasions when the sum of PCR and BCR were within ±30% of the measured respiration rates, a constraint that reduced the number of data points from 38 to 24. Corresponding net production rates were estimated, PNP as the difference between GPP and the phytoplankton respiration, and BNP using BCR in Equation 2 (Figure 20). During 2019-20 the respiration and net production attributable to bacteria was small relative to the phytoplankton with BNP on average providing 20% of the combined net production.

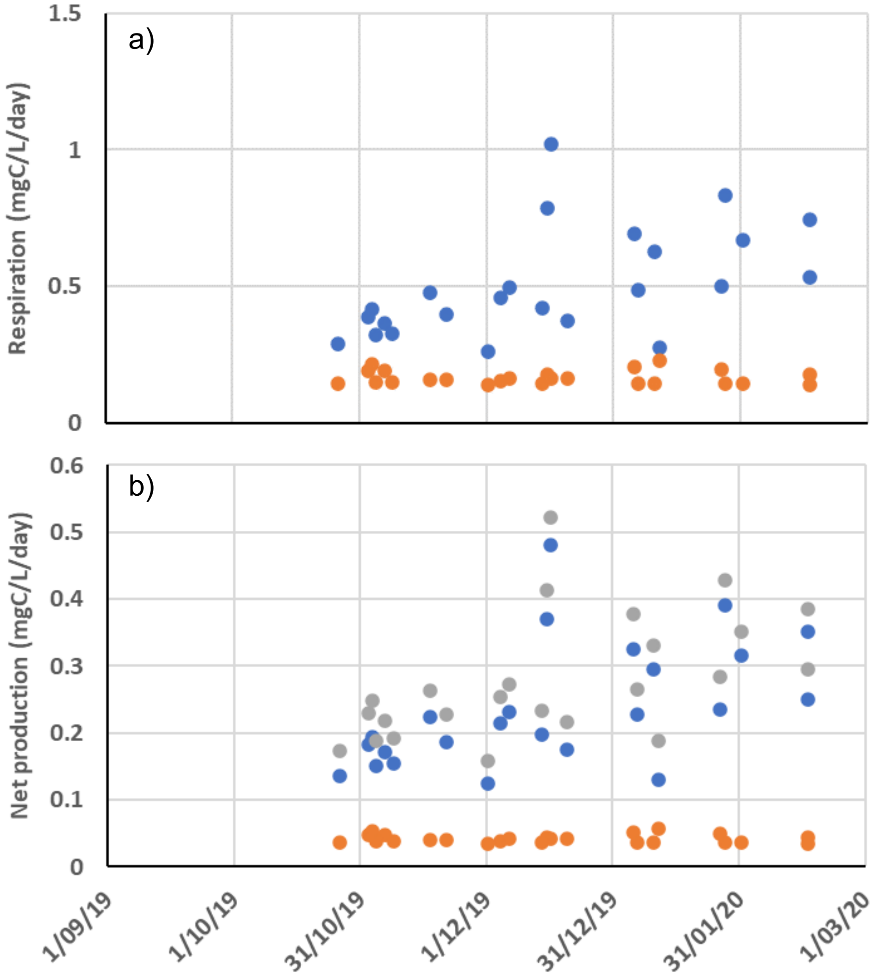


Figure 20. Respiration (a) and net production rates (b) of phytoplankton (●), bacteria (●) and net production of both combined (●) for all sites during 2019-20.

Water quality

Concentrations of DOC averaged 3 mg/L across the three sites during the sampling period, and showed little change except for an increase at Lock 1 to 4.6 mg/L during January, which appeared related to flow from Lake Victoria (Figure 21c), but this cannot be confirmed from measurements in the Selected Area alone and awaits a broader basin scale analysis.

Turbidity also changed in response to flow, increasing when additional flows arrived from upstream both during October/November and in early January (Figure 21b). There were no inflows from the Darling River, historically a major source of turbidity in the Lower Murray, and the turbidity fluctuations appeared to be associated with flows from either Lake Victoria or further up along the Murray River. Again, analyses of the sources of the turbidity fluctuations requires basin scale data. The importance of turbidity to the vertical attenuation coefficient for PAR (*kd*) is evident from the matched responses (Figure 21b).

Although nutrients can influence rates of metabolism and phytoplankton concentrations, there was no clear evidence that this occurred during 2019-20. Both TP and TN responded to flows in a similar manner to turbidity and increased substantially (Figure 22) but did not result in marked changes in metabolism. This suggested that phytoplankton metabolism was largely controlled by light, and heterotrophic metabolism by DOC concentrations. It is likely that these resources constrained population sizes such that nutrients were not reduced to limiting concentrations.

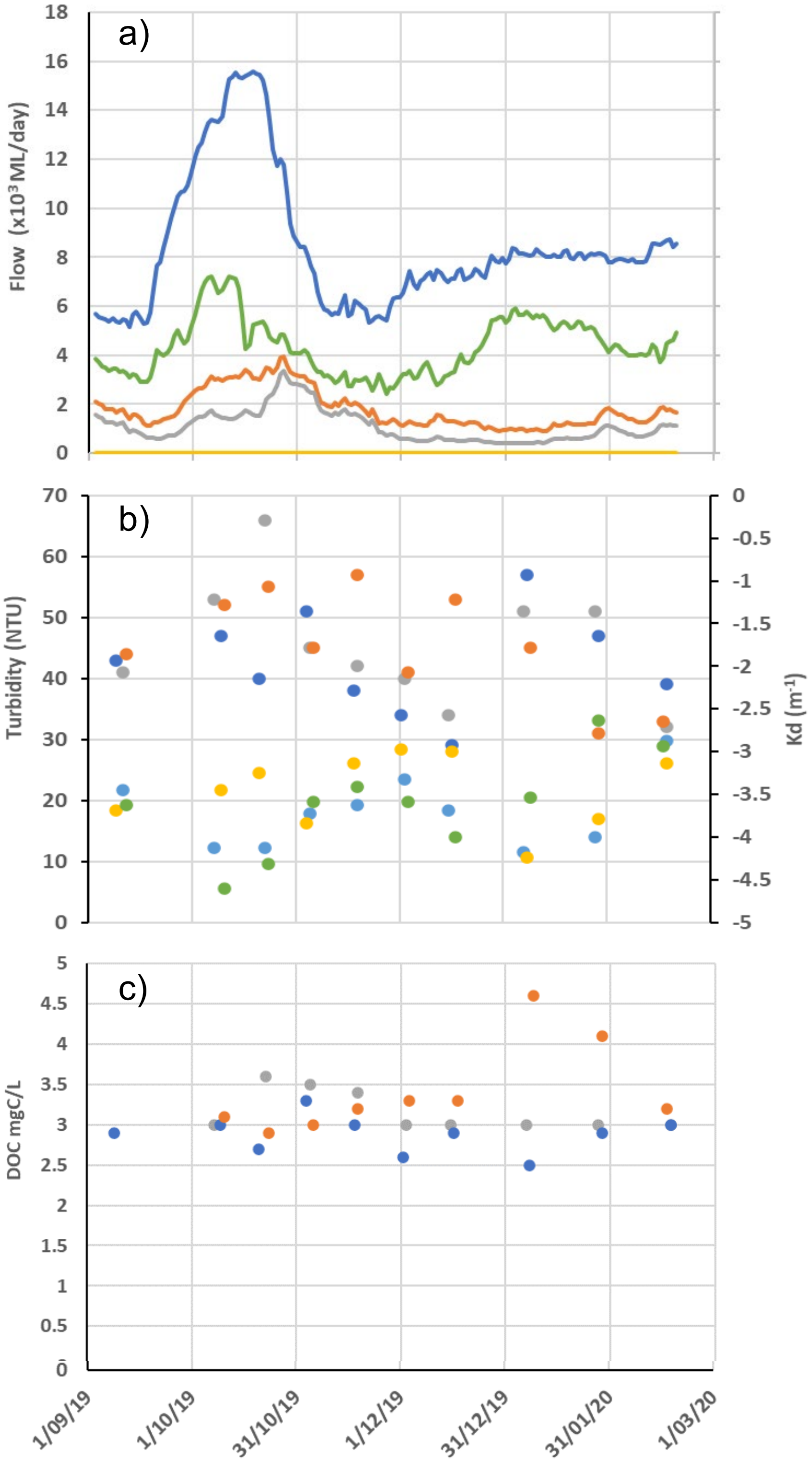


Figure 21. (a) Cumulative contributions of water sources to total flow at the South Australian border with additional flow due to Darling River (●), Victorian Tributaries (●), Murrumbidgee River (●), Lake Victoria (●) and the Murray River (●). (b) Turbidity at Lock 6 (●), Lock 4 (●) and Lock1 (●) and *kd* at Lock 6 (●), Lock 4 (●) and Lock1 (●). (c) Dissolved organic carbon at Lock 6 (●), Lock 4 (●) and Lock1 (●).

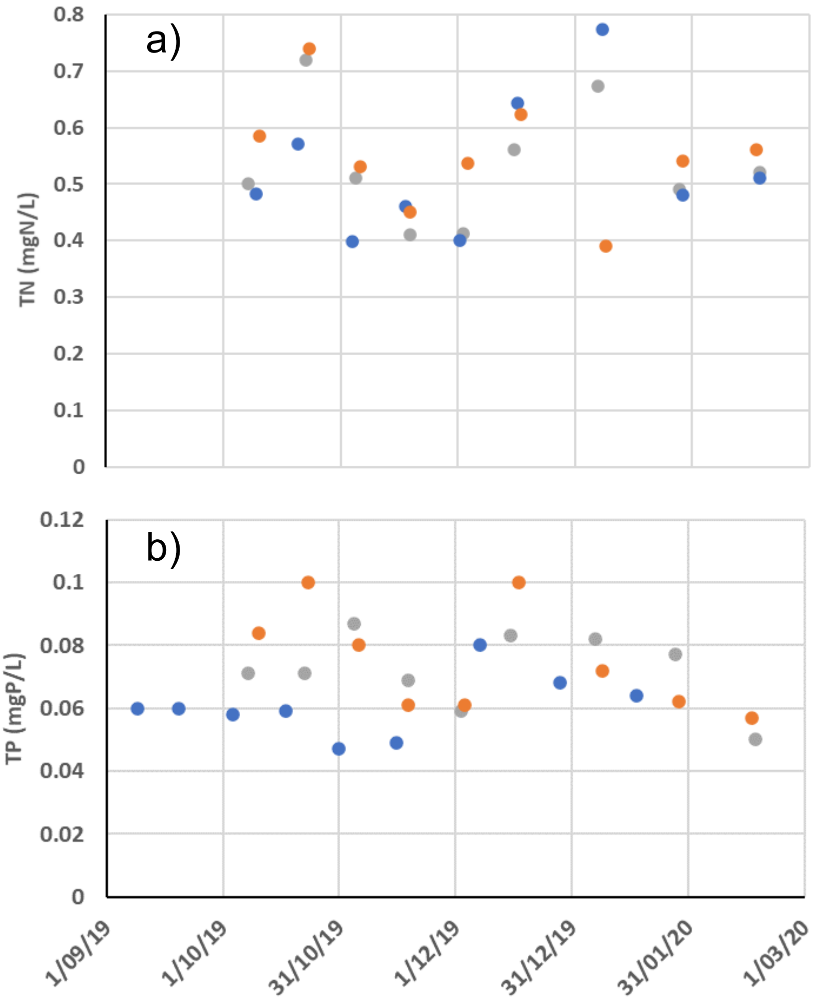


Figure 22. (a) Total nitrogen and (b) Total phosphorus at Lock 6 (●), Lock 4 (●) and Lock1 (●).

Evaluation

The contributions of Commonwealth environmental water to environmental outcomes associated with changes in dissolved oxygen and metabolism were assessed from a set of constructed metrics describing potential benefits from the evaluation questions. Significance levels were assigned to each evaluation question based on the extent that Commonwealth environmental water contributed towards the observed outcome, in one case the length of time of an enhanced effect, and in the other cases a percentage estimate of the extent of influence. The thresholds for significance levels among the indicators are based on expert opinion and may be adjusted in response to developing understanding of these processes.

Table 4. Stream Metabolism evaluation questions and answers relating to Commonwealth environmental water (CEW) and environmental water (eWater). The Lock 6 site has been used to answer the evaluation questions.

CEWO evaluation questions	**Outcomes of CEW delivery**						
	eWater type	**2014-15**	**2015-16**	**2016-17**	**2017-18**	**2018-19**	**2019-20**
What did CEW contribute to dissolved oxygen levels?	All eWater	31	69	35	68	45	40
	CEW	0	53	21	50	25	30
	eWater decreased the likelihood of low DO by increasing water mixing and oxygen exchange at the surface. This was assessed as the extra days per year with water velocities >0.18 m/s due to eWater. A substantial contribution was considered greater than 30 days, moderate 15–30 days, minor 7–14 days and negligible < 7days.						
What did CEW contribute to patterns and rates of primary productivity?	All eWater	1	2	2	2	2	1
	CEW	1	2	2	2	2	1
	Increased flows generally reduced the volumetric rate of primary production but increased the cross-sectional rate. This increased the overall "carrying capacity" of the river, although the implications of changes in the ratios of these two measures are unknown. At the LMR sites, the percentage increases in cross-sectional GPP due to eWater were negligible due to the largely stable water levels induced by weirs. A substantial contribution was considered an increase in cross-sectional GPP of 20% or greater, moderate 11–19%, minor 5–10%, negligible <5%.						
What did CEW contribute to patterns and rates of decomposition?	All eWater	3	9	9	7	6	4
	CEW	3	9	9	7	6	4

CEWO evaluation questions	Outcomes of CEW delivery						
	eWater type	2014-15	2015-16	2016-17	2017-18	2018-19	2019-20
	Bacterial respiration (BCR), a measure of decomposition, is directly related to DOC concentrations. Modelling of the influence of flows on BCR assumed that for any given day, DOC concentrations were the same with and without eWater. Percentage changes in river cross-sectional BCR due to the addition of CEW at Lock 6 were small due to the constant water level maintained by the weirs. A substantial contribution was considered an increase in cross-sectional BCR of 20% or greater, moderate 11–19%, minor 5–10%, negligible <5%.						

Contribution (to what extent CEW contributed towards the outcome, with the significance of the outcome considered):
⬜ Unknown 🟥 Negative ⬜ None/negligible 🟩 Minor 🟩 Moderate 🟦 Substantial

Discussion

The dissolved oxygen (DO) concentration is influenced by flow, especially in low flow areas such as weir pools of the LMR, where surface oxygen exchange is reduced. Increased flows due to environmental water can improve the DO conditions in low flow reaches by increasing mixing and enhancing oxygen exchange at the surface. The gas exchange coefficient is close to zero below 0.18 m/s and increases at higher velocities which helps minimise DO depletion. The environmental water contributions to improving gas exchange conditions were assessed from the additional days that they increased velocities above 0.18 m/s. DO concentrations remained at saturation levels throughout the 2019-20 monitoring period, despite periods of low flows, indicating low heterotrophic respiration rates. This aligned with the low DOC concentrations throughout the period. Environmental water delivery contributed to the maintenance of DO concentrations by helping to sustain the surface gas exchange for 30–40 days longer than otherwise would have occurred. These are substantial contributions to improving gas exchange, helping to avoid potential DO water quality impacts and the associated potential for fish kills. The periods of influence of these flows are depicted by the 0.2 m/s average velocity timeline for each weir pool (Figure 7) and averaged over the whole river length between Lock 1 and Lock 6 (Figure 6), for each of the different flow scenarios.

The linear correlation between GPP(b), the gross primary production per unit of phytoplankton biomass, and the mean irradiance (Im) was comparable with those previously reported for the Murray River (Oliver and Merrick 2006; Oliver and Lorenz 2010; Ye *et al.* 2020). This relationship, in conjunction with measurements of channel morphometry, enabled estimation of volumetric and cross-sectional rates of GPP(b) and GPP with and without environmental water. Water quality was considered the same across the flow regimes as it was not possible to determine independently the water quality of flows without environmental water. In part this was because the water source composition of the modelled flows, without Commonwealth environmental water and without environmental water are unknown. However, even if the composition of the counterfactual flows could be determined it would still be difficult to predict their water quality as reliable models are not available.

The effects of environmental flows on volumetric and cross-sectional GPP(b) and GPP were small, only a few percent, due to the weirs maintaining relatively constant water levels. Previously the influence of flow on metabolism at a river site unregulated by weirs was estimated and showed significant increases in the cross-sectional production of up to 30% or more as flows increased (Ye *et al.* 2020). Conversely, although a reduction in depth reduced the cross-sectional rate of GPP, it increased the volumetric rate by almost 20% in the modelled reach. It is evident that the weirs have a major effect on metabolism because of their disruption of the relationships between flow, water level and cross-sectional area. A fundamental question relates to the relevance of this disruption to the functioning of food webs and whether the impacts are detrimental. Analyses are continuing to address this question.

In addition to photosynthetic production, the food web is also supported by the supply of heterotrophic production through DOC utilisation. Rates of ER were partitioned into respiration by phytoplankton (PCR) and by the heterotrophic bacteria (BCR), and this enabled individual estimates of their net production. Throughout most of the monitoring period, the bacterial contribution to ER through decomposition was of similar magnitude but smaller than the phytoplankton respiratory contribution (PCR). Previously, during the flood in early 2016-17 it was found that the BCR could be larger than PCR when DOC was high, as in the flood waters.

The combined net production (CNP) of the phytoplankton and bacteria estimated their supply of organic carbon to the food web in the 2019-20 period and ranged between 0.16 and 0.52 mgC/L/day, with a mean of 0.3 mgC/L/day. These rates were similar to those reported previously for the previous five years of monitoring (Ye *et al.* 2020). These estimates of net production provide a different view of the carbon supplies to the river food webs compared to the traditional analyses of NEP which are close to zero over the monitoring period, as previously reported (Oliver and Merrick 2006; Gawne *et al.* 2007). The CNP estimates demonstrate that both heterotrophic and phytoplankton production are important sources of organic carbon to the river. Improved supplies of DOC are critical to providing food webs with organic carbon food resources through the heterotrophic pathway.

The effects of environmental flows on cross-sectional rates of decomposition were estimated from BCR, but as this rate was the same between the flow regimes because DOC concentrations were considered equivalent, they changed in proportion to the relative changes in cross-sectional area with flow. As with GPP, these differences were small at the monitoring sites due to the weirs restricting changes in cross-section. In contrast, at an unregulated site the modelled changes were large with relative increases of up to 1.54 due to increased flows (Ye *et al.* 2020).

Cross-site differences in metabolism were large and mainly related to changes in-channel morphology and the relationships between water depth and cross-sectional area. Due to the weirs the influence of flow on these characteristics was small, but they indicate the extent of variation in metabolism along river reaches and the variation that could occur in response to flows in the absence of weirs. The significance of this to river health is currently unknown.

Metabolism at the monitoring sites is influenced by water quality, especially turbidity and DOC which have fundamental effects. However, the data have not so far demonstrated a major influence of nutrients with metabolism unresponsive to changing concentrations that have resulted from altered flows. This suggested that light and DOC were limiting the development of microbial populations so that nutrient limitation was not being induced.

Analyses of the monitoring data have provided important insights into river metabolism and its significance in supplying food resources to the river ecosystem. In general, these findings support the hypotheses regarding the influences of flow on river metabolism and DO concentrations. Currently these findings are restricted to the monitoring sites, but the modelled relationships provide an opportunity to extend this understanding to the Murray River more generally to further our understanding of river metabolism and the influence of flows, including those due to environmental water.

Management implications

Environmental flows can help reduce the likelihood of low DO concentrations in the LMR, if they increase water velocities above a level of ~0.18m/s, below which surface oxygen exchange is poor. This critical velocity will vary with channel morphology and further investigation is required in other river sections. The level of flow required for surface exchange to offset de-oxygenation is also influenced by the DOC concentration, and if this is high then surface exchange may not be able to re-supply the respiratory oxygen depletion. Models assessing the level of DOC concentrations that lead to “black water” events are developing (Baldwin *et al.* 2016; Whitworth and Baldwin 2016), but a holistic approach that considers the full implications of river metabolism and surface exchange could further benefit management of environmental flows.

Increased flows interact with channel morphometry altering the average depth and the cross-sectional area of the flow and affecting the rate of volumetric and cross-sectional GPP. It has been demonstrated, using the developed models, that large changes in flow greatly influence metabolic rates in unregulated channels undergoing typical responses in water depth and cross-section (Ye *et al.* 2020). Depending on channel shape, changes in flow are likely to be more beneficial at some water levels in the channel than at others, particularly where the channel broadens. Using environmental flows to target these water levels could increase river productivity.

It is evident that the weirs have a major effect on metabolism through their disruption of the relationships between flow, water level and cross-sectional area. The small flow-induced changes in water level that occur in the weir pools result in minor changes in metabolism in response to environmental flows. If modifying metabolism is identified as a beneficial target for the weir pools, then larger water level changes will be required, perhaps through manipulation of water levels. However, large, regular changes in water depth have not been considered suitable within the weirs. With such constraints there is a requirement to identify the need for enhancing metabolism within the weir pools by assessing the relative importance of their local production, and whether changes in production due to their influence are detrimental. Based on the modelling, by holding water levels above natural low flow levels the weirs might be increasing total cross-sectional production but reducing volumetric rates compared to natural variations. The implications of this are unclear, but interpretation could be assisted by the analysis of comparative time series of net production from upstream unregulated sites. This would enable a comparison of local production at weir pool sites with that at unregulated sites. Also, with suitable selection of upstream sites, an assessment could be made of the influence of longitudinally transported production. Of particular interest are sites associated with major water sources including the Darling River, Lake Victoria and the upstream Murray River.

Environmental flows which alter the attenuation of light through increased turbidity have a major influence on GPP. Turbidity in the LMR is influenced substantially by flows from the Darling River, that may move directly to South Australia, but are often diverted into Lake Victoria for later supply downstream. The impact of turbidity delivery on river metabolism raises system scale questions of flow management involving the storage of Darling River and Murray River water in Lake Victoria, the timing of direct passage of turbid flows to South Australia, the timing and volumes of releases from Lake Victoria, and the benefits of mixing flows of different water quality. The metabolism models outlined in this report provide a basis for assessing the influence of the timing and duration of turbidity events on river production.

Based on historical recollections, it is suspected that the turbidity of the Murray River is generally higher than it was historically, and the models provide a basis for investigating the impact of this influence. However, the interactions between hydrology, water quality and metabolism are complex, and the models developed to describe individual processes such as gas exchange, vertical attenuation, GPPb, and ER interact dynamically, both in time and space. The individual models can be applied by managers to identify the scale of responses in metabolism that environmental flows may produce, but the value of these models will be greatly enhanced by their incorporation into a dynamic framework. The Source model provides such a framework, but such modelling activities are outside the scope and resources of the current project.

In general, Commonwealth environmental water deliveries increased the average water depth and reduced volumetric GPP, but simultaneously increased cross-sectional areas which increased the cross-sectional GPP. These opposite shifts in food production are likely to have fundamental effects on the composition and functioning of food webs, especially at unregulated sites where these changes are large. Data from upstream, unregulated sites that influence the weir pool responses would help to assess the impact of these production shifts on biotic community composition.

A major source of metabolic activity is the external supply of organic carbon to the river system. The estimates of the individual and combined net production (CNP) by autotrophs and heterotrophs demonstrated that both were important sources of organic carbon in the weir pools, with autotrophs generally providing a larger proportion. However, the external DOC supply was critical to the response of the heterotrophic pathway and during floods when DOC increased this became the major source of organic carbon (Ye *et al.* 2019). These changes influence the quantity, quality, and characteristics of the food supply to the food web but the influence on the riverine community structure, including higher trophic levels, of the changes in these various resources are not well known. Current research in MER is aimed at providing a better understanding of these links.

Measurements from unregulated sites are required to assess the cumulative effects of flow variation on metabolism and to underpin whole river accounting. Although such sample sites could not be incorporated into the current project, a recent demonstration project funded by the MDBA and utilising sites along the length of the Murray River (Biswas *et al. pers comm*) showed the large variation in metabolism that can occur longitudinally, with noticeably higher production upstream during the 2019-20 measuring period. Unfortunately, no general conclusions can be drawn from the small data set because of the highly variable hydrology and water quality that markedly influenced the metabolic responses; longer periods of data collection are required to reliably identify response patterns.

Conclusion

The analyses identified key environmental influences on GPP and ER including the:

- reliance of GPP on the mean light encountered by phytoplankton;
- effects on GPP of interactions between flow and channel morphometry;
- reliance of the mean light on turbidity and DOC;
- individual respiration rates of phytoplankton and bacteria;
- contributions of phytoplankton and bacteria to net production and their reliance on the mean light and DOC concentrations;
- effect of water velocity on surface oxygen exchange; and
- contribution of flow to improving velocities and reducing the likelihood of low DO.

Understanding these types of interactions is critical to predicting the likely changes in metabolism and net production within a particular river reach due to the delivery of environmental flows of given volume and water quality. Further, such understanding begins to provide a means for assessing the effects of environmental flows on the basal river food resources, a vital target for management. The dynamic interactions identified between many of the processes highlighted the need for measurements at a range of different sites, and the incorporation of the models into an interacting framework. The findings in relation to weir pools reflect their effect on river hydrology with a major impact on metabolism through disruption of the relationships between flow, water level and cross-sectional area. Assessing the impact of weir pool disruption on river metabolism and food resources requires comparative measurements at unregulated river sites and would be especially useful if these sites were upstream of the weir pools so that longitudinal influences could be incorporated into the analyses.

2.3 Matter Transport and Coorong Habitat

2.3.1 Matter Transport

Background

Modification to the flow regime can alter the biogeochemistry of rivers and their adjacent floodplain systems, including the estuarine and lagoon areas at their terminus near the ocean. For example, reduced flow may increase the intrusion of salt into the system from the ocean whilst simultaneously decreasing the export of salt from inland reaches. As there is continual deposition of salt onto the landscape (predominately from rainfall), it can accumulate if not transported by flow and exported from the system. Additionally, a change in the flow regime will alter the mobilisation of nutrients from the floodplain and change the subsequent primary productivity within the river. Environmental flows can be used to reinstate some of the natural processes or to increase the magnitude of the processes that control the availability and transport of dissolved and particulate matter. Salinity, dissolved and particulate organic nutrients, and chlorophyll *a* are often measured or modelled to understand the influence of flows on the concentrations and transport of matter.

In general, restoring river flow to an estuary is an important tool for salinity management by limiting seawater intrusion into the estuary. In the case of the Murray River, it is further complicated as seawater entering the Murray Mouth is highly dependent on how river water is released from the barrages, and these dynamics impact salinity in the connected Coorong lagoon. The Coorong is a 120 km long shallow reverse estuary that runs parallel to the coast and is separated from the Southern Ocean by Young Husband Peninsula. As water evaporates from the North and South Lagoons it is replaced with water coming from the north. When river water is exiting the barrages it reduces sea-water intrusion and maintains fresher conditions around the Murray Mouth which enables fresher water to replenish water evaporated in the Coorong. When there is more seawater intrusion into the Murray Mouth, this transports considerable amounts of salt into the Coorong as this water travels south-east to replenish water evaporated from the lagoons. The salinity of the Coorong, particularly in the South Lagoon is a key determinant of habitat suitability for macrophytes, invertebrates and fish.

Nutrients drive system productivity and so understanding how they are transported between the various components of riverine ecosystems can offer insights into river and estuary productivity. Dissolved inorganic nutrients are essential resources for the growth and survival of biota and are readily assimilated (Poff *et al.* 1997). In healthy ecosystems, mobilisation of nutrients can enhance productivity and support ecosystems, however, in poorly flushed or over-enriched systems, then addition of nutrients leads to eutrophication and numerous undesirable consequences.

Nitrogen, phosphorus and silica are of particular interest because they are generally the most limiting biologically active elements that control the productivity of aquatic ecosystems. River flows result in the mobilisation and transport of dissolved nutrients through the leaching of nutrients from dried sediments and dead organic matter in the river and floodplain catchment. Particulate organic nutrients (phosphorus and nitrogen) are those nutrients incorporated into the tissue of living and dead organisms. Flow can influence particulate organic nutrient concentrations and transport through several mechanisms, including through increased productivity associated with elevated dissolved nutrient concentrations. Chlorophyll *a* is the key measure of phytoplankton biomass used since it is indicative of the amount of primary production in riverine ecosystems. Flow can influence chlorophyll *a* concentrations and transport through increased phytoplankton productivity, by providing nutrients, or by flushing and diluting phytoplankton biomass.

To assess the contribution of environmental water delivery to matter transport in the LMR from 1 July 2019 to 30 June 2020, a hydrodynamic-biogeochemical model was applied for the region below Lock 1 to the Murray Mouth (Figure 1). The model has been validated with water quality data, and is used to understand salt, nutrient and phytoplankton movement.

Major hypotheses

Commonwealth environmental water will:

- Increase the mobilisation of salt from the Basin and increase the transport of salt passing from Lock 1 through the Murray River Channel and through the Lower Lakes and Murray Mouth;
- Increase the mobilisation of nutrients from the Basin and increase nutrient loads passing from Lock 1 through the Murray River Channel and through the Lower Lakes and Murray Mouth;
- Increase suspended solid loads (including phytoplankton biomass) passing from Lock 1 through the Murray River Channel and through the Lower Lakes and Murray Mouth; and
- Decrease the rate of salt and nutrient accumulation in the North and South Coorong lagoons.

Methods

The contribution of environmental water to the transport of salt, nutrients and phytoplankton was assessed with a coupled hydrodynamic–biogeochemical model for the reach below Lock 1 to the Murray Mouth. Salt, nutrient and phytoplankton transport was predicted for three different flow scenarios: with all environmental water (i.e. the observed flow), flow without Commonwealth environmental water, and flow without any environmental water (i.e. counter-factual simulations assessing what would have happened if flows were not augmented with environmental water).

When modelling, it is necessary to make assumptions on the relationships between flow and nutrients or salt, nutrient dynamics in sediments and floodplain habitats, and the utilisation of nutrients by phytoplankton. This leads to a degree of uncertainty in model outputs; however, given previous model development and validation initiatives over the past decade, it is considered that this uncertainty is within reasonable bounds (Aldridge *et al.* 2013) and the results can be used to reliably assess the general response attributable to environmental water.

Water quality sampling and analyses

Water temperature, electrical conductivity, dissolved oxygen, pH, filterable reactive phosphorus (hereafter referred to as phosphate), total phosphorus (TP), combined nitrate and nitrite (NOx), ammonium, total Kjeldahl nitrogen (TN), dissolved silica and chlorophyll *a* were monitored at multiple sites in the Murray River and form initial inputs to the biogeochemical models. Organic nitrogen was calculated as the difference between total Kjeldahl nitrogen and ammonium. These nutrient concentrations are representative of conditions in the river and Coorong, however, salinity was measured more frequently in the river and Coorong, enabling use of this data for the 2019-20 modelling and analysis.

Hydrodynamic–biogeochemical modelling

The model platform used to assess the effects of environmental water delivery on salt and nutrient transport was the coupled hydrodynamic–biogeochemical model "TUFLOW-FV – AED" (referred to as FV-AED for short), developed by BMT Global Pty Ltd. and the University of Western Australia. TUFLOW-FV has been used extensively in the region for hydrological assessments, and was previously used to assess the contribution of environmental water to dissolved and particulate matter for water years 2013-14 to 2018-19 (Ye *et al.* 2016a;2020).

In this assessment, two model domains were applied spanning: (1) Lock 1 to the Southern Ocean, including the Coorong (Figure 23), and (2) a high-resolution Coorong only model (described further below). The TUFLOW-FV model adopts an unstructured-mesh to resolve spatio-temporal changes in water velocity, temperature and salinity dynamics, in response to changing meteorological and inflow conditions. Superimposed on the hydrodynamics, the Aquatic EcoDynamics (AED) water quality modules were configured to simulate the dynamics of light, oxygen, nutrients, organic matter, turbidity and phytoplankton. Both model domains mentioned above were configured to simulate the same hydrologic and biogeochemical processes, however the Coorong only model had a higher resolution mesh for better resolving the water quality conditions, and it was also required for the habitat assessment of *Ruppia* and the various fish species of interest (Section 2.3.2).

The first model runs with the full domain were initialised with data from a range of sources. Inflow data (Lock 1), used to drive the main river domain, were provided by the MDBA for the three scenarios (Figure 24), i.e. with all environmental water ("***all water***", representing observed conditions), without Commonwealth environmental water ("***no CEW***"), and without any environmental water ("***no eWater***"). These simulations extend previous model runs that started 1 July 2014 (Ye *et al*. 2020), and were run for this assessment for the period between 1 July 2019 and 30 June 2020.

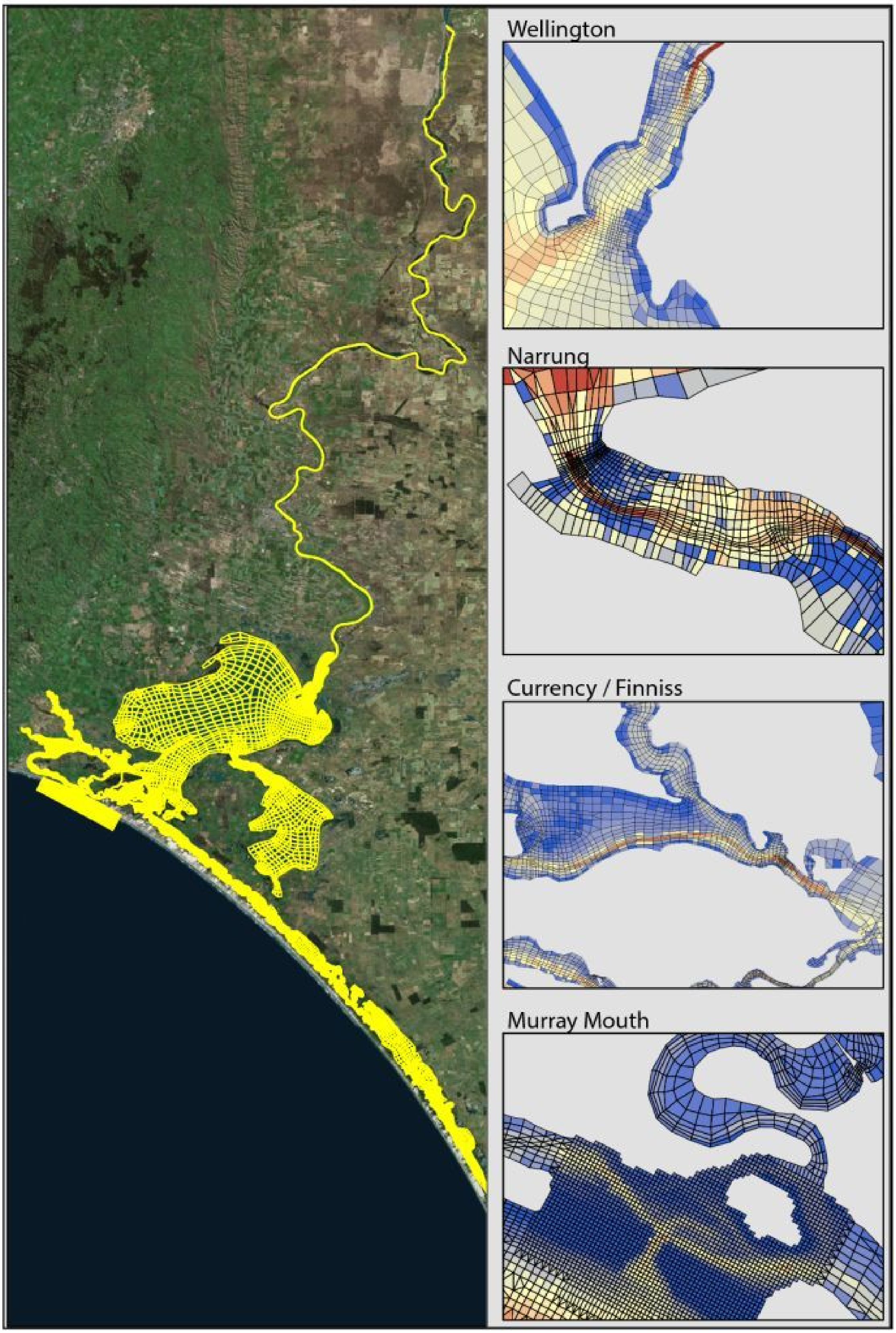


Figure 23. Overview of model domain applied in the Matter Transport study of LTIM using TUFLOW-FV. Grid provided courtesy of DEW. Coloured grids in maps on the right-hand side represent depths, i.e. increasing depth from shallow (blue) to deep (red).

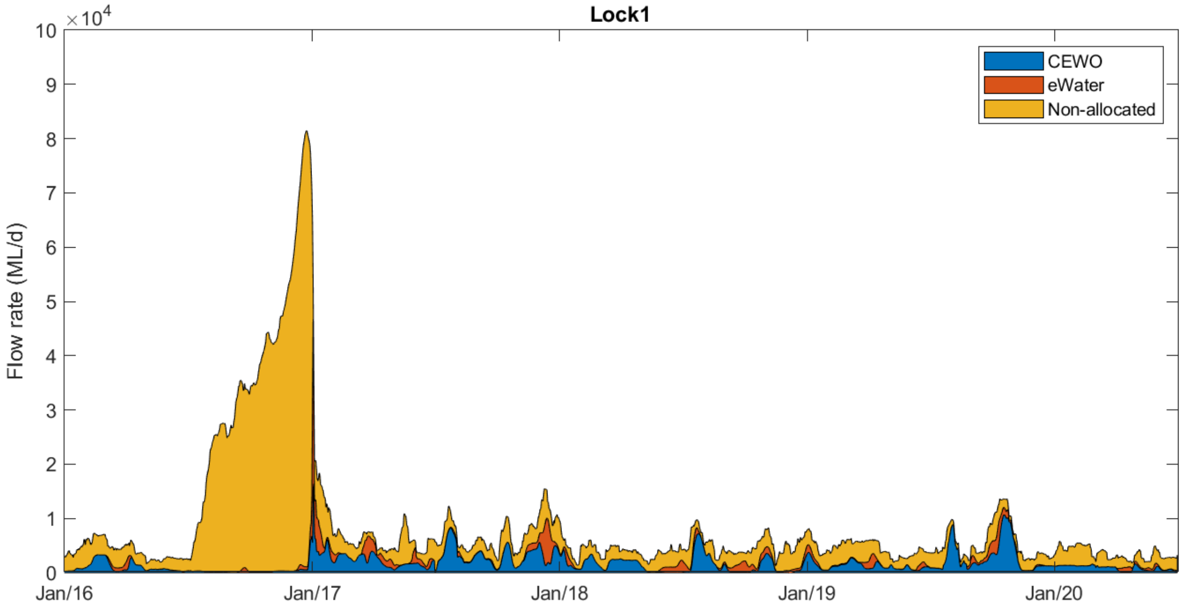


Figure 24. Overview of the flow rates from three water sources assessed by the model simulations. The area plots show the proportion of flow that was considered non-allocated ("no eWater" scenario), the proportion of Commonwealth environmental water (CEWO) and the proportion of non-CEW environmental water (eWater, together with non-allocated is the "no CEW" scenario). The cumulation of all these flows represents the 'all water" scenario. Flows were applied to the full domain matter transport model at the upstream boundary, which is at Lock 1.

Additional flow specifications for SA Water off-takes were also included. Irrigation return flows were assumed to be negligible over this period and were not included in the model. Similarly, flows from Eastern Mount Lofty Ranges were not included since their contribution to the Lower Lakes was considered to be relatively minor (Cook *et al.* 2010). Meteorological conditions were based on data from Narrung. Between Lake Alexandrina and the Coorong, five barrages were included (Goolwa, Mundoo, Boundary Creek, Ewe Island and Tauwitchere) and set with a spill-over height of 0.72 m AHD. The barrage operation was set to include gate operation based on operational information provided through discussions with representatives of DEW. At the bottom of the domain, two open boundaries were specified, one at the Murray Mouth and one at Salt Creek. Murray Mouth water level was based on Victor Harbor tidal data, which were available at 10 minute intervals. Salt Creek flow data were set based on available flow data from the WaterConnect website (DEW).

Water quality conditions for both boundary points were set based on a linear interpolation of the measured nutrient and salinity data used for this study. Water quality conditions for the river inflow at Lock 1 were determined based on interpolation of available data from Lock 1 or Morgan. For water quality properties for the without environmental water scenarios, rating curves were developed for flow and concentration. Based on the daily flow difference, a scaled concentration was estimated for water quality parameters including salinity, phosphate, ammonium, nitrate, total nitrogen and silica. The physico-chemical information at other sites was used to validate the model.

The influence of environmental water on the concentrations of matter was assessed through a comparison of modelled concentrations for the various scenarios for the

Barrages and Coorong. Modelled concentrations are presented as medians of modelled cells within Murray Mouth area surrounding sampling sites (Figure 25). It is well established what occurs at the lake and river sites in response to environmental water and these sites are no longer reported. A range in concentrations within the Murray Mouth cells is also presented for the 'all water' scenario.

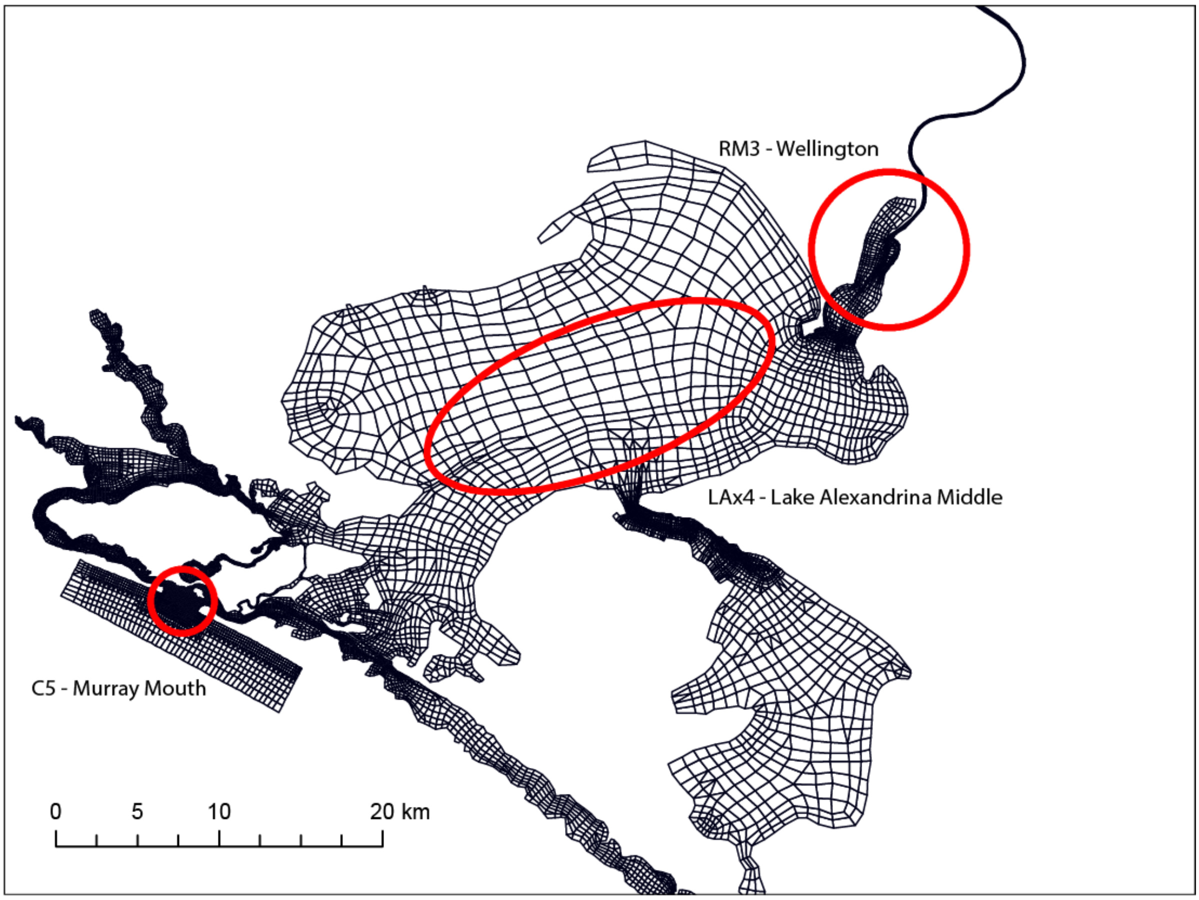


Figure 25. Modelled cells (circled) used for calculating the modelled concentration of nutrients or salt at the Wellington, Lake Alexandrina Middle and Murray Mouth sites.

The transport of matter was assessed through modelled exports from the Murray River Channel (Wellington), Lower Lakes (Barrages) and Coorong (Murray Mouth). Findings are presented for salinity, ammonium, phosphate, dissolved silica, organic nitrogen, organic phosphorus and chlorophyll *a*. Salinity is presented as practical salinity units (PSU), a measurement of the measured conductivity to standard potassium chloride (KCl) conductivity. PSU was used for validating model outputs as it overcomes observed differences in electrical conductivity caused by changes in water temperature. One PSU is approximately equal to one part per thousand.

Given the increasing efforts to improve and restore the Coorong, a high-resolution model of the Coorong was developed, which is considered to be more accurate in resolving the salt, nutrients and habitat across the system. The Coorong only domain (Figure 26) was run under the same scenarios above, but due to the long residence time, these simulations were run from 1 July 2017 to 30 June 2020, i.e., a period of three years; this window was chosen as it began after the high flow event in 2016 which "reset" salinity levels in the Coorong. The simulation included gauged Salt Creek inputs and measured ocean water levels based on the Barker Knoll telemetered site. Weather data were also used as a boundary condition in order to predict the effect of wind and evapo-concentration effects. Because of the importance of salinity movement in this system, a detailed salt and nutrient flux analysis was undertaken to understand the rate of salt and nutrient accumulation in both the North and South of the Coorong.

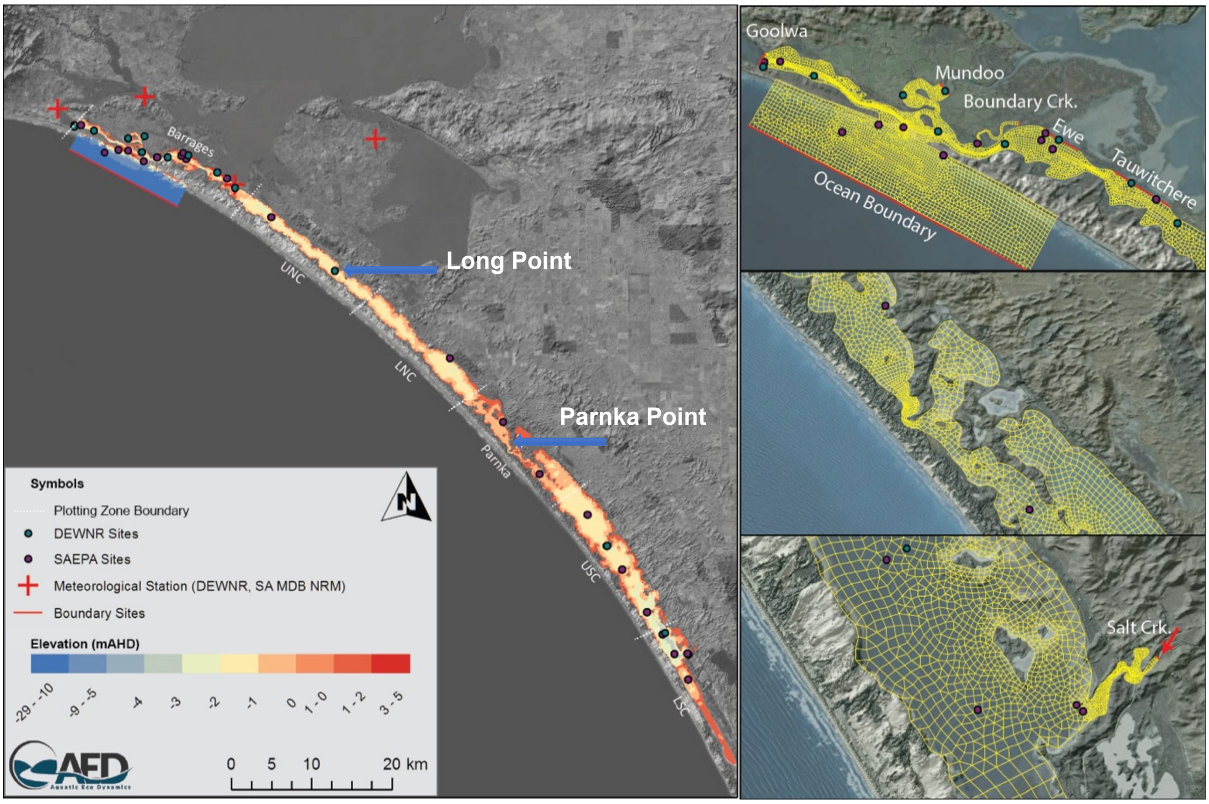


Figure 26. Fine resolution of the Coorong only domain, used for detailed Coorong salt and nutrient flux calculations, and habitat assessment of *Ruppia* and key fish species (Section 2.3.2).

The salt load exported from the Murray River is a function of the flow volume and the salt concentration and is unidirectional. Salinity in the Coorong is more complicated and needs to consider flow from the river, salt concentrations already in Coorong, which varies with distance from the Murray Mouth, salt input from the South East which is a function of volume and salt concentration of water draining from the large South East catchment, and input of water from the ocean which varies with the seasonal fluctuation in sea-level, daily tide variations and the volume of water exiting the Murray Mouth which acts to prevent seawater ingress. In addition to the "**All water**" and "**no eWater**" scenarios, we ran a "**no eWater 3yrs**" scenario to demonstrate the cumulative effects of water delivery to the lagoon conditions.

Results

Salt flux through the Murray River domain

Commonwealth environmental water was the only water that exited the barrages for the 2019-20 water year and so all salt export over the barrages is attributable entirely to this (Table 5). A total salt load of 623,999 tonnes was exported from the river basin for 2019-20.

If there was no environmental water in 2019-20 then no salt would have been exported from the basin which implies salt would be accumulating upstream in wetlands and floodplains.

In 2019-20, there was a net import of salt into the Coorong (through the Murray Mouth), which is consistent with all low flow years. In 2019-20 the net salt import to the Murray Mouth was 335,926 tonnes (Table 5). Without environmental water, the net import of salt would have been much larger at 2.3 million tonnes. Environmental water decreased salt import by approximately 2 million tonnes (Table 5), all of which is specifically attributable to Commonwealth environmental water.

Table 5. Six-year record of modelled salt export (tonnes) over the barrages to the Coorong estuary and through the Murray Mouth into the Southern Ocean.

Scenario	2014-15	2015-16	2016-17	2017-18*	2018-19*	2019-20*
Barrages						
All water	446,855	288,516	1,504,541	496,936	532,333	623,999
No CEW	161,791	36,884	1,383,674	60,088	0	0
No eWater	152,406	31,031	1,317,791	60,088	0	0
Murray Mouth						
All water	-157,852	-1,850,028	3,679,277	-497,342	-16,807	-335,926
No CEW	-3,202,552	-6,441,297	3,159,985	-2,168,279	-1,864,080	-2,332,963
No eWater	-5,048,511	-6,649,380	1,958,989	-2,168279	-1,864,080	-2,332,963

* The salt export and salinity data reported for years 2017-18 to 2019-20 are from the new high-resolution Coorong only model, which is different from the large domain model previously used for 2014-15 to 2016-17 and has updated barrage flow specification. The full domain model is estimated to underpredict the salinity and salt flux at the barrages, and hence the 2014-15 to 2016-17 are systematically lower than the other years. The post 2017 predictions with the Coorong model are considered to be more accurate, based on our recent validation assessment (Appendix D).

Salinity dynamics within the Coorong

The results of the three Coorong flow scenarios (“All water”, “no eWater” and “no eWater 3yrs"), were compared with the salinity measured continuously at monitoring stations in the Coorong. For the base-case, there was a good fit between sensor data and modelled data at all sites (Figures D3 and D4, Appendix D). The general trend is seen as the system getting more saline and more seasonal increases in salinity as you move further south (Figure 27; Figure 28). Transition zones between the river and ocean (Murray Mouth) and between lagoons show a greater salinity range as there is more exchange of water and salt at these zones.

The analysis shows the model can predict the salinity in the Coorong and the relative differences that would have occurred if environmental water was not provided. The dynamic nature of salt flux is further evident in the panels illustrating the salt mass flux between the various Coorong locations (Figure D5, Appendix D) for the base-case scenario, which has environmental water flow over the barrages. Salt flux over the barrages is unidirectional and acts to freshen the Murray Mouth region. Ocean input to the Coorong is variable but net positive over the period July 2017–July 2020 (Figure D5b, Appendix D). The role of freshwater flowing from the river is to limit this salt influx from the ocean and maintain appropriate salinity in the Coorong. Salt from the Murray Mouth can travel southward as it replenishes water that is evaporating in the Coorong. The dominant direction of salt flux is southward although it can move northward (negative flux in Figure D5c, Appendix D) when river flows over the barrages cease, the head of water decreased and the net flow of water is northwards.

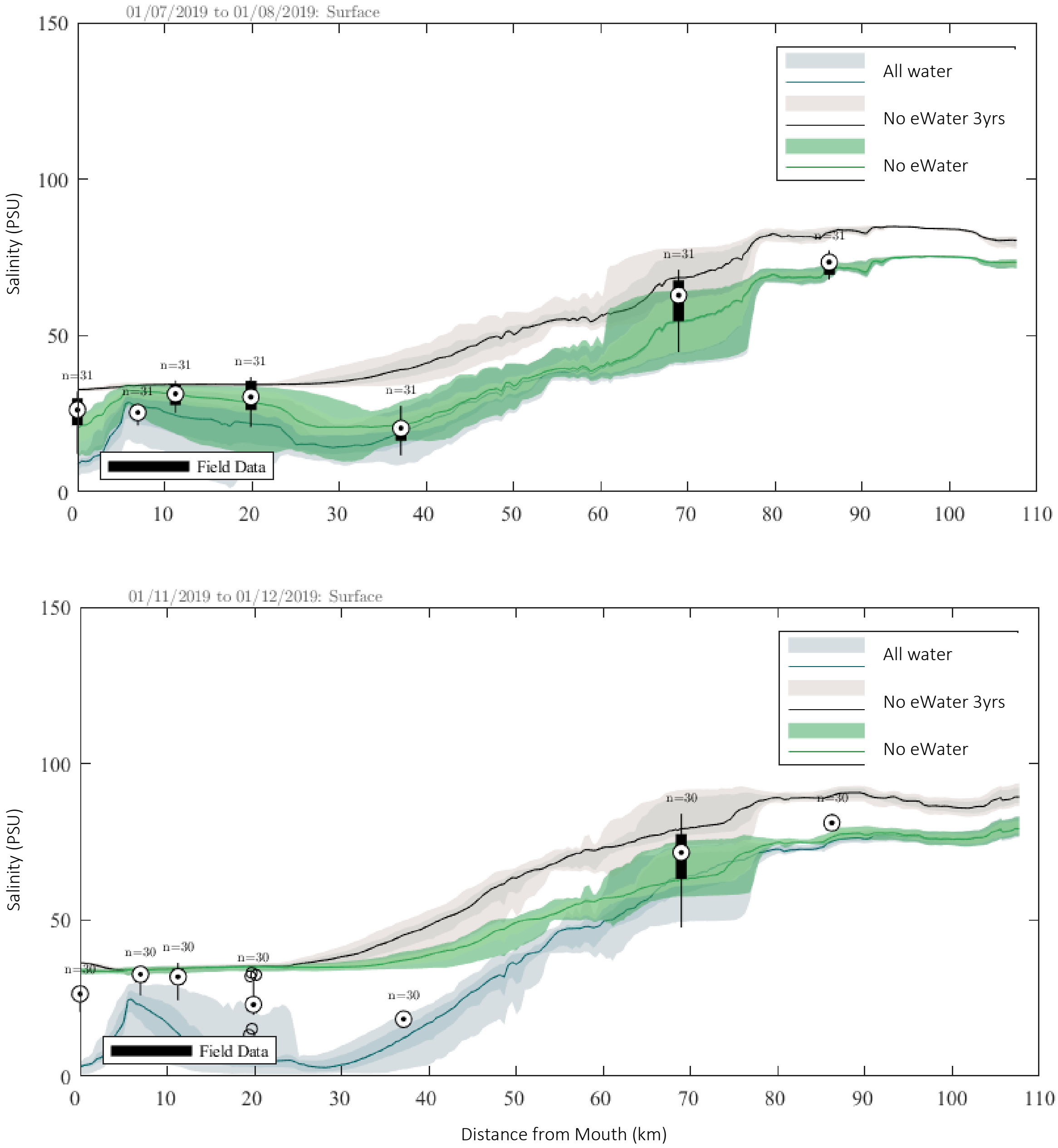


Figure 27. Comparison of measured and simulated salinity along the length of the Coorong lagoon (box-whisker), moving from the Murray Mouth into the South Lagoon. Model simulations for the "All water" (base-case observed conditions), and the no environmental water scenarios ("no eWater" and "no eWater 3yrs") are shown for July (top) and November 2019 (bottom).

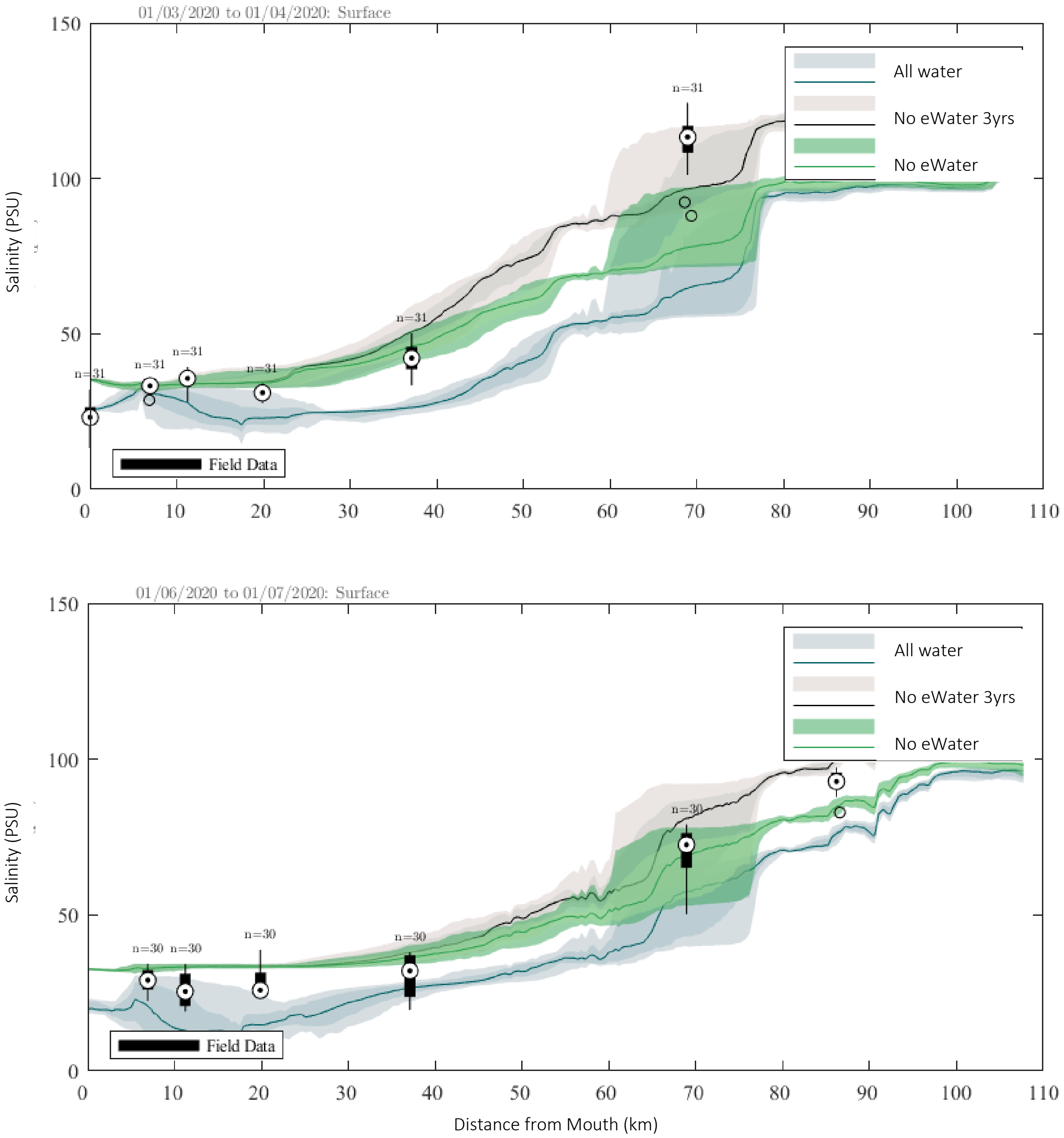


Figure 28. Comparison of measured and simulated salinity along the length of the Coorong lagoon, moving from the Murray Mouth into the South Lagoon. Model simulations for the "All water" (base-case observed conditions), and the no environmental water scenarios ("no eWater" and "no eWater 3yrs") are shown for March (top) and June 2020 (bottom).

Further to the South at Parnka Point there are much higher flux rates as the salt concentrations are higher and so the rate of salt movement is higher (Figure D5d, Appendix D). This is evident in the cumulative flux of salt at Parnka Point which is considerable higher in both the northward and southward vectors (Figure 29) than salt flux at Long Point.

Without environmental water delivered, river flow over the barrages would only have occurred in 2017-18 not in 2018-19 or 2019-20. The flow would have only transported ~40% of salt from the river catchment (Figure D6a, Appendix D) that would be exported with additional Commonwealth environmental water. Due to the low unregulated flow over the barrages in 2017-18 and no flow in subsequent years, there was considerably more salt imported from the ocean into the Murray Mouth region, resulting in greater southward salt flux (Figure D6, Appendix D). Without environmental water the cumulative southward salt flux in 2019-20 (Figure 29) would be three times greater than with environmental water (Figure 29). Environmental water reduced the salt load to the South Lagoon, measured as salt flux southward at Parnka Point, by over 3.244 million tonnes over the three-year period between July 2017 and July 2020.

Environmental water is required in every year to reduce excess salt accumulation in the Coorong. For the 'no eWater 3yrs' scenario when assuming environmental water was delivered in the first two years (2017-18 and 2018-19) but not in 2019-20, salt seems to have accumulated at a slower rate but at the end of the simulation period, the net accumulated flux is still more than twice what would have occurred had environmental flows been delivered in 2019-20 (Figure 29).

Monthly salt exports with and without environmental water delivery for July 2017–July 2020 show how seasonally dynamic salt export was in the Coorong (Figure D8, Appendix D). Figure 29 shows the cumulative net southward amount of salt mass into the North and South lagoons of the Coorong. Environmental water delivery maintained this flux to be close to zero over the period of interest.

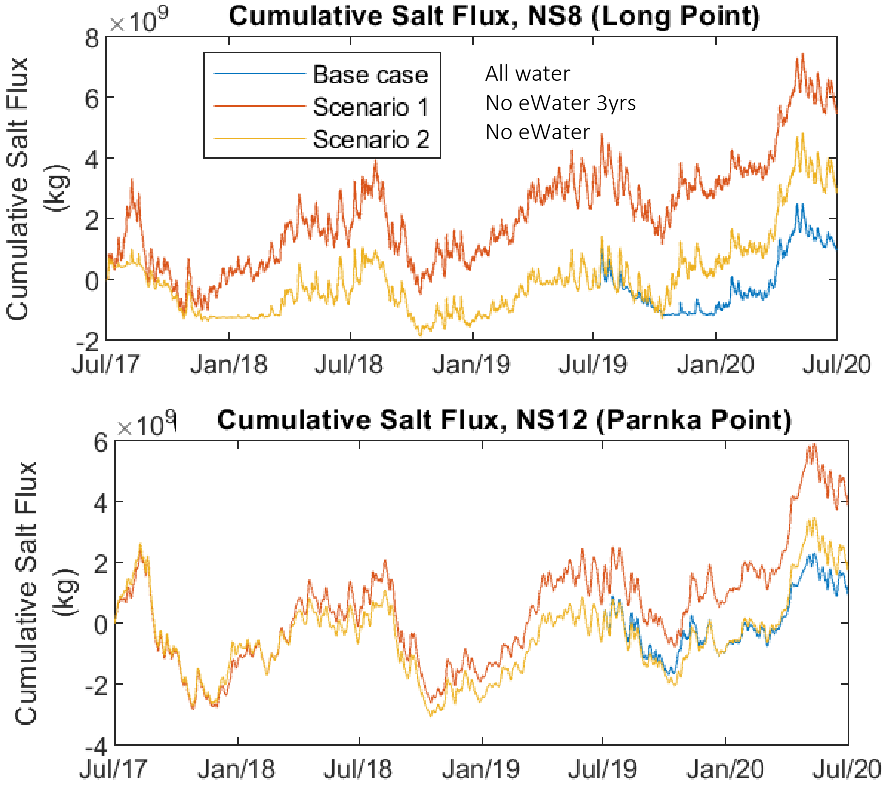


Figure 29. Cumulative net southward amount of salt mass into the North Lagoon (through Long Point) and South Lagoon (through Parnka Point) in the Coorong from July 2017–June 2020. Scenarios include with "All water", without environmental water ("no eWater 3 yrs") and without any environmental water in 2019-20 ("no eWater"). eWater delivery maintained this flux to be close to zero over the period of interest; even one year of no environmental water over the barrages contributes to salt accumulation in the North and South Lagoon. Note that only CEW water contributed to the eWater entering the Coorong (see Appendix D for barrage flow amounts).

Nutrients

In 2019-20, the total nitrogen (TN) export was 1,411 tonnes and total phosphorus (TP) export was 139 tonnes (Table 6). Commonwealth environmental water contributed to 100% of barrage flows during this year, and therefore 100% of the export of particulate organic nutrients.

Three-year model runs for three flow scenarios indicated that without any environmental water (no eWater 3yrs) and without environmental water only in 2019-20 (no eWater), there was less nitrogen transported to the Coorong than in the base-case (All water) which had environmental water delivery in each year (Figure 30). However, environmental water showed negligible effect on nitrogen transport to the South Lagoon.

Table 6. Total loads of dissolved and particulate matter (tonnes) in 2019-20.

Site	Scenario	Ammonium	Phosphate	Silica	Total nitrogen	Total phosphorus	Chlorophyll *a*
Barrage	All water	11.58	0.49	12693.26	1410.71	139.44	5.31
	No CEW	0	0	0	0	0	0
	No eWater	0	0	0	0	0	0
Murray Mouth	All water	0.94	1.56	-12321.45	-1224.00	-123.38	-2.10
	No CEW	9.03	3.93	-248.34	182.80	13.07	2.54
	No eWater	9.03	3.93	-248.34	182.80	13.07	2.54
South Lagoon (Parnka)	All water	0.11	-0.16	911.05	131.67	12.17	-0.18
	No CEW	0.40	-0.05	154.48	66.41	4.76	0.74
	No eWater	0.40	-0.05	154.48	66.41	4.76	0.74

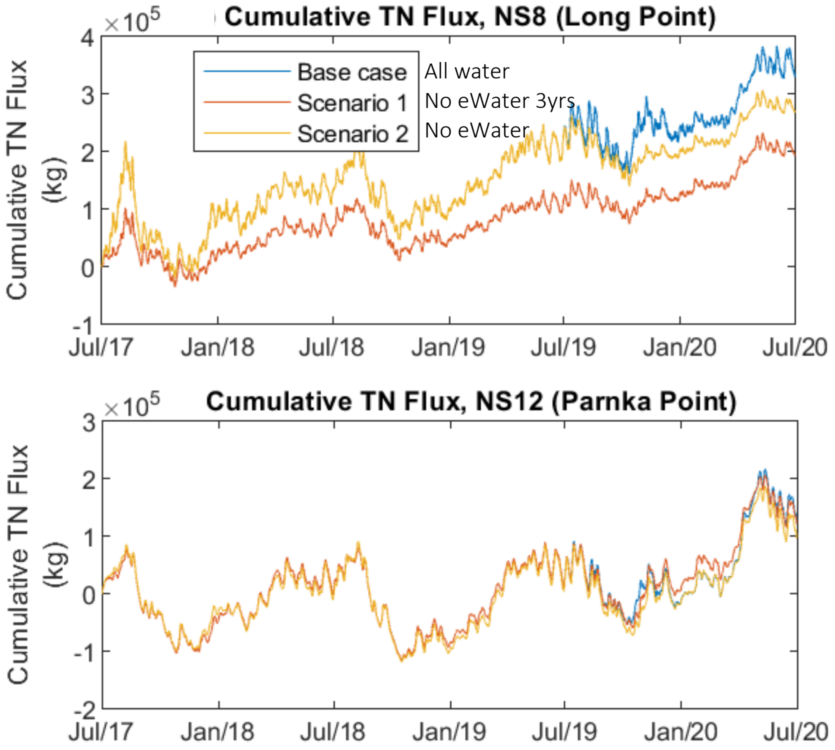


Figure 30. Nitrogen flux (cumulative net amount) through the Coorong with and without environmental water delivery for July 2017–June 2020. Scenarios included are "All water", without any environmental water ("no eWater 3 yrs") and without environmental water for 2019-20 ("no eWater").

Chlorophyll a

In 2019-20, barrage flows resulted in the export of 5.5 tonnes of phytoplankton biomass (measured by chlorophyll *a*) (Table 7). This was 100% attributable to Commonwealth environmental water.

Table 7. Six-year record of modelled phytoplankton (chlorophyll) export expressed as carbon, (tonnes) over the barrages to the Murray estuary and Coorong.

Scenario	2014-15	2015-16	2016-17	2017-18	2018-19	2019-2020
All water	27	23.5	2,939	221.5	5.8	5.5
No CEW	21	1.9	2,748	63.9	1.6	0
No eWater	19	1.5	2,629	25.7	0	0

Evaluation

The evaluation approach, including assessment criteria, is described in the evaluation section for Hydraulic Regime (Section 2.1).

Table 8. Matter Transport evaluation questions and answers. CEW = Commonwealth environmental water.

<table>
<tr><th colspan="2" rowspan="2">CEWO evaluation questions</th><th colspan="6">Outcomes of CEW delivery</th></tr>
<tr><th>2014-15</th><th>2015-16</th><th>2016-17</th><th>2017-18</th><th>2018-19</th><th>2019-20</th></tr>
<tr><td rowspan="2">What did CEW contribute to salinity:</td><td>Levels?
Reduction at the Murray Mouth due to CEW (Median salinity presented, PSU)</td><td>From 34.6 to 24.0 *</td><td>From 35.1 to 29.7 *</td><td>From 21.6 to 11.3 *</td><td>From 28.9 to 15.9</td><td>From 31.9 to 21.1</td><td>From 31.9 (without CEW) to 21.3 (with CEW)</td></tr>
<tr><td>Transport?
Additional export over barrages due to CEW (tonnes salt per year)</td><td>285,064*</td><td>251,632*</td><td>120,867*</td><td>436,848</td><td>532,222</td><td>623,999</td></tr>
<tr><td colspan="2">What did CEW contribute to the salinity regime?</td><td colspan="6">CEW increased salt export over the barrages; and reduced salt intrusion into the Murray Mouth from the ocean, which reduced salinity in the Coorong.
CEW played a key role in delivering flow to the Coorong, particularly during dry years with 80–100% of barrage releases being CEW.
CEW has played a key role in salt export from the Basin, accounting for 64, 87, 69, 70%** and 100% of salt export, during the five years of low flow (2014-15, 2015-16, 2017-18, 2018-19 and 2019-20), respectively. In these years, the total salt export ranged 228,293* – 623,999 tonnes, which is well below the Basin Plan target of 2 million tonnes of salt per year.
In the high flow year (2016-17), 1.5 million tonnes* was exported and CEW contributed 8% (120,867 tonnes) of salt export.</td></tr>
</table>

CEWO evaluation questions		Outcomes of CEW delivery					
		2014-15	2015-16	2016-17	2017-18	2018-19	2019-20
What did CEW contribute to nutrient and phytoplankton transport?	Nitrogen	609	1,007	123	1,508	816	1,145
	Export through barrages due to CEW (tonnes)	CEW increased nitrogen export as nitrogen load was largely a function of flow volume. Nitrogen fuels phytoplankton growth and food and so is important for web productivity					
	Phosphorus	54	90	11	137	77	103
	Export over barrages due to CEW (tonnes)	CEW increased phosphorus export as phosphate load was largely a function of flow volume.					
	Silica	3,551	6,836	0	8,787	5,469	10,787
	Export over barrages due to CEW (tonnes)	CEW increased silica export as silica load was largely a function of flow volume and CEW contributed 100% of flow. 2016-17 was a flood year and so the silica export that year may be a function of the concentrations used to model. Total silica export was 70,207 tonnes, although not attributed to CEW.					
	Phytoplankton	8	6	14	12	6	5.6
	Export over barrages due to CEW (tonnes)	CEW increased phytoplankton export as chlorophyll load was a function of flow volume and 100% of flow over the barrages is attributable to CEW. However, the significance of the outcome (load of phytoplankton export) was low in 2019-20 and so the overall contribution was negligible.					

* The salt export and salinity data reported for years 2014-15 to 2016-17 are from the large domain model previously used in the LTIM reporting. The numbers for 2017-18 to 2019-20 are from the new high-resolution Coorong only model, which uses a different method for barrage flow calculation and has a more accurate specification of salinity and salt flux. When we compared the two models, salt flux at the barrages was underestimated by the full domain model, and hence the 2014-15 to 2016-17 are systematically lower than the other years. The post 2017 predictions are considered to be more accurate, based on our recent validation assessment (Appendix D).

**Matter transport results are based on different modelled data and may not necessarily reflect the CEWO accounted data.

Contribution (to what extent CEW contributed towards the outcome, with the significance of the outcome considered):

⬜ Unknown 🟥 Negative 🟩 None/negligible 🟩 Minor 🟩 Moderate 🟦 Substantial

Discussion

Salinity

Environmental water dilutes salt in the LMR channel, Lake Alexandrina the Murray Mouth estuary and along the Coorong. The salinity was maintained well within the range required for potable water in the river and lake from 2014-15 to 2019-20, but water was about 10% fresher with environmental flows. It is conclusive that environmental flows are beneficial to river salinity and helped maintain river salinity below 800 EC at Morgan, which is a river management target of the MDBA and SA Water. Given the confidence in the fact that environmental water dilutes salinity in the main river channel and the Lower Lakes, the focus has shifted, since 2019-20, to how environmental water affects transport of material over the barrages and affects conditions within the Coorong.

The median salinity in the Murray Mouth in 2019-20 was 21.3 PSU which was lower than 2018-19 (median salinity 30.9 PSU), similar to 2017-18 (26.2 PSU) but higher than in 2016-17 (11.3 PSU). The 2016-17 fresher conditions reflected high river flows where flow into South Australia peaked at 94,600 ML/d. Commonwealth environmental water created fresher conditions at the Murray Mouth in 2019-20, compared to the without environmental water scenario.

Salinity in the Coorong is primarily a function of riverine inflows and tidal movement. When barrage flows are low, seawater enters the Murray Mouth and more salt is then transported to the Coorong where it is subject to evapo-concentration. Environmental water made up 100% of flow over the barrages and reduced the salt load to the South Lagoon measured as salt flux southward at Parnka point, by over 3.244 million from July 2017 to June 2020. It is also evident that environmental water flowing over the barrages is required in every year to reduce excessive salt accumulation within the Coorong. If environmental water had not been delivered in 2019-20 an additional 1.7 million tonnes of salt would have accumulated in the South Lagoon.

During the Millennium Drought, and particularly from 2007-08 to 2009-10, flow over the barrages ceased and the import of salt into the Coorong resulted in salinity in the South Lagoon that was five times seawater salinity, and demise of much of the aquatic life (Brookes *et al.* 2009). Environmental water provides freshening flows but also acts to inhibit seawater intrusions, thereby maintaining more appropriate salinity conditions in the Coorong. Given that barrage releases almost entirely (up to 100%) depend on Commonwealth environmental water in dry years, environmental water is critical for limiting salt flux to the South Lagoon. Even one year without barrage flow can result in a large flux of salt southwards (1.7 million tonnes net southwards flux). Salinity is a key determinant of *Ruppia* and fish habitat and is addressed in the following section.

Nutrients

During the LTIM monitoring period (2014–2019), the median concentrations of nitrogen and phosphorus did not vary considerably with or without environmental water. Environmental water, however, contributed considerably to the transport of nutrients, and this was primarily due to additional volume not a change in the nutrient concentrations. It was evident that environmental flows contributed a considerable load of nutrients to the Murray Mouth between 2014-15 and 2019-20. From this evidence, it can be concluded that environmental flows are a key driver in promoting estuarine productivity.

Commonwealth environmental water is the only water that flowed through the barrages into the Murray Mouth region in 2019-20 and so it accounts for all nutrient delivery to the estuary. If there was no environmental water, the nutrient load over the barrages would have been zero.

For the current MER modelling the focus has shifted to delivery of nutrients to the Murray Mouth and the implications of nutrient flux to the Coorong. The South Lagoon of the Coorong shows very high primary productivity, with problematic filamentous algae forming thick mats that detach and smother *Ruppia* plants, breaking off the flowering stems. From 2019/20, approximately 70 tonnes of additional nitrogen entered the South Lagoon when environmental flows were present than if no river water flowed through the barrages. These filamentous algae are supported by nutrients but it is unclear if their recent emergence is attributable to nutrients, a change in the salinity, or freshwater flows or inoculum from the South East drainage scheme. This then raises the question – is this flux of nitrogen to the Coorong problematic or beneficial? Nutrients *per se* are not the problem, but they can be problematic if present in excessive concentrations or when they enter problematic pathways such as algal blooms. In the case of the Coorong, the benefits from river flows reducing sea water ingress, reducing salinity in the South Lagoon and promoting favourable habitat for *Ruppia* and fish far exceed any potential issues caused by nutrients.

Chlorophyll a

Chlorophyll *a* is a photosynthetic pigment that is ubiquitous in the phytoplankton, so is often used as a measure of the relative size of the phytoplankton community. A considerable amount of the total organic nutrients is likely to be bound within phytoplankton, and so the chlorophyll loads reflect the loads of particulate organic nitrogen and phosphorus. Chlorophyll export can be interpreted as a transfer of food resources from one site to another. In 2019-20 flow was low and there was limited transfer of food from the river to the Coorong and coastal environments. Without environmental water this transfer of resources would have been even lower. River discharge likely played a significant role in promoting estuarine productivity (Giatas *et al.* 2018), contributing 5.6 tonnes of chlorophyll via barrage releases to the Murray Mouth. The Southern Ocean is oligotrophic and consequently nutrients and phytoplankton from the river are likely to play a significant role in coastal marine productivity.

Management implications

There are approximately 10^{11} tonnes of salt in groundwater in the MDB and an additional 1.5 million tonnes of salt is deposited in the basin each year by rainfall (Herczeg *et al.* 2001). Unless salt is exported from the basin with flow, there will be a net accumulation of salt within the basin. The Basin Plan sets out a salt export objective (section 9.09) to ensure adequate flushing of salt from the Murray River system into the Southern Ocean. The Basin Plan's indicative target for salt export from the Basin is 2 million tonnes per year. The five years of salt export modelling enable the contribution of environmental flows to salt export to be scrutinised (Table 5). Flow has been relatively low in fiver of the six years of LTIM/MER monitoring. In these years (2014-15, 2015-16, 2017-18, 2018-19 and 2019-20), Commonwealth environmental water played a key role in salt export from the basin, accounting for 64–100% of total salt export. In the high flow year (2016-17), 1.5 million tonnes were exported and Commonwealth environmental water contributed 8%.

Maximum exports of matter from the Murray Mouth are likely to be achieved by delivering environmental water during periods of low oceanic water levels (e.g. summer). In contrast, environmental water delivery to the Murray River Channel at times of high oceanic water levels, which peak in the Austral winter, is likely to increase the exchange of water and associated nutrients and salt through the Coorong, rather than predominately through the Murray Mouth. This may decrease salinities and increase productivity within the Coorong more than what would occur if water is delivered at times of low oceanic water levels.

The load of nutrients exported from the basin over the barrages is an interesting issue; on one hand nutrient export drives estuarine productivity, but on the other hand it is desirable to maintain an appropriate level of nutrients in the estuary and Coorong where they can support aquatic productivity.

Freshwater flows act to freshen the Coorong but also transport nutrients into the South Lagoon. The benefits from river flows reducing salinity in the South Lagoon and promoting favourable habitat for *Ruppia* and fish and a more resilient ecosystem far exceed any potential issue caused by nutrients.

Conclusion

The contributions of environmental water appear to have significantly increased the export of dissolved and particulate matter through the LMR to the Southern Ocean. In low flow years, environmental flow delivery can play a key role in salt export from the Basin. In 2019-20, Commonwealth environmental flow was responsible for 100% of barrage flows and thus all salt export from the basin. Environmental water is also critical in reducing salt import into the Murray Mouth from the ocean, lowering salinity in the Coorong and maintaining estuarine habitat to support ecological functions and biodiversity. Environmental flow deliveries during periods when there would otherwise be negligible water exchange between the Lower Lakes and Coorong is critical for maintaining the connectivity between freshwater and marine environment and promoting fresher conditions in the Coorong with more favourable habitat for estuarine fish and plants.

2.3.2 Coorong Habitat

Background

The Coorong is an estuarine lagoonal system with a natural salinity gradient ranging from freshwater to hyper-saline at the extremity. Freshwater flows are important in maintaining estuarine habitat and ecosystem health and preventing extreme hyper-salinity (Brookes *et al*. 2009). *Ruppia tuberosa* is an important macrophyte in the Coorong that provides habitat for fish and food for herbivorous birds in the Coorong (Phillips and Muller 2006), and it can tolerate a salinity higher than natural seawater. The germination and growth of *R. tuberosa* is known to be governed in large part by changes in salinity and water level regimes, which are influenced by flows through the barrages (Kim *et al.* 2013). Other factors that influence *R. tuberosa* growth include nutrient availability, water temperature, sediment quality and interactions with algae, including shading of light and interference with flowers and fruits on the surface (Collier *et al*. 2017). Early summer flows are likely to be particularly beneficial as they delay the drop in water level in the South Lagoon and can prevent extreme salinities emerging, thereby improving chances of completion of the reproductive cycle. In addition, salinity has also been identified as a key driver of fish distribution and assemblage structure by influencing the extent of estuarine fish habitat in the Coorong (Ye *et al.* 2011; 2016a; Bice *et al.* 2018). This sub-indicator aims to assess the benefits of environmental flows for the enhancement of *R. tuberosa* habitat, particularly those that are delivered in summer, as well as the improvement of estuarine fish habitat throughout the year for several key species with different levels of salinity tolerance.

Major hypothesis

Increased freshwater flow through the barrages and into the Coorong due to environmental watering will prevent areas of extreme salinity and increase water levels in the Coorong, thus expanding the extent of *R. tuberosa* and fish habitats.

Methods

This assessment applied the high-resolution Coorong model reported in the previous section, to simulate water level, salinity and key water quality attributes, spanning from the barrages to the southern end of the South Lagoon. The hydrodynamic model was forced by daily barrage flows (accounting for barrage operation logic), and oceanic and meteorological conditions. Salinities and water level along the North and South lagoons of the Coorong are calculated at a fine spatial resolution (Figure 27), allowing analysis of suitable areas of habitat. For fish and *R. tuberosa* (“*Ruppia*”), suitable habitat was determined according primarily to the salinity tolerances of biota, and does not take into account other variables (e.g. sediment type and food resources) that may also influence habitat. The model has been validated in detail against available data from water level and salinity monitoring across the Coorong; validation against 2019-20 data is shown in the previous section (Appendix D).

Results of salinities and water levels from scenarios with and without environmental water were used to estimate habitat extent of *Ruppia* and fish using previously reported environmental thresholds (Ye *et al*. 2016a; Collier *et al*. 2017). The *Ruppia* ecological response model has capability to account for habitat suitability of critical life stages, and is designed to estimate the probability of replenishing the sediment seed-bank, turion sprouting, seedling development to juvenile plants, and adult plant flowering and seed setting (Collier *et al*. 2017). Each stage (adult, seed, flowering) is assigned a suitability based on cell specific light, depth, salinity and temperature, which in the end results in a combined probability of sexual life-cycle completion. A suitability index for asexual reproduction is being considered but it is not considered sufficiently robust to include. Where the index was above 0.4, we computed a suitable area of habitat, by multiply the habitat score by the cell area, and summing over all cells within both lagoons.

A basic fish model was also implemented which calculates probabilities of habitat suitability for juveniles of key species based on known salinity thresholds. This was computed based on data for mulloway (*Argyrosomus japonicus*), black bream (*Acanthopagrus butcheri*), greenback flounder (*Rhombosolea tapirina*), yelloweye mullet (*Aldrichetta forsteri*), congolli (*Pseudaphritis urvillii*), Tamar goby (*Afurcagobius tamarensis*) and smallmouth hardyhead (*Atherinosoma microstoma*). The model adopts a seasonal effect by account for temperature sensitivity to the salinity thresholds, according to functions and parameters described in Table 9 and Table 10.

Table 9. Salinity suitability at different temperature levels for a range of Coorong fish species used to develop a habitat suitability index (HSI).

$HSI = \begin{cases} 1, 0 \le S < S_{c10} \\ \frac{S_{c50}-S}{S_{c50}-S_{c10}}, S_{c10} \le S < S_{c50} \\ 0, S \ge S_{c50} \end{cases}$	where : $S_{c10} = \begin{cases} LC_{10}^{14}, 0 \le T < 14 \\ (\frac{LC_{10}^{23}-LC_{10}^{14}}{23-14})(T-14) + LC_{10}^{14} \quad, 14 \le T < 23 \\ LC_{10}^{23}, T \ge 23 \end{cases}$ $S_{c50} = \begin{cases} LC_{50}^{14}, 0 \le T < 14 \\ (\frac{LC_{50}^{23}-LC_{50}^{14}}{23-14})(T-14) + LC_{50}^{14} \quad, 14 \le T < 23 \\ LC_{50}^{23}, T \ge 23 \end{cases}$

S: salinity; S_{c50}: LC50; S_{c10}: LC10; T: temperature

Table 10. Summary of LC_{50} and LC_{10} estimates (expressed as ppt) for six species at 14°C (LC^{14}) and 23°C (LC^{23}) temperatures.

Common name	LC_{50}^{14}	LC_{50}^{23}	LC_{10}^{14}	LC_{10}^{23}
Mulloway	64	59	60	51
Tamar goby	73	71	68	66
Black bream	85	88	79	82
Greenback flounder	88	79	81	73
Yelloweye mullet	91	82.4*	84	68
Congolli	100	94	90	87
Smallmouth hardyhead	108		100	97

*possibly affected by another unknown water quality factor.

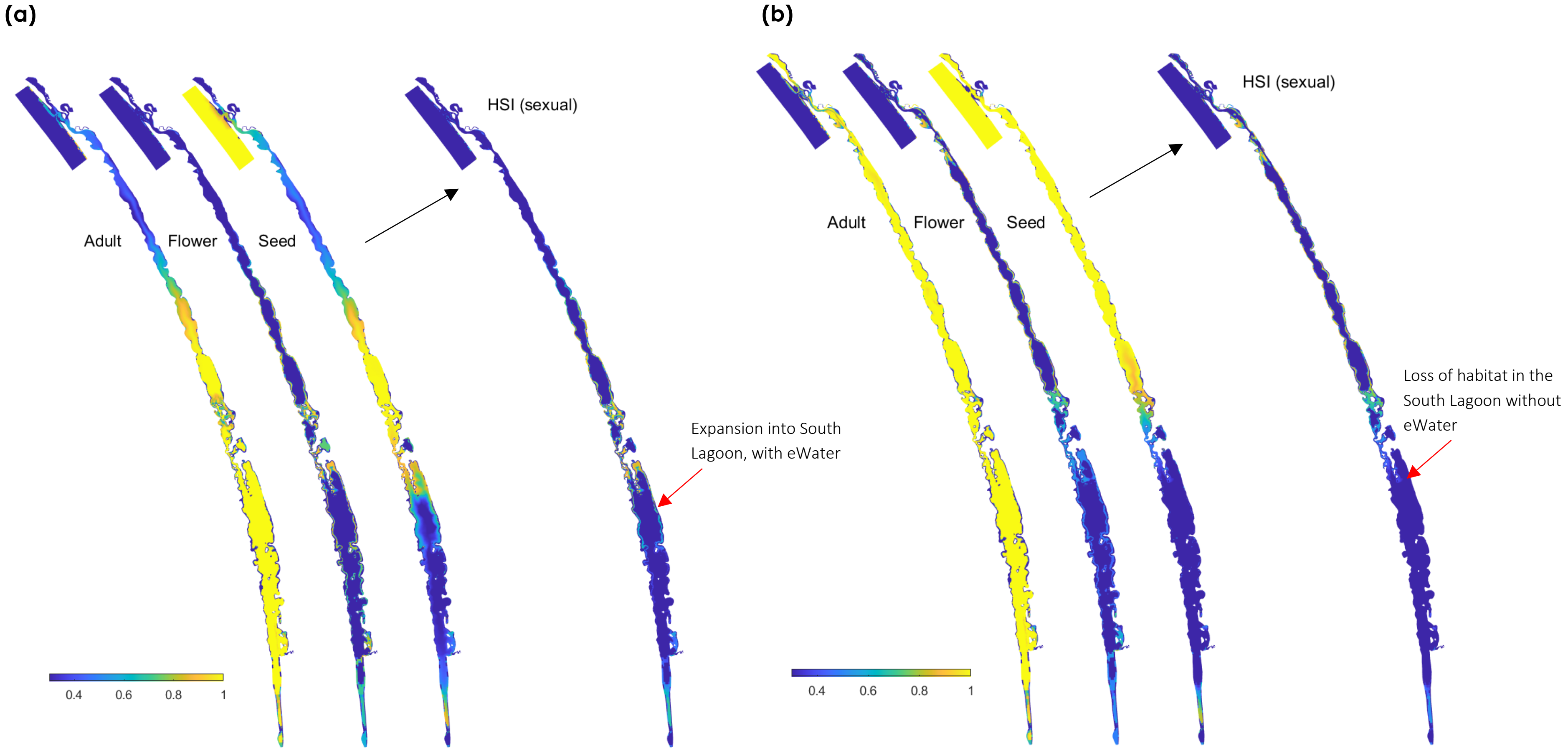
The Coorong has a long residence time and to account for the longer timescales of water and solute flux, three-year simulations were undertaken to assess habitat. The period simulated was from July 2017 to June 2020 which was chosen as it begins after the large flow event in 2016 "reset" salinity levels in the Coorong and covers three water years (and growing seasons). Rather than dynamically predicting barrage flows based on water levels either side of the structures, this simulation specifies the daily barrage flows based on flow volumes obtained from SA Water (see Section 2.3.1).

Results

Ruppia habitat

The *Ruppia* ecological response model predicts habitat suitability of critical life stages, in response to light, depth, salinity and temperature, which in the end results in a combined probability of sexual life-cycle completion. The requirements for each life stage are quite different, and when they are superimposed together, the areas where life-cycle completion is most likely becomes apparent on the margins of the Coorong lagoons, and the shallow areas around Parnka Point (Figure 31). By comparing the scenarios with and without environmental water, we note that there is a significant expansion of favourable habitat in the north of the South Lagoon, and along the South Lagoon margins, which is a priority area of *Ruppia* restoration.

The overall quality-weighted area of habitat (the suitable area weighted by the cells overall quality) that is able to complete a full sexual reproduction life-cycle, changes over the three years between approximately 25 km^2 and 40 km^2, and this appears to be mainly limited to the area suitable for successful flowering as the suitable area for plant growth is much higher (~150 km^2) (Figure 32). When integrated over the entire Coorong and Murray Mouth region, there was not a large difference between scenarios, and it was noted that suitable habitat area for adults appears to be slightly higher in the scenario which has no environmental water delivery (Figure 32), which was counter-intuitive considering the obvious benefits in the expansion of suitable habitat into the South Lagoon in Figure 31. On closer inspection of these results it shows that this was due to freshening of salinity around the barrages, which reduced the suitability of habitat in this location, though we note this is not a traditional area of *Ruppia* meadows and it is highlighted that in the area that is the target of management and restoration, there is a notable expansion of suitable area.



Expansion into South Lagoon, with eWater

Loss of habitat in the South Lagoon without eWater

Figure 31. Comparison of *Ruppia* HSI (habitat suitability index) values under (a) “All water” (base-case) conditions and (b) no environmental water since 2017 (No eWater 3yrs). The index is computed for each stage of the life-cycle, and the overall suitability is based on where all life-stage requirements are met. More saline conditions in ‘no eWater’ scenario leads to a loss of good habitat in the South Lagoon that meets the requirements of all life-stages.

The performance of seed production and overall sexual reproduction is moderately better in 2018-19 and 2019-20 with environmental flows (as seen in the "All water" and "No eWater" simulations: Figure 31).

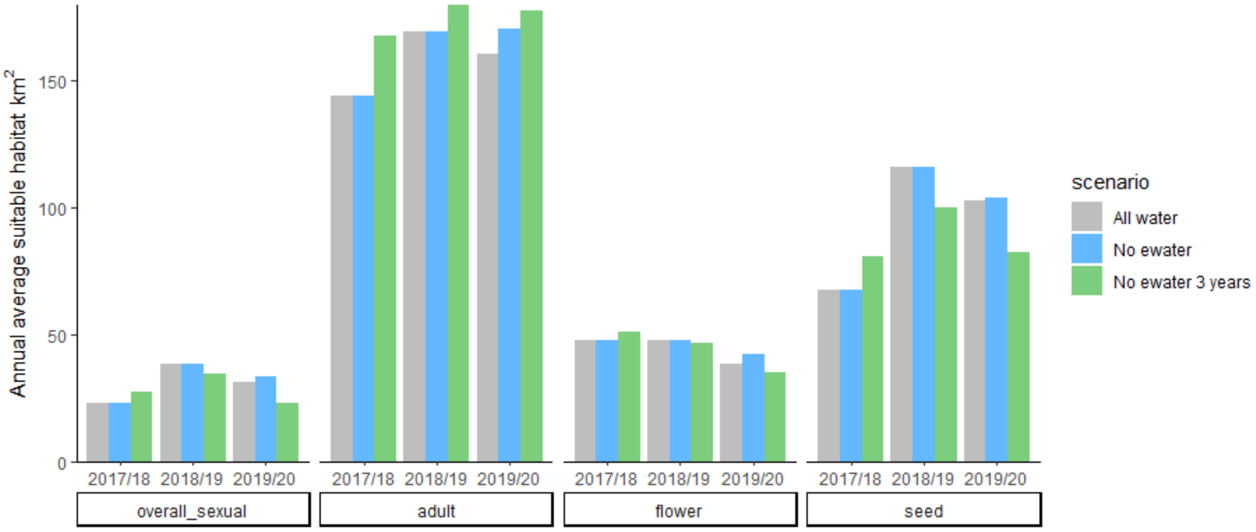


Figure 32. Habitat area suitable for *Ruppia tuberosa* (overall and individual life stages), comparing the "All water" (base-case) and without environmental water ("No eWater" and "No eWater 3 yrs).

Fish habitat

Environmental flows led to fresher conditions in the Coorong and an expansion of suitable fish habitat area; an example of the new habitat area created by environmental water is shown as a map for mulloway (Figure 33). Maps for other species have different ranges but similar overall patterns. This analysis shows the ΔHSI, that is the change between scenarios; in this case a value of 1 represents an area that was unsuitable under the "No ewater" scenario becoming fully suitable under the "All water" scenario.

To summarise the suitable habitat area for each species, the sum of viable habitat for each scenario was computed. Figure 34 shows the suitable fish habitat averaged over all months in each year. Without environmental water (No eWater 3yrs) the habitat suitable for mulloway would have contracted by 17% in 2017-18, 34% by 2018-19 and 40% in 2019-20 (Figure 34). Note that care should be taken in interpreting these habitat areas, bearing in mind other ecological constraints on population recovery are not captured in the index formulation.

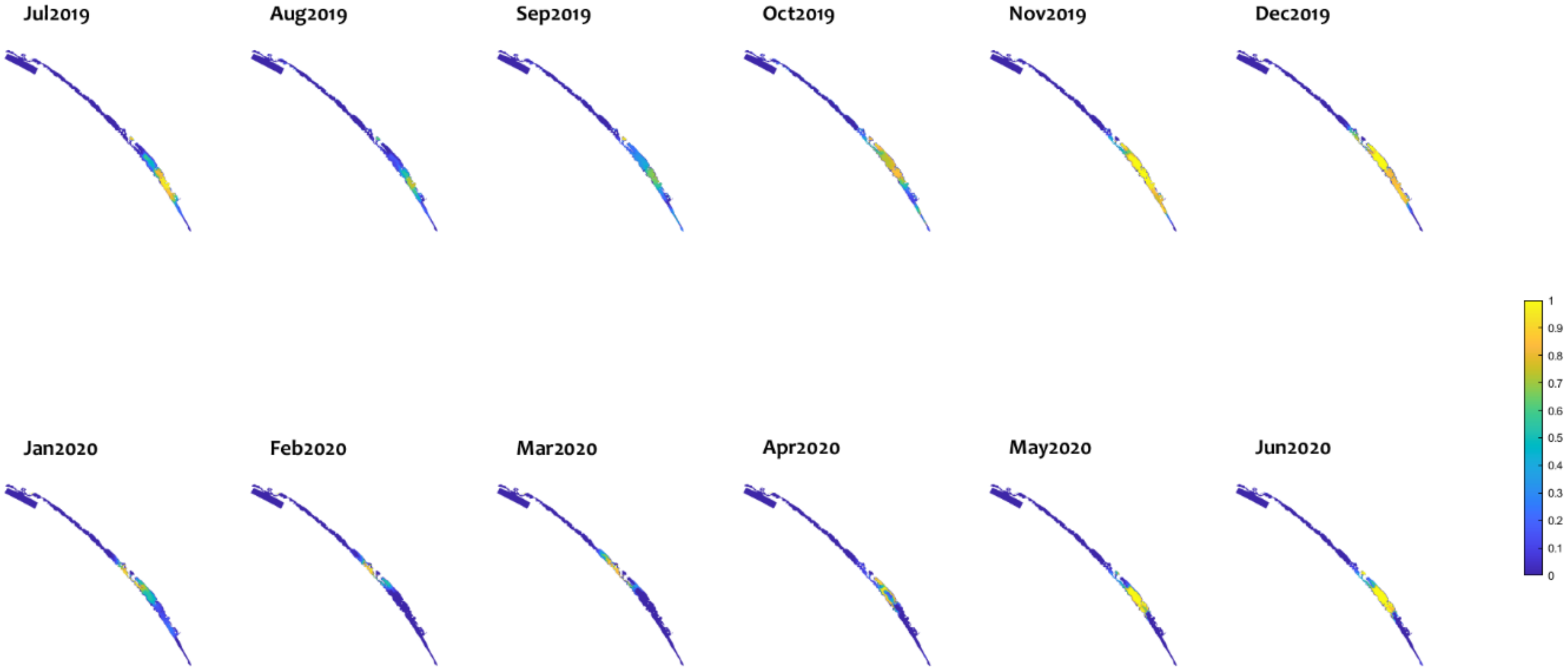


Figure 33. Monthly habitat area "gained" for mulloway due to environmental water delivery (calculated as the difference between habitat in the "All water" (base-case) and "no eWater 3yrs" scenario). Large areas of the South Lagoon have an increase in habitat quality of a maximum of 1, highlighting areas that would not be viable without environmental water, but became suitable due to the ongoing water delivery since 2017-18. The improvement in habitat score means salinity and temperature conditions are suitable for mulloway. Other features of the environment such as food resources, appropriate sediment will also determine whether mulloway expand into a habitat. It may take some time for ecosystem restoration to reach a point where fish populations are supported in the expanded habitat.

A similar trend is evident for black bream, Tamar goby, greenback flounder, yelloweye mullet, congolli and smallmouth hardyhead. Three years without environmental water reduced suitable black bream habitat by 45% (Figure 34). This scenario commenced after high flow in 2016-17. If the starting conditions were more saline then we could expect a more rapid contraction in suitable habitat if no environmental water was available.

If we consider just one year (2019-20) without environmental water ("No eWater"), the suitable habitat area contracted by between 5 and 17% for the seven fish species within that period. Mulloway habitat was the most sensitive to a single one-year reduction in environmental water. Smallmouth hardyhead can tolerate very saline conditions and their habitat was least affected by the one-year reduction in environmental water.

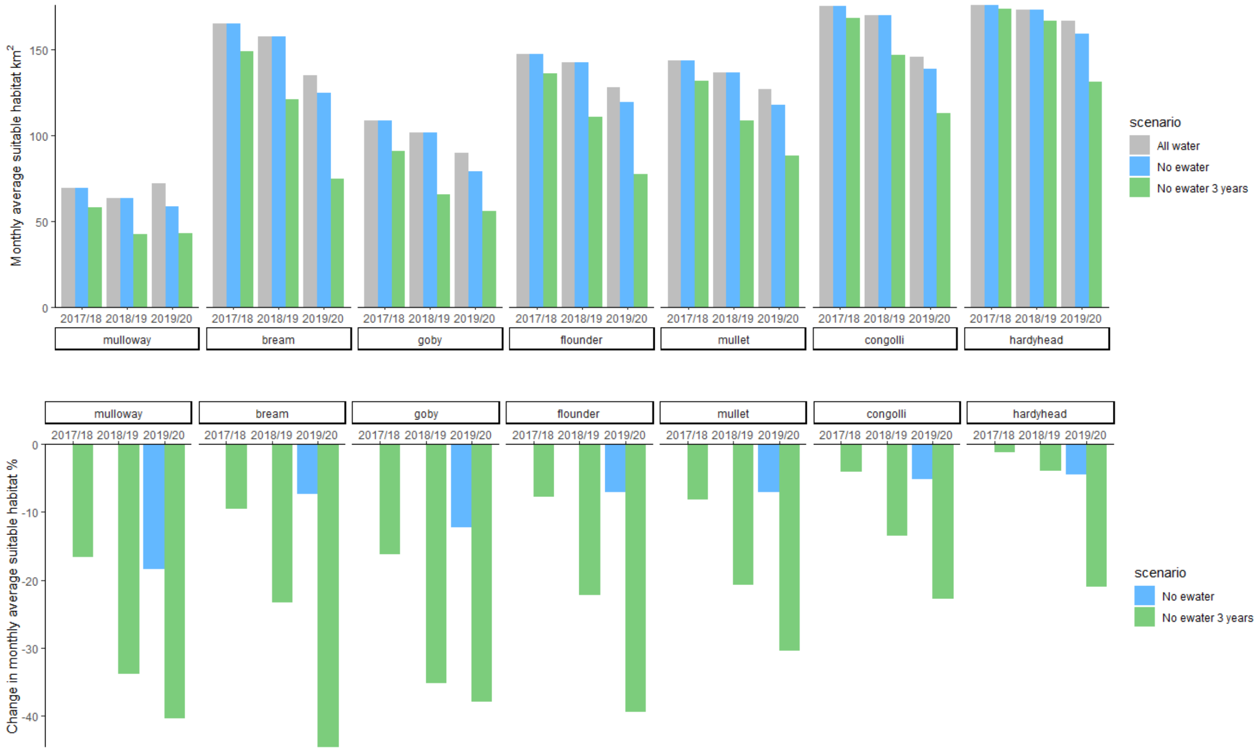


Figure 34. Habitat area of juvenile stages of key fish species for the three scenarios (top). Change in area (%) that would have been in the case of no environmental water is shown in the bottom panel. Environmental water provides a large habitat expansion for all species and this increases year on year. The environmental water modifies the salinity conditions to be expanding the habitat that the fish can exploit. Other features such as food resources and appropriate sediment will also influence whether fish are able to exploit the available habitat.

Evaluation

The evaluation approach, including assessment criteria, is described in the evaluation section for Hydraulic Regime (Section 2.1).

Table 11. Coorong habitat evaluation questions and answers. CEW = Commonwealth environmental water, eWater = environmental water.

CEWO evaluation questions	Outcomes of CEW delivery		
	2017-18	**2018-19**	**2019-20**
What did CEW contribute to improving *Ruppia tuberosa* habitat in the Coorong?	Habitat suitable for overall sexual life-cycle completion of *Ruppia* was significantly improved with eWater. It was estimated that adult *Ruppia* were not favoured by eWater, however, this was due to model sensitivity to the minimum salinity threshold value, and a loss of potential area in the Murray Mouth, which is not generally a management target. Habitat suitable for seed production was 20% greater with eWater than without eWater. When looking at the maps of suitable area, the benefit of eWater is pronounced in critical areas of the South Lagoon, highlighting the importance of eWater to the ecological restoration of the Coorong.		
What did CEW contribute to improving fish habitat in the Coorong?	eWater lead to fresher conditions in the Coorong and an expansion of suitable fish habitat area throughout the year. Without eWater, the habitat suitable for mulloway would have contracted by 17% in 2017-18, 34% by 2018-19, and 40% in 2019-20. A similar trend is evident for black bream, Tamar goby, greenback flounder, yelloweye mullet, congolli and smallmouth hardyhead. A major expansion of suitable habitat into the South Coorong is critically important to restore biodiversity in this recovering ecosystem.		

Contribution (to what extent CEW contributed towards the outcome, with the significance of the outcome considered):

⬜ Unknown 🟥 Negative ⬜ None/negligible 🟩 Minor 🟩 Moderate 🟦 Substantial

Discussion

Salinity is a key determinant of habitat suitability in the Coorong, and is primarily a function of riverine inflows, seawater ingress and tidal movement. When barrage flows are low, seawater enters the Murray Mouth and more salt is then transported to the Coorong. Environmental water contributed 100% of flow over the barrages in 2018-19 and 2019-20. It is evident that environmental water is critical for maintaining appropriate salinity in the Coorong and maintaining suitable habitat. If environmental water had not been delivered in 2019-20, an additional 1.7 million tonnes of salt would have accumulated in the South Lagoon.

The area of suitable habitat for *Ruppia* sexual reproduction increased over the three-year period from approximately 20 to 40 km^2, largely attributable to an anticipated improvement in flowering success. The salinity at the commencement of the simulation is likely to have an impact on the habitat suitability area. The large flows in 2016-17 would have 'reset' salinity in the Coorong which progressively became more saline moving the salinity to above the minimum threshold ranges set for habitat suitability. With additional environmental water, this return above the minimum salinity happened quicker. Care needs to be taken when interpreting this result as prolonged conditions with no barrage flow are detrimental to the system and the habitat suitability will be highly dependent on the antecedent flow. When looking at the habitat maps, it was noted the overall habitat area hadn't changed significantly, but that there was a significant improvement in the South Lagoon, which is a critical target for restoration.

The suitable habitat area for adult plants appears to be slightly higher with no environmental water delivery. This contradicts the hypothesis that environmental flows favour *Ruppia* growth, and the observations noted above about the expansion of suitable area into the South Lagoon. The fact that the area of favourable habitat increases 2018-19 over the 2017-18 area in the base case scenario suggests that it is the minimum salinity threshold that is leading to this result. Following the large flow of 2016-17 the Coorong was fresher but steadily became more saline with the lower river flow over the barrages in subsequent years. This appears to have slightly improved the condition for *Ruppia*. Further sensitivity testing of the model to the minimum salinity threshold concentration will occur in subsequent years when more flow scenarios are available. This result highlights the importance of using multiple years of flow in the Coorong modelling as the Coorong has 'memory' and salinity is a function of historical salt loading as well as annual inputs and outputs.

The estuarine fish species that inhabit the Coorong vary slightly in their tolerance to salinity with yellow-eye mullet, congolli and smallmouth hardyhead able to tolerate more saline conditions. Fish differ from *Ruppia* in that they can move in response to changing salinity and habitat suitability. It is generally considered advantageous to have a greater area of habitat with suitable water quality (e.g. salinity) and abundant food resources to support the maintenance of fish populations. Estuaries are important feeding, spawning, nursery and refuge grounds for many estuarine dependent fish species (Bice *et al.* 2018). Without environmental water, fish habitat contracts quickly and significantly. Even after the high flow year in 2016-17, if there was no environmental water in 2017-18, significant habitat contraction would have occurred. As mulloway and Tamar goby have the smallest area of suitable habitat, this contraction would have the most profound impact on these species, followed by black bream, greenback flounder and yellow-eye mullet. Habitat for the more salinity tolerant congolli and smallmouth hardyhead would not have changed as significantly in 2017-18 as for the other species. Consecutive years of no environmental water reduced suitable habitat by up to 45% within three years and even the highly salt-tolerant smallmouth hardyhead experienced a >20% contraction of suitable habitat.

Management implications

Flow over the barrages is critical to maintain sexual reproduction of *Ruppia*. The starting salinity conditions are important and environmental flows could be tailored to improve *Ruppia* habitat suitability. These environmental flows would need to consider the health of *Ruppia* in the system, the extent of *Ruppia* cover and the prevailing salinity. The model could potentially be used to optimise environmental water delivery through the barrages to maximise the area of suitable *Ruppia* habitat based on the volume of water available.

The area of suitable estuarine fish habitat is very sensitive to river flow over the barrages. Environmental flows lead to fresher conditions in the Coorong and an expansion of suitable fish habitat area. Without environmental flows, the suitable habitat contracts for a range of fish species (e.g. mulloway, black bream, Tamar goby, greenback flounder, yellow-eye mullet and congolli). Smallmouth hardyhead are more salt-tolerant and their habitat also contracts with no environmental water but the major impact on habitat suitability took one year longer to manifest than the other species. To maintain suitable habitat for the range of estuarine species, environmental flows should be maintained to support barrage releases every year. Without environmental flows in 2019-20, there would have been an 18% contraction in suitable habitat for mulloway in the Coorong. Three years with environmental flows would have reduced mulloway habitat by 40%.

Conclusion

Environmental water contributed to a significant increase in the area of suitable habitat for estuarine fish and improve habitat suitability for *Ruppia* to complete sexual life-cycle in the Coorong. Environmental flows reduce salt import into the Murray Mouth and provide fresh water to the Coorong, which together maintain salinity conditions in the Coorong that expand the area of habitat suitable or fish. Environmental flows create conditions suitable for sexual reproduction of *Ruppia*. The suitable habitat for adult *Ruppia* was overall neutral with environmental water than without environmental water. This may be a function of the starting conditions which were relatively ‘fresh’ following the high flows of 2016-17, and uncertainty around the lower salinity tolerance given the fresher conditions with environmental water. Habitat becomes more suitable in subsequent years. From the Millennium Drought, there is clear evidence that prolonged periods with no flow over the barrages create extreme hyper-saline conditions in the South Lagoon that are detrimental to *Ruppia,* and the scenarios with environmental water show a large expansion of *Ruppia* habitat in the South Lagoon.

Environmental flows lead to fresher conditions in the Coorong and an expansion of suitable fish habitat area. Without environmental water there is a contraction of fish habitat. The rate at which the suitable habitat contract will vary for the different fish species and is dependent upon the salinity at the commencement of the period of interest.

2.4 Littoral Vegetation Diversity and Productivity

Background

Littoral (streambank) vegetation is an important component of the biota of riverine ecosystems. It is an important primary producer for both the riverine and terrestrial ecosystem (e.g. Roberts and Ganf 1986; Froend and McComb 1994), can improve water quality (e.g. Kadlec and Wallace 2009; Maddison *et al.* 2009; Li *et al.* 2010; Borin and Salvato 2012), oxygenate the sediment and water column (e.g. Blom *et al.* 1990; Sorrell and Hawes 2010; Dickopp *et al.* 2011), provide habitat for water birds (e.g. Jansen and Robertson 2001; Kapa and Clarkson 2009) and invertebrates (e.g. Papas 2007; Walker *et al.* 2013) and stabilise banks (e.g. Abernethy and Rutherfurd 1998). Littoral zones are also hot spots for biodiversity because they contain a specialised group of species adapted to wetting and drying not found in aquatic or terrestrial systems and increase the species pool at the landscape scale (Sabo *et al.* 2005).

Littoral vegetation responds rapidly to changes in water level (e.g. Nicol *et al.* 2018a) as inundation changes the physicochemical environment and acts as an environmental sieve (*sensu* van der Valk 1981) producing conditions that are hostile to some species but favourable to others (e.g. Nicol *et al.* 2003). In riparian zones, inundation often acts as disturbance, removing the existing vegetation and providing an environment almost free of competition with high soil moisture (e.g. Pettit and Froend 2001; Bagstad *et al.* 2005; Beauchamp and Stromberg 2008). Due to the unpredictable nature of flooding regimes in arid Australian systems (Puckridge *et al.* 1998; Puckridge *et al.* 2000), many littoral species are short-lived annuals that are adapted to take advantage of these brief periods of favourable conditions germinating as water levels recede, completing their life cycle whist soil moisture is high and competition is low and replenishing the seed bank (e.g. Nicol 2004; Capon 2007). These species persist in the soil seed bank whilst conditions are unfavourable and are examples of Grime's (1979) r-selected species and have more in common with desert annuals than aquatic species (e.g. Nicol 2004; Capon 2007). Emergent and amphibious species (*sensu* Brock and Casanova 1997; Casanova 2011) are also present and are typically perennial taxa (Cunningham *et al.* 1992) adapted to persist or respond to inundation and exposure and often have a requirement of variable water levels to establish and/or reproduce (e.g. Brock and Casanova 1997; Casanova 2011; Nicol *et al.* 2018b).

Management of water levels in the LMR has typically focussed on maintaining stable water levels in lower weir pools, which can result in small increases in flow causing large water level rises in tailwaters immediately downstream of weirs (Maheshwari *et al.* 1995). Therefore, tailwaters represent areas where flood dependent and amphibious vegetation can persist in the absence of overbank flows over a wider range of the elevation gradient (Blanch *et al.* 1999; Blanch *et al.* 2000). These water level changes make tailwaters an ideal location to assess the influence of Commonwealth environmental water as the volumes available for delivery will have a significant impact on water levels. The response of the vegetation along the elevation gradient in tailwaters can be exploited to evaluate the benefit of Commonwealth environmental water.

In 2019-20, Commonwealth environmental water was delivered to produce a spring pulse peaking at 15,569 ML/d in the LMR on 14 October. The aim of this study is to investigate the response of littoral vegetation to Commonwealth environmental water delivery by testing the hypotheses in the following section. This will be undertaken by comparing species composition and biomass between areas inundated and not inundated by the spring flow pulse.

Major hypotheses

- Increases in flow above regulated entitlement flow will result in increased water levels in the littoral zone that will facilitate the recruitment of floodplain and amphibious understorey species and in turn increase biological and functional diversity of the littoral zone plant community.
- The increase in water level due to increases in flow above regulated entitlement flow will result in increased productivity of littoral vegetation compared to non-inundated areas.

Methods

Study sites and hydrology

Vegetation surveys were undertaken on the banks of the LMR in weir pool tailwaters downstream of Lock 1, Lock 4 and Lock 6 (referred herein to as reaches) to correspond with the locations of other indicators (Figure 1).

Flow in 2019-20 was generally low with an in-channel flow pulse peaking in mid-October 2019 that was comprised almost entirely of Commonwealth environmental water (Figure 35). The flow pulse resulted in an increase in water levels between 65 cm (downstream of Lock 6) and 107 cm (downstream of Lock 4) (Figure 35). For a detailed description of the hydrology and environmental water delivery see Sections 1.4 and 2.1.

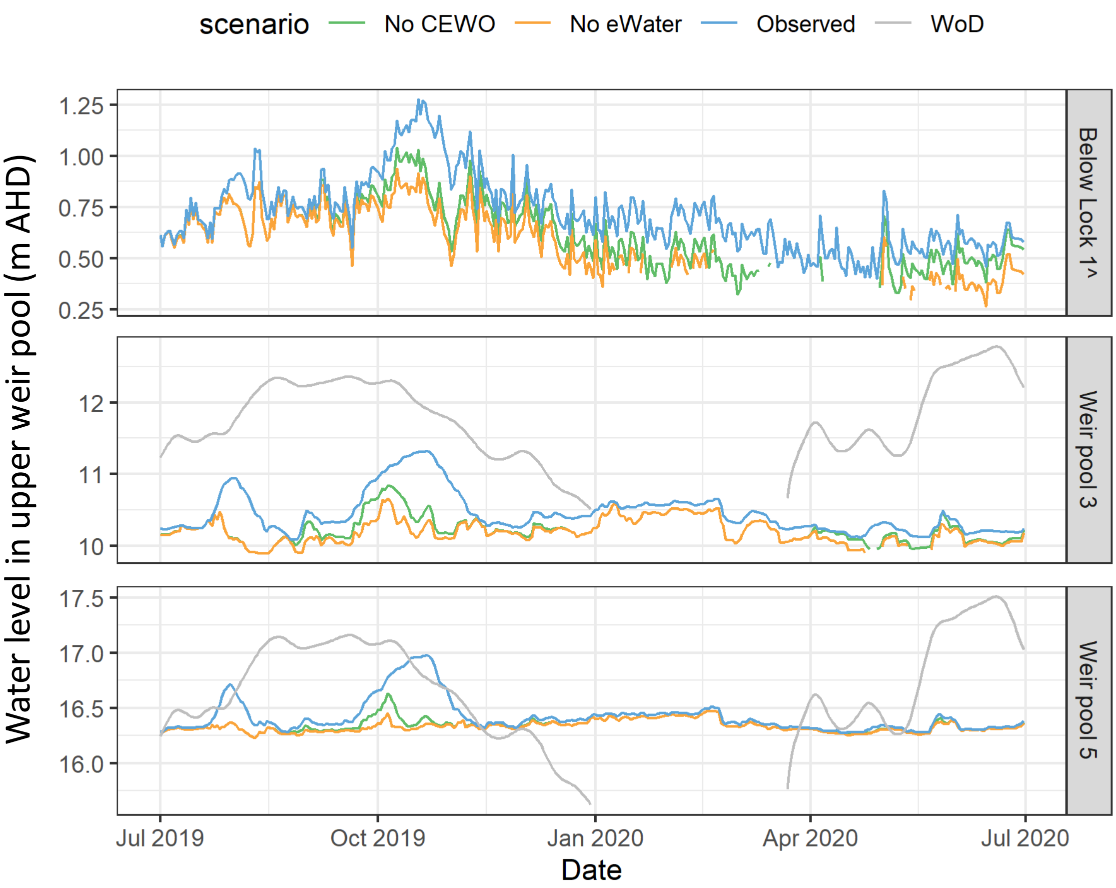


Figure 35. Observed and modelled (without development, without Commonwealth environmental water and without any environmental water) water levels in upper weir pools (tailwaters) below Lock 1, Weir Pool 3 (below Lock 4) and Weir Pool 5 (below Lock 6).

Vegetation surveying protocol

Sampling of littoral vegetation occurred in December 2019, four to five weeks after the peak of the flow pulse. Six sites were established in each reach, which comprised of a transect running perpendicular to elevation contours from normal pool level to the elevation inundated by flows of 40,000 ML/d. The position of the lowest elevation of each transect was recorded by GPS (Table D1 in Appendix D) to enable the same transects to be surveyed in subsequent years.

Quadrats with dimensions of 15 x 1 m were positioned on each transect (the long side parallel to elevation contours) in relation to the maximum water level in spring 2019; one quadrat was located at normal pool level (herein referred to as *Pool level*), two in the area inundated by the spring flow pulse (*Inundated*), one at the spring 2019 high water mark (*Inundation extent*) and two above the high water mark (*Not inundated*) (Figure 36). Each quadrat was divided into 15, 1 x 1 m cells to determine species frequency in each quadrat (i.e. each species will have a score of zero (not present) and 15 (present in each cell). In addition, a 50 x 50 cm (0.25 m^2) section of each quadrat was harvested and dried at 40^{o} C to a constant weight to determine above ground biomass.

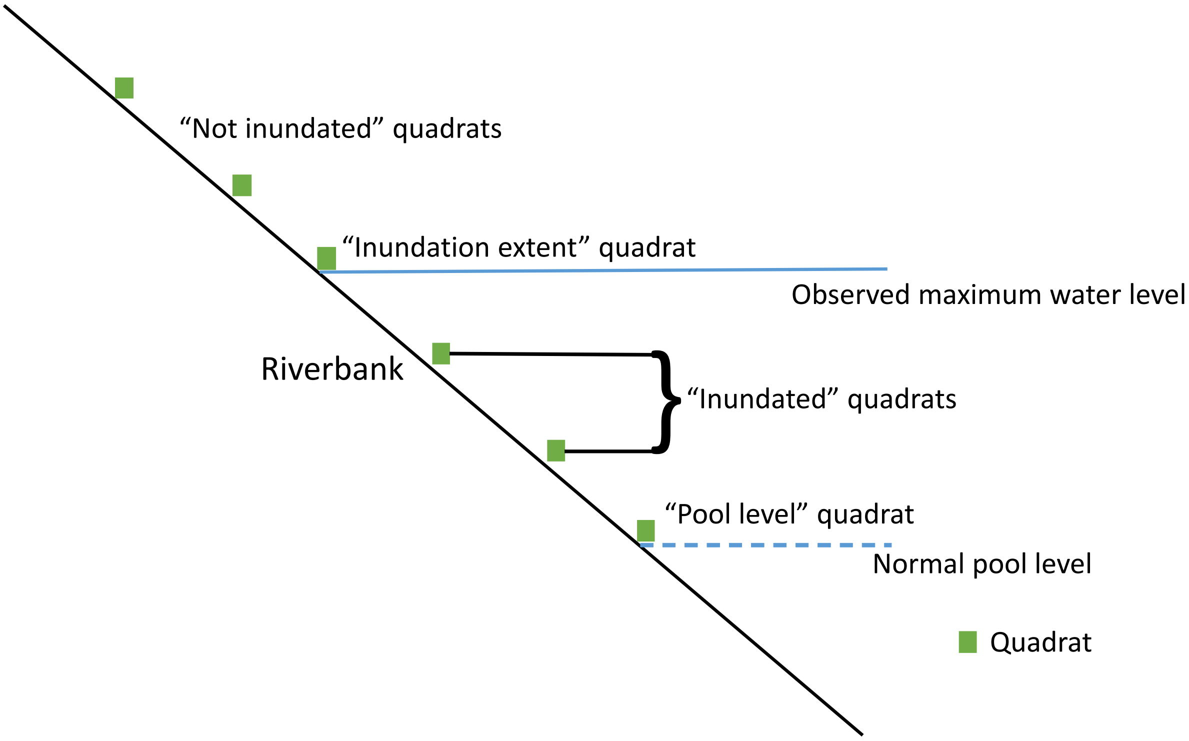


Figure 36. Position of quadrats in relation to maximum water level in when sampled in December 2019.

The *Inundated* and *Inundation extent* zones were impacted by the delivery of Commonwealth environmental water. Modelling indicated that without Commonwealth environmental water, the *Inundated* zone would have remained dry and there would have been no increase in soil moisture in the root zone at the *Inundation extent* elevation (Figure 35). The *Pool level* quadrats would have been the only quadrats inundated with the delivery of other environmental water (Figure 35).

<u>Plant identification and nomenclature</u>

Plants were identified using keys in Jessop and Toelken (1986), Cunningham *et al.* (1992), Sainty and Jacobs (2003) and Jessop *et al.* (2006). In some cases, due to immature individuals or lack of floral structures, plants were identified to genus only. Nomenclature follows the Centre for Australian National Biodiversity Research and Council of Heads of Australasian Herbaria (2020). A comprehensive list of all species surveyed, their functional classification, growth form, life history strategy and conservation status are presented in Table D2 in Appendix D.

<u>Plant functional groups</u>

The functional classification used to assess targets for the Chowilla condition and intervention monitoring programs was used in this study to assess the response of the littoral vegetation to environmental water. The functional classification was based on the classification framework devised by Brock and Casanova (1997), which was based on species present in wetlands in the New England Tablelands region of New South Wales and modified by Nicol *et al.* (2010) to reflect the vegetation of the Chowilla system.

The functional classification used for this study splits species into four groups (terrestrial, flood dependent, amphibious and emergent) based on their response to inundation:

- "Terrestrial" species are intolerant of either partial or complete inundation;
- "Flood dependent" species are intolerant of complete inundation as mature or juvenile plants but germinate on newly exposed soil after inundation but not in response to rainfall. Many of these species are short-lived annuals that take advantage of the bare soil (lack of competition) and high soil moisture after inundation and can complete their life cycle in a matter of weeks (Nicol 2004);
- "Amphibious" species require fluctuating water levels and are adapted to survive inundation and exposure as juvenile or adult plants. Brock and Casanova (1997) split this group into five different groups based on their anatomical responses to inundation; however, they all have similar water regime requirements; and
- "Emergent" species have a requirement for permanent shallow water or continuous high soil moisture in the root zone to survive. Unlike amphibious species these species are well adapted to static water levels and will displace amphibious species under these conditions.

Data analysis

Species composition between the different inundation zones (*Not inundated*, *Inundation extent*, *Inundated* and *Pool level*) and reaches (Lock 1, Lock 4 and Lock 6) were compared using nMDS Ordination, Multivariate PERMAOVA and Indicator Species Analysis. Bray-Curtis (1957) similarities were used to calculate the similarity matrix for nMDS Ordination and Multivariate PERMAOVA analyses, which were undertaken using the Package PRIMER version 7.0.12. (Clarke and Gorley 2015) and PERMANOVA+ for PRIMER (Anderson *et al.* 2008). Indicator Species Analysis (Dufrene and Legendre 1997) was undertaken using the package PCOrd version 5.12 (McCune and Mefford 2006). Above ground biomass was compared between the different inundation zones and reaches using univariate PERMANOVA using the package PERMANOVA+ for PRIMER (Anderson *et al.* 2008). Euclidean distances were used to calculate the similarity matrix for the univariate PERMANVA analysis.

Indicator Species Analysis

Dufrene and Legendre's (1997) indicator species analysis combines information on the concentration of species abundance in a particular group (inundation zone) and the faithfulness of occurrence of a species in a particular group (McCune *et al.* 2002). A perfect indicator of a particular group should be faithful to that group (always present) and exclusive to that group (never occurring in other groups) (McCune *et al*. 2002). This test produces indicator values for each species in each group based on the standards of the prefect indicator. Statistical significance of each indicator value is tested by using a Monte Carlo (randomisation) technique, where the real data are compared against (in this case) 5,000 runs of randomised data (Dufrene and Legendre 1997). For this study, the groups were assigned according to inundation zone within a reach (planned comparison). A species that is deemed not to be a significant indicator of a particular group is either uncommon or widespread. An uncommon species is only found in one group but in low numbers and a widespread species is found in more than one group in similar numbers (Dufrene and Legendre 1997). Whether a species was classed as a widespread or uncommon non-significant species was determined by examination of the raw data.

Results

Littoral plant communities

Native species richness was variable between reaches, with the reach downstream of Lock 1 having the highest native species richness in all zones (Figure 37). Despite the variability among reaches, there were common patterns of native species richness within reaches. The *Not inundated* zone had the lowest native species richness regardless of reach (Figure 37). Inundation and increased soil moisture in the root zone resulted in increased species richness with the highest species richness in the Lock 1 and Lock 4 reaches being in the *Inundated* zone (areas inundated by Commonwealth environmental water) (Figure 37). In the Lock 6 reach, native species richness was highest at *Pool level*, with the *Inundated* zone being the second highest (Figure 37).

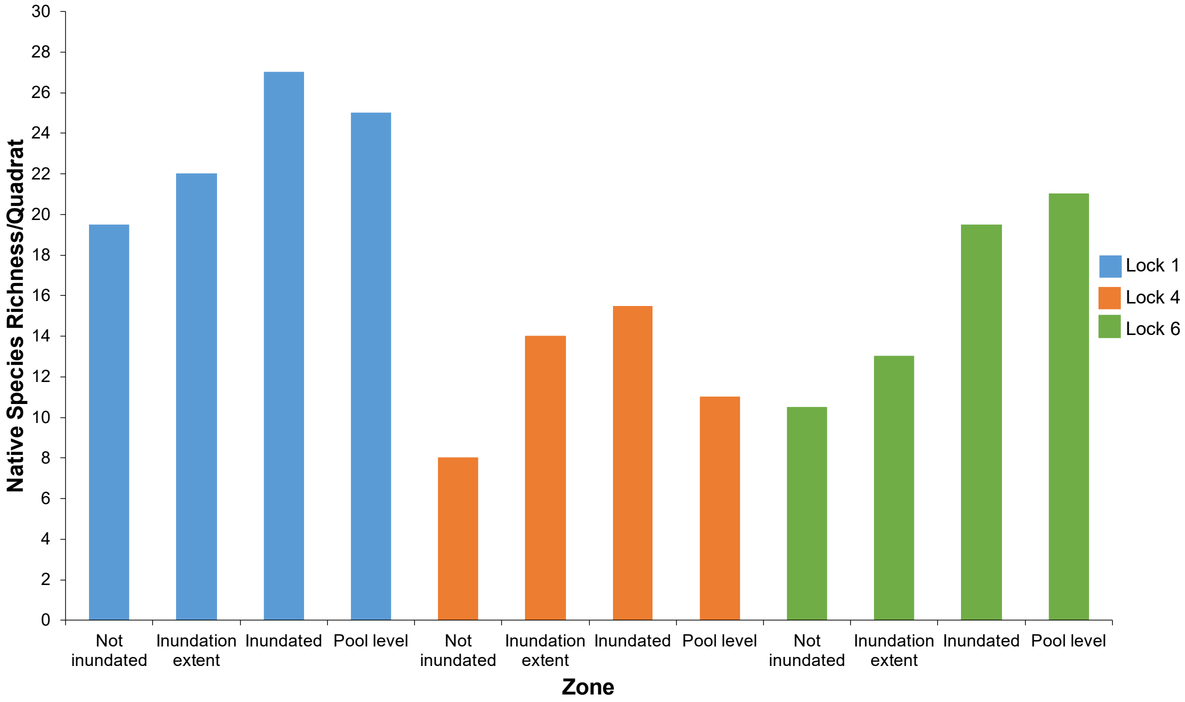


Figure 37. Native species richness (species/quadrat) in each inundation zone in each reach in December 2019.

The abundance (mean number of cells occupied by species from each functional group) of functional groups was variable between reaches (similar to species richness) with more of every functional group present downstream of Lock 1 (Figure 38). Nevertheless, there were similar patterns among reaches regarding the influence of inundation; terrestrial taxa were more abundant in non-inundated areas (the *Not inundated* and *Inundation extent* zones) and there was an increase in emergent taxa with decreasing elevation (Figure 38). Flood dependent taxa were widespread but were most abundant in the *Inundated* zone in all reaches (Figure 38). Amphibious species were also widespread and most abundant at *Pool level* in the Lock 1 and Lock 6 reaches and in the *Inundated* zone downstream of Lock 4 (Figure 38).

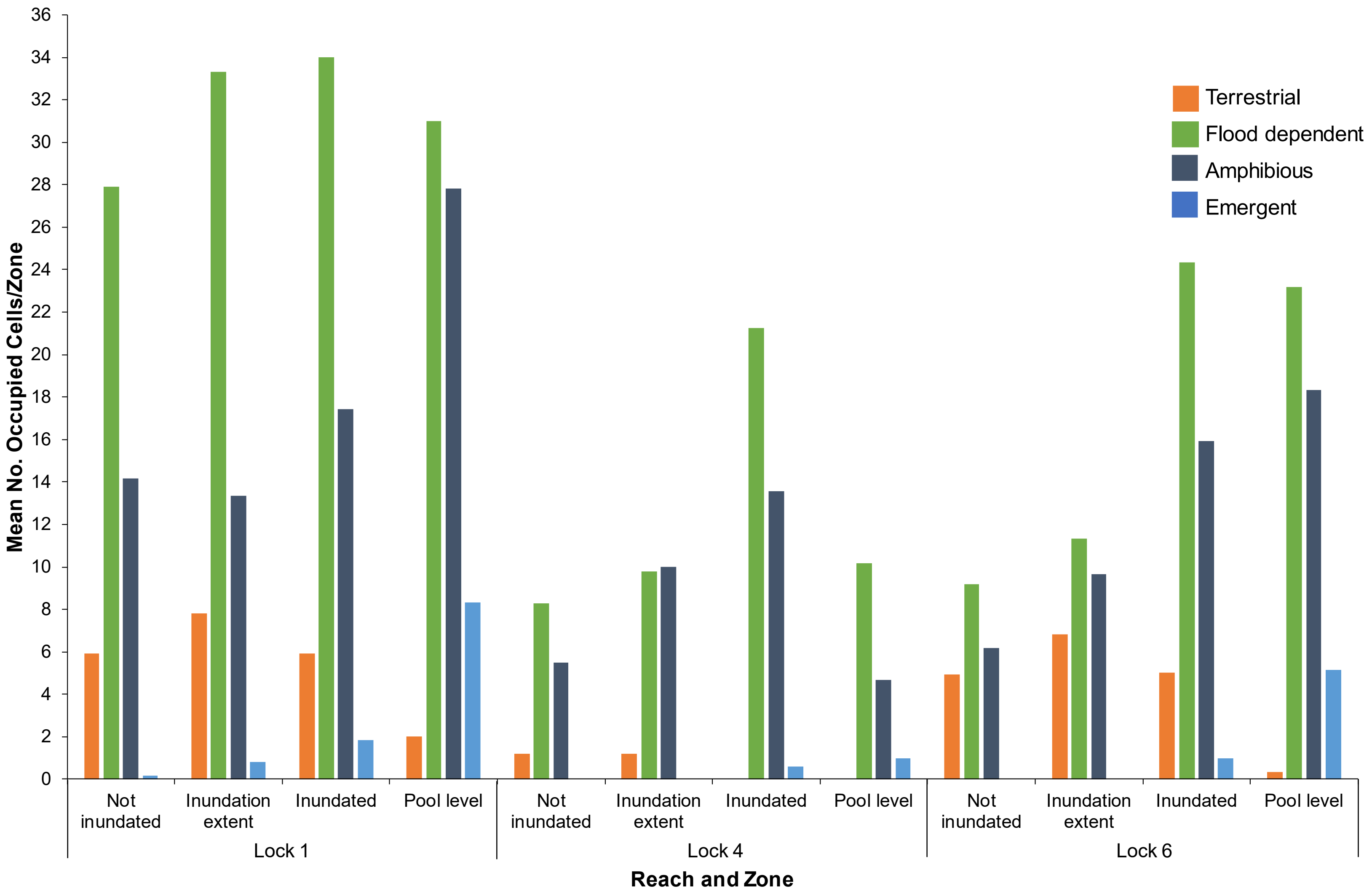


Figure 38. Number of cells occupied by each functional group in each inundation zone in each reach in December 2019

nMDS ordination showed that the vegetation downstream of Lock 1 was different to the two upstream reaches (Figure 39). This was supported with PERMANOVA detecting significant differences in the plant community between reaches (with multiple comparisons showing each reach was significantly different) and inundation zones (each zone was also significantly different), with no significant interaction (Table 11). This provides evidence that the differences in the plant community between inundation zones (and, in response to environmental water delivery) was similar across reaches despite the plant communities being significantly different at each reach.

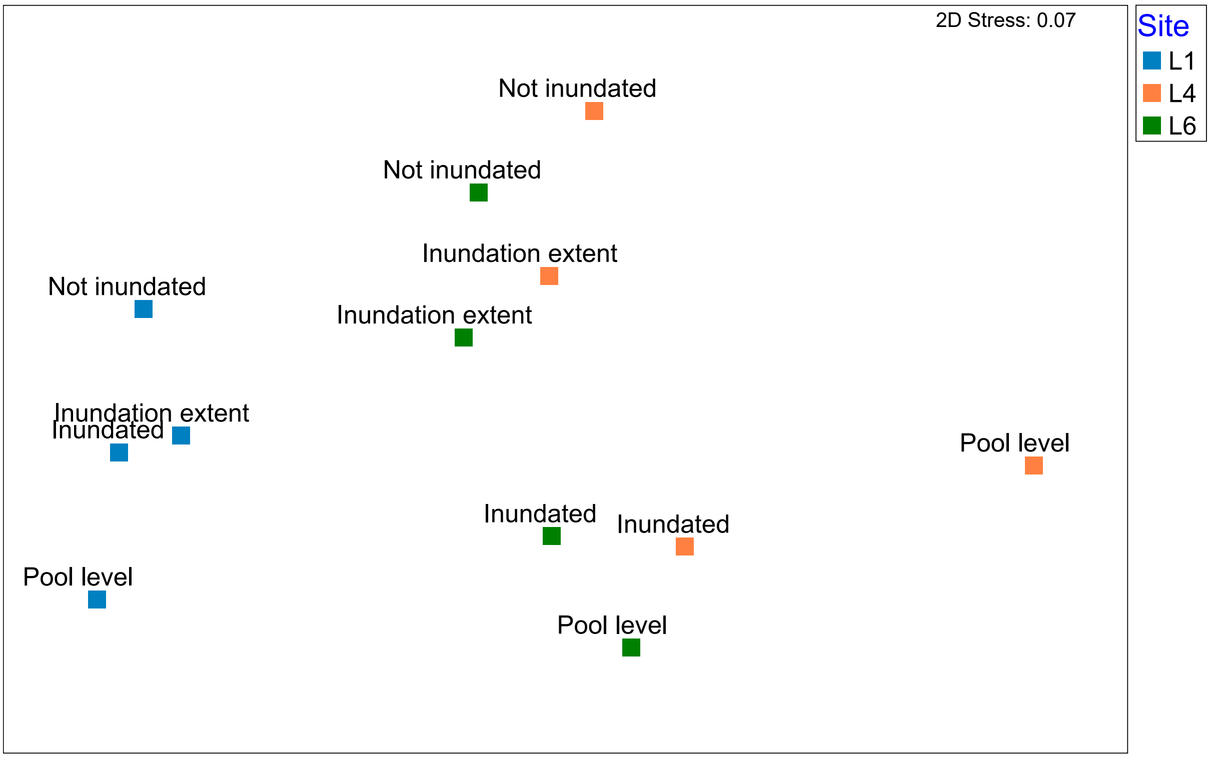


Figure 39. nMDS Ordination comparing the plant community in each inundation zone in each reach in December 2019.

Table 11. PERMANOVA results comparing plant community in each inundation zone in each reach in December 2019.

Factor	DF	*Pseudo F*	*P*
Reach	2,107	8.81	0.001
Inundation Zone	3,107	3.27	0.001
Site x Inundation Zone	6,107	1.11	0.266

Despite the significant differences between inundation zones, *Pool level* (with the exception of *Xanthium occidentale*, in the reach downstream of Lock 1) was the only zone with significant indicators in the Lock 1 and Lock 6 reaches (with no significant indicators in the Lock 4 reach) (Table D3, Appendix D). However, many of the species not deemed significant indicators of one inundation zone were present in two zones in similar abundances. In particular, several amphibious and flood dependent taxa were present in the *Pool level* and *Inundated* zones and absent at the *Inundation extent* and *Not inundated* zones (Table D3, Appendix D).

Several exotic species recruited in the *Inundated* and *Pool level* zones in response to environmental water delivery. *Xanthium occidenale* and *Symphyotrichum subulatum* were present in all reaches exclusively in the *Inundated* and *Pool level* zones and *Dittrichia graveolens* was present at Lock 6 in the same zones (Table D3, Appendix D).

Whilst not a significant indicator and widespread across the different inundation zones, *Eucalyptus camaldulensis* seedlings were observed throughout the inundated zones of the Lock 4 and Lock 6 reaches (Figure 40). All seedlings in the aforementioned zones in the lock 4 and Lock 6 reaches were small (similar in size to the one pictured in Figure 40) and clearly had germinated in response to the spring pulse.

Figure 40. Newly germinated *Eucalyptus camaldulensis* (river red gum) seedling in the Lock 4 reach.

<u>*Above ground biomass*</u>

Above ground biomass was highly variable among reaches but unlike species richness, abundance of functional groups and plant communities, there were no consistent patterns in relation to inundation zone among reaches (Figure 41); hence, the significant interaction between reach and inundation zone detected by PERMANOVA (Table 12). Biomass downstream of Lock 1 decreased with decreasing elevation, in the Lock 4 reach was highest in the *Inundation extent* zone and in the Lock 6 reach was highest in the *Inundated* zone (Figure 41).

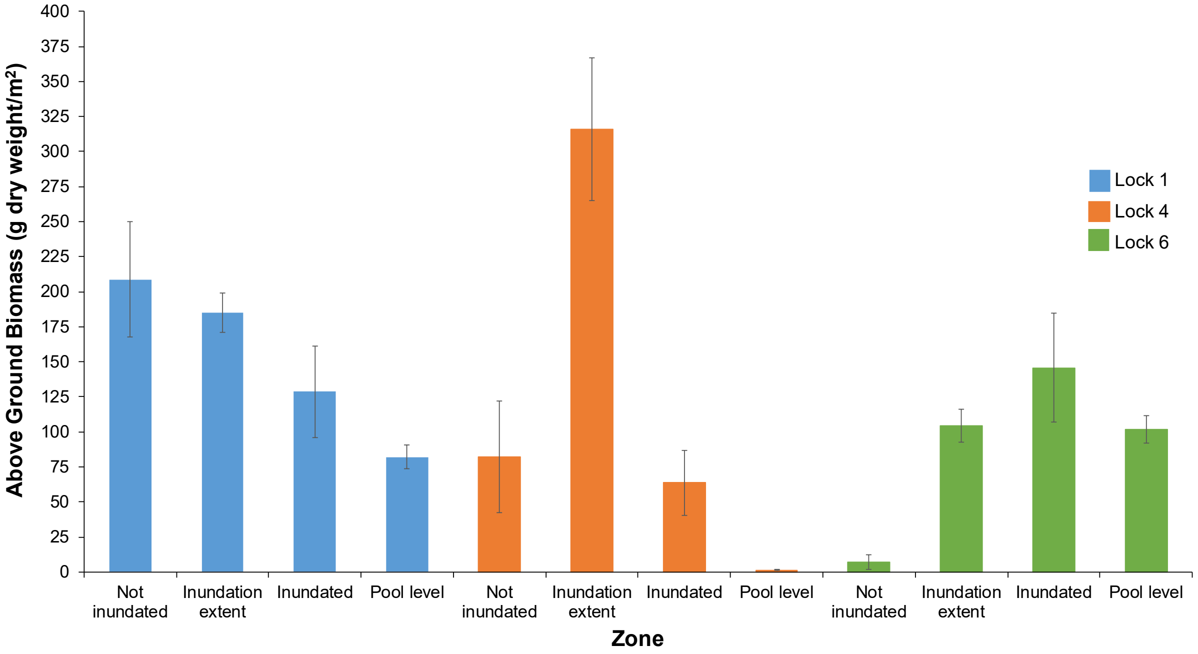


Figure 41. Above ground biomass in each inundation zone in each reach in December 2019 (error bars = ± 1 SE).

Table 12. PERMANOVA results comparing above ground biomass in each inundation zone in each reach in December 2019.

Factor	**DF**	***Pseudo F***	***P***
Site	2,107	2.09	0.112
Inundation	3,107	2.69	0.041
Site x Inundation zone	6,107	2.49	0.02

Evaluation

The evaluation approach, including assessment criteria, is described in the evaluation section for Hydraulic Regime (Section 2.1).

Table 13. Littoral Vegetation Diversity and Productivity evaluation questions and answers. CEW = Commonwealth environmental water, eWater = environmental water.

CEWO evaluation questions	Outcomes of CEW delivery (2019-20)
What did CEW contribute to littoral understorey vegetation diversity and productivity?	CEW delivery increased plant species diversity at multiple spatial scales in the LMR. This was evidenced by consistently higher native species richness in inundated zones in each reach compared to non-inundated areas, higher native species richness (albeit lower than inundated zones) on the edge of the inundation footprint, where there was increased soil moisture due to capillary action, and different plant communities in the inundated zones (despite there being few significant indicators) compared to non-inundated zones. CEW delivery in the form of a spring pulse produced conditions suitable for the recruitment of specialised riparian species that are adapted to fluctuating water levels and do not inhabit terrestrial or truly aquatic habitats, increasing plant functional diversity. Native species (*n* = 24) that responded positively to delivery of CEW either by recruitment or increased abundance: • *Alternanthera denticulata* • *Bolboschoenus caldwellii* • *Centipeda minima* • *Cyperus gymnocaulos* • *Eleocharis acuta* • *Eucalyptus camaldulensis* • *Isoetopsis graminifolia* • *Isolepis australiensis* • *Juncus usitatus* • *Lachnagrostis filiformis* • *Limosella australis* • *Ludwigia peploides* • *Lythrum hyssopifolia* • *Myriophyllum papillosum* • *Paspalum distichum* • *Persicaria lapathifolia* • *Phragmites australis* • *Pseudognaphalium luteoalbum* • *Polygonum plebeium* • *Schoenoplectus pungens* • *Senecio runcinifolius* • *Sphaeromorphaea australis* • *Stemodia florulenta* • *Typha domingensis*

CEWO evaluation questions	**Outcomes of CEW delivery (2019-20)**
What did CEW contribute to above-ground biomass produced by understorey littoral vegetation?	Biomass production due to CEW was inconclusive and varied depending on reach with no clear pattern in relation to CEW delivery. The only reach that responded as hypothesised was Lock 6 with greater biomass in the *Inundated* and *Inundation extent* zones.

Contribution (to what extent CEW contributed towards the outcome, with the significance of the outcome considered):
⬜ Unknown 🟥 Negative 🟩 None/negligible 🟩 Minor 🟩 Moderate 🟦 Substantial

Discussion

Results from monitoring in December 2019, after the spring flow pulse had transited the LMR, supported one of the hypotheses:

- Increases in flow above regulated entitlement flow will result in increased water levels in the littoral zone that will facilitate the recruitment of floodplain and amphibious understorey species and in turn increase biological and functional diversity of the littoral zone plant community.

As the spring pulse was almost entirely the result of the delivery of Commonwealth environmental water, this provided evidence that its delivery increased biological and functional diversity of the plant community.

The spring pulse (and in turn the delivery of environmental water) significantly changed the plant communities in areas that were inundated and where soil moisture in the plant root zone increased. This was not unexpected as inundation changes the physicochemical environment and acts as an environmental sieve (*sensu* van der Valk 1981) producing conditions that are hostile to some species but favourable to others (e.g. Nicol *et al.* 2003). There was evidence that many terrestrial taxa were extirpated as a result of inundation and, as water levels receded, there was recruitment of flood dependent and amphibious species. In the case of perennial amphibious species (e.g. *Cyperus gymnocaulos, Juncus usitatus, Ludwigia peploides*), there may not have been recruitment from seed but increases in cover and number due to growth or clonal expansion (rhizomes of *Cyperus gymnocaulos* and *Juncus usitatus* were widespread throughout sites).

Whilst there were consistent patterns of floristic composition between zones in each reach there were large differences between reaches, in particular between Lock 1 and the other two reaches (Figure 37; Figure 39). The reach below Lock 1 had higher species richness across all zones, which is probably due to the more variable water levels in comparison to the Lock 4 and Lock 6 reaches (Figure 35). The reach below Lock 1 is connected to the Lower Lakes and wind driven water level changes (seiches) driven by lakes Alexandrina and Albert impact this weir pool.

Despite having higher species richness, *Eucalyptus camaldulensis* (river red gum) seedlings were absent in the Lock 1 reach. Areas where *E. camaldulensis* seedlings were present were typically bare or sparsely vegetated sand bars, which were not present downstream of Lock 1. In the Lock 4 and Lock 6 reaches, the flow pulse not only provided hydrological conditions favourable for *E. camaldulensis* seed germination but also the patches of bare substrate these species require for establishment in their early life stages (*sensu* Petit and Froend 2001). The presence of *E. camaldulensis* seedlings is important because they are generally not recorded in the monitoring programs of other watering interventions such as pumping (Nicol *et al.* 2010, Nicol 2012), weir pool manipulation (Gehrig *et al.* 2015, Gehrig *et al.* 2016) and environmental regulator operation (Nicol *et al.* 2020). In future vegetation surveys the locations of up to 100 *E. camaldulensis* seedlings in each reach will be recorded by GPS and their survivorship in the short to medium-term monitored.

Whilst the highest species richness in all reaches occurred in areas that were inundated by the spring pulse, amphibious and flood dependent species were present across all zones in all reaches (Figure 37; Figure 38). Many amphibious species, such as *E. camaldulensis*, *Duma florulenta* and *Cyperus gymnocaulos* are long-lived perennials that are tolerant to low soil moisture (Cunningham *et al.* 1992) and are widespread across the entire floodplain. In addition, there were many flood dependent and amphibious species present in the *Not inundated* zone that had recruited in response to the 2016 flood (J. Nicol *pers. obs.*), which may have resulted in there being less difference in plant communities between inundated and non-inundated areas. In future surveys the impact of the 2016 flood in the *Not inundated* zone will become less evident in the absence of large overbank floods.

The second monitoring hypothesis was not supported by the results:

- The increase in water level due to increases in flow above regulated entitlement flow will result in increased productivity of littoral vegetation in areas inundated compared to non-inundated areas.

Patterns of above ground biomass at the different inundation zones varied between reaches. The patterns at the Lock 6 reach generally supported the hypothesis but it is unlikely that the differences between zones, with the exception of the *Not inundated* zone, are significantly different (Figure 41). The pattern downstream of Lock 4 was completely different with low biomass at *Pool level* and the highest on the edge of the inundation footprint (Figure 41). Downstream from Lock 1 biomass decreased with decreasing elevation (Figure 41).

It is unclear why above ground biomass patterns were different at each reach. High biomass in the *Not inundated* zone was probably the result of the 2016 flood; however, this was not evident in the Lock 6 reach (Figure 41). There was evidence to suggest that increased root zone soil moisture at the *Inundation extent* zone resulted in increased growth of the extant vegetation. This was clear in the Lock 4 reach, with relatively high biomass also recorded in the other two reaches (Figure 41).

Finally, the results (in particular, diversity and plant communities) showed that the elevation gradient can be used to demonstrate the benefit of environmental water when the water is used to create a flow pulse. In future years, the approach will be modified for different patterns of water delivery as it does not require the use of fixed position quadrats. In other monitoring programs where fixed position quadrats were used to monitor the change in vegetation before and after an intervention (e.g. Chowilla intervention monitoring) (Nicol *et al.* 2010; Nicol 2012), the zones of highest diversity and biomass after the intervention were often not sampled because there were no quadrats present. Comparing vegetation along an elevation gradient and positioning quadrats in relation to water delivery ensures that all important zones are monitored. The main drawback to this approach is that change through time is not measured but this can be overcome by having a sub-set of quadrats at fixed elevations surveyed every year.

Management implications

Results from the 2019-20 water year showed that delivery of a spring pulse was beneficial for littoral vegetation diversity, but the influence was unclear with regards to above ground biomass in the LMR. Delivering environmental water to support a spring flow pulse in the river channel reinstates part of the natural hydrograph. The increase in water level is coupled with an increase in water velocity and in turn hydraulic complexity and lateral and longitudinal connectivity. This is in contrast to other watering interventions commonly undertaken in the LMR such as weir pool raising and environmental regulator operation, where the water level rise is decoupled from the increase in flow velocity, or pumping where there is no connectivity with the river channel. Whilst these differences probably have less consequences for plants compared to other biotic groups, as water level is the key driver of plant communities and species typically have persistent (*sensu* Thompson 1992) or aerial seed banks (Nicol 2004), they may be significant. Although it is impossible to make direct comparisons between this study and other vegetation monitoring programs undertaken in the LMR; the presence of *Eucalyptus camaldulensis* seedlings is one significant outcome that is different between this intervention and other watering interventions, such as weir pool raising, regulator operation and pumping where they were not observed (Nicol *et al.* 2010; Nicol 2012; Gehrig *et al.* 2015; Gehrig *et al.* 2016; Nicol *et al.* 2020). Another benefit of water delivery as a flow pulse is that it supports hydrochory (dispersal of propagules by water), which is a significant source of propagule input to inundated areas (Gibbs *et al.* 2020). These results provide evidence that water delivery as a pulse is preferable compared to using environmental water to support weir pool raising, regulator operation or pumping. Whilst the aforementioned interventions have positive outcomes for vegetation, they should be seen as measures for maintaining perennial vegetation during extended periods of low flow.

One drawback of the flow pulse was the recruitment of several exotic species, in particular the proclaimed pest plant *Xanthium occidentale*. This species is a common amphibious pest plant on stream banks, floodplains and in temporary wetlands (Hocking 1983; Nicol *et al.* 2018b) and is an unavoidable consequence of flooding or watering interventions in the LMR. The proximity to water prohibits the use of herbicides, which makes control of pest plants difficult in littoral zones, but the benefits to native species as observed in this instance outweigh the negative impacts of pest plants.

Conclusions

The 2019-20 flow pulse (and in turn environmental water delivery) resulted in changes to the littoral plant community in areas that were inundated resulted in increased species and functional diversity. Water delivery also provided hydrological conditions to support *Eucalyptus camaldulensis* germination in areas where vegetation is sparse or absent. Patterns of biomass could not be related to water delivery and were inconclusive in 2019-20.

2.5 Microinvertebrate Assemblage

Background

To date, more than 400 species of planktonic and littoral microinvertebrates (protists, rotifers and microcrustaceans) have been identified in the LMR and Lower Lakes. This assemblage provides an important food resource for a range of higher order consumers (Shiel *et al*. 1982; Shiel and Aldridge 2011; Shiel and Tan 2013a, 2013b). Different habitat types support different assemblages of microinvertebrates within riverine ecosystems, and at the simplest level can be classified as either littoral, limnetic, lotic or benthic.

Microinvertebrates are rapid responders to environmental flows. Within habitats that undergo wetting and drying cycles (e.g. the littoral zone and floodplains), these organisms start to emerge from an egg-bank and begin to reproduce within hours of inundation (Tan and Shiel 1993). Therefore, a healthy egg-bank, which is primarily a result of a long-term flooding regime, is an important driver of the magnitude of response to inundation (Boulton and Lloyd 1992). Once inundated, longer water residence times (WRT) will result in higher density and biomass of organisms and result in a shift from rotifer to crustacean dominated communities (e.g. Basu and Pick 1996; Baranyi *et al*. 2002; Obertegger *et al*. 2007). Therefore, habitats such as littoral zones and floodplains favour the development of abundant microinvertebrate communities which can then be transferred between habitats, through hydrological mixing and exchange. Once in the main river channel, only some organisms will survive, with a component of the persistent community, reproducing within areas of the main river channel. Which component persists will largely depend on factors such as season and hydraulics.

Therefore, Commonwealth environmental water can facilitate the maintenance and development of microinvertebrate assemblages within the Lower Murray region by:

- Inundating areas adjacent to the main river channel and therefore supporting populations that can act as a source to the main river channel community,
- Improving lateral and longitudinal hydrological connectivity which promotes the dispersal of organisms, and
- Improving the flow regime over the long-term to promote a more diverse and abundant egg-bank and thus more diverse and abundant community dispersed through lateral and longitudinal connectivity.

To determine the responses of the microinvertebrate community to Commonwealth environmental water deliveries in the LMR, their diversity and density is being assessed every year over the spring–summer period. These data can be used to model the response of different microinvertebrate taxa to flow, floodplain inundation and water quality. The microinvertebrate response models can then be used with different modelled flow scenarios to evaluate the contribution of Commonwealth environmental water to microinvertebrate outcomes (for more details on the modelling approach see Appendix D).

Major hypotheses

- H1: Overall microinvertebrate density, and taxonomic richness, will increase with environmental water delivery due to the combined effects of longitudinal transport and improved lateral connectivity with off-channel habitats;
- H2: The density will increase for 'floodplain associated'[e] microinvertebrates during periods of greater floodplain inundation (H2a) and for those that are likely to be transported to the LMR from upstream during periods of high longitudinal connectivity (H2b); and
- H3: Environmental water delivery during spring will increase the density of preferred microinvertebrate prey species for large-bodied fish larvae.

Methods

Sampling sites and procedure

Microinvertebrate sampling was conducted three times during spring and three times during summer approximately two weeks apart between September and February at three core MER sites, concurrent with stream metabolism sampling (Figure 1). This sampling regime was similar to that undertaken during the LTIM Project. See (Ye *et al.* 2020) for more details.

Table 14. Microinvertebrate sampling dates from 2019-20 in the Lower Murray River.

Trip	Sampling dates
1	10/10/2019
2	21/10/2019
3	4/11/2019
4	18/11/2019
5	2/12/2019
6	17/12/2019
7	8/01/2020

A Perspex Haney plankton trap (4.5 L capacity) was used mid-channel (by boat) to collect surface, middle and bottom volumes (9 L), which were filtered through a 37 µm-mesh plankton net suspended in a bucket and rinsed into a 200 ml PET bottle screwed to a purpose-built ferrule at the net end. The filtrate was then preserved in the field (100% ethanol) to a final concentration of ~75%, and a volume of <200 ml. In the laboratory, the sample was decanted into a measuring cylinder, the volume noted, the cylinder agitated, and a 1 ml aliquot withdrawn using a Gilson auto pipette. This aliquot was run into a Pyrex 1 ml Sedgewick-Rafter cell, and the microinvertebrates present were counted and identified. Counts for each sample were based on a single subsample.

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[e] 'Floodplain associated' microinvertebrates are those that are thought to benefit from floodplain environments.

Table 15. Details of microinvertebrate sampling sites in the Lower Murray River.

Zone	**Site**	**Latitude**	**Longitude**
Floodplain	5–15 km DS Lock 6	S-34.0190	E140.87572
Floodplain	15 km DS Lock 4	S-34.3892	E140.59477
Gorge	5–15 km DS Lock 1	S-34.4052	E139.61723

Modelling and statistical analyses

Both categorical and continuous predictors were used to build a model to predict the response of microinvertebrates to flow. Categorical predictors were month, water year, lock and site, while continuous predictors quantified: (1) flow, (2) flow variability, (3) water quality and (4) inundated floodplain area (Appendix D). For flow, we calculated both short-term (mean flow on the date of sampling) and long-term flow (mean flow over the preceding 12-week period) predictors. Flow variability was quantified as the change in flow over the preceding 10-days prior to sampling (denoted 'flow trend' or 'dQ10'). Water quality parameters used were daily temperature and electrical conductivity. Floodplain inundated area was quantified using the DEW MIKE model outputs (M. Gibbs, pers comm; see Appendix D for details). Values were summed over 28- and 60-day periods to estimate inundated floodplain area corresponding to an estimated minimum period required for the majority of taxa to emerge from the egg bank and complete one or two life cycles. A global model including all predictors was fit first, then optimised using an automated procedure. Model predictive performance was assessed prior to use in modelling scenarios (see Appendix D for details).

To assess the influence of Commonwealth environmental water on microinvertebrate density, taxonomic richness and community assemblage structure (including rotifers, cladocerans and copepods) over the long-term (5 years), variation between sampling years (i.e. 2014-15, 2015-16, 2016-17, 2017-18 and 2019-20, presented in Appendix D) and sites (2019-20, presented below) was investigated. In testing all hypotheses, a common methodology was used. First, models were fit to all existing data using observed flow conditions and other predictors (following section). Second, the predictors were re-calculated using the modelled flow data for scenarios of no Commonwealth environmental water contributions (hereafter **"No CEW"**) and no environmental water at all (**"No eWater"**). Models were re-run using the predictors calculated on modelled scenarios and the predicted microinvertebrate metrics were compared between the three scenarios using paired t-tests to determine whether predicted changes in microinvertebrates differed from observed variability (i.e., whether such a result would be expected at random). The overall contribution of environmental water was calculated as the difference between the observed and modelled No eWater scenarios. The difference between the No CEW and observed scenarios was the contribution by Commonwealth environmental water. Models were built to test each hypothesis as follows:

- H1: (a) density of microinvertebrates (estimated as the mean of three replicates) and (b) taxonomic richness (total taxa within the three replicates);

- H2: Density of microinvertebrate taxa with assumed greater dependence on: (i) lateral connectivity (due to the longer period of time to complete their life cycle), and, (ii) longitudinal transport. Category (i) taxa were littoral and pelagic cladocerans, and calanoid and cyclopoid copepods, comprising 32 taxa (Table 16a). Category (ii) taxa (Table 16b) were primarily pelagic rotifers, with their dependence inferred from their known biogeographical range (R. Shiel, pers comm, 2020) along with prior reports indicating the importance of upstream taxa in determining the turnover of lower Murray microinvertebrate assemblages (Ye *et al*. 2020);
- H3: Density of microinvertebrate taxa identified in the Long-Term Intervention Monitoring technical report (Ye *et al*. 2020) as preferred fish prey species (Table 17).

Table 16. Taxa used to quantify the influence of environmental water on.

Dependence group	Taxa
(i) Lateral connectivity	*Acanthocyclops* cf. *vernalis, Armatalona macrocopa, Australocyclops australis, Boeckella triarticulata, Bosmina meridionalis, Calamoecia ampulla, Calamoecia sp.*, calanoid and cyclopoid copepodites, *Ceriodaphnia cornuta, Ceriodaphnia* sp., *Chydorus* cf. *eurynotus, Daphnia carinata s.l., D. galeata, D. lumholtzi, Daphnia sp., Diaphanosoma excisum, Gladioferens* sp., *Ilyocryptus* sp., *Leberis diaphanus, Macrothrix* sp., *Mesocyclops notius, Microcyclops varicans, Moina cf. australiensis, Moina* cf. *tenuicornis, Moina micrura, Neothrix sp.,Pseudochydorus globosus, Pseudomonospilus diporus, Simocephalus* sp., *Thermocyclops* sp.
(ii) Longitudinal connectivity	*Anuraeopsis coelata, Anuraeopsis fissa, Brachionus bidens, B. calyciflorus amphiceros, B. diversicornis, B. falcatus, Cephalodella catellina, Collotheca cf. tenuilobata, Conochilus dossuarius, C. natans, C. unicornis, Filinia longiseta, F. pejleri, F. terminalis, Keratella australis, K. cochlearis, K. lenzi, K. procurva, K. slacki, Polyarthra remata, P. vulgaris, Synchaeta oblonga, S. pectinata, Trichocerca pusilla complex, T. similis, T. similis grandis*

Table 17. Microinvertebrate prey species for fish larvae of large-bodied native species identified from gut content analysis used in modelling fish prey species density.

Taxon	Family	Phylum/Order
Boeckella triarticulata	Centropagidae	Calanoida
Bosmina meridionalis	Bosminidae	Cladocera
Brachionus spp (22 taxa)	Brachionidae	Rotifera
Calamoecia spp (2 taxa)	Centropagidae	Calanoida
Ceriodaphnia spp (2 taxa)	Daphnidae	Cladocera
Daphnia (4 taxa)	Daphnidae	Cladocera
Filinia (7 taxa)	Trochosphaeridae	Rotifer
Gladioferens sp.	Centropagidae	Calanoida
Ilyocryptus sp.	Ilyocryptidae	Cadocera
Keratella australis	Brachionidae	Rotifera
Keratella procurva	Brachionidae	Rotifera
Lecane spp. (14 taxa)	Lecanidae	Rotifer
Neothrix sp.	Neotrichidae	Cladocera

Results

Density patterns

Mean density across the three replicates for all sampling events over the 2019–20 water year ranged from 190–3,387 ind/L (mean [± 95% CI] = 1,057 [698, 1,416]). Generally, sites tended to have comparable densities (e.g. near overlap in confidence intervals; Figure 42a), except for an unusually high density recorded at Lock 4 in the final sample of the year. Lock 1 site samples tended to have higher density than upstream sites early in the season, but this pattern reversed after December. The 21 October 2019 samples at all sites were collectively of higher density than other sampling times.

Diversity patterns (taxonomic richness)

Taxonomic richness ranged from 12–27 taxa (18.3 [16.3, 20.3]) and was similar at all locks early in the water year (Figure 42b). All sites had similar richness early in the year, but concurrent with the decline in density, taxonomic richness declined at the Lock 1 site, remaining lower than the two upstream locks, which both generally increased in richness over the year.

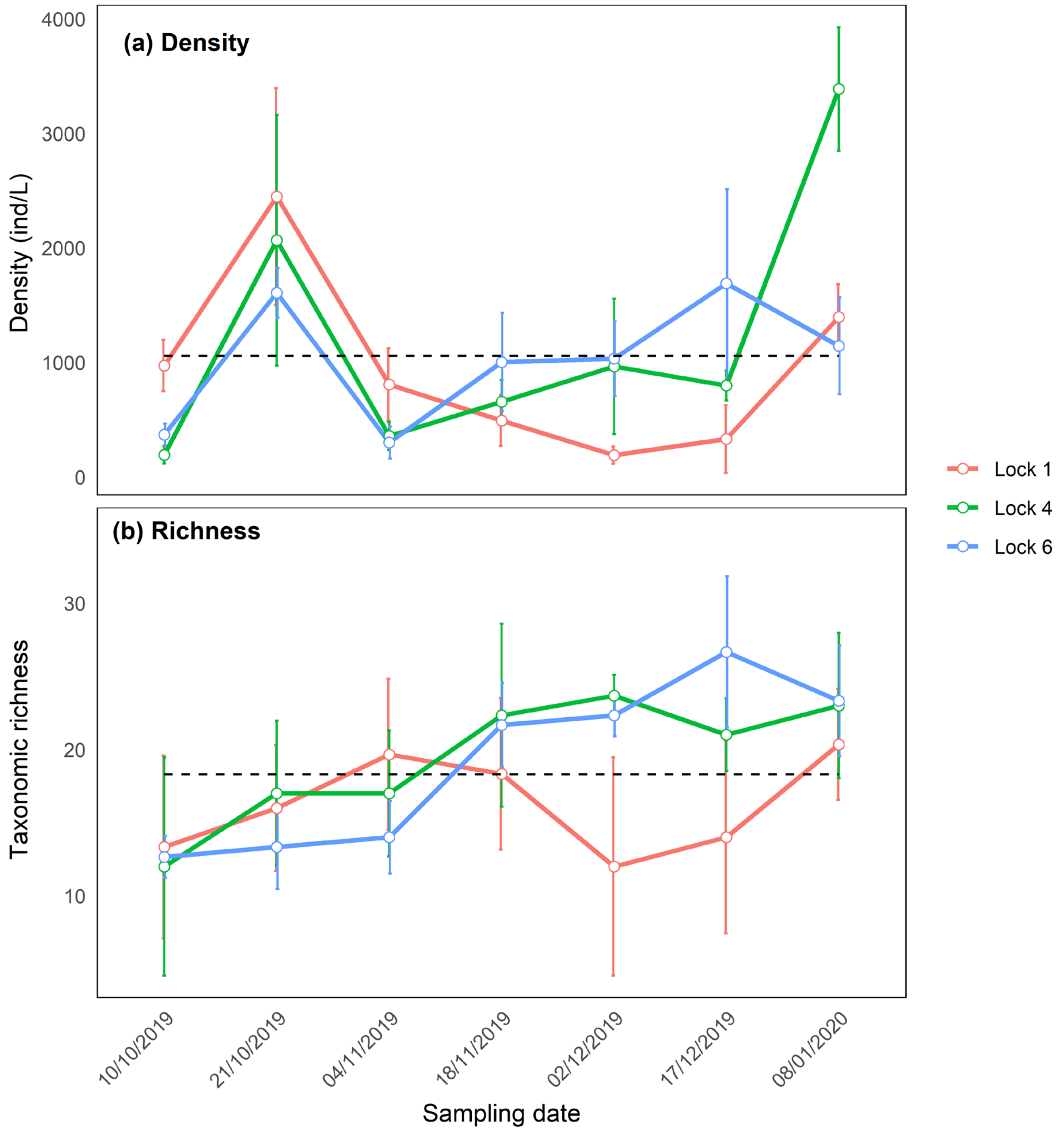

Figure 42. **Observed density and taxonomic richness for 2019-20 sampling at each lock. (a) mean density of three sub-samples at each site (lock, indicated by colour) and date. Error bars show 95% confidence limits and horizontal black lines show mean for the water year.**

Functional group density

The distribution of functional group density among sampling sites during 2019-20 suggests dominance of pelagic rotifers throughout the year, and pelagic and littoral rotifers on 21 October 2019 (Figure 43). All three functional group classes identified in Figure D10 (Appendix D) were observed at least once during 2019-20 at each of the locks.

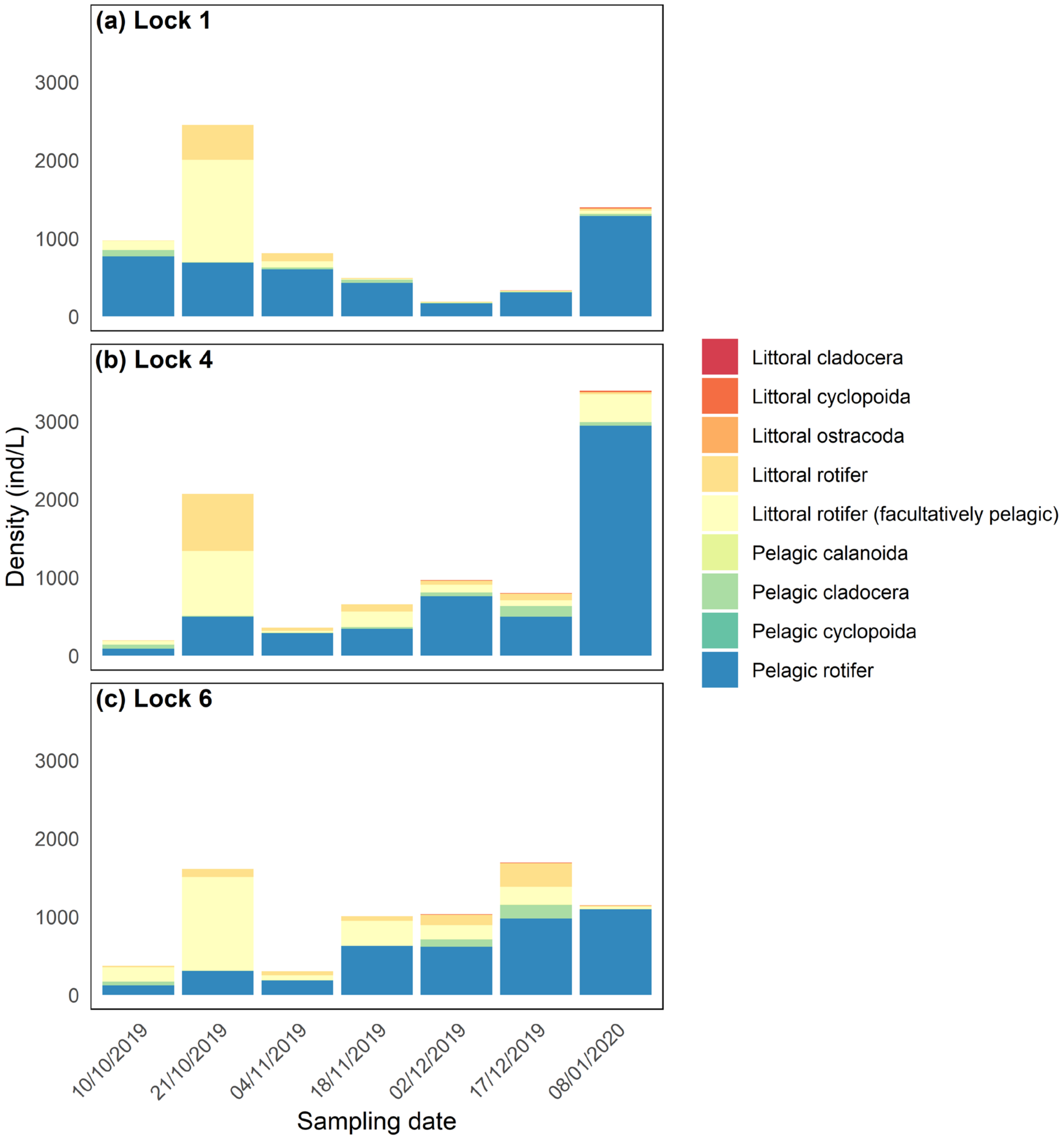

Figure 43. Functional group density by sampling date and lock for the 2019-20 season. Three sub-samples were collected at a single sample site located 5 km downstream of each lock shown in the panel caption.

<u>*Modelled responses to environmental flows*</u>

H1: Density and taxonomic richness

The modelling for 2019-20 estimated 28% of observed microinvertebrate density was due to environmental water delivery (mean difference between observed and no environmental water scenarios ±[95% CI] = 342.2 [127.2, 557.3] ind/L; *t* = 3.32, df = 20, *p*-value = 0.003) (Figure 44a). Commonwealth environmental water was estimated to have accounted for 83% of this (mean difference ± [95% CI] between observed and no Commonwealth environmental water scenarios = 284.7 [80.6, 488.8] ind/L; *t* = 2.91, df = 20, *p* = 0.008).

Around 11% of taxonomic richness for 2019-20 was predicted to have been due to environmental water (mean difference = 4.1 [1.3, 6.9]; *t* = 3.09, df = 20, *p* = 0.006), with Commonwealth environmental water estimated to have contributed around 68% of this (mean difference = 2.8 [0.1, 5.6]; *t* = 2.09, df = 20, *p* = 0.04) (Figure 44b).

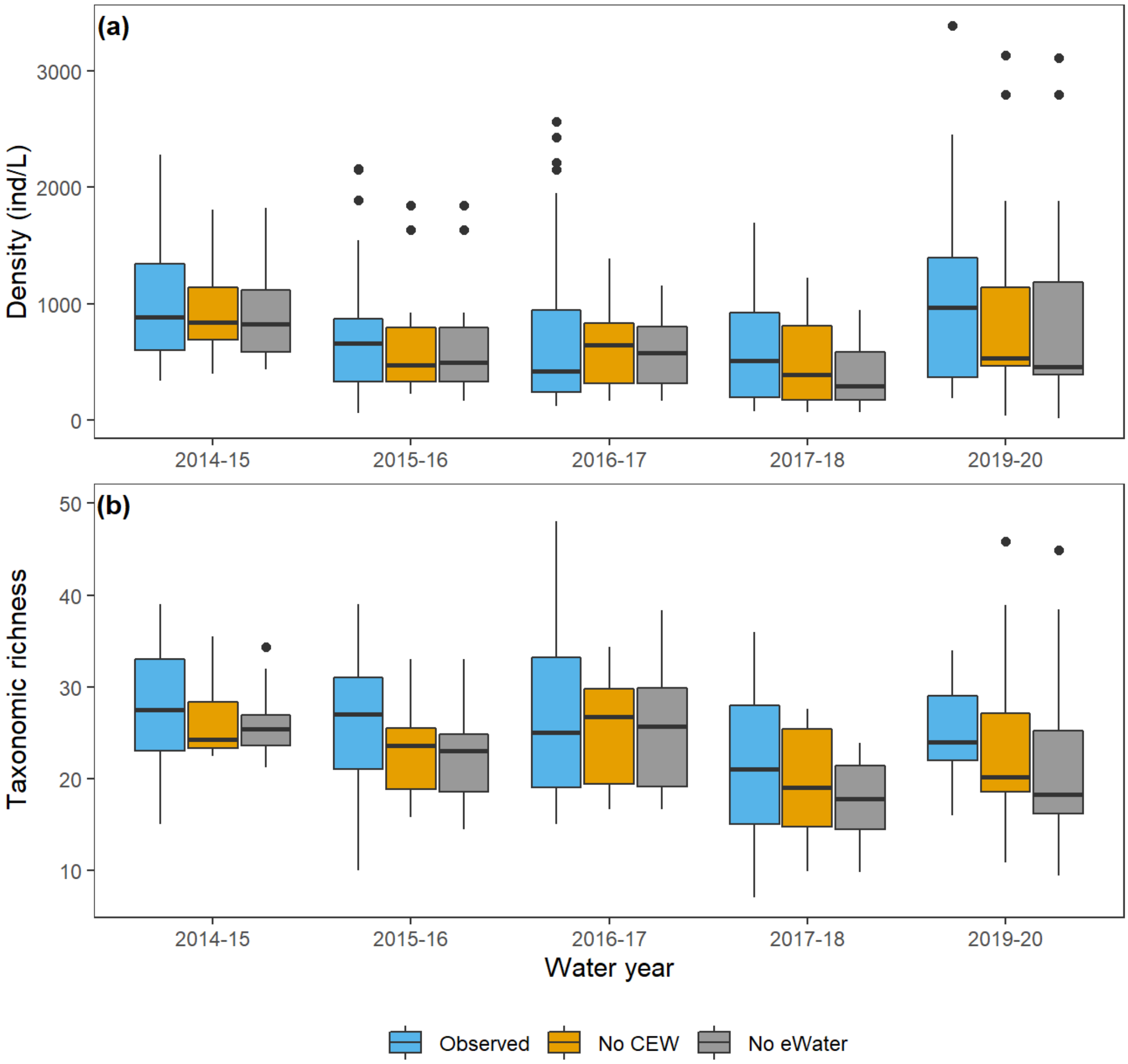

Figure 44. Observed and modelled (a) microinvertebrate density (individuals/litre, ind/L) and (b) taxonomic richness (number of taxa) by water year over the period 2014–2020 (2018-19 not sampled). In each panel, observed data ('Observed'; blue bars) for all samples in the water year shown on the x-axis are compared with modelled expectations under two environmental flow scenarios: Under no Commonwealth environmental water delivery ('No CEW'; orange bars) and if no environmental water were provided at all ('No eWater'; grey bars). Results of the generalised linear models used to run the scenarios are given in Tables D4 (density) and D5 (taxonomic richness) in Appendix D.

H2: Lateral and longitudinal connectivity

In 2019-20, modelling indicated that the density of floodplain dependent taxa increased 43% due to environmental water delivery, leading to increased lateral connectivity (mean difference = 18.1 [8.2, 28.0] ind/L; *t* = 3.81, df = 20, *p* = 0.001) (Figure 45a). Commonwealth environmental water was predicted to have contributed 72% of this increase (mean difference = 13.0, [3.7, 6.5] ind/L; *t* = 5.07, df = 20, *p* < 0.001).

Similarly, in 2019-20, modelling indicated that the density of taxa common to upstream areas increased 32% due to environmental water delivery, improving longitudinal connectivity (mean difference = 219.9[127.8, 312.0] ind/L; *t* = 4.98, df = 20, p < 0.001) (Figure 45b). Commonwealth environmental water was estimated to account for 83% of this (mean difference = 181.7 [102.5, 260.9] ind/L; t = 4.78, *df* = 20, p < 0.001).

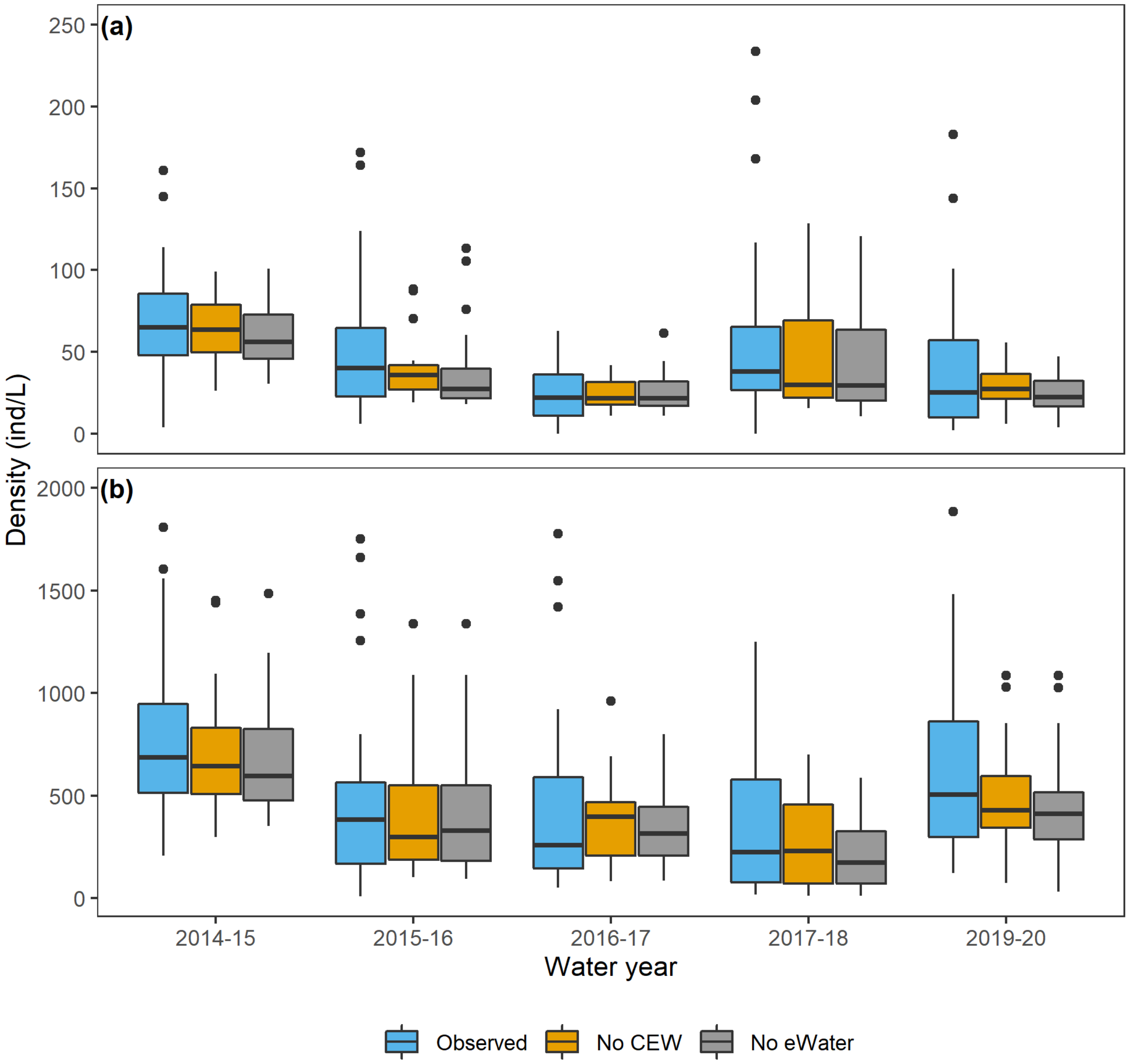

Figure 45. Observed and modelled density (individuals per litre, ind/L) with and without environmental water (a) microinvertebrate taxa dependent on lateral connectivity to floodplains and backwaters and (b) microinvertebrate taxa dependent on longitudinal transport and connectivity with upstream areas by water year over the period 2014–2020 (2018-19 not sampled). In each panel, observed data ('Observed'; blue bars) for all samples in the water year shown on the x-axis are compared with modelled expectations under two environmental flow scenarios: Under no Commonwealth environmental water delivery ('No CEW; orange bars) and if no environmental water were provided at all ('No eWater'; grey bars). Results of the generalised linear models used to run the scenarios are given in Tables D4 (density) and D5 (taxonomic richness) in Appendix D.

H3: Spring microinvertebrate prey species density

Although 2019-20 spring prey species density (defined as all individuals of the taxa in Table 17 in each sample) was the lowest recorded at 73.9 [48.3, 99.5] (mean ± [95% CI]), differences in density between years were not statistically significant (Kruskal Wallis X^2 = 5.84, df = 4, p = 0.21), due to the high variability within sites in all sampling years.

Modelled spring prey species density in the absence of any environmental water (Figure 46) was 62% lower (mean difference ±[95% CI] between observed and modelled no environmental water scenarios = 40.3 [17.5, 62.8] ind/L; t = 3.93, df = 11, p = 0.002). Commonwealth environmental water was estimated to account for ~59% of this difference (23.6 [-7.4, 54.6] ind/L per sampling event) and although not statistically significant (because of the small number of samples involved; t = 1.68, df = 11, p = 0.12), such an increase in prey-species density was likely of biological importance.

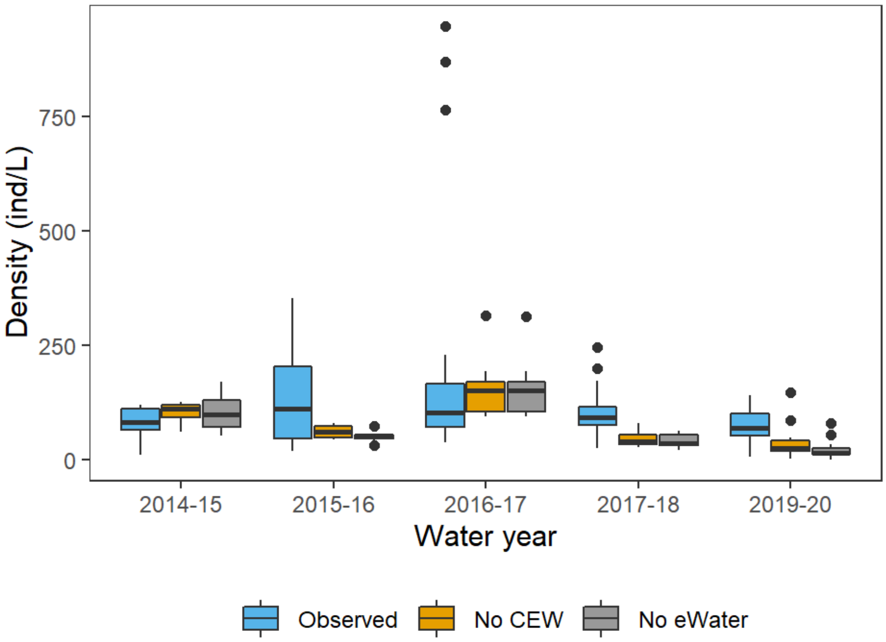

Figure 46. Observed and modelled density (individuals per litre, ind/L) of microinvertebrate prey taxa for large-bodied fish larvae in the Lower Murray during spring months.

Evaluation

The evaluation approach is described in the evaluation section for Hydraulic Regime (Section 2.1).

Table 18. Microinvertebrate Assemblage evaluation questions and answers. CEW = Commonwealth environmental water, eWater = environmental water. Hypotheses: H1a, H1b, H2a, H2b and H3. The mean [95% CI] and % attributable to CEW contributions for each water year are detailed for each evaluation question and each water year. The significance of the contribution was classified as none-negligible if the contribution was 0–9%, minor = 10–19%, moderate =20–29% and substantial = ≥30%.

CEWO evaluation questions	**Outcomes of CEW delivery**				
	2014-15*	**2015-16***	**2016-17**	**2017-18***	**2019-20***
What did CEW contribute to microinvertebrate density (H1a)?	72.6 [47.9, 97.3] (+7%)	58.7 [33.4, 84.0] (+8%)	25.9 [10.3, 41.6] (+4%)	131.3 [83.8, 178.7] (+21%)	284.7 [80.6, 488.8] (+23%)
	eWater increased overall microinvertebrate density by an average of 18% across all years, with CEW accounting for 64% of this.				
What did CEW contribute to microinvertebrate diversity (taxonomic richness) (H1b)?	0.8 [-0.3, 2.0] (+3%)	1.0, [0.4, 1.6] (+4%)	0.9 [-0.2, 2.0] (+3%)	24.4 [1.6, 3.4] (+11%)	2.8 [0.0, 5.6] (+11%)
	eWater increased overall microinvertebrate taxa richness by an average of 9% across all years, with CEW accounting for 65% of this.				
What did CEW contribute to microinvertebrate communities of the LMR via lateral connectivity (H2a)?	1.8 [-0.8, 4.5] (+3%)	11.5 [8.6, 14.4] (+22%)	-0.67 [-1.4, <0.1] (-3%)	15.08 [9.2, 20.9] (+24%)	13.0 [3.2, 22.8] (+30%)
	eWater increased the density of taxa dependent upon lateral connectivity by an average of 18% across all years, with CEW accounting for 86% of this.				
What did CEW contribute to microinvertebrate communities of the LMR via longitudinal connectivity (H2b)?	52.0 [34.6, 68.7] (+6%)	22 [7.1, 36.4] (+4%)	19 [7.3, 30.3] (+5%)	70 [44.9, 94.3] (+19%)	182 [102.5, 260.9] (+27%)
	eWater increased the density of taxa dependent upon longitudinal connectivity by an average of 17% across all years, with CEW accounting for 65% of this.				
What did CEW contribute to spring microinvertebrate fish prey species density (H3)?	17 [10.0, 24.7] (+15%)	59 [47.4, 69.7] (+49%)	0.4 [0.25, 0.59] (0%)	65 [53.7, 75.7] (+59%)	24 [-7.4, 54.6] (+37%)
	eWater increased the density of microinvertebrate fish prey species during spring by an average of 37% across all years, with CEW accounting for 86% of this.				

* = additional management levers were also used (Appendix B)

Contribution (to what extent CEW contributed towards the outcome, with the significance of the outcome considered):

⬜ Unknown 🟥 Negative ⬜ None/negligible 🟩 Minor 🟩 Moderate 🟦 Substantial

Discussion

A combination of trend analyses in observed data and ecological modelling was used to assess the microinvertebrate community and its response to environmental water delivery across all years, with specific interest in the first year of MER sampling (2019-20). Raw data comparison of diversity (taxonomic richness and evenness) and functional group (density) showed that the microinvertebrate community in 2019-20 had high density and taxonomic richness during peak environmental water delivery. These increases in taxonomic richness and density at a time of increased flow, suggest that organisms were also being dispersed downstream, a vital process in the protection of species and genetic diversity. Through increases in local richness and the supply of new recruits (in the form of diapause eggs and live organisms) downstream, these flow induced responses also support successional processes following future disturbance events (resilience). The increases in density are also likely to have broader outcomes for the aquatic food web, as microinvertebrates provide food for a range of higher trophic organisms. The greater the density of microinvertebrates, the higher the rate of predator-prey encounters. This means, predators do not need to utilise as much energy to obtain food to meet their own physiological and behavioural requirements (Cooper and Goldman 1980; Vinyard 1980). Assemblages in 2019-20 were characterised by Group 2 taxa (littoral/pelagic rotifers), a group primarily comprised of taxa from the *Trichocerca pusilla* complex[f]. Previous studies have indicated that these taxa are positively associated with increased flow and longitudinal connectivity during spring (see Furst *et al*. 2017, 2018, 2020). Therefore, these increases are likely to be related to the timing and longitudinal connectivity achieved through environmental water delivery in 2019-20.

Results of modelling using all data (that is, including LTIM and MER samples), showed clear benefits for microinvertebrate density and richness from environmental water delivery. These benefits were highest for floodplain dependent taxa and during 'dry' years (2015-16, 2017-18 and 2019-20). This was particularly apparent in spring prey species density estimates, where Commonwealth environmental water alone were predicted to have attributed to between 36 and 59% of observed values. Numerically, this represents between 23 and 64 ind/L of prey species density, a substantial contribution to riverine productivity during low flow years.

<u>*Diversity (taxonomic richness and evenness) patterns*</u>

In interpreting the 2019-20 data against the LTIM period, changes in the spatial sampling design warrant a degree of caution. However, the similarity with water year 2014-15 increases confidence, as both years had comparable statistics, featuring similarly high richness, high densities and low evenness. Taxonomic richness was the highest of any year in 2019-20 (18.3 ± 0.59 ind/L) (indicative of high resilience), while taxonomic evenness was low (suggesting a community where most individuals belong to one or a few taxa and many taxa are rare and indicative of low resistance) in comparison to all other years (0.69

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[f] '*Trichocerca pusilla* complex' is a group of morphologically similar species which are highly difficult to discriminate, including *Trichocerca pusilla, T. agnatha* and one or more undescribed *Trichocerca* species.

± 0.02), except for 2014-15 (0.66 ± 0.02). See Appendix D for further explanation of resistance and resilience.

Tellingly, a similarity between the 2014-15 and 2019-20 sampling periods was the occurrence of large spring/summer floods in the prior two to three water years. Large spring/summer floods are likely to drive dispersal and productivity simultaneously, replenishing the microinvertebrate eggbank throughout the system (e.g. Junk *et al*. 1989; Jenkins and Boulton 2003). Over time however, eggbanks degrade, where some taxa are more prone to degradation than others (Brendonck and De Meester 2003). Therefore, without new recruits, eggbanks become less diverse over time (e.g. Boulton and Lloyd 1992). Consequently, large spring/summer floods are likely to have a temporally declining residual effect (due to the degradation of the eggbank) on the active community. Disturbance can often increase evenness as it removes dominant taxa (Hillebrand *et al*. 2008), as we observe in the 2016-17 data. Thus, we theorise that the 2014-15 and 2019-20 sampling periods reflect more stable microinvertebrate communities, benefiting from a legacy effect of replenished and diversified floodplain egg banks from the high flow periods throughout much of 2011-12 and 2012-13 and the large flood event during 2016-17. This result highlights one of the many important aspects of frequent large-scale spring/summer floods. While such flood events are not a direct outcome of environmental water, the latter clearly plays a crucial role in maximising the benefits over subsequent years, by maintaining lateral connectivity and transporting microinvertebrates back to the main channel of the river. This effect was evident in the modelling, where in the low flow years following the flood events, 21–31% of the observed density of floodplain-dependent taxa was estimated to be attributable to Commonwealth environmental water.

Functional group density

There were clear patterns in the relative dominance of functional groups, with Group 2 (high density of littoral/pelagic rotifers) being most dominant in 2019-20. Group 2 samples were primarily comprised of taxa from the *Trichocerca pusilla* complex along with *T. similis* and *T. similis grandis*. Group 2 samples were present under all sets of conditions, but most favoured during long-term flows (mean daily flow over 12-weeks) exceeding 5.4 GL/d but with relatively constant (or slowly decreasing) flow, or during the month of December. This finding is consistent with previous studies which have found these taxa to occur in high density during periods of higher longitudinal connectivity, water velocity and discharge, particularly during the months of October and November within the main channel of the LMR (Furst *et al.* 2017, 2018; Gibbs *et al.* 2020). Under these conditions, it is possible that a proportion of these organisms are swept from littoral habitat (their preferred habitat, e.g. Chengalath and Mulamoottil 1975), entrained within the flowing water and moved downstream. Increases in density may then occur due to in-channel reproduction and/or the constant entrainment of these organisms into an envelope of water as it passes downstream. In fact, interruptions to longitudinal connectivity related to diversions and structures that cause decreases in water velocity appear to disrupt longitudinal trends (suggesting interference in transportation) of these organisms (Furst *et al.* 2018). This and previous studies suggest that peaks in the density of these taxa frequently occur between October and November when water temperatures are between ~19–21 °C (Furst *et al.* 2017, 2018). However, this study also highlighted that peaks also commonly occurred in December in the LMR. One possible explanation for this is that the later peaks may be due to transportation of microinvertebrates from upstream areas (e.g. near Tocumwal) (accounting for travel times), where the required flow and temperature conditions are being met as in the LMR in November. The specific species of higher trophic levels that predate upon *Trichocerca* species in the LMR have not been identified. However, a number of international studies have found this genus to be an important food resource for both fish (e.g. Van Den Avyle and Wilson 1980; McCullough and Stanley 1981; Dev and Rahmatullah 1998; Sampson *et al.* 2009) and shrimp (e.g. Grossnickle 2001; Haskell and Stanford 2006). For more discussion relating to patterns in Group 1 and Group 3 microinvertebrates in previous years see Appendix D.

Modelled effects of environmental flows for microinvertebrates

Overall, modelling demonstrated that environmental water delivery has consistently provided benefits for the microinvertebrate community in the LMR. All three hypotheses under test were clearly supported.

The first hypothesis was that *overall microinvertebrate density, and taxonomic richness, will increase with environmental water due to the combined effects of longitudinal transport and improved lateral connectivity with off-channel habitats*. Modelling indicated that in 2019-20, there was a higher than usual benefit from Commonwealth environmental water, representing a predicted 23% increase in density, compared with the all-years average of 18%. As discussed above, we speculate this could be a legacy effect of large flood events in the recent flow history. Predictably, because of its correlation with density, taxonomic richness also increased due to Commonwealth environmental water, although the proportional increase was only 9% across all years and 10% in 2019-20. Due to the high correlation between density and richness, the testing of other hypotheses focused only on estimating changes in density.

The second hypothesis was that *density will increase for 'floodplain associated' microinvertebrates during periods of greater floodplain inundation (H2a) and for those that are likely to be transported to the LMR from upstream during periods of high longitudinal connectivity (H2b)*. Both H2a and H2b were supported, with similar level of effect to that found for all taxa (~19% overall) but a greater contribution attributable to Commonwealth environmental water for lateral connectivity dependent taxa (~85%). This dispersal, both longitudinally and laterally, are important processes in the preservation of species and genetic diversity, through the supply of new recruits to downstream habitats (spatial dispersal) and the egg bank (temporal dispersal).

The third hypothesis was that *environmental water delivery during spring months will increase the density of preferred microinvertebrate prey species for fish.* This hypothesis had the strongest support, particularly for low flow years. Modelling estimated that in 2019-20, the density of prey species in spring would have been ~37% lower without Commonwealth environmental water.

In 2019-20, environmental water was delivered in November/December, resulting in an in-channel flow pulse of ~15,000 ML/d. It is likely that the increases were driven by a combination of the flushing of weir pools and the inundation of temporary habitats adjoining the river such as backwaters, low-lying wetlands and floodplain, triggering emergence from the egg-bank and providing habitat for reproduction. Additionally, improved lateral connectivity due to environmental water probably increased the access to floodplain egg-banks replenished in 2016-17, a flood year, therefore greater microinvertebrate increases were observed in 2017-18 than 2019-20.

Unsurprisingly, models selected several common predictors, including hydrological, inundated floodplain areas and physiochemical predictors along with some two-way interactions. Firstly, despite a high correlation with flow being evident for all periods, the influence of flow tended to be more predictive using longer periods (e.g. 12 weeks) than mean daily flows. The positive coefficient estimates are evidence that delivery of more water over a longer period is generally going to achieve better outcomes. Microinvertebrates are well recognised for their ability to respond rapidly to environmental water delivery (i.e. within hours) due to emergence from the egg-bank (e.g. Tan and Shiel 1993). Therefore, despite the importance of greater flows over longer periods seeming intuitive, as they are required for emergent communities to develop into stable and highly dense communities, this can often be overlooked (e.g. Obertegger *et al.* 2007). This finding emphasises that the duration of flow should be a key consideration when delivering environmental water for meeting objectives related to the microinvertebrate community.

Secondly, as expected, greater area of floodplain inundated had a positive effect, however, it was involved in a negative interaction with electrical conductivity in all models. This suggests that positive outcomes achieved with floodplain inundation (including high microinvertebrate densities, longitudinal and lateral dispersal and food production for fish), are likely to be greatest during periods of lower electrical conductivity. The effect of electrical conductivity on taxonomic richness is not surprising as salinity is well documented as having a negative relationship with microinvertebrate taxonomic richness in freshwater and coastal habitats (e.g. Marc *et al.* 2003; Nielsen *et al.* 2008; Gutierrez *et al.* 2018). Therefore, these results suggest that electrical conductivity had important, but indirect, multiplicative effects on taxonomic richness, particularly for specific subsets associated with lateral connectivity in this part of the system. Within the LMR, electrical conductivity commonly increases throughout summer, reaching its highest values during minimum flow periods, typically in late summer. Therefore, the delivery of environmental water that aims to inundate temporary habitats, including floodplain, may have less value late in the season due to increased electrical conductivity. In support of this, floodplain taxa and fish food both exhibited lower densities in later months. Notably, temperature and electrical conductivity had a negative interaction, suggesting that when both values are high, the combined effect on microinvertebrates is detrimental.

Another notable predictor was the flow trend over the past ten days, which showed an increasing trend in flow also increased density but, after accounting for this, taxonomic richness had a negative association. This suggests that for a given density, taxonomic richness increased more as water levels decreased rather than increased. This supports the conceptual understanding that as water levels decrease, microinvertebrates from the floodplain, wetland, backwater and littoral habitats drain back into the main river channel, transferring new taxa and thus providing a wider variety of food resources for higher trophic organisms.

Management implications

This project has highlighted a number of key findings that may be used to inform future environmental water deliveries, aiming to promote a response in the microinvertebrate community in the LMR.

Environmental water delivery consistently provided benefits for microinvertebrate densities (including prey densities) across years, however there appeared to be limited benefits in high flow years and greater benefits in subsequent years following a large unregulated flood. This, however, was largely due to the smaller proportion of environmental water in comparison to overall flow in high flow years in comparison to low flow years. The value of similar volumes of environmental water as different percentages of overall flow might warrant exploration. For example, environmental water delivery might be prioritised in the first year or two following large floods, specifically to increase in-channel productivity by leveraging higher floodplain microinvertebrate richness and density.

There were clear patterns in the relative dominance of functional groups under different flow conditions. Investigations such as those being undertaken as part of the research task of this project will provide insights into the quality, as a food resource for higher trophic organisms, of different communities (dominated by different functional groups). Environmental water delivery can then aim to promote specific communities characterised by certain functional groups at appropriate times.

Increases in prey items appeared to be driven by both the flushing of organisms from the lake like conditions that develop in weir pools and through the inundation of temporary habitats during pulse flows and the flushing of backwater habitats and littoral zones. Again, investigations such as those being undertaken as part of the research task of this project may provide insights into whether these similar food resources, yet from different habitats, provide comparable quality food for higher trophic organisms.

There were numerous predictors that were consistently important in modelling, both hydrological and physicochemical and could potentially help inform future environmental water delivery for microinvertebrate communities. For example: (1) the influence of flow tended to be more predictive using longer periods (e.g., 12 weeks) than mean daily flows, suggesting that more water delivered over a longer period could be more likely to achieve better outcomes; and (2) greater area of floodplain inundated had a positive effect, but in all models the benefits decreased with increasing salinity (electrical conductivity), therefore, positive outcomes of floodplain inundation are likely to be greatest during periods of lower salinity.

Conclusion

Data collected as part of LTIM and MER have generated a unique multi-year microinvertebrate dataset. This dataset has allowed an approach that identified functional groups and the hydrological conditions in which they were commonly present. Additionally, it has allowed a modelling approach to be undertaken which identified several hydrological and physicochemical predictors influential on observed microinvertebrate density, taxonomic richness and composition. Moving forward, future work should aim to identify and incorporate additional predictors (e.g. the density of higher and lower trophic levels) and further refine those identified here to increase explanatory power of the models and better inform alternative management interventions. Concurrent research being conducted under the research task of this project, aims to provide insights into the ecological value of various planktonic (including microinvertebrate) communities within the broader food web, such as their quality as a food resource (i.e. fatty acid composition). These investigations combined are likely to provide a more quantifiable guide for managers to better inform future environmental water delivery.

2.6 Flow-cued Spawning Fish Recruitment

Background

In the southern MDB, spawning and recruitment of golden perch corresponds with increases in water temperature and discharge, either in-channel or overbank (Mallen-Cooper and Stuart 2003; Zampatti and Leigh 2013a; 2013b). Silver perch display similar life history characteristics and population dynamics, although in the lotic reaches of the Murray River, silver perch may spawn circa-annually (Tonkin *et al.* 2019). Annual increases in flow (spring flow pulses) were a distinct hydrological feature of the unregulated Murray River (Mallen-Cooper and Zampatti 2018). In regions where these features remain intact (e.g. the mid-Murray River), golden perch display more consistent recruitment (Zampatti *et al.* 2018). In the Murray River downstream of the Darling junction, however, spring flow pulses are compromised by river regulation. Commonwealth environmental water may be used, at least in- part, to restore these pulses.

Since 2012, >500 GL/year of Commonwealth environmental water has been delivered to the LMR to enhance the flow regime and rehabilitate the health of aquatic ecosystems. In the LMR, Commonwealth environmental water has been primarily used to contribute to increased base flows and freshes (i.e. increases in flow contained within the river channel) (Section 1.4). One of the ecological objectives of flow augmentation was to contribute to increased spawning and/or recruitment of flow-dependent fish species. Since 2012-13, there has been no substantial recruitment of golden perch, despite some spawning of golden perch coinciding with periods when environmental water was used to promote flow pulses in the LMR. This has led to a population dominated by a few distinct and ageing cohorts. Evidence from these investigations suggest that greater flow rates (>20,000 ML/d) are likely required to significantly influence golden perch spawning and recruitment in the LMR. During the MER Project, the recruitment of golden perch and silver perch in the LMR is investigated through age structure analysis to assess the impact of the flow regime (including environmental water) on populations. Data collected as part of the LTIM Project (Ye *et al.* 2020) is integrated in the reporting and evaluation for this indicator.

Hypotheses

- Increased spring–summer flow (nominally >20,000 ML/d), either in-channel or overbank, will promote the spawning and recruitment (to YOY) of golden perch and silver perch; and
- Multiple years of enhanced spring–summer flow will increase the resilience of golden perch and silver perch populations in the LMR by promoting a more diverse age structure.

Methods

Under the current core fish assemblage (Category 1) monitoring (Section 2.6), there is limited capacity to evaluate the effect of Commonwealth environmental water on the reproduction (spawning and recruitment) of golden perch and silver perch. Fundamental to this evaluation is knowledge of hydrological conditions at the time and place of spawning to explicitly relate spawning of flow-cued fishes to flow. For the purposes of this project, comprehensive evaluation of the reproduction (including spawning) of golden perch and silver perch in response to Commonwealth environmental water is contingent upon spring–summer flows exceeding 20,000 ML/d (see Section 5.3 of the Lower Murray MER Plan, SARDI *et al.* 2019).

In 2019-20, to evaluate golden perch and silver perch recruitment and its association with flow, including environmental water, we used electrofishing to: (1) assess temporal variability in the relative abundance of golden perch; and (2) collect a representative sub-sample of the golden perch and silver perch populations in the LMR to determine population age structure. Due to low numbers of silver perch collected during this project, we only present findings for golden perch in this report.

Sampling YOY and population age structure

From 2015–2020, adult and juvenile golden perch were sampled using a 7.5 kW Smith Root (Model GPP 7.5) boat electrofishing unit at approximately 16 sites in the LMR (Ye *et al.* 2020). Annual sampling was undertaken in April–May to complement Category 1 Fish Assemblage sampling and to maximise the likelihood of collecting YOY from the previous spring–summer spawning season.

Electrofishing was conducted during daylight hours and all available littoral habitats were surveyed. At each site, the total time during which electrical current was applied (on-time effort) ranged from approximately 676 to 2,880 seconds. All individuals were measured to the nearest mm (total length, TL) and a sub-sample of golden perch proportionally representing the length-frequency of golden perch collected was retained for ageing.

Abundance

Temporal variability in the relative abundance of golden perch was investigated by assessing changes in Category 1 electrofishing catch-per-unit-effort (CPUE) data (See Section 2.8, Fish Assemblage). Differences in the relative abundance (individuals per 90 seconds of electrofishing on-time effort) between years were analysed using univariate single-factor PERMANOVA (permutational ANOVA and MANOVA) in the software package PRIMER v. 6.1.12 and PERMANOVA+ (Anderson *et al.* 2008). These analyses were performed on untransformed, standardised relative abundance data.

Ageing

We used age-frequency distributions to assess the age structure and year-class strength of golden perch. Golden perch retained for ageing were euthanised and sagittal otoliths were removed. Whole otoliths were embedded in clear casting resin and a single 400 to 600 μm transverse section was prepared. Sections were examined using a dissecting microscope (x25) under transmitted light. Estimates of age were determined independently by three readers by counting the number of discernible opaque zones (annuli) from the primordium to the otolith edge. YOY (<1 year old) fish were defined as individuals lacking clearly discernible annuli.

Results

Abundance

Relative abundance of golden perch declined from 2015 (0.57 ± 0.08 ind./shot) to 2020 (0.27 ± 0.03 ind./shot, Figure 47). This was supported by PERMANOVA which demonstrated significant differences between years (Pseudo $F_{5,54}$ =5.6435, $P \leq 0.001$). Pair-wise comparisons revealed significant differences between 2015 and 2019, 2015 and 2020, 2016 and 2019, and 2016 and 2020, but not between any other years (Table D11 in Appendix D).

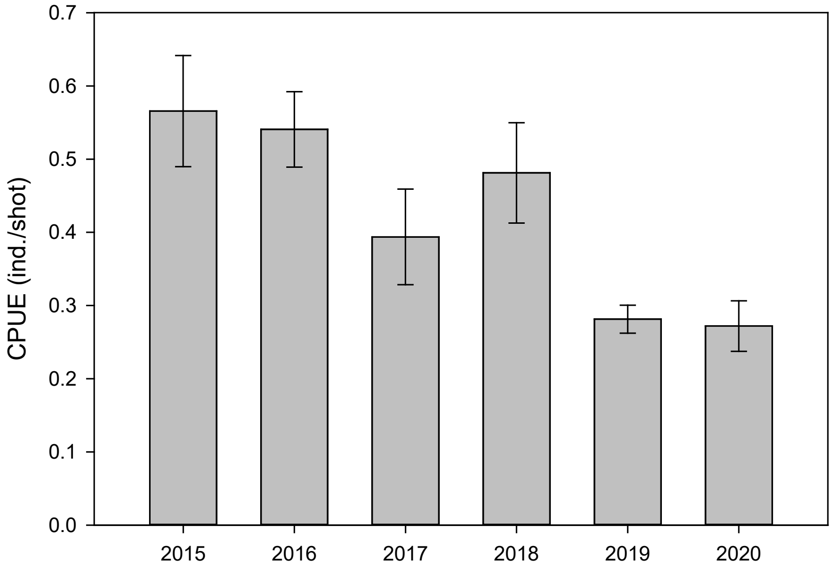

Figure 47. Mean catch-per-unit-effort (CPUE) ± standard error of golden perch captured during Category 1 Fish Assemblage electrofishing (individuals per 90 second shot) in the gorge geomorphic zone (10 sites) of the Lower Murray River in autumn from 2015–2020. CPUE data from five sites are presented for 2017 as other sites were sampled during winter 2017.

Age structure

From 2015–2020, YOY (age 0+) golden perch were absent from electrofishing samples, although in 2017, two individuals were collected in fyke nets. In 2015, the sampled population of golden perch ranged in age from 2+ to 18+ years, with dominant cohorts of age 4+ (30% of population), 5+ (35%), 14+ (9%) and 18+ (10%) representing fish born in 2010-11, 2009-10, 2000-01 and 1996-97, respectively (Figure 48). These four cohorts persisted in the population from 2015–2020, but their relative contribution varied. In 2020, golden perch collected in the LMR ranged in age from 1+ to 23+ years, with dominant cohorts of age 6+ to 10+ fish, born in 2013-14 to 2009-10, collectively comprising 87% of the sampled population.

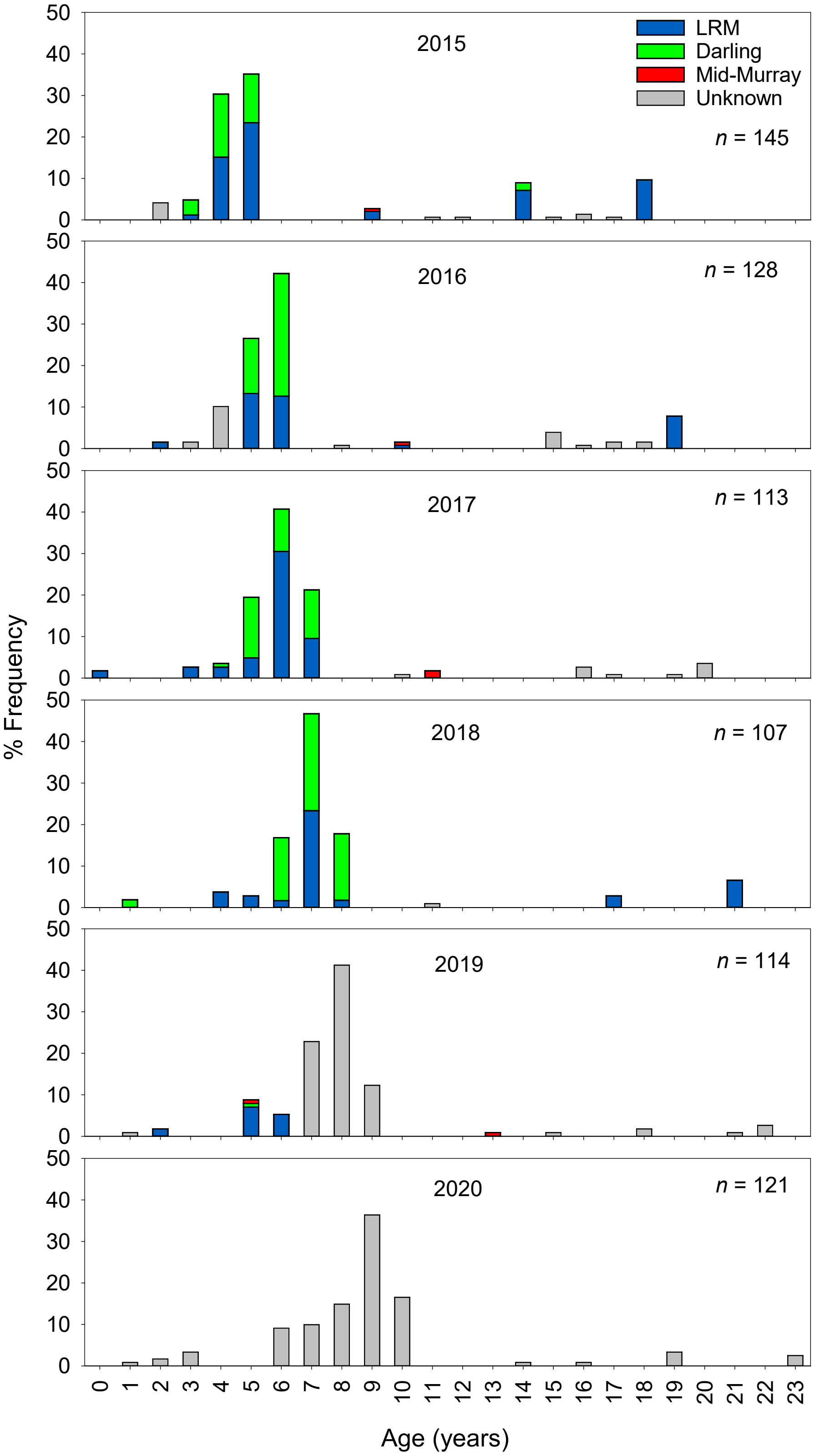

Figure 48. Age frequency distribution of golden perch from the Lower Murray River from 2015–2020, also showing the natal origins of dominant cohorts inferred from otolith core $^{87}Sr/^{86}Sr$ signatures of the sampled fish in comparison to the water sample reference collection (Ye *et al.* 2020). LRM = Murray River, below the Darling confluence. Percentage of origin for each cohort are based on the sub-sampled population. Age cohorts with grey bars were not assessed for natal origin.

Evaluation

The evaluation approach, including assessment criteria, is described in the evaluation section for Hydraulic Regime (Section 2.1).

Table 19. Flow-cued Spawning Fish Recruitment evaluation questions and answers. YOY = young-of-year.

CEWO evaluation questions	**Outcomes of flow delivery**	
	2014-15 – 2018-19	**2019-20**
Did the flow regime (including environmental water) contribute to recruitment of golden perch and silver perch?	Negligible recruitment	In the LMR during 2019-20, the flow regime (including environmental water) contributed to negligible recruitment of golden perch and silver perch (to YOY, age 0+).
Did the flow regime (including environmental water) contribute to the resilience of golden perch and silver perch populations?	From 2014-15 to 2019-20, the flow regime (including environmental water) in the LMR did not promote resilience in golden perch or silver perch populations.	

Contribution (to what extent CEW contributed towards the outcome, with the significance of the outcome considered):
⬜ Unknown 🟥 Negative ⬜ None/negligible 🟩 Minor 🟩 Moderate 🟦 Substantial

Discussion

Over the LTIM and MER projects, we aimed to identify potential associations between recruitment of golden perch and silver perch and environmental water delivery. We hypothesised that (1) increased flow (nominally >20,000 ML/d) in spring–summer would promote the spawning and recruitment to YOY, and (2) multiple years of enhanced spring–summer flow would increase the resilience of golden perch and silver perch populations in the LMR by promoting a diverse age structure.

From 2015–2020, golden perch recruitment in the LMR was poor, with no strong age classes added to the population. In 2015, the sampled population of golden perch ranged in age from 2+ to 18+ years, with dominant year classes from 2010-11, 2009-10, 2000-01 and 1996-97. These cohorts persisted in the population to 2020, but the relative proportions of older age cohorts (e.g. 2009-10, 2000-01 and 1996-97) declined. In association with an ageing population, abundance of golden perch in the LMR steadily declined by almost half from 2015 to 2020. Over the same period, declines in golden perch abundance were also observed in other monitoring programs in the region, for example, in TLM *condition monitoring* at the Chowilla icon site (Fredberg *et al*. 2019). Potential factors contributing to decline are a lack of recruitment, mortality (fishing and natural) and upstream emigration of adults (Zampatti *et al*. 2018). Ultimately, a lack of younger cohorts, reduced abundance and diminished age structure diversity (e.g. population dominated by few cohorts) lead to a population that lacks resilience to environmental perturbations and other impacts (e.g. fishing).

Management implications

From 2014-15–2019-20, >500 GL/year of Commonwealth environmental water was delivered to the LMR for a range of ecological objectives, including increasing spawning and recruitment of flow-dependent species. These deliveries generally increased discharge at the South Australian border from base (entitlement) flows (~3,000–7,000 ML/d) to small freshes (~12,000–17,800 ML/d) during spring/summer. Nonetheless, the delivery of these flow pulses commonly involved re-regulation by Lake Victoria, which may have impacted the longitudinal integrity of flow. Over the past six years golden perch recruitment was negligible. Evidence from the current investigation and allied studies suggest that greater flow (>20,000 ML/d) is likely required to significantly influence local golden perch spawning and recruitment in the LMR. Annual spring–summer in-channel flow pulses of this magnitude were a key feature of the hydrograph of the LMR prior to regulation (Mallen-Cooper and Zampatti 2018), and would have occurred in recent years under natural, unregulated conditions.

Ultimately, fish behaviour and life history processes are influenced by the hydraulic characteristics of flow (i.e. hydrodynamics), rather than volumes of water and rates of discharge (hydrology). Riverine hydrodynamics likely provide cues for movement and spawning and facilitate the downstream dispersal of early life stages. Contemporary flow management in the MDB mainly focuses on volume and discharge, whereas a hydraulic perspective is more relevant to understanding ecological processes and eliciting ecological outcomes (Bice *et al*. 2017; Mallen-Cooper and Zampatti 2018). This is particularly relevant in the LMR, where contiguous weir pools result in a highly modified hydraulic environment. Indeed, the river only begins to substantially regain its lotic character at flows >20,000 ML/d (Bice *et al*. 2017). Decreasing weir height reinstates lotic characteristics at lower discharges and, in conjunction with restoration of spring pulses, may improve the recruitment and population structure of riverine fishes such as golden perch. However, a key knowledge gap remains – the role that water velocity and turbulence plays in the suspension, transport and retention of golden perch eggs and larvae, which is a critical process during early life history of this species.

Conclusion

There has been no substantial recruitment of golden perch in the LMR since 2013-14, leading to a population dominated by a few distinct and ageing cohorts. To improve the resilience of golden perch populations in the LMR, it would be pertinent in the coming years to provide flows that may facilitate golden perch spawning and recruitment. Specifically, Commonwealth environmental water could contribute to spring/early summer in-channel flow >20,000 ML/d in the LMR or spawning, recruitment and subsequent downstream dispersal from upstream reaches (e.g. the lower Darling and mid-Murray rivers).

2.7 Murray Cod Recruitment

Background

Murray cod has great recreational and cultural significance in the MDB. Nonetheless, populations have declined as a result of human impacts including alteration of flow regimes, barriers to movement, overharvesting and habitat (hydraulic and physical) degradation (Lintermans and Phillips 2005), and the species is currently considered vulnerable under the Australian EPBC Act (1999). In the Murray River downstream of the Darling River junction, the fragmentation of the river by sequential weirs, alteration to hydraulics and loss of lotic habitats are considered primary threats to the persistence of Murray cod populations (Zampatti *et al.* 2014; Mallen-Cooper and Zampatti 2018). Indeed, under low flows, lotic environments that provide favourable juvenile and adult habitat, and support key life history processes (spawning and recruitment) are restricted to select anabranch systems (e.g. Chowilla) and lock and weir tailwaters. During elevated flows (e.g. >20,000 ML/d), however, lotic conditions are returned to considerable reaches of the Lower Murray (Bice *et al.* 2017).

Murray cod spawn annually over a well-defined period from October–December, irrespective of flow (Davis 1977; Rowland 1998), but recruitment in the LMR main channel is positively associated with flow (Ye and Zampatti 2007; Zampatti *et al.* 2014). Indeed, from 2003–2010 during the Millennium Drought, discharge in the LMR was predominantly <10,000 ML/d, and Murray cod recruitment, measured as abundances of YOY in autumn, was limited in the predominantly lentic main channel. Subsequently, recruitment was observed in association with spawning that occurred in high flow years from 2010–2013 (Zampatti *et al.* 2014). In recent years (2015–2019), annual recruitment of Murray cod occurred in the LMR main channel, following flow conditions including an in-channel flow pulse (15,000–18,000 ML/d) and an overbank flow (>90,000 ML/d) (Ye *et al.* 2016a; 2017; 2018; 2019; 2020), but also during three years of modest in-channel flows (<12,000 ML/d). Furthermore, these recent cohorts have generally persisted in the population (Ye *et al.* 2019).

The mechanisms that facilitate recruitment of Murray cod (to YOY) in the Lower Murray likely relate to enhanced spawning habitat and survival of early life stages associated with improved riverine hydraulics and productivity. Survival is likely mediated by enhanced habitat availability, and growth rates and condition, and ultimately determines recruitment and population abundance. This indicator will explore these mechanisms, together with a related research project (see SARDI *et al.* 2019), by assessing aspects of Murray cod recruitment (e.g. abundance, growth, condition) in association with flow. Understanding the magnitude of recruitment, and causal links between recruitment and flow, is critical for informing future environmental flow management and will help evaluate ecological outcomes of Commonwealth environmental water.

Major hypotheses

- Elevated spring–summer flow, either in-channel or overbank, and associated increase in lotic habitat, will enhance recruitment (to YOY) of Murray cod;
- Elevated spring–summer flow, either in-channel or overbank, and associated increase in lotic habitat, will be associated with enhanced growth rates and morphometric condition of Murray cod; and

- Multiple years of enhanced spring–summer flow will result in broad size/age distributions of Murray cod and increased population resilience in the Lower Murray.

Methods

To evaluate the contribution of flow, including Commonwealth environmental water, to the morphometric condition, growth and recruitment of Murray cod in the LMR during 2019-20, we used: (1) length–weight relationships and associated metrics to assess body condition; (2) otolith microstructure analysis (including daily otolith growth increments) to determine the time of spawning, along with seasonal and daily growth rates; and (3) electrofishing to quantify the abundance of YOY to assess recruitment, and to establish a length frequency distribution of the Murray cod population in the LMR to determine population structure. The data used and presented in this indicator section were collected from a number of past projects including: CEWO Short-Term Intervention Monitoring (STIM) and LTIM projects; TLM Chowilla Fish Condition and Intervention Monitoring; Goyder Ecological Connectivity of the River Murray project; and MDB Fish Surveys.

Sampling

Murray cod were collected from the main channel of the Lower Murray at several stages during early ontogeny, from larvae through to YOY (>120 d of age) (Table 20). Sampling for larvae (<30 mm) in November 2020 comprised a combination of passive (light traps and drift nets) and active (bongo net tows) methods. Sampling effort varied across sites, but included setting approximately 20–30 quatrefoil light traps (225 x 225 x 255 mm, 5 mm mesh) and 2–4 drift nets (1.5 m long x 0.5 m diameter, 500 µm mesh) each night over 2–3 nights (set time of 14–18 hours). Larval tows were performed opportunistically during day and night with a paired bongo net (0.5 x 0.5 m x 3 m long, 500 µm mesh) (Ye *et al.* 2018). Larvae were preserved (70–95% ethanol) in the field and returned to the laboratory for processing.

Electrofishing was used specifically to collect juveniles (~30–150 mm) and sample the broader population (Table 20). All sampling was performed using a 7.5 kW Smith Root (Model GPP 7.5) boat electrofishing unit during daylight hours. Sampling in January and February 2020 was targeted towards preferred habitat of early juveniles (e.g. root complexes of snags and rock crevices). Two complementary types of sampling were used to assess YOY abundance and population structure: 1) autumn (March/April) non-targeted Fish (Channel) electrofishing (see Section 2.8 Fish Assemblage) and 2) autumn (May) habitat-specific (targeted) electrofishing. Electrofishing during May was performed in a consistent manner, and is thus comparable, with previous sampling in LTIM (2015–2019) for Category 3 golden perch YOY sampling (see Ye *et al.* 2020) and was targeted towards a wider range of Murray cod habitats, including snags in flowing habitat extending beyond the river bank (preferred by sub-adults and adults). At each site, electrofishing 'on-time' effort was variable, and depended on the available habitat. All individuals were measured for TL (nearest mm) and weight (to 0.5 kg), and a sub-sample of juvenile (<160 mm) fish were retained for morphometric and otolith analyses, as well as diet and tissue analyses for the allied research project (not presented in this report).

Table 20. Sampling trips targeting Murray cod in the Lower Murray River during 2019-20.

Date range	Methods	Indicator
5–18/11/19	Light traps, drift nets, larval tows	Growth, condition
6–9/1/20	Light traps, electrofishing	Growth, condition
24–27/2/20	Electrofishing	Growth, condition
30/3–8/4/20*	Electrofishing	Condition, YOY abundance, population structure
7–8/5/20	Electrofishing	Condition, YOY abundance, population structure

* Fish (Channel) sampling in the Gorge zone (see Section 2.8 Fish Assemblage for details)

Sampling for larvae (November) and early juveniles (January and February) was conducted in the tailwaters (<15 km downstream) of Lock 3 (Gorge zone) and Lock 4 (Floodplain zone) (Figure 1). Sampling to assess YOY abundance and population structure in May occurred at sites in the tailwaters of Locks 3 (Gorge zone), 4, 5 (Floodplain zone), and was supplemented by non-targeted sampling in March/April at ten sites between Locks 1 and 3 in the Gorge zone (Fish (Channel) sampling). In addition to the sampling described above, Murray cod larvae and juveniles were opportunistically collected through other field sampling (e.g. Section 2.6 Flow-cued Spawning Fish Recruitment) and included in growth and condition analyses.

<u>*Otolith preparation and increment counts and measurements*</u>

Larval/juvenile Murray cod were measured for TL to the nearest millimetre, weighed to 0.001 of a gram, and sagittal otoliths were removed. Sagittae were embedded in crystal bond™, then ground and polished from the anterior margin towards the core. For larvae and early juveniles (from January), we used 9 and 3 µm lapping film, while for otoliths of larger juveniles from February, used 240-grit sandpaper and 15, 9 and 3 µm lapping film sequentially. The ground surface was adhered to the centre of a microscope slide with crystal bond™ and then further ground and polished from the posterior side, to produce sections of approximately 50 µm thickness.

Prominent hatch marks were evident on most sectioned otoliths, providing a reliable reference point to begin increment counts. As such, daily increment counts were made from the hatch mark along the maximum growth axis towards the ventral apex. Two readers examined each otolith on separate occasions and each reader performed two counts of the increments. Counts from each reader were compared and if they differed by more than 10%, the otolith was rejected; but if count variation was within 10%, the mean of all counts was accepted as the best estimate of daily increment number. Hatch dates were determined by subtracting estimated age from capture dates. Prior to increment counts, otolith readers were calibrated by reading a reference collection of otoliths from known-age Murray cod larvae and achieving 90% agreement. Otolith sections were examined using a compound microscope (x 600) fitted with a digital camera and Optimas image analysis software (version 6.5, Media Cybernetics, Maryland, USA).

The Individual widths (µm) of the otolith increments (10–60 increments) in a single-plane focus were measured along the widest radius on the distal section of the otolith using the image analysis software Stream (Olympus) (see Bice *et al.* 2016). YOY Murray cod collected in May were omitted from age analyses due to unreliability in daily increment estimates.

<u>*Data analyses*</u>

Morphometric condition

The length–weight relationships for juvenile (0+ year old, <160 mm) Murray cod were described using linear regression, following the methods outlined in Tonkin *et al.* (2008). Linear and non-linear relationships were examined with non-linearity modelled by fitting polynomial terms of increasing order (up to 4) to length. The relative support for each of the models was assessed using Akaike's Information Criterion, corrected for small sample size (AICc) (Burnham and Anderson 2002). AICc values were rescaled as differences between the model and the model with the lowest AICc value, with the likelihood of the model calculated (see Tonkin *et al.* 2008 for equation). Morphometric condition was estimated by calculating the relative condition factor (K*rel*) of an individual fish, using the estimated weight calculated from the selected model equation: $K{rel} = Wt_{actual}/Wt_{estimated}$. To determine if the condition of Murray cod differed among different weir pools and years (i.e. 2015–2020) of different flow regimes, differences in K*rel* of individuals sampled between March and August were assessed using a two-factor univariate PERMANOVA. A significance value of $\alpha = 0.05$ was adopted for all statistical comparisons, acknowledging an increased likelihood of type 1 errors for multiple comparisons.

Growth rate

Mean seasonal growth was described by fitting linear and nonlinear models to length-at-age data. Three models were considered to model the fish growth rate: linear, Gompertz and Von Bertalanffy. Model selection procedures using AICc, described above, were used to select the model with most support. Mean seasonal growth rate was calculated as the function of TL and age (days) at capture. To determine if the seasonal growth rates of Murray cod differed among different weir pools, differences in mean seasonal growth rate were assessed using a single-factor univariate PERMANOVA.

Measurements of daily increment widths were undertaken to assess daily growth of larval and juvenile Murray cod, following methods described in Bice *et al.* (2016). For each individual, each increment was assigned to a 'growth day' (Julian day), using a back calculated hatching date.

Recruitment

To compare recruitment of Murray cod across years, temporal variability in the relative abundance of YOY Murray cod (i.e. <160 mm TL) was assessed using two datasets: 1) March/April non-targeted Category 1 electrofishing CPUE data (see Section 2.8 Fish Assemblage); and 2) May targeted electrofishing CPUE data. For the first dataset, sites were grouped and represented as two reaches, namely downstream of Lock 3 and downstream of Lock 2. Differences in relative abundance (individuals per minute of electrofishing on-time effort) among years at each reach were analysed using univariate single-factor PERMANOVA (permutational ANOVA and MANOVA) in the software package PRIMER v. 6.1.12 and PERMANOVA+ (Anderson *et al.* 2008). These analyses were performed on untransformed, standardised relative abundance data. Comparisons of CPUE between weir pools were not explored as they were not of interest in this study.

Results

Catch summary

During sampling in the LMR from 5 November 2019 to 8 May 2020, a total of 173 Murray cod were collected. Catch details in tailwater sites that were consistently sampled throughout the spring–autumn period are provided in Table 21. The majority of likely age 0+ juvenile Murray cod (*n* = 147) were retained for morphometric, age and growth assessments.

Table 21. Catch summary of Murray cod in the tailwater (<15 km) below Lock 3 and 4 in 2019-20.

Date range		**Lock 3**			**Lock 4**	
	n***	**TL (mm)**	**Age (days)**	***n******	**TL**	**Age (days)**
5–18/11/20	12	10–11	10–14	20	10–21	11–14
6–9/1/20	3	28–43	60–73	28	23–63	57–86
24–27/2/20	7	53–96	79–116	30	60–104	103–127
30/3– 8/4/20	8	54–113	136–145			
7–8/5/20	4	95–118		13	76–123	

*Sample numbers for otolith analyses are less than what is shown and are presented below in Figure 50. A total of 48 individuals (not presented here) were also captured collectively from below Lock 4 (*n* = 1) in December 2019, and below Locks 2 (*n* = 6), 3 (*n* = 40) and 5 (*n* = 1) from March–May 2020.

Length and hatch dates

Murray cod larvae and age 0+ juveniles sampled from the LMR ranged in TL from 10–124 mm. From November 2019 to May 2020, length frequency distributions indicated progression in length between sampling events (Figure 49). Estimated ages during November, January, February and April ranged from 10–14, 57–86, 79–127 and 136–152 days, respectively (Table 21), and corresponded to hatch dates from 13 October to 9 December 2019. Assuming an incubation period of 7 days (Ingram *et al.* 2012), peak spawning is estimated to have occurred from 17 October to 11 November 2019, with spawn date frequency distributions influenced by the timing of larval sampling (5–7 and 13 November 2019, Figure 50a).

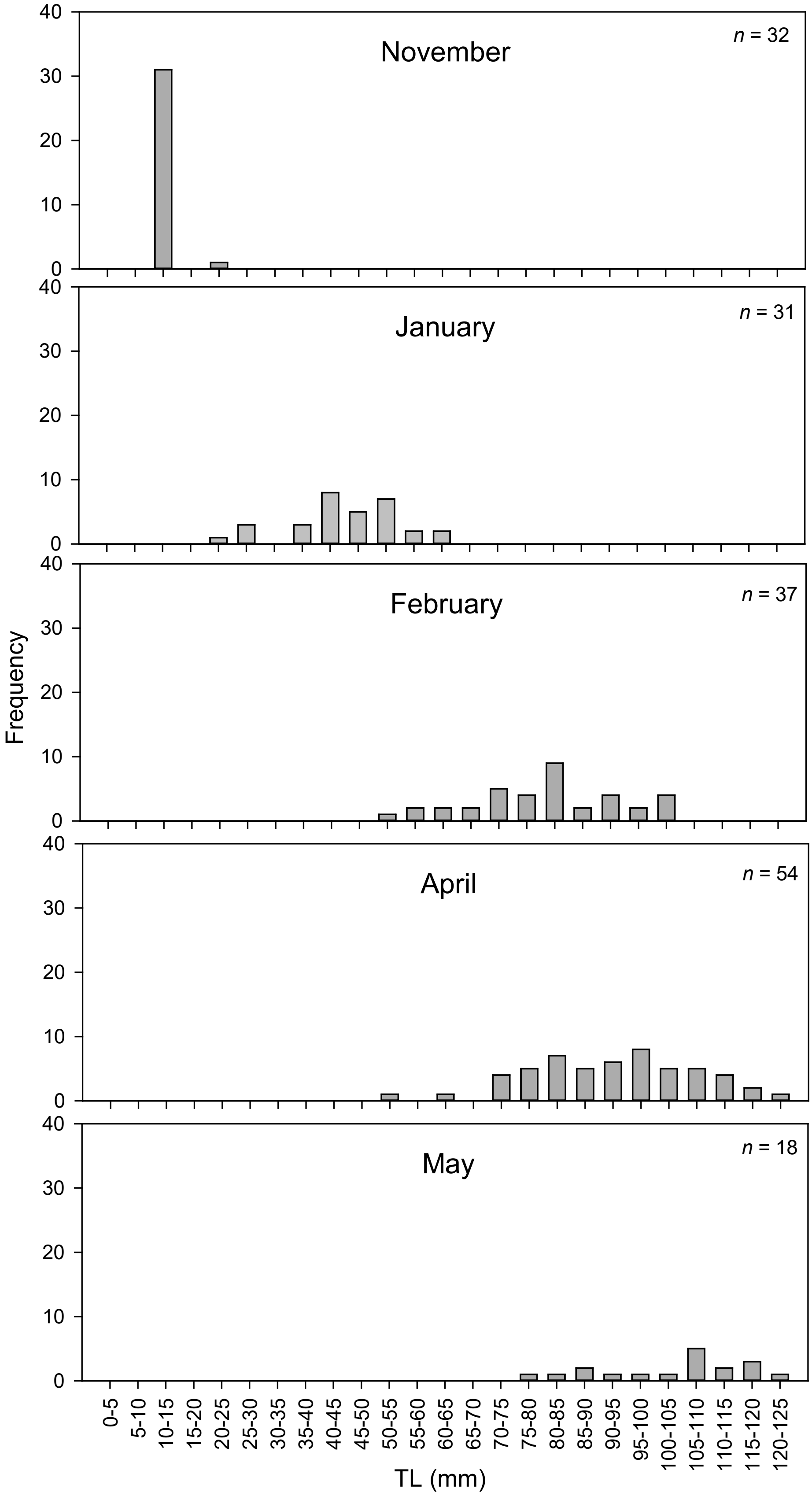

Figure 49. Length frequency distributions of larval/juvenile Murray cod collected in the Lower Murray River (all sites pooled) during the five sampling trips in 2019-20.

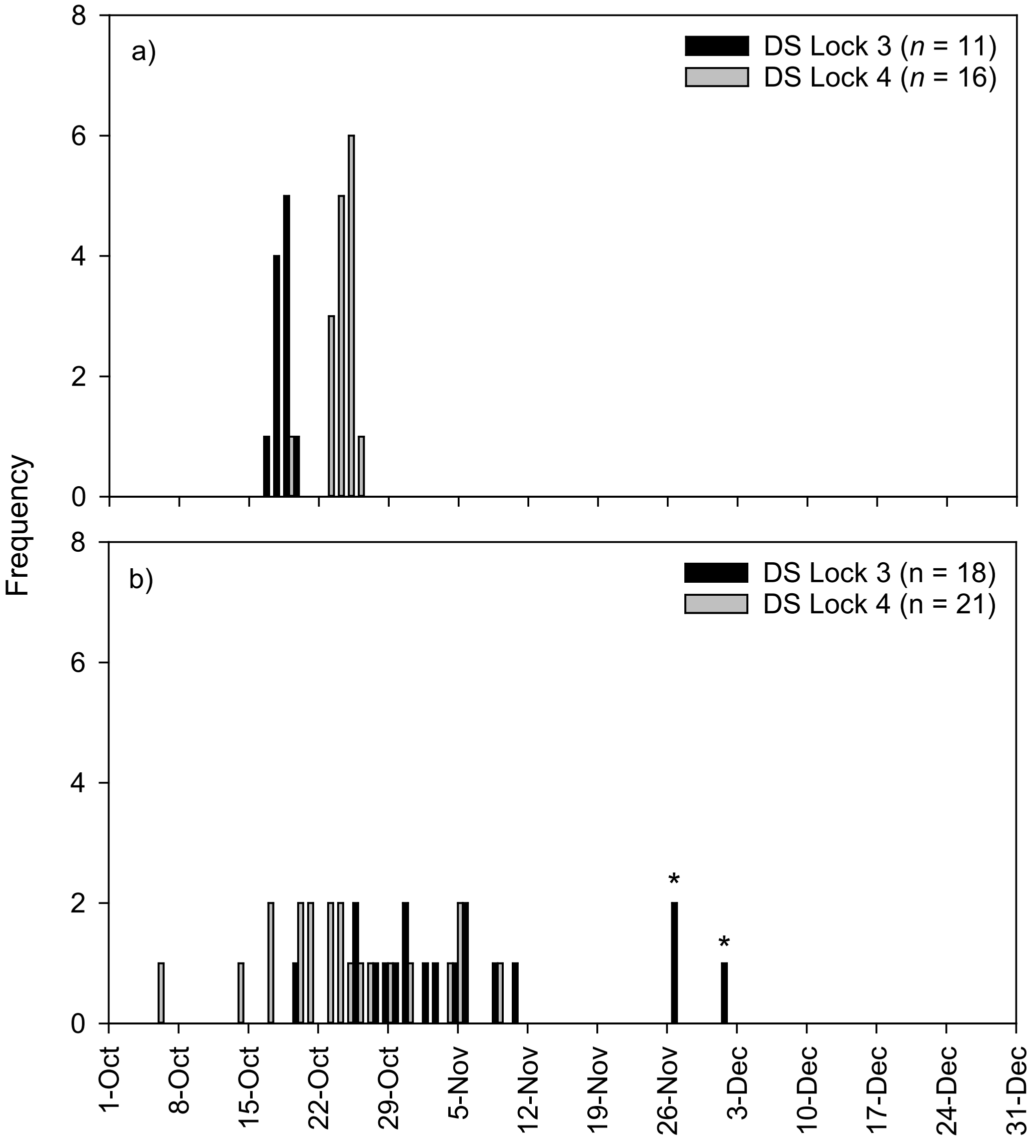

Figure 50. Estimated spawn date frequency distributions of (a) larval Murray cod captured in November 2019 and (b) juvenile Murray cod captured from January to April 2020, in the Lower Murray River (LMR). *late November/early December spawn dates correspond with spawn dates of known stocked fish in the LMR from February 2020.

Morphometrics and seasonal growth

Using the model selection procedure, a cubic polynomial of TL was applied to model length–weight relationships for larval/juvenile (<160 mm) Murray cod for all years (baseline data, 2004–2020), and for 2019-20 (Figure 51). Comparison of the 2019-20 length–weight relationship to the baseline curve indicates that the condition of Murray cod during 2019-20 was similar to or slightly below the ‘average’ condition. To further investigate this, differences in the relative condition index (*Krel*) of YOY (sampled autumn/winter) Murray cod among weir pools and sampling years (2015 to 2020) (Figure 52) were assessed using PERMANOVA. There was a significant effect of sampling year on *Krel* (PERMANOVA, $Pseudo\text{-}F_{5,96} = 2.283$, $p = 0.047$), but not of weir pools (PERMANOVA, $Pseudo\text{-}F_{5,96} = 1.3552$, $p = 0.251$) or the interaction between sampling year and weir pools (PERMANOVA, $Pseudo\text{-}F_{11,96} = 1.399$, $p = 0.192$). This suggests morphometric condition was significantly different across 2015–2020, and temporal differences were consistent among weir pools. Pairwise comparisons revealed that *Krel* of YOY was significantly greater in 2017 than all other years, and significantly greater in 2019 compared to 2018 (Figure 52).

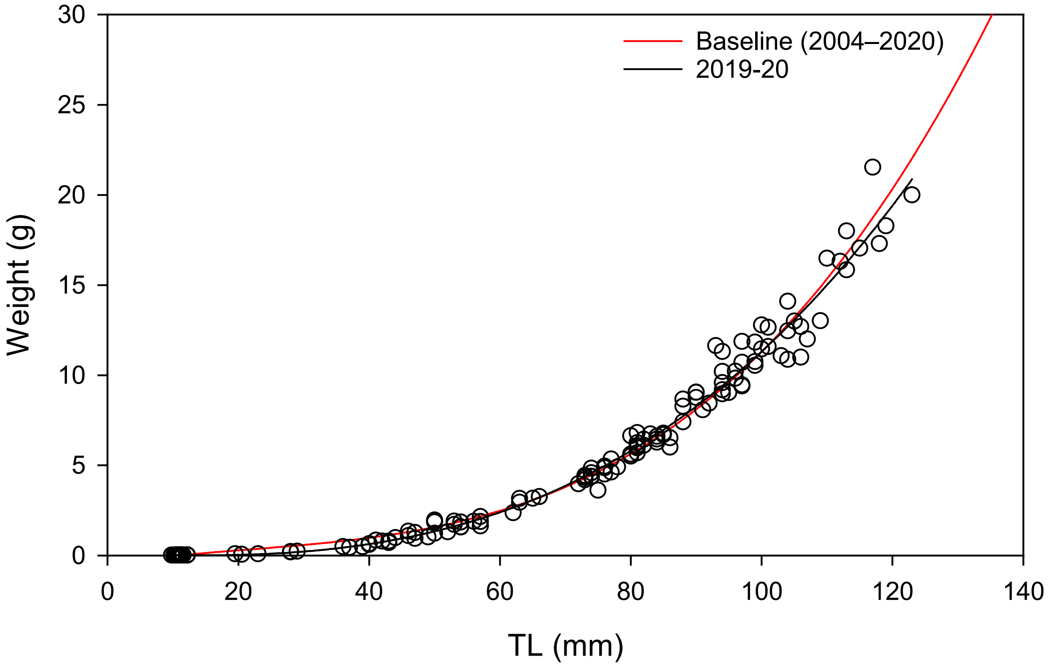

Figure 51. The relationship between weight and total length (TL) of YOY (<160 mm) Murray cod in the Lower Murray River predicted by a cubic linear regression model ($y=-0.4588+0.0527x-0.011x^2+0.000017791x^3$) for 2019-20 (black line and circles, *n* = 154) and all years (2004–2020, 'stocked' fish removed, red line). See Table D12 in Appendix D for statistics.

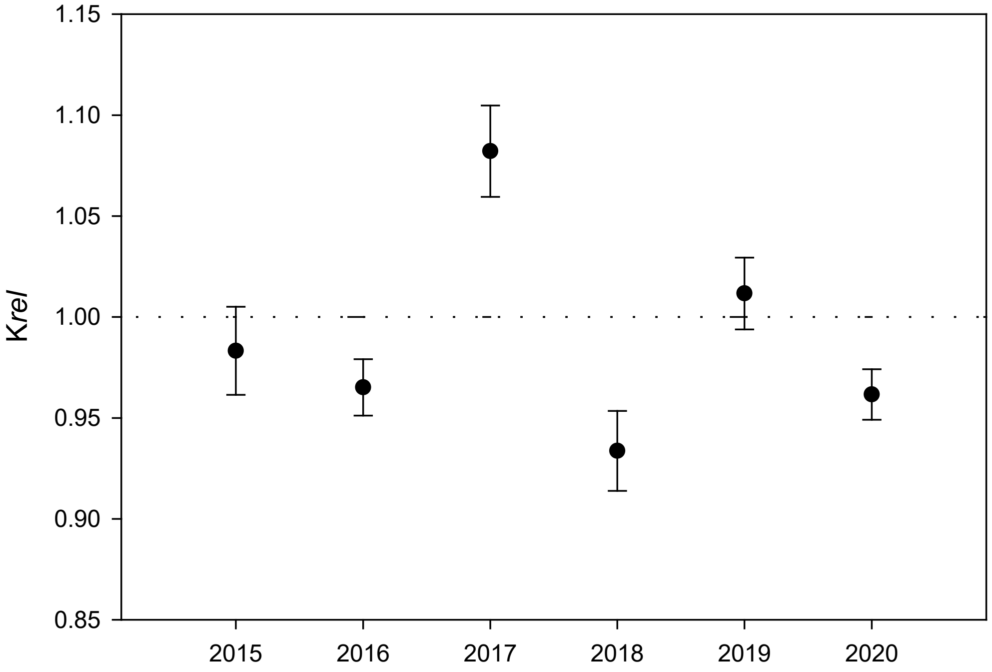

Figure 52. Mean relative condition index (K*rel*) ± S.E. of YOY (<160 mm) Murray cod in the Lower Murray River during autumn/winter 2015 (*n* = 11), 2016 (*n* = 14), 2017 (*n* = 6), 2018 (*n* = 11), 2019 (*n* = 25), 2020 (*n* = 51). See Table D13 in Appendix D for statistics.

Model selection indicated that the Gompertz model was the best fit for the 2019-20 Murray cod age–length data (Figure 53). Relationships between length and age for Murray cod collected from each weir pool in 2019-20 were similar. Mean seasonal growth rate in the weir pool below Lock 4 (0.791 mm/d ± 0.023 S.E.) was slightly higher than below Lock 3 (0.769 ± 0.029), yet this difference was not significant (PERMANOVA, $Pseudo\text{-}F_{1,\,59} = 1.249$, $p = 0.260$).

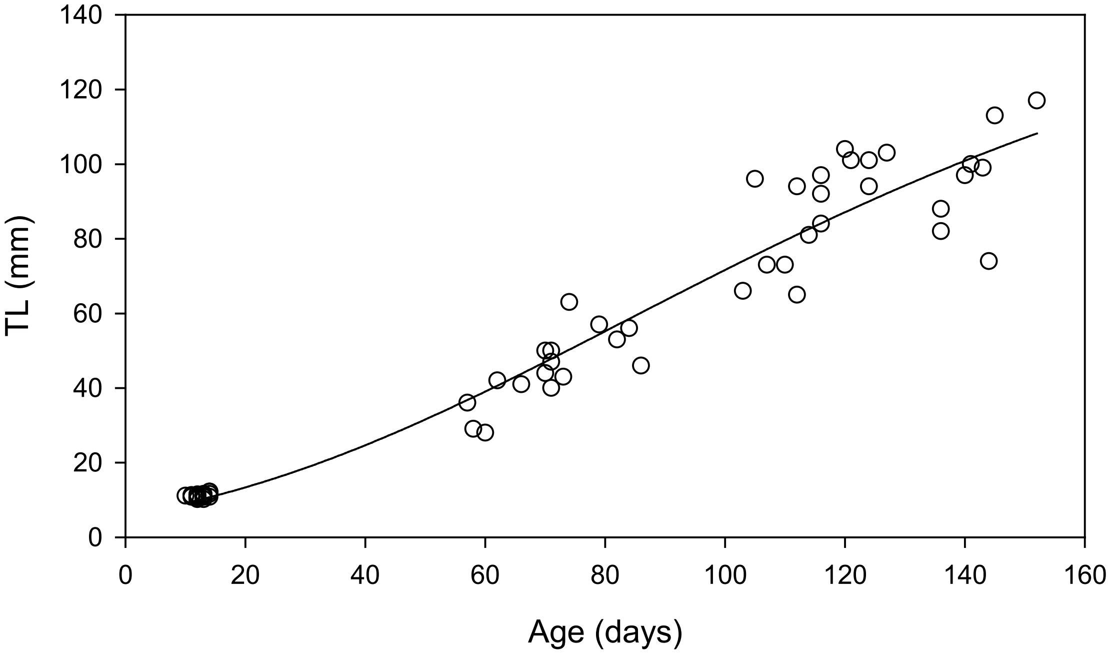

Figure 53. The relationship between total length (TL) and estimated age of YOY (<160 mm) Murray cod in the Lower Murray River during 2019-20, predicted by the Gompertz model. See Table D14 in Appendix D for statistics.

Population structure and recruitment

Mean relative abundances (non-targeted CPUE) of Murray cod in the weir pools below Locks 2 and 3 remained relatively consistent from 2015 to 2019 (<0.05 fish/min). In 2020, there was a four-fold increase in mean abundance below Lock 3, driven by the abundance of YOY fish (Figure 54 and Figure 55). Due to the large variability associated with this increase, however, there was no significant effect of year on CPUE (PERMANOVA, $Pseudo\text{-}F_{5,\,48} = 2.229$, $p = 0.066$). The proportional abundance of YOY from these sites throughout the six-year period was relatively high (>50%).

Relative abundances (targeted CPUE) of Murray cod in the tailwaters of Locks 4 and 5 varied among years (PERMANOVA, $Pseudo\text{-}F_{6,\,36} = 3.973$, $p = 0.006$) and showed a general increasing trend in abundance from 2013 to 2016, before a decline in abundance in 2017. This was followed by a significant increase in relative abundances to a peak in 2020 (PERMANOVA, $t = 3.139$, $p = 0.011$). Consistent with the non-targeted CPUE data from below Lock 3, this increase in relative abundance below Lock 4 was driven by an increase in abundance of YOY (Figure 54 and Figure 55).

Excluding years (2013 and 2014) with low sample sizes (*n* < 10), Murray cod generally exhibited broad length frequency distributions, comprised of juveniles (including YOY), sub-adults (300–600 mm) and adults (>600 mm) (Figure 55). YOY fish (i.e. <150 mm) dominated the sampled population during 2019 (69 %) and 2020 (67 %). Furthermore, from 2015, new (YOY) cohorts can be seen to persist and progress through the population each year to adult size (>600 mm).

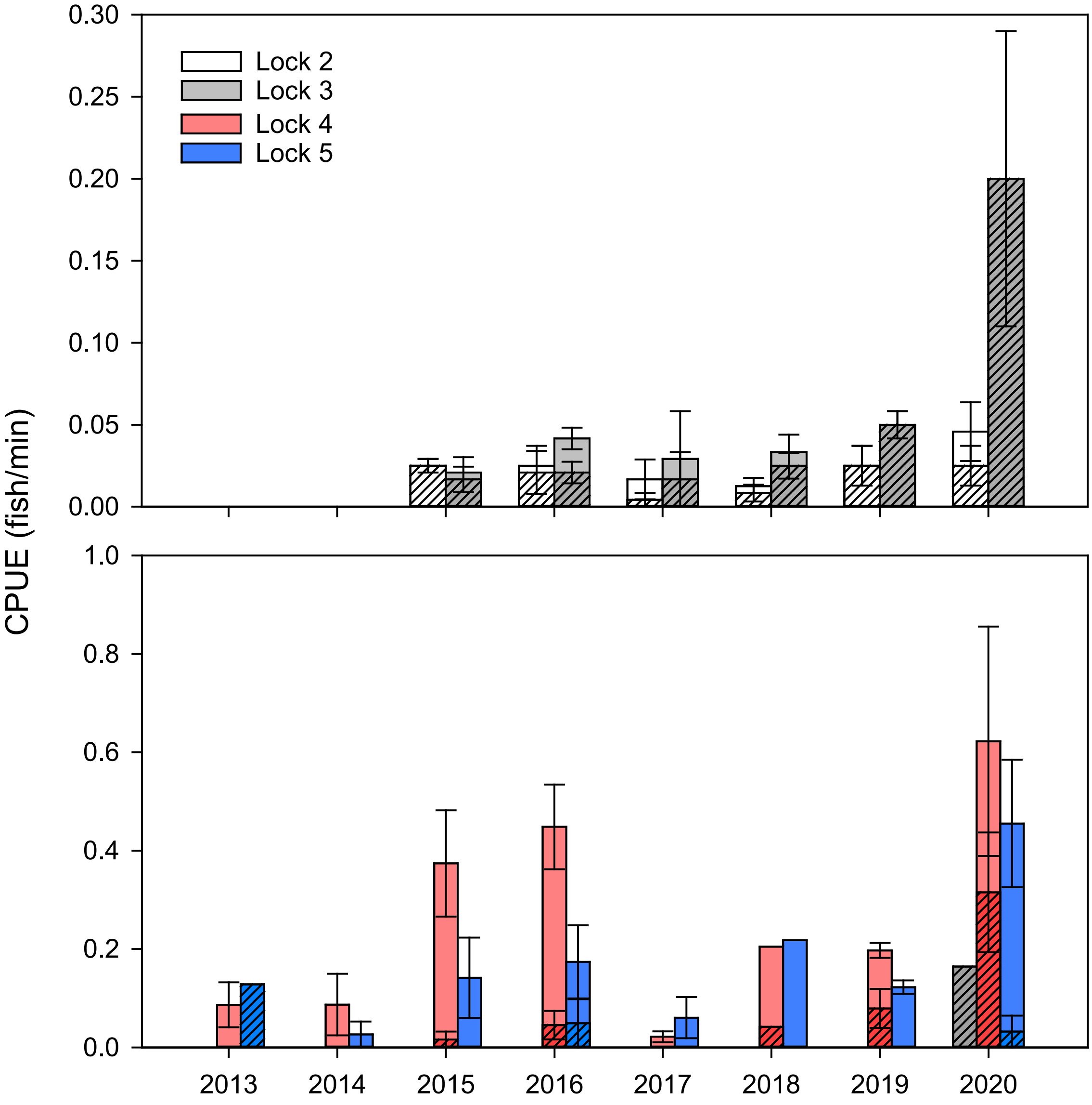

Figure 54. Mean non-targeted (top) and targeted (bottom) electrofishing catch-per-unit-effort (CPUE) ± S.E. of Murray cod in the weir pools downstream of Locks 2, 3, 4 and 5 of the Lower Murray River. Dashed bars represent the proportion of YOY to the total CPUE. Note the different scales on the CPUE-axis for the different methods of electrofishing. See Tables D15 and D16 in Appendix D for statistics.

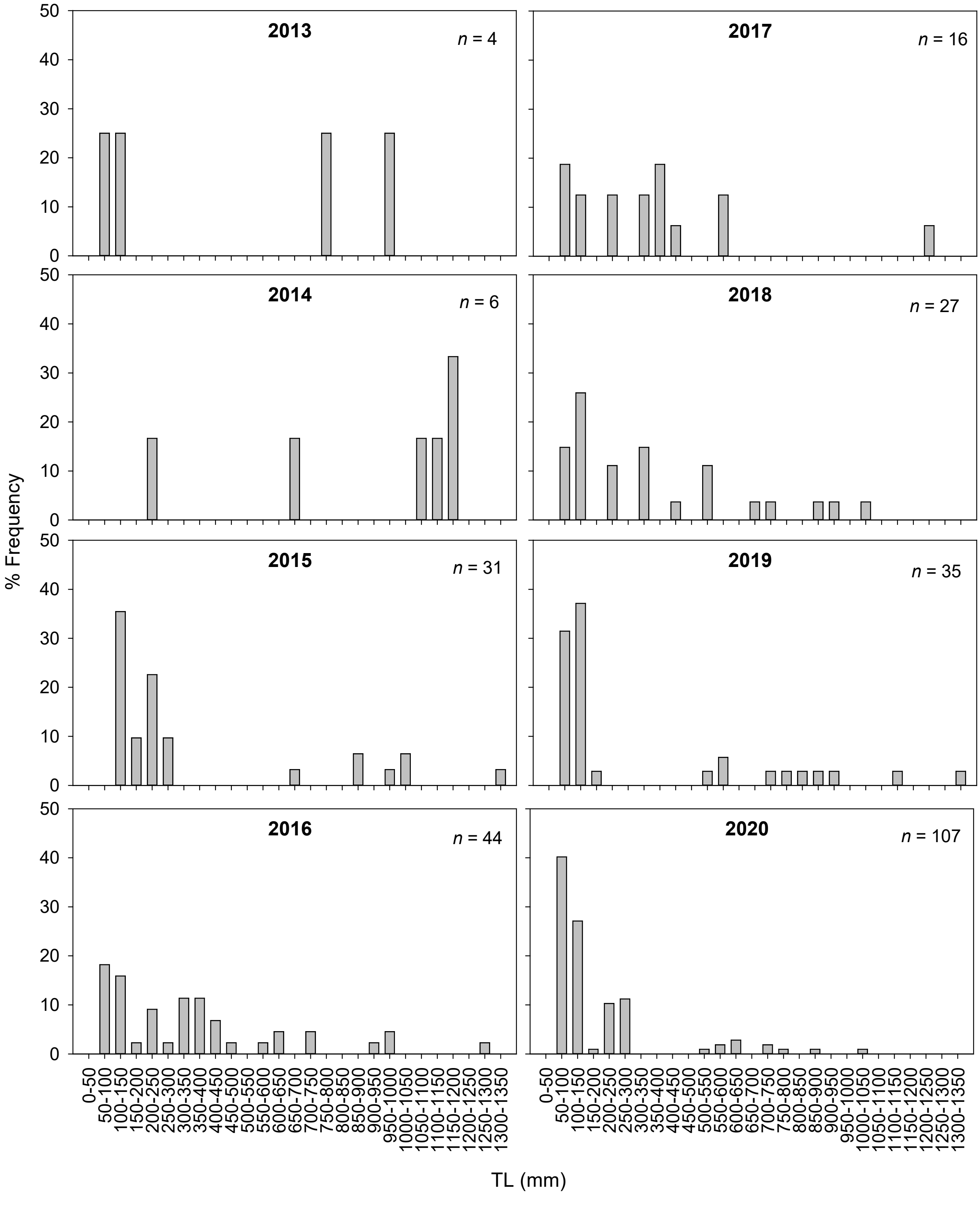

Figure 55. Total length (TL) frequencies of Murray cod in the main channel of the Lower Murray River during autumn/winter from 2013–2020.

Evaluation

The evaluation approach, including assessment criteria, is described in the evaluation section for Hydraulic Regime (Section 2.1). The assessment for Murray cod recruitment in 2019-20 took a conservative approach and is likely to vary in future based on further analysis as additional annual data are added and findings from the allied research project are integrated.

Table 22. Murray Cod Recruitment evaluation questions and answers. CEW = Commonwealth environmental water, eWater = environmental water.

CEWO evaluation questions	Outcomes of CEW delivery (2019-20)
What did CEW contribute to the growth and morphometric condition of Murray cod?	The influence of CEW on the growth and condition of Murray cod in 2019-20 is unknown. Increased flows (including CEW) during the spawning/early larval period, however, may have improved food resources (e.g. microinvertebrates) that in part supported survival of larvae. Nonetheless, CEW likely had negligible or minor influence on later growth as there was minimal CEW delivery post-November 2020.
What did CEW contribute to recruitment of Murray cod?	The influence of CEW on the recruitment of Murray cod in 2019-20 remains unquantified. Nevertheless, increased flows (including CEW) during the spawning/early larval period may have benefited Murray cod by increasing the extent and duration of lotic habitat, potentially enhancing spawning habitat area and survival of early life stages.
What did CEW contribute to the resilience of Murray cod populations?	N/A*

*The influence of CEW on the resilience of Murray cod populations will be assessed based on its contribution to length/age structure in future years.

Contribution (to what extent CEW contributed towards the outcome, with the significance of the outcome considered):

⬜ Unknown 🟥 Negative ⬜ None/negligible 🟩 Minor 🟩 Moderate 🟦 Substantial

Discussion

Over the course of the MER Project (2019-20 to 2021-22), we aim to identify associations between hydrology and hydraulics, and the recruitment of Murray cod in the LMR, and ultimately to evaluate the benefit of environmental water delivery. Flow may influence the spawning and early life history of Murray cod in the MDB via several mechanisms, but responses to flow likely differ among river systems (Tonkin *et al.* 2018). For the LMR, we hypothesise that elevated spring–summer flow, either in-channel or overbank, and an accompanying increase in lotic habitat, may: (1) enhance the area of suitable spawning habitat; (2) be associated with enhanced growth rates and morphometric condition of Murray cod; and (3) enhance recruitment (to YOY). Further, we hypothesise that multiple years of enhanced spring–summer flow will result in broad size/age distributions of Murray cod and increased population resilience in the Lower Murray.

Murray cod spawn annually over a well-defined period from October–December, irrespective of flow (Davis 1977; Rowland 1998), but recruitment in the LMR main channel is positively associated with flow and lotic habitats (Zampatti *et al.* 2014). In recent years (2015–2019) under predominately in-channel flows <18,000 ML/d, regular recruitment of Murray cod was observed in the LMR. During 2019-20, there was a strong recruitment event, indicated by an increase in the relative abundance of YOY throughout the LMR. In the Upper Murray River, Tonkin *et al.* (2018) found support for spring flows and antecedent maximum (e.g. bankfull) flow conditions (i.e. flows preceding and during the spawning period) increasing recruitment strength (Tonkin *et al.* 2018). Potential mechanisms driving this could be increased extent and quality of spawning habitat (e.g. Koehn 2009; Baumgartner *et al.* 2014; Stuart *et al.* 2019) or provision of additional food resources for larvae. During 2019-20, the majority of environmental water was delivered to South Australia during the spawning season and early larval period from late September to early November 2019, as a spring fresh, peaking at 15,600 ML/d in mid-October (Figure 5). The delivery of Commonwealth environmental water during this period greatly increased the length of weir pools (e.g. >20 km in Weir Pools 2 and 3) experiencing lotic (i.e. water velocities >0.3 m/s) conditions (Figure 56), thereby increasing the extent of habitat favoured for spawning and early life history processes (e.g. drift and residence) (Gibbs *et al.* 2020). Multiple years of data and further modelling is required to identify the potential drivers of increased recruitment in 2019-20, and to better evaluate the influence of Commonwealth environmental water on Murray cod recruitment.

During 2019-20, Murray cod were sampled at various stages of their early life history from larvae (November) through to YOY (April/May). To assess morphometric condition and growth during this year, and the influence of the flow regime (including Commonwealth environmental water) on these parameters, data from other projects including LTIM were used to develop ‘baselines’ for comparison. Since 2019-20 was the first year early juvenile Murray cod were successfully sampled in summer (January and February), additional years of data are required to provide reliable baselines for robust comparison and evaluation. While body condition of YOY Murray cod sampled in April/May was relatively similar from 2015–2020, the condition in 2017, following overbank flows in 2016-17, was significantly greater. This suggests that condition may be influenced by large-scale (i.e. overbank) flow events, while small-scale seasonal flow deliveries (e.g. winter or spring–summer freshes <18,000 ML/d) may have small/negligible influence. Further support of this was provided by the observed similarity in condition among weir pools, suggesting major changes in body condition are not driven by localised changes (e.g. weir pool) and are operating at a broader (river) scale. Potential mechanisms driving this may be related to increases in productivity/food resources (allochthonous sources) and/or hydraulic changes that improve habitat quality and increased feeding efficiency.

Interestingly, our results on morphometric condition, in part, contrast those presented on growth rates of juvenile Murray cod in association with discharge in other areas of the MDB (the Gwydir, Lachlan, Edward-Wakool, Murrumbidgee and Goulburn rivers) by Stoffels *et al.* (2020). They suggested maximum early juvenile growth occurs in years of median annual discharge and proposed the maximisation of slackwater habitats – favoured by early juvenile Murray cod – under these flows (Vietz *et al.* 2013) as a likely mechanism. Further, in some rivers, high discharge can reduce the surface area of slackwaters (Price *et al.* 2012), leading Stoffels *et al.* (2020) to associate this mechanism with reduced growth under high discharge in their study. We believe similar mechanisms – enhancement/reduction of favoured juvenile habitat (e.g. slackwaters) – are likely operating in the LMR; however, the large size and highly regulated nature of the LMR, relative to the rivers studied by Stoffels *et al.* (2020) may confer different relationships between discharge and condition/growth. For instance, the hydraulic impact of multiple sequential weirs likely dictates that increases in discharge are required to promote hydraulic diversity, including slackwaters (Bice *et al.* 2017; Gibbs *et al.* 2020), while the impact of high discharge on slackwater area may be less pronounced.

Abundance (CPUE) of Murray cod in the LMR exhibited variability through 2013–2020. A decline in abundance, notably adults (>600 mm TL), during 2017 was associated with a hypoxic blackwater event in the LMR (Ye *et al.* 2018). Following this widespread blackwater event, declines in the abundance of adult Murray cod were also observed throughout the Murray River and some of its tributaries (e.g. Lachlan River, Murrumbidgee River, Edward–Wakool River system) (Dyer *et al.* 2017; Wassens *et al.* 2017; Watts *et al.* 2017; Stoffels *et al.* 2018). After 2017, abundance of Murray cod increased to a peak in 2020, driven predominantly by newly recruited YOY. Length frequency distributions showed a broad size distribution of Murray cod from 2015–2020, comprised of juveniles (including YOY), sub-adults and adults. In 2019 and 2020, YOY dominated the sampled population (≥67%) and cohorts from recent recruitment events (e.g. age 1+ and ~3+) were also apparent in 2020. This contrasts with the Millennium Drought period (2003–2009) when length frequency distributions in the main channel of the LMR were dominated by adults >800 mm TL, indicating a lack of recent recruitment (Zampatti *et al.* 2014). The contemporary length frequency distributions in the LMR main channel indicate population resilience to environmental perturbations such as hypoxic blackwater events.

Management implications

Murray cod show a preference for lotic habitats and in the LMR, the extent of lotic habitat available during spawning and early life stages may be important for the magnitude of spawning and subsequent recruitment. In 2019-20, environmental water delivered as freshes during October–early November increased the longitudinal extent of lotic habitats - defined as mean cross-sectional velocity >0.3 m/s - in the LMR by 10% for at least 30 days. Specifically, downstream of Locks 4 (Weir Pool 3) and 3 (Weir Pool 2), the peak of the spring pulse was associated with an increase of lotic habitat area, relative to without Commonwealth environmental water, two-fold and five-fold, respectively (Figure 56).

Murray cod condition appeared to be influenced by large-scale flow events (e.g. floods) rather than localised or smaller scale deliveries (e.g. small freshes). As such, flow pulses may support enhanced recruitment of Murray cod more so through expansion of lotic habitat favoured for spawning and by early life stages rather than productivity benefits. Nonetheless, further analysis and comparison of otolith daily growth increments will inform on finer temporal-scale influence of flow on growth.

In addition to the delivery of environmental water, other actions that support the maintenance/enhancement of lotic habitats in the LMR (e.g. weir pool lowering) may further benefit Murray cod recruitment and population resilience.

Conclusion

Strong recruitment of Murray cod to YOY was observed in the LMR during 2019-20. While environmental water likely had negligible influence on the growth and condition of Murray cod, as the major volumes of water were delivered outside the period of growth (November–May), an increase in the extent of favourable (lotic) habitat during the spawning and early larval period may have played a key role in supporting spawning and recruitment in 2019-20. Given the early stage of our investigations for this indicator, evaluation of the contribution of Commonwealth environmental water towards growth, condition and recruitment in 2019-20, was conservatively designated as unknown. In future years, a more robust evaluation will be informed by multiple years of data (including otolith increment analysis), and results of the Selected Area research project and Basin-scale research and evaluation.

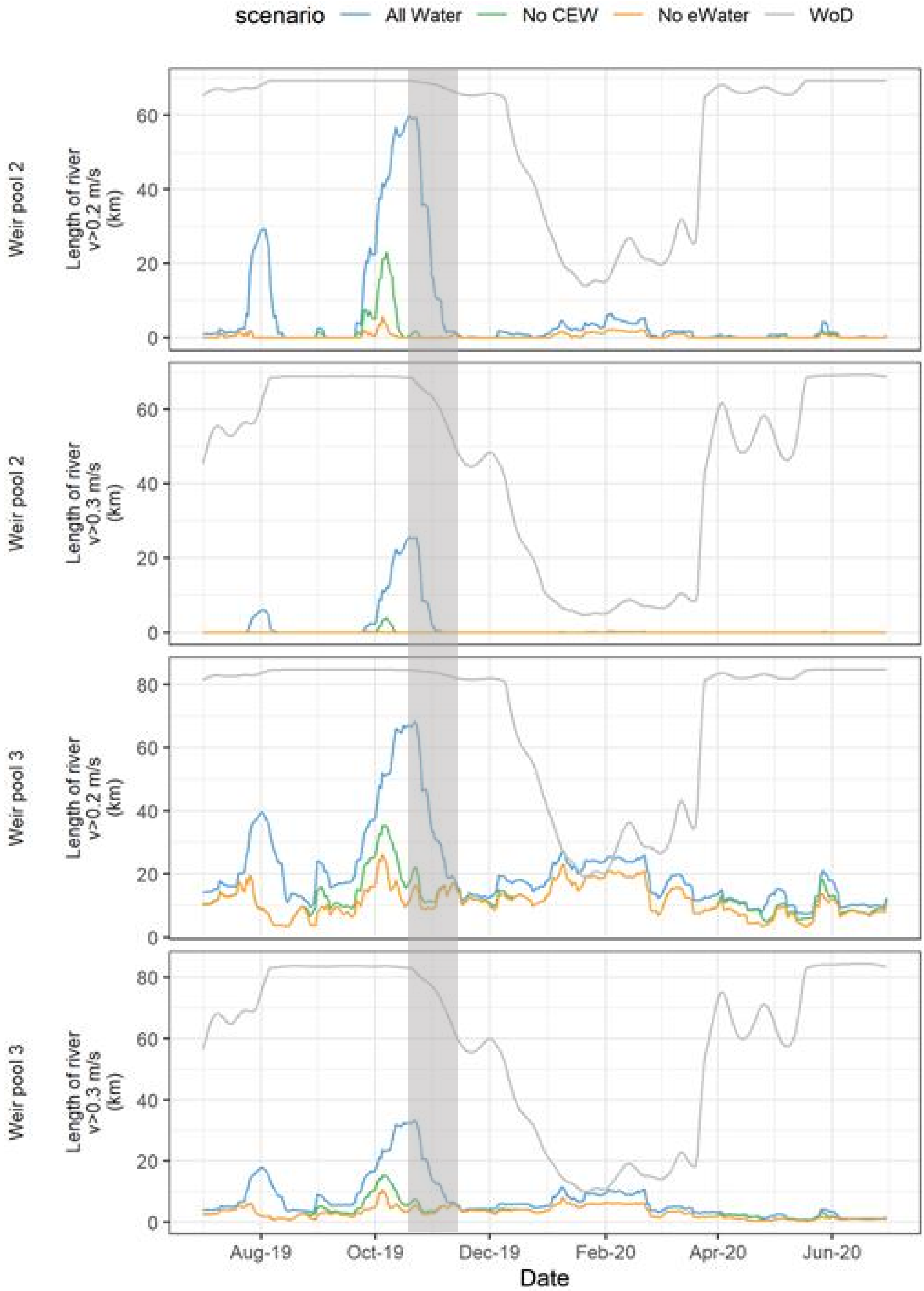

Figure 56. Increases in lotic habitat in the weir pools below Lock 3 and Lock 4 (refer to Section 2.1 for details) with the peak spawning period of 17 October–11 November 2019, based on back-calculated daily ages, indicated by a grey shaded bar.

2.8 Fish Assemblage

Background

In 2020, we collected fish assemblage data in the main channel of the LMR to inform Basin-scale evaluation of fish community responses to Commonwealth environmental water. The evaluation of these responses under the LTIM Project (2015–2019) was undertaken by the Centre for Freshwater Ecosystems at La Trobe University (King *et al.* 2020), while the evaluation under the MER Project (2020–2022) is being undertaken by a team of fish ecologists and biometricians, led by the Arthur Rylah Institute for Environmental Research (ARI).

Objectives

In this section, our objectives are to: (1) provide summary statistics of the catch rates and population demographics for nominated species; (2) describe temporal variation in fish assemblage and population structure from 2015–2020; and (3) discuss key findings based on published research and a contemporary understanding of fish life histories and population dynamics in the LMR. Our interpretations of the data for this indicator do not infer association with environmental water delivery.

Methods

During March–April 2020, small- and large-bodied fish assemblages were sampled from the gorge geomorphic zone of the LMR (Figure 1) using fyke nets and electrofishing, respectively. Prescribed methods (Hale *et al.* 2014) were used to obtain population structure data for seven target species (Figure 57). Refer to SARDI *et al.* (2019) for detailed sampling design and methodology.

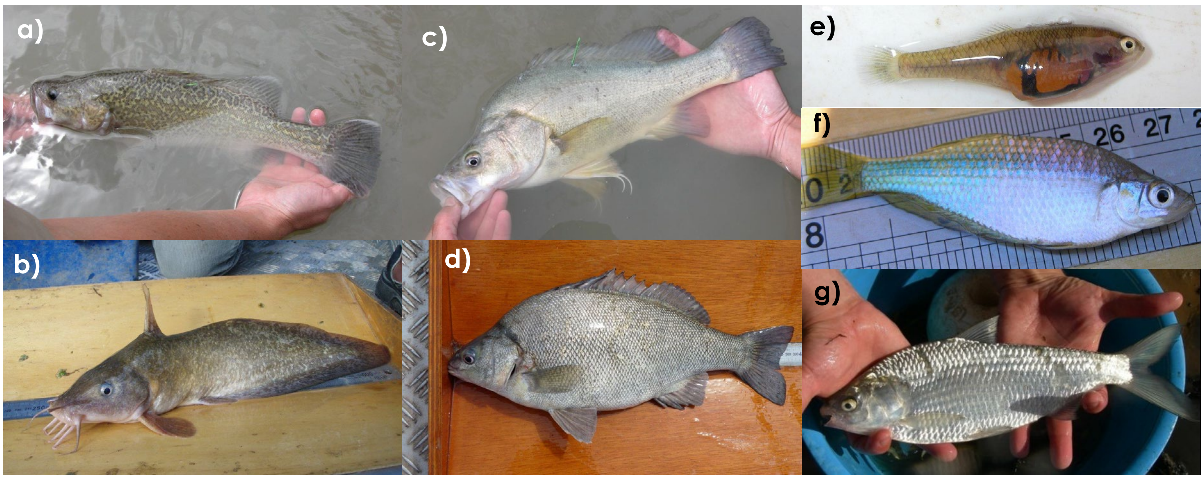

Figure 57. Target species for the Lower Murray River: (a) Murray cod and (b) freshwater catfish (equilibrium life history); (c) golden perch and (d) silver perch (periodic life history); and (e) carp gudgeon, (f) Murray rainbowfish and (g) bony herring (opportunistic life history).

Temporal variation in fish assemblage structure (species composition and abundance), between sampling years (i.e. 2015–2020), was investigated using Non-metric Multi-Dimensional Scaling (MDS), permutational multivariate analysis of variance (PERMANOVA) and Similarity Percentages (SIMPER) analysis in the software package PRIMER v. 6.1.12 (Clarke and Gorley 2015) and PERMANOVA + v.1.02 (Anderson *et al.* 2008). To determine temporal variation in population structure, length frequency histograms were qualitatively compared among sampling years.

Results

Catch summary for 2020

A total of 11,773 individuals (ind.) from eight large-bodied fish species were collected by electrofishing. Bony herring (*Nematalosa erebi*) was the most abundant species (97% of the catch by number), followed by common carp (*Cyprinus carpio*) (1%) (Figure 58a).

A total of 13,729 individuals from seven small-bodied species were collected using fyke nets. Carp gudgeon (*Hypseleotris* spp.) was the most abundant species (87% of catch by number), followed by Murray rainbowfish (*Melanotaenia fluviatilis*) (5%) and unspecked hardyhead (*Craterocephalus stercusmuscarum fulvus*) (4%) (Figure 58b).

Temporal variability in fish assemblage structure

MDS ordination of electrofishing data demonstrated separation of 2017 samples from all other years (Figure 59a). PERMANOVA indicated that large-bodied fish assemblages were significantly different among years ($Pseudo\text{-}F_{5,54}$ =4.9401, $P \leq 0.001$). Pairwise comparisons revealed significant differences between 2017 and 2015, 2017 and 2018, 2017 and 2019, and 2017 and 2020, but not for any other pairs of years (Figure 59a).

For small-bodied fish assemblages, there were significant differences among years ($Pseudo\text{-}F_{5,59}$ =4.0628, $P \leq 0.001$). PERMANOVA pair-wise comparisons revealed significant differences in small-bodied fish assemblages between 2017 and 2016, 2017 and 2018, and 2017 and 2020, but not for any other pairs of years (Figure 59b).

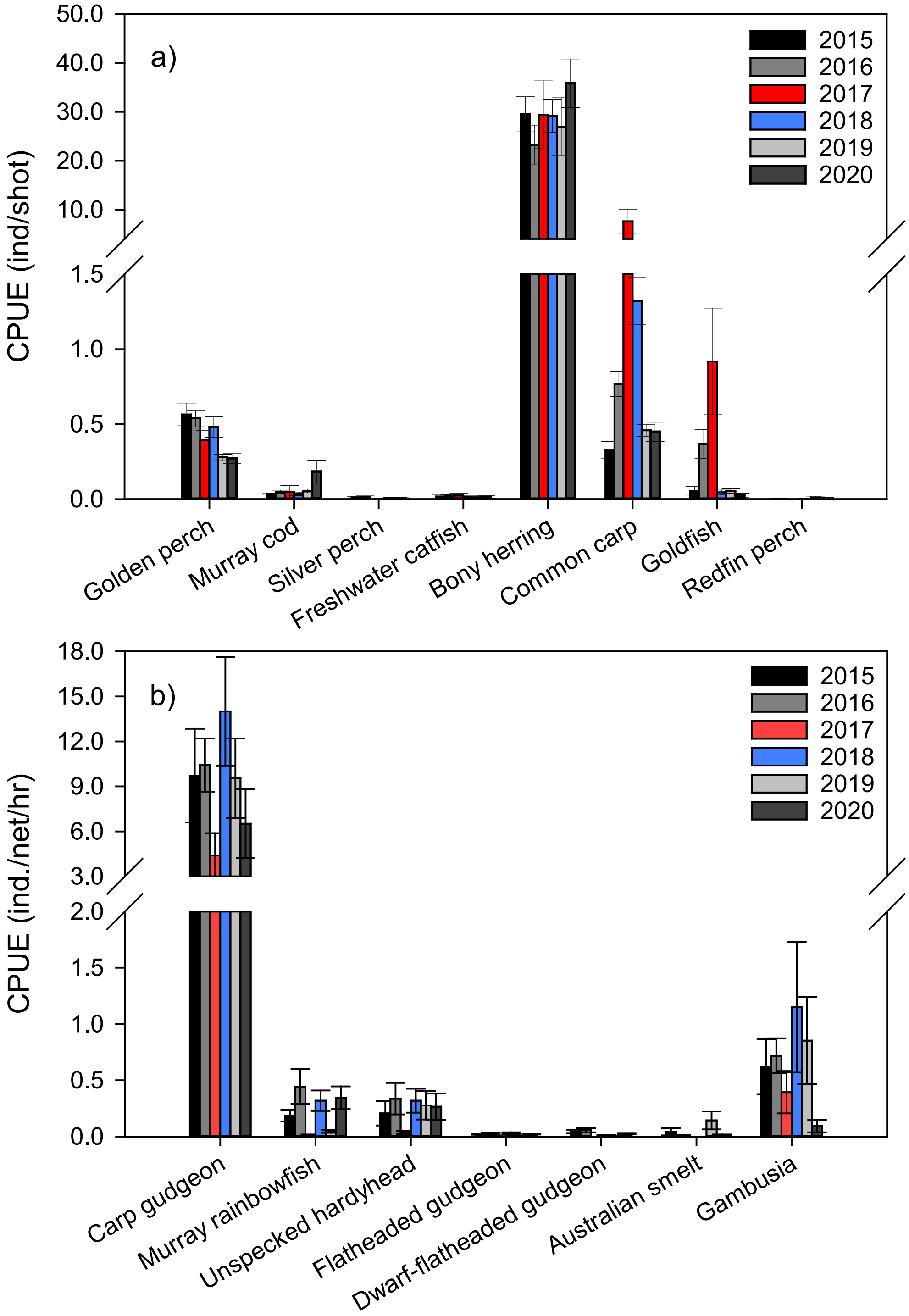

Figure 58. Mean catch-per-unit-effort (CPUE) ± standard error of (a) large-bodied fish species captured using electrofishing (individuals per 90 second shot) and (b) small-bodied fish species captured using fine-mesh fyke nets (individuals per net per hour) in the gorge geomorphic zone (10 sites) of the Lower Murray River in autumn from 2015–2020. Electrofishing CPUE data from five sites are presented for 2017 as other sites were sampled during winter 2017. See Table D17 in Appendix for statistics.

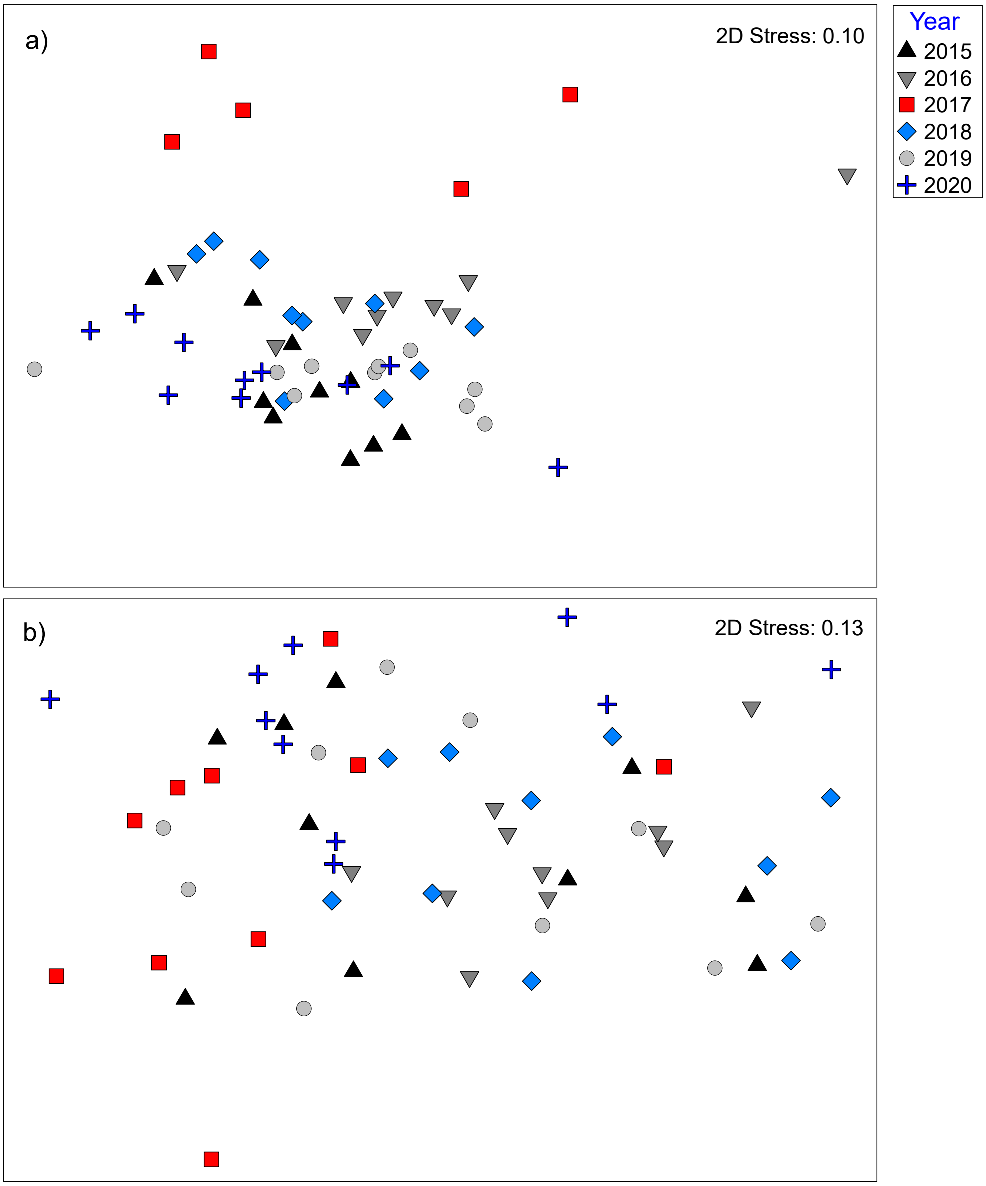

Figure 59. Non-metric multi-dimensional scaling (MDS) plot of (a) large-bodied fish assemblages sampled by electrofishing and (b) small-bodied fish assemblages sampled by fyke netting in the gorge geomorphic zone of the Lower Murray River from 2015–2020. Sites (*n* = 5) sampled in winter 2017 were removed from the ordination.

SIMPER indicated that differences between years for large-bodied fish assemblages were primarily driven by higher abundance of common carp and goldfish in 2017 (Figure 58). SIMPER indicated that differences between 2017 and other years (i.e. 2016, 2018 and 2020) for small-bodied fish assemblages were driven by a lower relative abundance of carp gudgeon in 2017 (Figure 58).

Temporal variation in length/age structure of large-bodied species

In 2018, the sampled golden perch population was mostly comprised of age 6+ (23%), 7+ (51%), 8+ (19%) and 21+ (4%) fish (Figure 60), while freshwater catfish (*Tandanus tandanus*) ranged in age from 8+ to 13+ years (Figure 62). The length distribution of golden perch (227–487 mm) and freshwater catfish (431–453 mm) in 2020 indicates the absence of new recruits and an ageing population. In 2018, only one silver perch (*Bidyanus bidyanus*) was sampled (age 1+, 151 mm fork length, FL) (Figure 61). In 2020, the capture of a small silver perch (97 mm FL) may suggest the presence of an age 0+ cohort.

In 2018, the sampled Murray cod population consisted of individuals 74–140 mm (age 0+) and 307–515 mm (not sacrificed, potentially age 2+ and/or older). In 2020, the sampled population consisted of individuals 59–124 mm (age 0+) and 511–720 mm (potentially age 3+ and/or older) (Figure 63). Similar to previous years (e.g. 2018), in 2020, the sampled bony herring population was dominated by age 0+ fish (95%), but comprised older fish and ranged in age from 0+ to 11+ years (Figure 64).

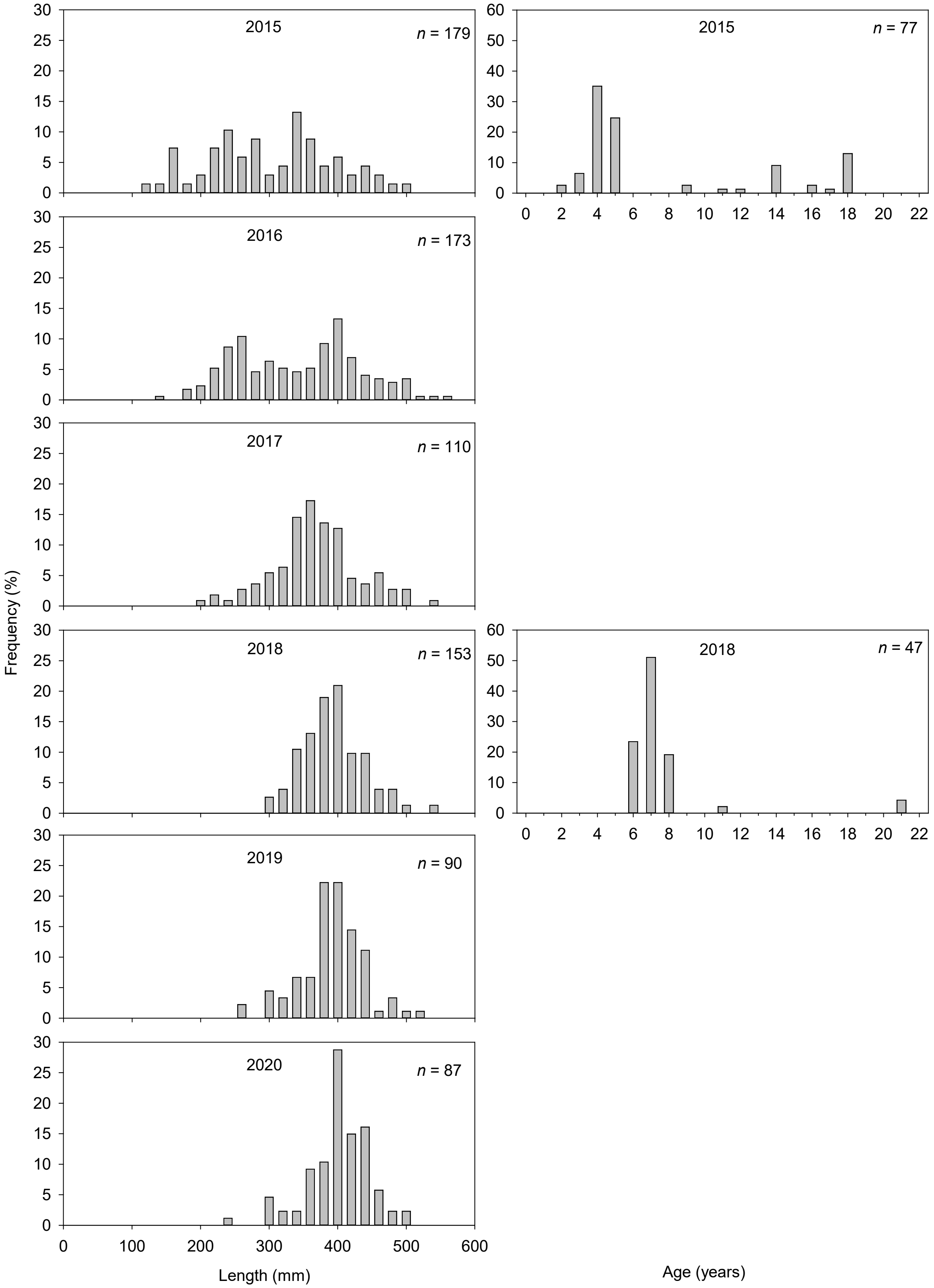

Figure 60. Length frequency distributions and age structures of golden perch collected from the gorge geomorphic zone of the Lower Murray River from 2015–2020.

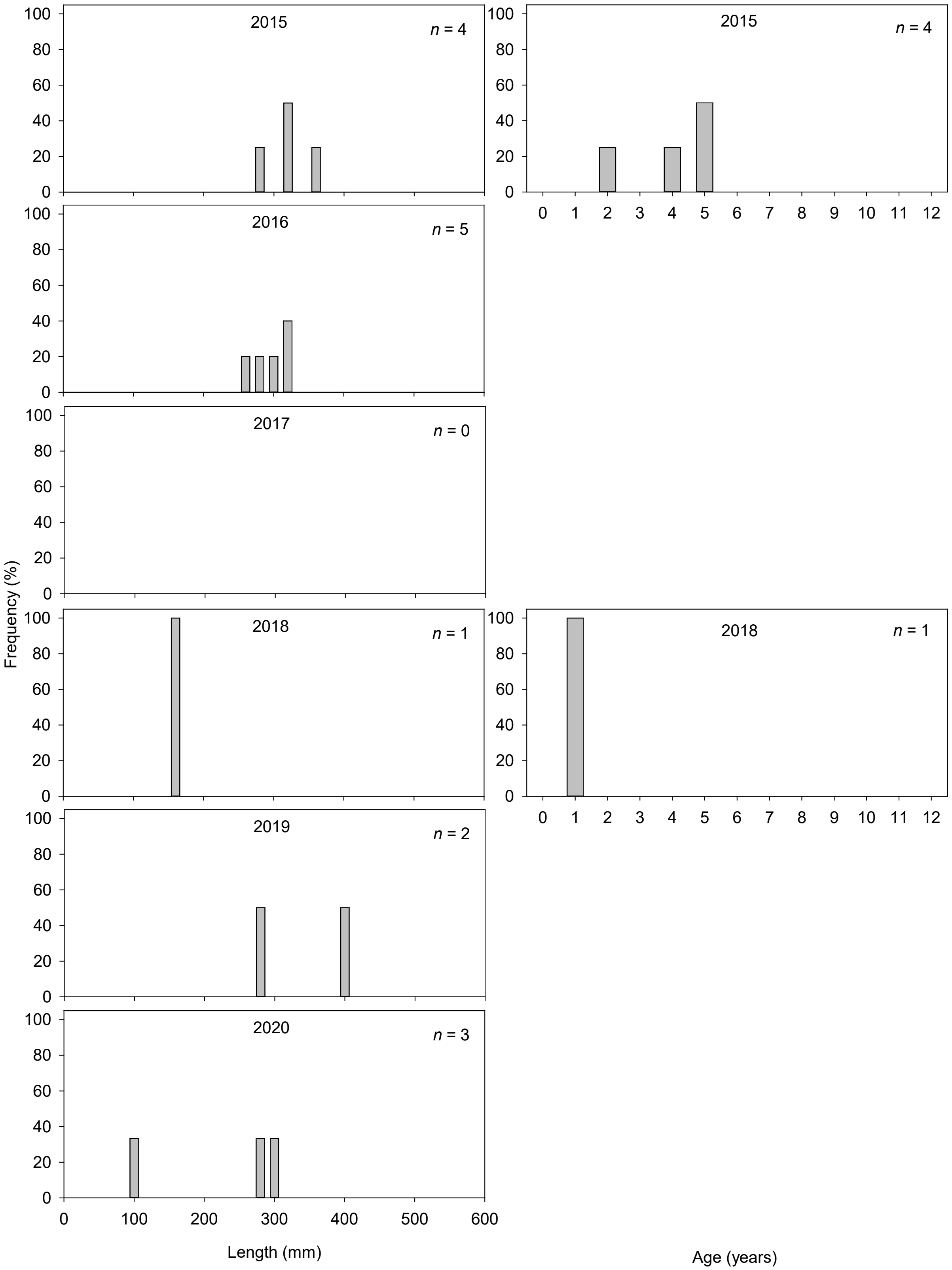

Figure 61. Length frequency distributions and age structures of silver perch collected from the gorge geomorphic zone of the Lower Murray River from 2015–2020.

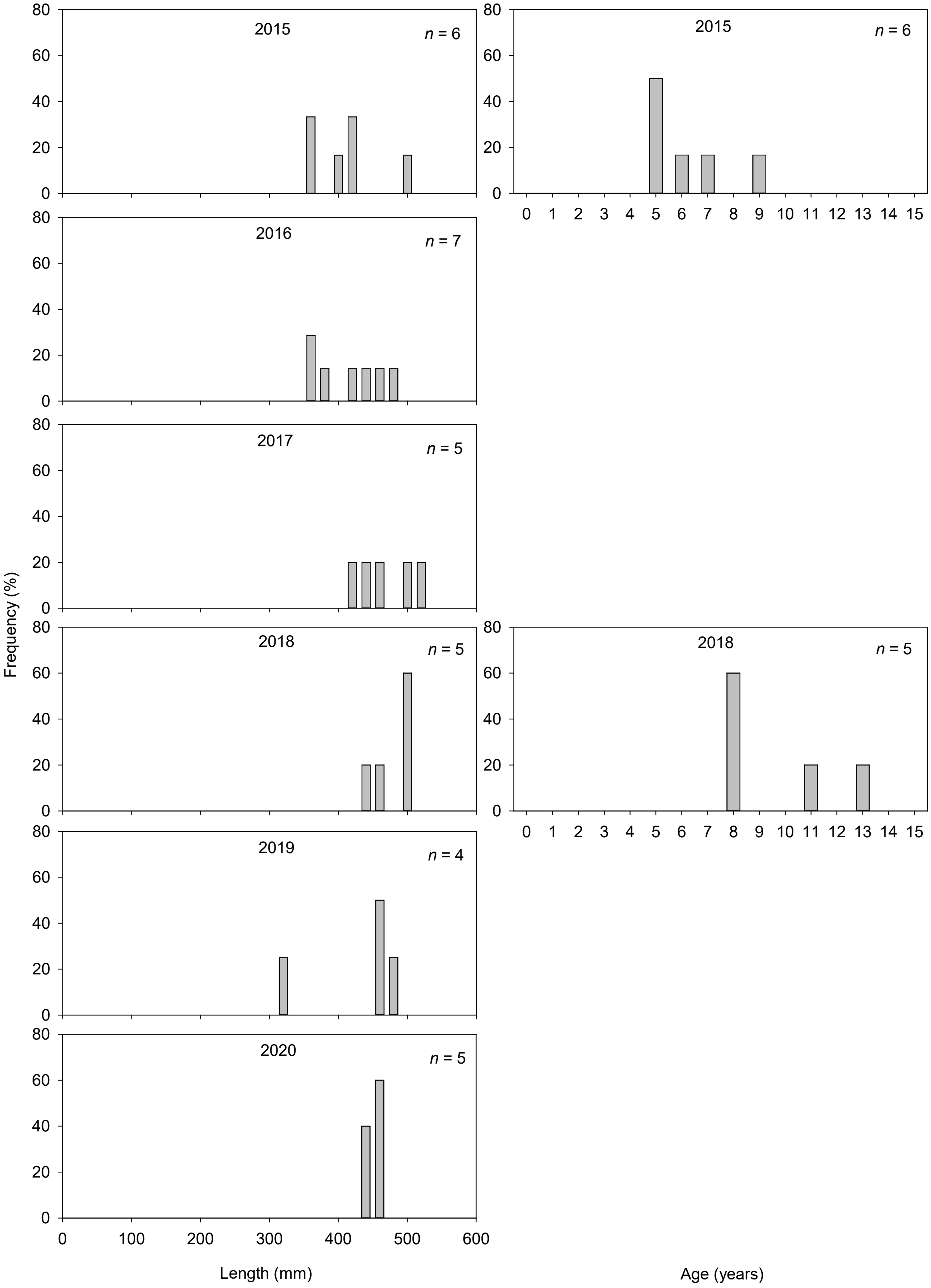

Figure 62. Length frequency distributions and age structures of freshwater catfish collected from the gorge geomorphic zone of the Lower Murray River from 2015–2020.

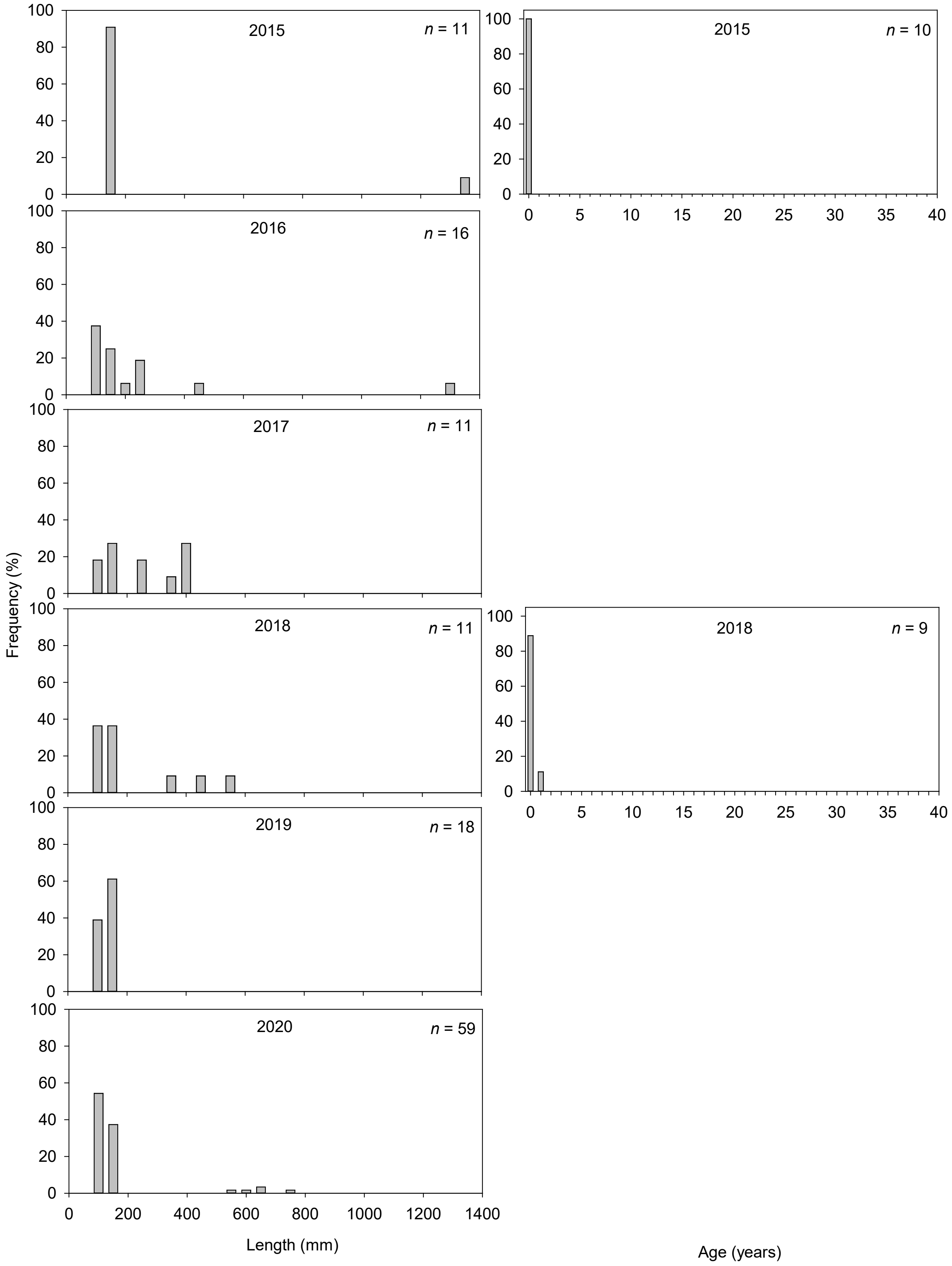

Figure 63. Length frequency distributions and age structures of Murray cod collected from the gorge geomorphic zone of the Lower Murray River from 2015–2020. Individuals >340 mm were not retained for ageing.

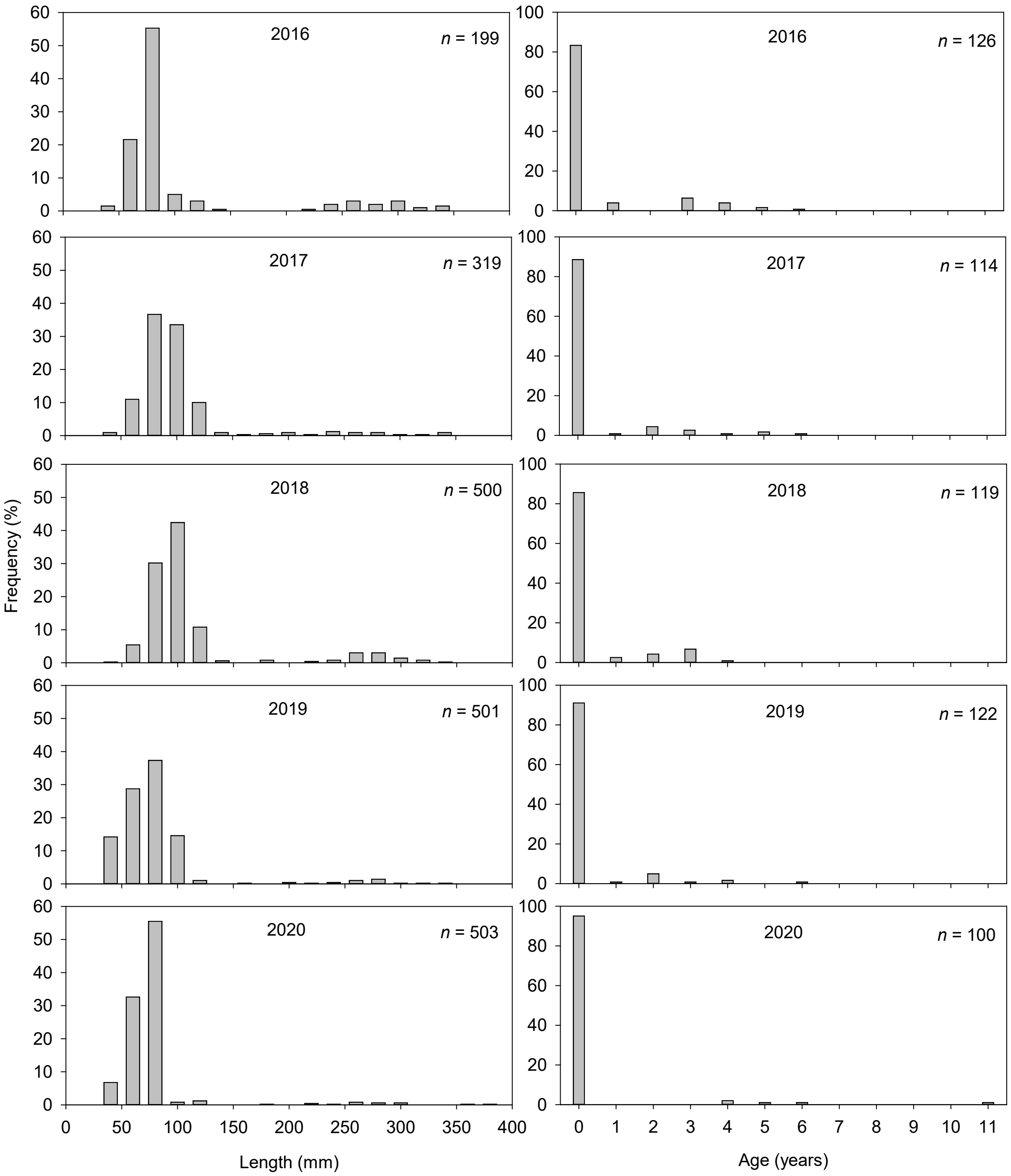

Figure 64. Length frequency distributions and age structures of bony herring collected from the gorge geomorphic zone of the Lower Murray River from 2016–2020.

Evaluation

There are no CEWO evaluation questions for this indicator for the Lower Murray Selected Area. The Basin-scale evaluation of fish community responses to Commonwealth environmental water is being undertaken by a team of fish ecologists and biometricians, led by the ARI. For this report, fish monitoring data from this standardised sampling, and additional targeted sampling for flow-cued spawners (Section 2.6) and Murray cod (Section 2.7), were consolidated to evaluate a number of fish targets of the Long-Term

Environmental Watering Plan (LTWP) for the South Australian Murray River (DEWNR 2015) and the Basin Plan Matter 8 Report (DEW 2020) in Appendix C.

Discussion

During 2014-15 and 2015-16, relatively low (<15,000 ML/d), stable flows predominated in the LMR. In these years, small-bodied fish abundance and diversity were high. Abundances of flow-cued spawning species (i.e. golden perch and silver perch) remained similar in both years and overall, fish assemblage structure was characteristic of low flows in the LMR and similar to that during drought in 2007–2010 (Bice *et al.* 2014).

In 2017, following flooding in spring–summer 2016 (peak flow ~94,600 ML/d), there was a significant change to the small- and large-bodied fish assemblages, with an overall decrease in the abundances of small-bodied species and an increase in the abundance of common carp. A reduction in submerged vegetation in the main channel of the LMR during 2016-17, due to a combination of increased water depth/decreased light penetration and physical scour, likely resulted in a decrease in habitat availability and decreased abundance of small-bodied fishes (Bice *et al.* 2014). In 2017, increased abundance of common carp was driven by a large recruitment event associated with flooding in 2016-17. Following a recession in water levels in summer 2017, large numbers of YOY (age 0+) common carp likely entered the main channel from off-channel floodplain and wetland habitats (their typical spawning and nursery habitat) and were captured during sampling in autumn and winter 2017.

In general, the fish assemblage in 2017 was typical of high flow conditions and similar to assemblages in high flow years from 2010–2012, with the exception of low abundances of golden perch (Bice *et al.* 2014). In 2016-17, recruitment of native, large-bodied flow-cued spawners (e.g. golden perch) was negligible, despite a flow regime that was potentially conducive to spawning of these species (Mallen-Cooper and Stuart 2003; Zampatti and Leigh 2013a; 2013b) (also see Section 2.6). It is possible, that recruitment in 2016-17 was compromised by hypoxia associated with blackwater during the spring–early summer spawning season (Ye *et al.* 2018), impacting the survival of eggs and larvae.

Following in-channel flows (up to 17,800 ML/d) in spring–early summer 2017-18, small-bodied fish species composition and abundance in 2018 reverted back to that of pre-flood conditions (i.e. 2016 and 2015), likely due to structural and hydraulic habitats (i.e. submerged vegetation and stable water levels) conducive to small-bodied fish recruitment in the main river channel. Species composition and abundances were similar in 2019 and 2020, following comparable hydrological conditions (i.e. low, in-channel flows <18,000 ML/d) in 2018-19 and 2019-20. From 2017 to 2020, the large-bodied fish assemblage trended back towards one typical of 'low flows' (e.g. 2016, 2015 and 2008, Bice *et al.* 2014) due to a reduction of common carp and goldfish abundance in 2019 and 2020 to levels similar to 2015 and 2016.

Based on electrofishing length frequency data, no recruitment (to YOY) was observed for freshwater catfish in the LMR from 2014–2020. In the LMR, the spawning biomass of freshwater catfish may be historically low (Ye *et al.* 2015) and their recruitment dynamics are poorly understood.

From 2015–2019, regular recruitment and similar inter-annual abundances of Murray cod (i.e. fish <150 mm TL) were observed in the LMR, during years following an in-channel flow pulse (15,000–18,000 ML/d) and a high, overbank flow (>90,000 ML/d), but also during three years of low, stable, in-channel flows (<12,000 ML/d). In 2020, during similar hydrological conditions to 2017-18 (in-channel spring flow pulse <18,000 ML/d), Murray cod abundance increased significantly, driven by the increased abundance of new recruits. Furthermore, based on length frequencies, there was evidence of some cohorts from 2015–2019 persisting in the population. These results contrast with data collected from 2003–2010, during the Millennium Drought, when Murray cod recruitment, measured as abundance of YOY in autumn, was limited in the predominantly lentic main channel habitats of the LMR. Subsequently, recruitment was observed in high flow years from 2010–2013 (Zampatti *et al.* 2014). The mechanisms that facilitate recruitment of Murray cod (to YOY) in the LMR are unclear and being explored as part of the Murray Cod Recruitment indicator (Section 2.7) and associated research project (not presented in this report).

Management implications

Prolonged low, in-channel flows (<20,000 ML/d) promote hydraulic (e.g. lentic) and structural (submerged plant) habitat conditions suitable for low flow generalist fishes such as small-bodied carp gudgeon and unspecked hardyhead. Conversely, in-channel flows >20,000 ML/d facilitate a shift towards more natural riverine hydraulic characteristics that benefit flow-dependant species such as golden perch and Murray cod. Increased variability in the annual flow regime that includes large, overbank (>45,000 ML/d) flows will lead to increased recruitment of a range of species with different life histories (e.g. flow-dependant), promoting diverse fish assemblages and resilience in populations.

Conclusion

In the main channel of the LMR during 2015 and 2016, fish assemblages were characterised by high abundances of small-bodied species and a lack of recruitment of native, large-bodied flow-cued spawners. Fish assemblage structure was similar to that during drought in 2007–2010 (Bice *et al.* 2014) and characteristic of a low flow scenario. Following high flows in 2016-17, the fish assemblage shifted towards one characterised by low abundances of small-bodied species and high abundance of a large-bodied species, common carp. This assemblage was more typical of high flows, similar to 2010–2012 (Bice *et al.* 2014). Nevertheless, recruitment of native, large-bodied flow-cued spawners (e.g. golden perch) was negligible in 2016-17, likely due to water hypoxia associated with the blackwater event. During 2018–2020, a return to low, in-channel flows (<18,000 ML/d) resulted in the fish assemblage trending back towards that of 2015 and 2016, following: an increase in small-bodied fish abundance; a lack of recruitment from native, flow-cued spawners; and a decrease in common carp and goldfish abundance.

3 CONTINGENCY MONITORING

During 2019-20, contingency monitoring supplemented existing monitoring of lamprey migration funded under the TLM Program. These additional resources ensured an appropriate level of monitoring to assess abundance and movement of lamprey, and the contribution of Commonwealth environmental water to end-of-system connectivity and migration of lamprey. A summary of the key findings and evaluation are presented below. Detailed methods, results and discussion are reported in Bice *et al.* (2020) (https://www.environment.gov.au/water/cewo/publications/lamprey-migration-lower-river-murray-cew-delivery-2019).

3.1 Lamprey migration in 2019-20: key findings and evaluation

Background

Pouched lamprey (*Geotria australis*) and short-headed lamprey (*Mordacia mordax*) are the only anadromous fishes native to the MDB. Their lifecycles are characterised by a parasitic marine phase, upstream spawning migrations into freshwaters followed by adult mortality, freshwater larval and juvenile development, and subsequent downstream migration to the ocean. Historically, lamprey were common in the Murray River with spawning migrations potentially extending up to 2,000 km upstream, but they are now rarely encountered, suggesting barriers to migration and flow regulation have impacted these species. In the past decade in the MDB, however, a focus on restoring connectivity (e.g. fishway construction) and ecologically relevant components of flow regimes, through the delivery of environmental water, have increased opportunities for migration that may aid recovery of lamprey populations. A key objective of the delivery of Commonwealth environmental water in the southern MDB is to improve end-of-system flow and connectivity through the Murray barrages and Murray Mouth to support fish movement and expanded distributions. The migratory life histories of pouched lamprey and short-headed lamprey make these species well suited to demonstrate the achievement of these outcomes.

The objective of this project in 2019 was to assess the abundance of adult lamprey migrating through fishways at the Murray barrages and to use passive integrated transponder (PIT) telemetry to investigate subsequent upstream spawning migrations in the MDB. Specifically, the project aimed to:

- Assess the abundance of migrating lamprey at multiple fishways on the Murray barrages in winter–spring 2019, and implant individuals with PIT tags;
- Compare abundance from winter–spring 2019 with previous years of targeted lamprey sampling in 2015–2018;
- Interrogate PIT telemetry data from fishways along the Murray River to describe the spatio-temporal characteristics of migration, including extent, timing and rate, as well as interaction with fishways; and
- Evaluate the contribution of Commonwealth environmental water to connectivity, and the abundance and migration of lamprey at the Murray barrages in 2019.

Methods

Targeted sampling for lamprey was conducted from 9 July–4 October 2019 (*n* = 11–20 sampling events per fishway) at seven vertical-slot fishways on Goolwa, Mundoo, Boundary Creek, Ewe Island and Tauwitchere barrages. Additional sampling occurred on 21–25 October and 18–22 November at five fishways on Goolwa and Tauwitchere barrages, and the Hunters creek causeway, as part of TLM Program condition/intervention monitoring, and data on lamprey captures from these events are also reported. During each sampling event, fishway traps were set overnight in fishway entrances, and deployed and retrieved using a truck fitted with an overhead crane. Upon retrieval, all trapped fish were removed and placed into aerated holding tanks. Lamprey were sorted from the catch for processing and all remaining fish released upstream. Prior to release, lamprey were anaesthetised and implanted with PIT tags (Texas Instruments RI-TRP-REHP half-duplex, 3.85 x 23 mm, 0.6 g). Upstream migration through fishways on main channel weirs at Locks 1–11 was subsequently monitored with KarlTek KLK5000 PIT reader systems (KarlTek Pty Ltd, Melbourne, Australia) and detection data transmitted to the MDBA PIT telemetry database.

Results

From May–December 2019, discharge to the LMR at the South Australian border (QSA) was characterised by flows <8,000 ML/d, punctuated by conspicuous in-channel flow pulses that peaked at ~11,000 and ~15,600 ML/d in August and October, respectively. These flow pulses comprised a substantial proportion (66–70%) of Commonwealth environmental water. During sampling (9 July–4 October), discharge from the Murray barrages ranged from 0–19,166 ML/d (mean = 3,236 ML/d). All water released from the barrages over this period, including water to operate fishways, was Commonwealth environmental water.

Totals of 45 pouched lamprey (43 PIT tagged) and 16 short-headed lamprey (15 PIT tagged) were captured from fishways during sampling at the Murray barrages in winter–spring 2019. Pouched lamprey were captured from early July to early September, but migration peaked in early August, while short-headed lamprey were captured from early July to mid-November, with peak migration in early October (Figure 65a). The abundance of pouched lamprey was moderate–high relative to preceding years with targeted lamprey monitoring (Figure 65b), whilst short-headed lamprey was sampled in greatest abundance since 2006. Both species were captured in greatest numbers from fishways on Goolwa and Mundoo barrages.

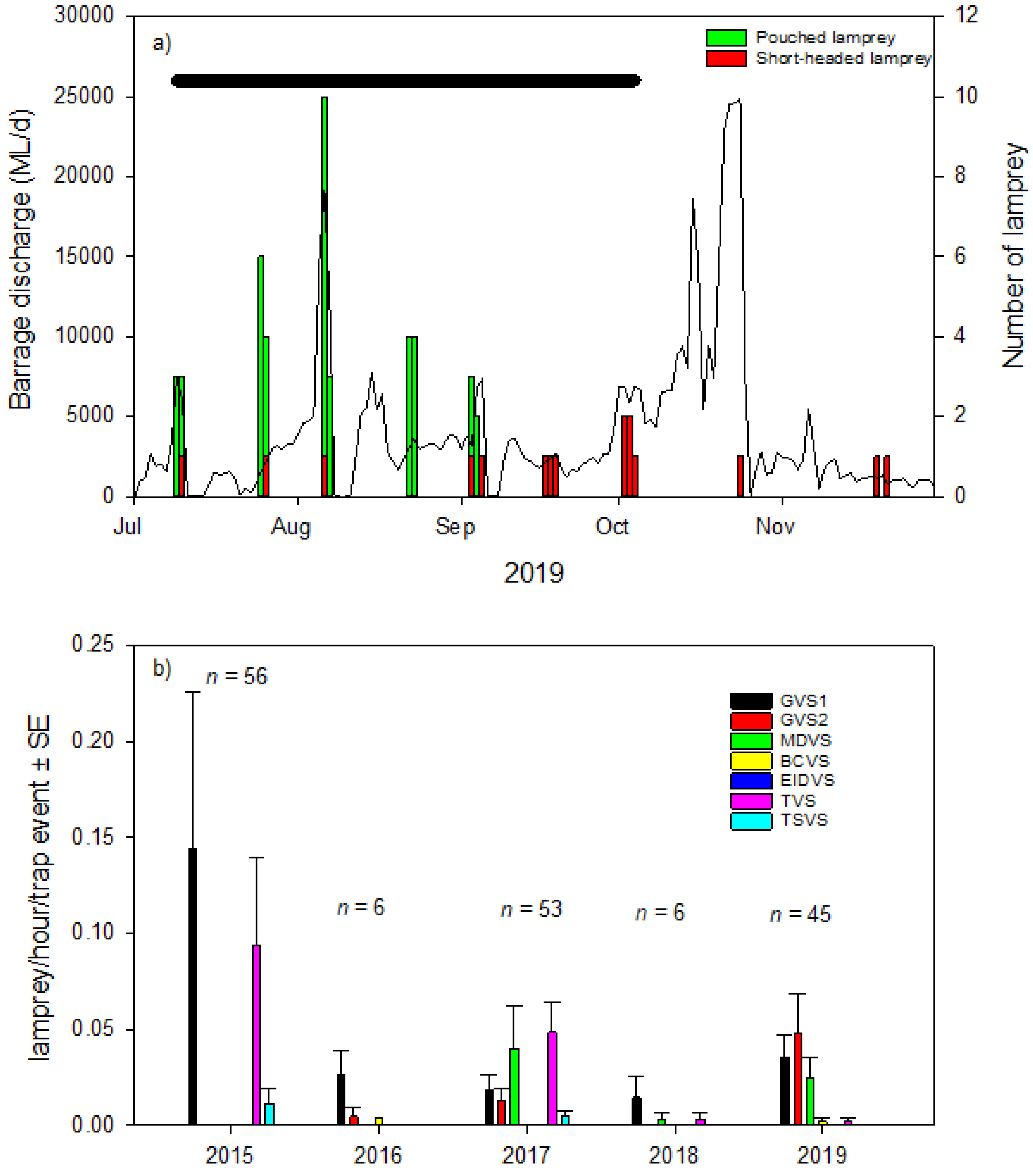

Figure 65. a) Numbers and date of capture for pouched and short-headed lamprey in 2019, presented with total daily barrage discharge (ML/d). Horizontal back bar represents period of targeted lamprey sampling; and b) relative abundance (lamprey per hour per trap event) of pouched lamprey sampled from various fishways from 2015–2019 (total numbers sampled in each year are also presented). GVS1 = Goolwa vertical-slot 1, GVS2 = Goolwa vertical-slot 2, MDVS = Mundoo dual vertical-slot, BCVS = Boundary Creek vertical-slot, EIDVS = Ewe Island dual vertical-slot, TVS = Tauwitchere vertical-slot, TSVS = Tauwitchere small vertical-slot. Note MDVS was not sampled in 2015 or 2016, while GVS2, EIDVS and BCVS were not sampled in 2015.

Of PIT tagged individuals, 44% of pouched lamprey were subsequently detected on one or more fishway PIT reader systems along the Murray River, with extent of migration ranging from 274–726 km (Locks 1–8). Migration rates varied among individuals and reaches but were at times rapid (up to 37 km/d). Just one PIT tagged short-headed lamprey (7%) was detected after release; nonetheless, this individual was detected passing eight main channel fishways, and was last detected exiting the Lock 10 fishway (825 km from the river mouth). This represents the first known tracking data on the upstream spawning migration of this species.

Evaluation

The evaluation approach, including assessment criteria, is described in the evaluation section for Hydraulic Regime (Section 2.1).

Table 23. Lamprey Migration evaluation question and answer. CEW = Commonwealth environmental water, eWater = environmental water.

CEWO evaluation questions	Outcomes of flow delivery 2019-20
What did Commonwealth environmental water contribute to connectivity and the migration of lamprey at the Murray barrages in winter–spring 2019?	In winter–spring 2019, releases of CEW from the Murray barrages represented 100% of discharge, including that for fishway operation. This facilitated connectivity between freshwater, estuarine and marine environments, and contributed substantially to beneficial migration outcomes for lamprey. This included moderate–high abundances of pouched and short-headed lamprey, relative to previous years, passing the Murray barrages on migrations that continued for 100's of kilometres upstream.

Contribution (to what extent CEW contributed towards the outcome, with the significance of the outcome considered):
⬜ Unknown 🟥 Negative ⬜ None/negligible 🟩 Minor 🟩 Moderate 🟦 Substantial

Discussion and management implications

Data collected in this and allied monitoring projects have improved knowledge on the migration of lamprey in the MDB and may be used to inform future Commonwealth environmental water delivery and infrastructure management. This includes the following:

- In a contemporary context, lamprey migrations in the Murray River still occur over vast distances (100s to >1,000 km), supported by fishways and Commonwealth environmental water delivery.
- At the Murray barrages, timing of pouched lamprey migration extends from June–September (peak July–August) and short-headed lamprey from July–November (peak September–October). These are key periods for flow management to promote lamprey migration.
- Greatest abundances of migrating pouched lamprey have been sampled during years of moderate winter–spring barrage discharge (mean daily discharge

≥2,500 ML/d) with short periods of peak discharge of ~15,000 ML/d. Abundance during higher winter barrage discharge (often unregulated) remains unknown mostly due to difficultly of sampling. Nevertheless, the hydrographs delivered in winter–spring 2015, 2017 and 2019 likely represent appropriate templates for future environmental water planning.
- Importantly, winter–spring 2015, 2017 and 2019 comprised multi-site watering events that were supported by return flows from delivery of Commonwealth environmental water in the Goulburn River. The delivery of Commonwealth environmental water is critical to maintaining barrage discharge and connectivity during years of low flow. Additionally, such water may comprise cues critical to the upstream migration of lamprey (e.g. ammocoete pheromones).
- Lamprey primarily pass upstream via fishways on Goolwa, Mundoo and Tauwitchere barrages. Abundances tend to be greater at the Goolwa and Mundoo fishways, likely due to their proximity to the Murray Mouth and greater influence of discharge from these barrages/fishways on downstream salinities. As such, during times of limited water availability, winter–spring releases that specifically target lamprey migration could be prioritised to Goolwa and Mundoo barrages to maximise attraction.

Despite advances in knowledge, the general life histories of pouched lamprey and short-headed lamprey in the MDB remain poorly understood. Specific knowledge gaps for future research to better support Commonwealth environmental water delivery include:

- Identifying specific localities (i.e. streams and river reaches) that provide spawning and nursery habitats for pouched lamprey and short-headed lamprey in the MDB;
- Investigating the influence of flow magnitude, source and longitudinal integrity of flows on lamprey upstream riverine migrations, including ultimate destination; and
- Investigating the downstream migrations of macrophthalmia (sub-adults) with regard to timing, distance, cues and interaction with potential barriers.

4 SYNTHESIS AND EVALUATION

To assess ecological response to Commonwealth environmental water in the Lower Murray, a series of evaluation questions were investigated, which were earlier adapted from Basin-scale questions (SARDI *et al.* 2019). The contribution of environmental water to Hydraulic Regime, and Matter Transport and Coorong Habitat was assessed throughout the year using a modelling approach, whereas indicators of specific ecological responses (Stream Metabolism and Water Quality, Littoral Vegetation, Microinvertebrates, Flow-cued Spawning Fish Recruitment, Murray Cod Recruitment and Lamprey Migration) were assessed through empirical monitoring during targeted seasons. This annual report for the MER Project focuses on the evaluation of Commonwealth environmental water delivery during 2019-20, with ecological outcomes for all years (2014-15 to 2019-20) presented, building on the findings from the LTIM Project (Table 24).

The year 2019-20 was a climatically and hydrologically dry year. Without environmental water, flow to South Australia would have been limited to entitlement flow (i.e. ~3,000–7,000 ML/d) throughout the year. A total of ~750 GL of Commonwealth environmental water was delivered to the LMR during this year, in conjunction with other sources of environmental water (~180 GL; i.e. TLM, VEWH and RMIF). Commonwealth environmental water represented a 32% increase in total flow volume in the LMR, which improved longitudinal hydrological connectivity and met the Basin-wide Environmental Water Strategy target (30% increase in flows in the Murray River). Environmental water created two distinct in-channel flow pulses: a winter flow pulse peaking in late July (11,000 ML/d) and a spring flow pulse peaking around mid-October (15,600 ML/d) 2019, and provided flows during summer–late autumn to maintain water levels of the Lower Lakes and support barrage releases.

Winter flow to South Australia between mid-July and mid-August 2019, supported by return flows of Commonwealth environmental water from the Goulburn River, maintained mean daily barrage releases of ~2,500 ML/d with intermittent pulses of up to ~15,000 ML/d. Barrage flows (including fishway releases), of which 100% was Commonwealth environmental water, continued into spring, which facilitated connectivity between freshwater, estuarine and marine environments. In winter–spring 2019, these releases promoted lamprey migration with moderate–high abundances of pouched and short-headed lamprey, relative to previous monitoring years, passing the Murray barrages on migrations that continued for 100's of kilometres upstream.

From late September–early November 2019, a spring flow pulse (~15,600 ML/d) was delivered to the LMR via multi-site environmental watering events, with return flows from the Murray and Goulburn rivers and Barmah–Millewa Forest. The increased flow in the LMR improved hydraulic diversity, with an extra 34 km (10%) of river characterised as lotic (mean water velocity >0.3 m/s) for at least 30 days due to the Commonwealth environmental water. The greater extent and duration of lotic habitat may have benefited Murray cod during their reproductive season (spring–early summer), potentially enhancing spawning habitat area and survival of early life stages. During this year, there was strong recruitment of Murray cod with a high abundance of YOY (age 0+) detected, continuing a trend of successful recruitment observed in recent years. The mechanisms that influence the recruitment success need further investigation and are currently being explored via the Selected Area research.

Environmental flows, in combination with weir pool raisings, also increased water level variability (IQR) by 0.13 m in the tailwaters across Weir Pools 1–5. Periodic increases in water levels could improve the condition of littoral vegetation (Gehrig *et al.* 2016) and increase biofilm diversity (Steinman and McIntire 1990), which is a key component of riverine food webs. Vegetation monitoring in December 2019 demonstrated increased native plant species diversity at multiple spatial scales following the inundation of littoral zones by spring flows in the LMR, supported by environmental water. Furthermore, river red gum germination was observed exclusively in inundated areas of the floodplain geomorphic zone. In addition, the spring flow pulse produced conditions suitable for the recruitment of specialised riparian species (e.g. Australian mudwort, lesser joyweed, spreading nut-heads) that are adapted to fluctuating water levels, therefore increasing plant functional diversity.

Increased flows supported by environmental water promoted water mixing (velocities >~0.2 m/s) and oxygen exchange at the surface, which made a significant contribution to reducing the risk of low dissolved oxygen (DO) in the LMR. The potential low oxygen period in the main channel was reduced for 30 days by Commonwealth environmental water, and 40 days by all environmental water during 2019-20, primarily associated with the spring flow pulse. Maintaining DO is particularly important during spring–summer as this period corresponds with high ecosystem respirations rates and is the primary reproductive season of many species that generally favour DO >5 mg/L. The consequences of low DO on survival of aquatic biota are evident by extensive fish kills during the 2016-17 hypoxic black water event in the Murray River, and more recent fish kills in the Darling River during 2018–2020.

In 2019-20, environmental flows moderately increased nutrient transport and export, which may have stimulated primary and secondary productivity in downstream ecosystems, potentially benefiting food webs of the LMR. However, the estimated increase in primary production by environmental water was only 1%, thus showing negligible effect on carrying capacity in the LMR. Such findings have been consistent over the last six years. In the heavily regulated LMR, the influence of environmental water delivery on riverine production may be restricted by the stable water levels that characterise weir pool environments during in-channel flows, However, in ‘unregulated’ river reaches of the Murray River (e.g. adjacent to the Hattah Lakes), environmental water could lead to substantial increases in riverine production (e.g. 31% in cross-sectional GPP, during 2017-18) (Ye *et al.* 2020).

Despite negligible recorded increases in primary production in the main channel due to environmental water during spring–summer 2019-20, environmental flows were estimated (through modelling) to increase microinvertebrate abundance and diversity in the LMR by 23 and 11%, respectively. This included a considerable increase, specifically, in the density of taxa dependent upon lateral connectivity (30%) and those transported downstream via longitudinal connectivity (27%). The misalignment with primary production and microinvertebrate responses to environmental water may be due to a number of factors, including the contribution of an ‘external’ source of microinvertebrates, which could be newly-hatched, transported from adjacent or upstream off-channel habitats. Lateral exchange from increased water levels and variability and returned flows of environmental water from upstream sources (e.g. Goulburn River/Upper Murray) likely facilitated the transport and increased microinvertebrate abundance and diversity in the main channel of LMR. These contributed to a more diverse food source for larger animals (e.g. fish). It was estimated that the density of preferred prey species of large-bodied native fish larvae increased by 37% during spring 2019 due to environmental water delivery in the LMR. This may have contributed to the enhanced recruitment of Murray cod via increasing food resources during early life stages.

During autumn 2019-20, no YOY golden perch were detected in the LMR, suggesting localised spawning/recruitment failure and a lack of immigration from spatially distinct spawning sources such as the lower Darling and mid-Murray rivers. With low in-channel flows (<18,000 ML/d) prevailing since the 2016-17 flood, the current (2020) fish assemblage in the main channel of the LMR represents one typical of low flows, with high abundances of small-bodied species, and a lack of recruitment of native, large-bodied flow-cued spawners.

Like other dry years, in 2019-20, Commonwealth environmental water supported continuous barrage flows and maintained connectivity throughout the entire year. The end-of-system flow increased salt export (estimated ~624,000 tonnes) out of the Basin and reduced salt import (estimated ~2 million tonnes) into the Coorong. Subsequently, reduced salinity levels in the Coorong increased favourable fish habitat for estuarine species. This is best exemplified by a modelled 40% increase in the area of suitable habitat for mulloway in 2019-20 due to environmental water delivery to this region from 2017-18 to 2019-20. Over the three years, environmental flows also led to some improvements in habitat suitability for *Ruppia* seed production and life-cycle completion in the southern Coorong.

Table 24. CEWO evaluation questions by indicators for the Lower Murray, which includes the Lower Murray River (LMR) and the Coorong, Lower Lakes and Murray Mouth (CLLMM). Evaluation questions are sourced or adapted from Gawne *et al.* (2014). Evaluation of CEW for hydraulic and matter transport questions is based on modelled data. CEW = Commonwealth environmental water. Refer to the evaluation in respective indicator sections (Section 2) for more detail. *Not all evaluation questions are presented here for this indicator.

CEWO evaluation questions	**Outcomes of CEW delivery**					
	2014-15	**2015-16**	**2016-17**	**2017-18**	**2018-19**	**2019-20**
What did CEW contribute to hydraulic diversity within weir pools? (LMR)	▨	▨	▨	🟩	▨	🟩
What did CEW contribute to variability in water levels within weir pools? (LMR)	▨	🟩	▨	🟩	▨	🟩
What did CEW contribute to hydrological connectivity (lateral and longitudinal)? (LMR)	▨	🟩	▫	🟩	▨	🟩
What did CEW contribute to dissolved oxygen levels? (LMR)	▫	🟦	🟩	🟦	🟩	🟩
What did CEW contribute to patterns and rates of primary productivity? (LMR)	▫	▫	▫	▫	▫	▫
What did CEW contribute to patterns and rates of decomposition? (LMR)	▫	▨	▨	▨	▨	▫
What did CEW contribute to salinity levels and transport? (CLLMM)	🟩	🟩	▨	🟩	▨	🟩
What did CEW contribute to the salinity regime? (CLLMM)	🟩					
What did CEW contribute to nitrogen and silica transport? (CLLMM)	⬜	⬜	⬜	⬜	⬜	⬜
What did CEW contribute to phosphorus transport? (CLLMM)	▫	▨	▫	▨	▨	▨
What did CEW contribute to phytoplankton transport? (CLLMM)	▫	▫	▨	▨	▫	▫
What did CEW contribute to improving *Ruppia tuberosa* habitat? (CLLMM)	⬛	⬛	⬛	🟩		
What did CEW contribute to improving fish habitat? (CLLMM)	⬛	⬛	⬛	🟦		
What did CEW contribute to littoral understorey vegetation diversity and productivity? (LMR)*	⬛	⬛	⬛	⬛	⬛	🟩
What did CEW contribute to microinvertebrate density? (LMR)*	▫	▫	▫	🟩	⬛	🟩
What did CEW contribute to microinvertebrate diversity? (LMR)*	▫	▫	▫	▨	⬛	▨
Did the flow regime (including CEW) contribute to recruitment and resilience of golden perch and silver perch populations? (LMR)	▫	▫	▫	▫	▫	▫
What did CEW contribute to growth, morphometric condition and recruitment of Murray cod? (LMR)*	⬛	⬛	⬛	⬛	⬛	⬜
What did CEW contribute to connectivity and the migration of lamprey at the Murray barrages in winter–spring 2019 (CLLMM)?	⬛	⬛	⬛	⬛	⬛	🟦

Contribution (to what extent CEW contributed towards the outcome, with the significance of the outcome considered). See respective indicator evaluation sections (Section 2) for selection criteria.

⬛ Not assessed ⬜ Unknown 🟥 Negative ▫ None/negligible ▨ Minor 🟩 Moderate 🟦 Substantial

5 GENERAL MANAGEMENT RECOMMENDATIONS

Environmental water has been used to re-establish key features of the natural flow regime to restore riverine ecosystems in the MDB (MDBA 2012; Koehn *et al.* 2014; Gawne *et al.* 2014; Webb *et al.* 2017; Watts *et al.* 2020), including the significant ecological assets of the main channel of the LMR and the CLLMM region (MDBC 2006). In the Lower Murray, this has typically involved contributing to base flows (~South Australian entitlement flows) and promoting or increasing the magnitude, duration and/or frequency of freshes (in-channel flow pulses) during dry years. Over the long-term, this is expected to make a significant contribution to achieving ecological outcomes in the LMR and the CLLMM, through restoring ecological processes and improving habitat for biota in the main channel and associated floodplain/wetlands and the Coorong.

General recommendations for flow management in the LMR are provided below, based on monitoring outcomes from the LTIM and MER projects, in conjunction with our contemporary understanding of flow-related ecology in the LMR. More specific management considerations are provided in Sections 2 and 3, based on ecological outcomes and findings from indicators. Our findings and recommendations on flow management are most relevant to the spring–summer period as this is the primary period driving biological response of selected indicators in the LMR. However, additional learnings from contingency monitoring of lamprey migration in 2019-20 are provided to inform winter flow delivery to the LMR, particularly barrage flows.

Overall, environmental flow, in 2019-20, represented 39% and 100% of the total volume delivered to the LMR and through the Murray barrages, respectively. This was significant in promoting longitudinal connectivity, including end-of-system flows. The outcomes from the MER Project in 2019-20 reinforce that in the LMR, environmental water can be used to increase flow variability, e.g. promote in-channel flow pulses.

Importantly, spring–early summer in-channel flow pulses were key features of the natural hydrograph in the LMR but are conspicuously absent from the contemporary flow regime. These flow pulses improve longitudinal connectivity, increase hydraulic diversity and water level variability (particularly in tailwaters) and contribute to a broad range of ecological processes and outcomes in riverine and estuarine ecosystems (e.g. lotic habitat, matter transport, food resources, spawning cues for riverine fishes, conditions suitable for recruitment of littoral vegetation). Such outcomes are well demonstrated by the ecological responses in the Lower Murray associated with the spring flow pulses in recent years, including that which occurred in 2019-20 (peaking at ~15,600 ML/d, with 11 days >15,000 ML/d) supported by return environmental flows in the LMR. However, due to the limited magnitude and/or duration of the flow pulse, the spatial/temporal scale of hydraulic improvement and overall riverine ecosystem responses were limited. This is exemplified by the absence of golden perch recruitment in this region. Despite the lack of recruitment of golden perch, a flow pulse of this magnitude resulted in recruitment of native littoral understorey vegetation and germination of river red gums. This was due to the ~0.5 m water level rise in tailwaters directly resulting from environmental water. Higher flow rates (>20,000 ML/d) and/or greater durations may be required to substantially reinstate flowing river characteristics to achieve greater ecological outcomes (e.g. improving riverine production and recruitment of flow-cued spawning fishes) in the LMR.

With existing volumes of environmental water and delivery constraints, during dry years, reaching and sustaining flows >20,000 ML/d in the LMR is largely reliant on coordinating flow deliveries across much of the southern MDB, including flows from the Goulburn, Murrumbidgee, Darling and Murray rivers. Under wetter scenarios, flows >20,000 ML/d may be achieved by delivering environmental water in conjunction with unregulated flows. Nevertheless, in recent years, flows of 10,000–18,000 ML/d during spring have been associated with recruitment of Murray cod. Flows of this volume result in improvements to the extent of lotic habitat in individual weir pools. Murray cod life history operates over these smaller spatial scales (10s of km), and as such, observed recruitment may be associated with improved lotic habitat resulting from environmental water delivery.

Additionally, winter flows from upstream source water (e.g. Murray and Goulburn rivers) can be delivered via return flows from multi-site watering events through the Southern Connected Basin to the LMR, ensuring barrage releases and fishway operations to facilitate spawning migrations of lamprey, among other species. More broadly, winter flows help maintain food resources, habitat and connectivity between habitats. Therefore, they are an important component to reinstating flow regimes to support native fish population recovery in the southern MDB, consistent with the MDBA's 'Water for the environment multi-year priorities' (https://www.mdba.gov.au/issues-murray-darling-basin/water-for-environment/water-environment-multi-year-priorities).

Improving riverine hydraulics (e.g. water velocity and turbulence) is fundamental for ecological restoration in the LMR. Flows of 20,000–45,000 ML/d can significantly improve hydraulic conditions, by transforming >50% of a weir pool from lentic (slower flowing water, median velocities ≤0.3 m/s) to lotic habitat (faster flowing water, >0.3 m/s) (Ye *et al.* 2018). Restoring such hydrodynamic conditions will underpin riverine ecological processes and support the rehabilitation of many declining biota that are adapted to a flowing environment in the LMR (Mallen-Cooper and Zampatti 2017). Pre-regulation, the LMR was characterised by lotic, riverine habitats, with water velocities ranging ~0.2–0.5 m/s, even at flows <10,000 ML/d (Bice *et al.* 2017). With limited volumes of environmental water, infrastructure management, such as weir pool lowering, could be considered to complement flows to achieve hydraulic restoration in this region (Figure 13). Hydraulic modelling suggests that at flows of 15,000 ML/d, lowering weir pools by 1 m could result in a similar extent of flowing water habitat with flows of 20,000 ML/d in the main channel of the LMR. Due to the impact of weirs, the direct influence of small in-channel flow pulses on the inundation area of littoral/riparian zones is limited in the LMR (unless it is via weir poor manipulations). However, in-channel flow pulses supported by environmental water can increase water level variability in the tailwater reaches (below the weirs), and benefit littoral vegetation.

The timing of flow delivery is important and should continue to align with ecological objectives and consider biological processes and species' life history requirements. For example, flow pulses during the spring/summer reproductive season are required to promote recruitment of flow-dependent species (e.g. Murray cod) and stimulate spawning in flow-cued species (e.g. golden perch); winter/spring flow pluses are needed to facilitate spawning migration of diadromous fish (e.g. lamprey); and summer/autumn flows are critical to reduce salinities and maintain water levels in the Coorong. To achieve outcomes for lamprey migration, the delivery of Commonwealth environmental water should be considered during the peak timing of migration for pouched (July–August) and short-headed (September–October) lamprey. Compared to previous years (e.g. 2017-18), the spring flow pulse in 2019-20 occurred earlier (peak in mid-October) in the LMR, coinciding with the spawning season (i.e. September to November) of Murray cod. This may have contributed to the strong recruitment of Murray cod (YOY) in the LMR during 2020, for which current research is exploring key drivers. Whilst the timing of flow is important, a large proportion of environmental water is delivered to the LMR as return flows (e.g. in winter/spring/early summer). A collaborative and coordinated approach to environmental water planning and management across the southern Basin is thus essential. This includes aligning the timing of water delivery to achieve multi-site ecological outcomes.

Overall, environmental water delivery that promotes longitudinal and lateral connectivity will enhance the productivity in the LMR through increased carbon and nutrient inputs, and matter transport. Water delivery, in conjunction with weir pool manipulation, to promote more natural water level changes are desirable to improve in-channel productivity, although the capacity of influence may be limited with current infrastructure and operational arrangements in the LMR. Management approaches that further promote lateral connectivity, such as wetland and floodplain inundation with return flows to the main channel, will deliver secondary productivity (e.g. microinvertebrates) to the LMR. Furthermore, longitudinal connectivity of river flow is important for the transport and dispersal of aquatic biota (e.g. microinvertebrates, fish larvae) to and throughout the LMR. Monitoring during LTIM showed the transport of microinvertebrate species from upstream catchments (e.g. upper Murray, Goulburn, Darling) to the LMR and contributed to diverse communities. Monitoring in 2019-20 also demonstrated increased abundances of microinvertebrates that are floodplain-associated or likely transported from upstream catchments due to improved connectivity (lateral and longitudinal) by environmental flows in the LMR. Increased diversity and abundance of microinvertebrates may improve productivity and community resilience in the LMR, which is important for aquatic food webs and may support diversity in higher trophic organisms (e.g. larval fish).

Managing environmental water releases across broad ‘riverscape’ scales is critical not only to achieve desired flow volumes in the LMR, but also with regard to water quality and the transport of biota. Indeed, water quality (e.g. turbidity, DOC, the amount and form of nutrients) and entrained biota (e.g. plankton, fish larvae/juveniles) may vary between different sources of water (and be dependent on ecological processes occurring at those sources), and thus, flows from different upstream sources can influence ecological outcomes (e.g. recruitment of golden perch or silver perch) in the LMR. Consequently, maintaining flow integrity from upstream (e.g. Darling River or mid-Murray) to the LMR is important to support broad-scale ecological processes and promote positive outcomes (e.g. improved productivity, enhanced lamprey migration, spawning and recruitment of flow-dependent fishes). In this regard, consideration needs to include: (1) maintaining hydrological integrity (i.e. magnitude, variability and source) of flow from upstream; and (2) the potential effects on water quality and biological attributes by river operations that re-route (e.g. through floodplains or wetlands) or fragment the flow (e.g. by diversions or water storages), which could lead to changes in ecological response and the structure and function of aquatic food webs.

During dry years, Commonwealth environmental water is pivotal in maintaining barrage flows and end-of-system connectivity to support a functioning river system and species' life history processes (e.g. migration of diadromous fish). Barrage flows increase salt export out of the Basin, contributing to the Basin Plan salt export objective/target. They also play a critical role in reducing salt import from ocean into the Coorong, lowering salinities and maintaining estuarine habitat (e.g. for *Ruppia*, fish) to support ecological functions and biodiversity. Barrage flows also reduce the risk of Murray Mouth closure.

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7 APPENDICES

APPENDIX A: EXPECTED OUTCOMES OF COMMONWEALTH ENVIRONMENTAL WATER IN THE LOWER MURRAY RIVER, LOWER LAKES AND COORONG

Table A1. Summary of specific watering actions and expected outcomes for the Lower Murray River, Lower Lakes and Coorong in 2019-20 (Source: CEWO). Volumes of Commonwealth environmental water (CEW) are given at the South Australian (SA) border.

Watering action and target	Expected outcomes	Delivery details
Winter pulse (July to August 2019) CEW volume: LMR 178,630 ML, barrages 195,555 ML.	• Fish habitat and condition; • Riverine function; • Lamprey and congolli migration; • Coorong water quality/habitat suitability; and • Increase lake levels in advance of spring pulse.	• Return flows from the Goulburn winter fresh reached South Australia in late July with a pulse above 10,000 ML/d QSA for about five days. Flows tapered off through the first two weeks of August. • As this water reached the lower lakes in early August, additional bays were opened alongside fishways at Tauwitchere and Goolwa to attract and encourage fish species including lamprey and congolli to migrate. Releases were generally around 1,400–5,000 ML/d (gate openings were generally 4 at Tauwitchere, 1 full + 2 partial gates at Goolwa) to end August, with periodic barrage closures due to reverse head conditions. • A single larger 'pulse' totalling 35 GL over the barrages (Goolwa, Mundoo and Tauwitchere) was delivered over three days in early August to stimulate fish responses and improve water quality in the North Lagoon. These releases were undertaken in response to local climatic conditions, taking advantage of anticipated swell and wind impacts to maximise water movement along the North Lagoon. • Lake levels rapidly increased from 0.65 m AHD at the start of July to exceed 0.8 m in mid-August.
Spring pulse (September to November 2019) CEW volume: LMR 344,093 ML, barrages 364,340 ML.	• Coorong water quality; • Estuarine habitat; • Fish habitat and condition; • Riverine function and productivity; • Lamprey and congolli migration; and • Lower lakes fish recruitment.	• Return flows from the Murray and Goulburn coordinated spring freshes began reaching the SA border in mid-September. QSA increased from 5,300 ML/d on 18 September up to a short peak of 11 days above 15,000 ML/d during 12–23 October. • Additional barrages were opened from late September in response to increasing flows from the river and rising lake levels as the spring pulse arrived. During mid-late October up to 37 gates were open, primarily at Tauwitchere and Ewe Island. • The Lower Lakes levels increased from 0.75 m on 24 September to ~0.85 m by the 25 October. • Once the peak had passed, barrage releases were reduced to retain spring pulse water in the lakes, in order to maintain steady low flows to the Coorong for as long as possible into summer.

Watering action and target	**Expected outcomes**	**Delivery details**
Summer base flows in river, fishway flow and connection through barrages (December 2019 to February 2020) CEW volume: LMR 125,553 ML, barrages 14,566 ML.	• Protect assets and avoid damage in the river channel; and • Lower Lakes and Coorong.	• The delivery of 100 GL direct trade commenced in December. This water allowed fishways to remain open however was not sufficient to provide for any additional open gates due to extremely hot and dry conditions causing the lake levels to drop rapidly. • Fishway-only releases were maintained despite the rapidly falling lake levels.
Base flows and opportunistic pulses through the barrages (March to June 2020) CEW volume: LMR 101,804 ML, barrages 110,708 ML.	• Coorong water quality; • Estuarine habitat; • Fish habitat and condition; and • Riverine function and productivity.	• Opportunistic releases to the Coorong (in addition to base releases) were made in April–June in response to water levels and weather conditions. Automated gates at Tauwitchere, Goolwa and Ewe Island were used to deliver pulses of additional water during 'windows' when conditions were most conducive to releases that would benefit the Coorong (ranging from a few hours to several days).
Lower Murray weir pool manipulations (July to October 2019) CEW volume: LMR 352 ML.	Ecological objectives under a 'Moderate' scenario: • Growth and expansion of littoral vegetation including *Juncus, Cyperus gymnocaulos, Schoenoplectus*; • Understorey plant community sustained and productive; • Create diverse and productive biofilm and macroinvertebrate communities; • Provide breeding habitat for small fish (in littoral vegetation) and reed-dependent waterbirds; • Groundwater exchange with river and relieve soil salinity stress in littoral zone; and • Contribute to in-channel flows during drawdown in late spring to assist in delivering water to the Coorong, Lower Lakes and Murray Mouth (CLLMM).	• The weir pool at Lock 2 was raised to 0.52 m above normal pool level during July/August. The fill volume was 5,639 ML. Raising commenced on 13 July and reached the maximum height on 19 August. • Raising was also attempted at Lock 6, commencing on 5 August. However flow rates were insufficient to support the action and after reaching a maximum of 0.28m above normal pool level on 13 August, the pool was returned to normal level by 18 August. 1,502 ML was used in the attempted raising, of which 1,499 was returned in the lowering (3 ML of net losses were accounted against this action). • The weir pool at Lock 2 was lowered from 20 September to reach normal pool level on 23 October. This was slightly earlier than planned to align with the receding spring pulse. The return volume was 5,290 ML (349 ML of net losses were accounted against this action).

APPENDIX B: OVERVIEW OF OTHER MANAGEMENT ACTIVITIES DURING 2019-20

In addition to environmental water deliveries to the Lower Murray in 2019-20 (Section 1.4), the following management actions are relevant to the analyses and interpretations in this report.

Other watering and management activities in the Lower Murray

Manipulation of water levels in Weir Pools 2 and 6

Raising of Weir Pool 2 occurred between early August and mid October 2019. Water levels within were raised to a maximum of 0.50 m above NPL in August/September before undergoing a drawdown in late September (Table A1; Figure B1). Raising of Weir Pool 6 commenced in early August, but curtailed as flow to South Australia declined. Approximately 352 ML of Commonwealth environmental water was delivered to account for losses (e.g. evaporation) during the manipulation of Weir Pools 2 and 6 (source, CEWO).

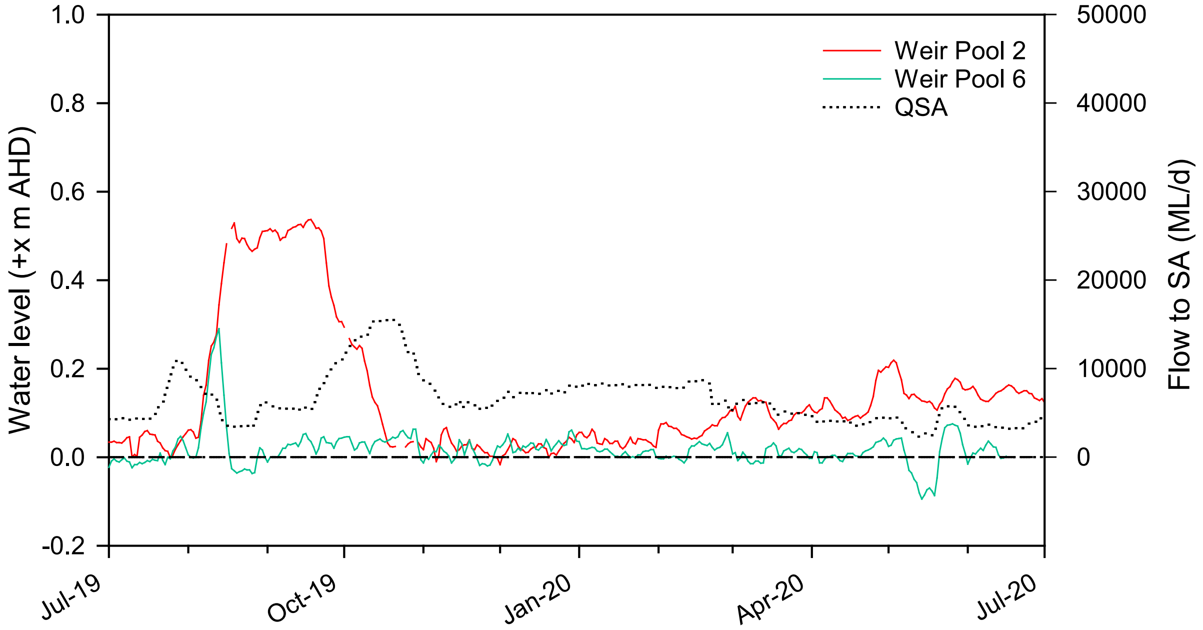

Figure B1. Water levels in the Lock 2 and 6 weir pools in 2019-20, showing weir pool manipulations (DEW). Water levels are measured at Lock 2 US (A4260518) + 6.1 m AHD and Lock 6 US (A4260510) + 19.25 m AHD sites.

Watering and management activities outside of the Lower Murray

Manipulation of water levels in Weir Pools 7, 8, 9 and 15

Water levels in Weir Pools 7, 8, 9 & 15 were raised and lowered, relative to their NPL during 2019-20 (Table B1). The expected outcomes of the raising and lowering are summarised. Approximately 775 ML of Commonwealth environmental water was used to account for 'net' use, i.e. combined loss from raising and savings from lowering for the duration of the environmental watering event and for all weirs involved in the event.

Table B1. Timing of water manipulation actions for weir pools upstream of the Lower Murray Selected Area during 2019-20 (source, MDBA). Raisings are above normal pool level (NPL).

	Weir pool			
	7	8	9	15
Raising details	Raising of weir pool to +60 cm: August to December 2019 Modelled use: 509 ML	Raising of weir pool to +95 cm: September to December 2019 Modelled use: 1,738 ML	Raising of weir pool to +20 cm: August 2019, May 2020 Modelled use: N/A*	Raising of weir pool to +60 cm: August to December 2019 Modelled use: 3,238 ML
Aims of raising	• Provide fast flowing habitat in off-channel creeks for large-bodied native fish species such as Murray cod, golden perch and silver perch.	• Provide temporary flowing water habitat in off-channel creeks to support dispersal, spawning and recruitment opportunities for native fish; • Generate improved biofilm diversity (more types of algae), thereby increasing their value as a food source for micro/macro invertebrates and fish; and • Provide spring flows to cue medium bodied fish spawning and movement.	For anabranches: • Provide refuge habitat; • Maintain fluctuating inundation levels on the littoral zone to improve wetland productivity; and • Control river red gum sapling encroachment.	Inundate high value ecological and cultural wetlands including Margooya Lagoon, Walshes Bend and Bumbang Island, in order to: • Enable feeding, breeding and recruitment opportunities for flood-dependent organisms; and • Promote cycling of carbon and nutrients within the river, floodplains and wetlands.
Lowering details	Lowering of weir pool to -55 cm: December 2019 to April 2020 Modelled gain: 357 ML	Lowering of weir pool to -100 cm: January to May 2020 Modelled gain: 2,335 ML	Lowering of weir pool to -10 cm: February to May 2020 Modelled gain: 1,450 ML	Lowering of weir pool to -30 cm: June-July 2019 and April-May 2020 Modelled gain: 568 ML

	Weir pool			
	7	8	9	15
Aims of lowering	• Increase velocity and hydraulic complexity for native fish; • Stabilise sediment and promote growth of vegetation on exposed floodplain and river banks; and • Export carbon from floodplain and benches to the Murray River.			

*Raising of Lock 9 was operational only in order to maximise diversions to Lake Victoria (not part of the weir pool variability program) and therefore resulted in no modelled 'use'.

Other watering events and management actions

During 2019-20, environmental water was delivered to the LMR channel, primarily as deliveries from return flows through coordinated watering events across the southern connected Basin, to achieve multi-site environmental outcomes. The major upstream watering events that were supported by environmental water and may be relevant to the evaluation in this report are summarised in Table B2. Refer to LTIM annual technical reports for more detail.

Table B2. Details for upstream watering events and management actions supported by environmental water (eWater) in 2019-20. CEW = Commonwealth environmental water, TLM = The Living Murray, VEWH = Victorian Environmental Water Holder, IVT = Inter-Valley Transfer, NSW DPIE = New South Wales Department of Planning, Industry and Environment, RMIF = River Murray Increased Flows.

Event	Event description and timing	Supporting eWater
'Southern Spring Flow'	• Between July and October 2019, nearly 330,000 ML of environmental water was delivered from Hume Dam targeting environmental outcomes along 2,000 km of the Murray River. • This water provided benefits at Barmah-Millewa Forest, Gunbower Creek, Koondrook-Perricoota Forest, Lake Kramen at Hattah Lakes, Chowilla, the Edward-Kolety-Wakool and the Lower Lakes and Coorong, as well as the Murray River channel itself. • Environmental flows targeted up to 15,000 ML/d downstream of Yarrawonga, about half of what would have occurred naturally in winter/spring 2019-20. Based on inflows over winter and spring, flows would have naturally reached about 30,000 ML/day, with overbank flows from July to October 2019. • The flows were designed to support a broad range of outcomes. In the Mid-Murray, this includes providing water to the wetland vegetation in Barmah-Millewa Forest (such as Moira grass), and improving habitat and food availability for native fish, such as Murray cod. • The flows were timed to align with environmental flows from the Lower Goulburn, creating a pulse down the Murray River. This pulse aimed to improve productivity in the river (that is, supporting the river food chain (or food web)).	CEW, TLM, RMIF

Event	Event description and timing	Supporting eWater
Goulburn winter and spring freshes	• Following base flows in June 2019, the winter fresh commenced at Murchison on 01 July 2019 and finished on 1 August 2019. Flows peaked at 9,549 ML/day at Murchison, boosted by natural inflows. At McCoys Bridge the winter flow commenced on 6 July with a lower peak of 8,503 ML/day. • Expected outcome of winter fresh: remove terrestrial vegetation and re-establish flood tolerant native vegetation; inundate benches to encourage plant germination; provide carbon (e.g. leaf litter) to the channel; and improve water quality and waterbug habitat. • The Goulburn spring fresh delivered 135,923 ML from Eildon with the pulse commencing on 21 September and ending 19 October at McCoys Bridge. Before and after the spring pulse there was additional environmental water delivered as part of maintaining increased base flows for the Goulburn River. • Expected outcome of summer fresh: inundate vegetation on benches and the lower banks to facilitate recruitment, sustain growth, and encourage flowering, seed development and distribution.	CEW, TLM, VEWH
Barmah-Millewa Forest inundation	• Given that Barmah forest received the majority of flows in 2018-19, Millewa forest was the priority asset to receive water in 2019-20. The objective was to completely fill the Gulpa Wetlands (Reed Beds Swamp, Coppingers Swamp and Duck Lagoon) to provide optimal bittern nesting habitat over October–November 2019 and into December 2019. • Spring pulse started 1 September 2019 targeting 6+ weeks at 15,000 ML/d downstream of Yarrawonga, with a planned completion date of 28 October 2019 (at Yarrawonga). An estimated 25% of Barmah-Millewa Forest was inundated as part of this flow event. • Forest regulators on the Barmah Forest (Victoria) side of the Murray River were closed on 31 October 2019 to control giant rush encroachment and discourage unwanted colonial bird breeding. A small number of regulators in Millewa Forest (NSW) remained open until 30 November to allow Murray Cod and other native fish to complete their breeding cycle and have time to exit the forest creeks. Environmental water was used to cover additional use in the forest during this period. • Further deliveries proceeded into December 2019 in the Gulpa Creek region (Coppingers swamp, duck lagoon and Reed Beds swamp) to support Australasian Bittern breeding. NSW TLM and NSW PEW covered this additional water use.	CEW, TLM, RMIF, NSW MAA (PEW)

APPENDIX C: DEW EVALUATION QUESTIONS

Table C1. DEW short-term (one-year) and long-term (five-year) evaluation questions for CEWO LTIM/MER indicators. Evaluation questions are based on ecological targets from the Long-Term Environmental Watering Plan (LTWP) for the South Australian Murray River and Basin Plan Matter 8 report. DEW evaluation questions serve as 'additional' questions as there may be some CEWO questions that are also relevant to DEW's targets from the LTWP and/or the Matter 8 report. CEW = Commonwealth environmental water; eWater = environmental water.

Contribution (to what extent CEW contributed towards the outcome, with the significance of the outcome considered):
⬜ Unknown 🟥 Negative ⬜ None/negligible 🟩 Minor 🟩 Moderate 🟦 Substantial

Hydrology (Channel) and Hydraulic Regime (modelling)

DEW evaluation questions	**Outcomes of CEW delivery (2014-15–2019-20)**					
	2014-15	**2015-16**	**2016-17**	**2017-18**	**2018-19**	**2019-20**
To what extent did CEW contribute to meeting the EWRs (all metrics) for the Lower Murray Channel?	**0/0**	**1/0**	**9/9**	**1/0**	**0/0**	**0 EWR met with CEW/0 EWR met without CEW**
	The 10,000 ML/d for 60 days Environmental Watering Requirement (EWR) was met in 2 of the 6 years. Without CEW contributions, this EWR would not have been met in these years. In the 2016-17 high flow year, 9 EWRs were met by the unregulated flow. Total number of EWRs is 7 for the channel + 5 floodplain = 12 (DEWNR 2015).					
To what extent did CEW contribute to meeting the expected outcome for velocity in the Lower Murray? *The majority of lower third weir pools will have median cross-sectional velocities of >0.3 m/s for at least 60 consecutive days between September–March.*	**0/0**	**0/0**	**1/1**	**0/0**	**0/0**	**0 met with CEW/0 met without CEW**
	Assuming significant weir pool manipulations are not occurring, analysis of velocity results and expert elicitation has identified that this evaluation question is met by the 20,000 ML/d for 60 days EWR. This EWR was only met in 2016-17, and in that high flow year the EWR would have been met without CEW contributions.					

Stream Metabolism and Water Quality

DEW evaluation questions	**Outcomes of CEW delivery (2014-15–2019-20)**					
	2014-15	**2015-16**	**2016-17**	**2017-18**	**2018-19**	**2019-20**
To what extent did CEW contribute to managing the concentrations of cyanobacteria?	As the concentrations of cyanobacteria were not measured in the MER Project, the extent to which CEW contributed to managing them is unknown.					
To what extent did CEW contribute to open-water productivity in the Lower Murray?*	1	2	2	2	2	1
	Increased flows generally reduced the volumetric rate of primary production but increased the cross-sectional rate. This increased the overall "carrying capacity" of the river, although the implications of changes in the ratios of these two measures are unknown. At the LMR sites, the percentage increases in cross-sectional GPP due to eWater were negligible due to the largely stable water levels set by weirs. A substantial contribution was considered an increase in cross-sectional GPP of 20% or greater, moderate 11–19%, minor 5–10%, negligible <5%.					
To what extent did CEW contribute to maintaining dissolved oxygen levels above 50% saturation throughout the water column at all times in the Lower Murray?*	0	53	21	50	25	30
	CEW decreased the likelihood of low DO by increasing water mixing and oxygen exchange at the surface. This was assessed as the extra days per year with water velocities > 0.18 m/s due to CEW. A substantial contribution was considered greater than 30 days, moderate 15–30 days, minor 7–14 days and negligible < 7days.					

*Refer to the evaluation in Section 2.2 for details.

Littoral Vegetation Diversity and Productivity

DEW evaluation questions	Outcomes of CEW delivery (2019-20)
To what extent did CEW (and other environmental water) contribute to littoral understorey vegetation diversity and productivity?	Environmental water delivery increased plant species diversity at multiple spatial scales in the LMR. This was evidenced by consistently higher native species richness in inundated zones in each reach compared to non-inundated areas; higher native species richness (albeit lower than inundated zones) on the edge of the inundation footprint, where there was increased soil moisture due to capillary action, compared to non-inundated zones (where soil moisture was not increased) was also observed. Environmental water delivery in the form of a spring pulse produced conditions suitable for the recruitment of specialised riparian species that are adapted to fluctuating water levels and do not inhabit terrestrial or truly aquatic habitats, increasing plant functional diversity. There were no clear patterns of above ground biomass with regards to environmental water delivery; therefore, the influence of environmental water delivery on understorey productivity is inconclusive.

Micro-invertebrate Assemblage

DEW evaluation questions	Outcomes of CEW delivery (2014-15–2019-20)					
	2014-15	2015-16	2016-17	2017-18	2018-19	2019-20
To what extent did CEW contribute to increased dispersal of organisms between river and wetlands in the Lower Murray?	3	22	-3	24	N/A	30
	eWater increased the density of taxa dependent upon lateral connectivity by an average of 18% across all years (with CEW accounting for 86% of this). The significance of the contribution was classified as none-negligible if the contribution was 0–9%, minor = 10–19%, moderate =20–29% and substantial = ≥30%. Refer to the evaluation in Section 2.5 for details.					

Fish (all indicators)

Fish data have been consolidated to evaluate a number of fish targets of DEW's LTWP. These questions and answers do not relate to evaluation of flow or Commonwealth environmental water. Furthermore, the LTIM Fish monitoring program is not designed to determine what is facilitating changes in population dynamics of fish species for DEW's LTWP evaluation questions, e.g. spawning and recruitment of freshwater catfish or common carp. NA = not applicable.

Outcome: 🟥 = negative; 🟩 = positive; ▨ = unable to be detected.

DEW evaluation questions	**Answers to evaluation questions (2015 to 2020)**					
	15	**16**	**17**	**18**	**19**	**20**
Did the population age structure of Murray cod include recent recruits (i.e. <300 mm TL), sub-adults (300–600 mm) and adults (>600 mm) in the Lower Murray?	X	√	√	√	√	√ (Figure 55)
Did the length-frequency distribution for Murray cod indicate a large recruitment event, demonstrated by a YOY cohort representing >50% of the population from the Lower Murray?	X	X	X	X	√	√ (67%, Figure 55)
Did the abundance of Murray cod in the Gorge zone increase by ≥20% over a 5-year period?	NA	NA	NA	NA	√	√ (269%, Figure 58)
Did the population age structure of golden perch include adults (age ≥4+) and sub-adults (age 1+–3+)?	√	√	√	√	√	√ (Figure 48)
Was there a large recruitment event of golden perch, demonstrated by a YOY cohort representing >30% of the population from the Lower Murray.	X	X	X	X	X	X (Figure 48)
Did the abundance of golden perch in the Gorge zone increase by >30% over a 5-year period?	NA	NA	NA	NA	X	X (-50%, Figure 58)
Did the abundance of silver perch in the Gorge zone increase by >30% over a 5-year period?*	NA	NA	NA	NA	X	X (-40%, Figure 58)
Did the abundance of freshwater catfish in the Gorge zone increase by ≥30% over a 5-year period?*	NA	NA	NA	NA	X	X (-29%, Figure 58)

DEW evaluation questions	**Answers to evaluation questions (2015 to 2020)**					
	15	**16**	**17**	**18**	**19**	**20**
Did the length-frequency distribution for bony herring** in the Gorge zone include size classes representing YOY?	NA	√	√	√	√	√
Did the length-frequency distribution for Murray rainbowfish and carp gudgeon, include size classes representing YOY in the Gorge zone?	√	√	√	√	√	√
Did the relative abundance of common carp in the Gorge zone increase during the current year, relative to the previous year, in the absence of an increase in the relative abundances of flow-dependent native species decreased?***	NA	√	√	X	X	X (Figure 58)
Did the estimated biomass of common carp in the Gorge zone increase during the current year, relative to the previous year, in the absence of an increase in the estimated biomass of flow-dependent native species decreased?***	NA	√	X	X	X	X

* These results should be interpreted with caution, given the large error estimate.

** Bony herring were not assessed as a target species during 2014.

*** In the 2019-20 report, the calculations used to answer the evaluation question were changed from an approach using ratios to better reflect the ecological target of the LTWP. To remove sampling season bias, only sites sampled during autumn 2017 were used in comparisons against 2018. Common carp were not weighed as part of the Fish (channel) sampling, so biomass was estimated by converting fork lengths to weights based on a FL–mass equation in Vilizzi and Walker (1999).

APPENDIX D: SUPPLEMENTARY INFORMATION FOR INDICATORS

Matter Transport and Coorong Habitat

Methods

The contribution of environmental water to the transport of salt, nutrients and phytoplankton was assessed with a coupled hydrodynamic–biogeochemical model for the reach below Lock 1 to the Murray Mouth. Salt, nutrient and phytoplankton transport was predicted for three different flow scenarios: with all environmental water (i.e. the observed flow), flow without Commonwealth environmental water, and flow without any environmental water (i.e. counter-factual simulations assessing what would have happened if flows were not augmented with environmental water).

When modelling, it is necessary to make assumptions on the relationships between flow and nutrients or salt, nutrient dynamics in sediments and floodplain habitats, and the utilisation of nutrients by phytoplankton. This leads to a degree of uncertainty in model outputs; however, given previous model development and validation initiatives over the past decade, it is considered that this uncertainty is within reasonable bounds (Aldridge *et al.* 2013) and the results can be used to reliably assess the general response attributable to environmental water.

Water quality sampling and analyses

Water temperature, electrical conductivity, dissolved oxygen, pH and turbidity were monitored in the Murray River Channel (at Morgan) between July 2019 and June 2020. In addition, integrated-depth water samples were collected and sent to the Australian Water Quality Centre, a National Association of Testing Authorities accredited laboratory. Samples were analysed for filterable reactive phosphorus (hereafter referred to as phosphate), total phosphorus (TP), combined nitrate and nitrite (NOx), ammonium, total Kjeldahl nitrogen (TN), dissolved silica and chlorophyll *a* using standard techniques. Organic nitrogen was calculated as the difference between total Kjeldahl nitrogen and ammonium. These nutrient concentrations are representative of conditions in the river and Coorong, however, salinity was measured more frequently in the river and Coorong enabling use of this data for the 2019-20 modelling and analysis.

Hydrodynamic–biogeochemical modelling

The model platform used to assess the effects of environmental water delivery on salt and nutrient transport was the coupled hydrodynamic–biogeochemical model "TUFLOW-FV – AED" (referred to as FV-AED for short), developed by BMT Global Pty Ltd. and the University of Western Australia. TUFLOW-FV has been used extensively in the region for hydrological assessments and was previously used to assess the contribution of environmental water to dissolved and particulate matter for water years 2013-14 to 2018-19 (Ye *et al*. 2016a; 2020).

In this assessment, two model domains were applied spanning: (1) Lock 1 to the Southern Ocean, including the Coorong (Figure 23), and (2) a high-resolution Coorong only model (described further below). The TUFLOW-FV model adopts an unstructured-mesh to resolve spatio-temporal changes in water velocity, temperature and salinity dynamics, in response to changing meteorological and inflow conditions. Superimposed on the hydrodynamics, the Aquatic EcoDynamics (AED) water quality modules were configured to simulate the dynamics of light, oxygen, nutrients, organic matter, turbidity and phytoplankton. Both model domains mentioned above were configured to simulate the same hydrologic and biogeochemical processes, however the Coorong only model had a higher resolution mesh for better resolving the water quality conditions, and it was also required for the habitat assessment of *Ruppia* and the various fish species of interest (Section 2.3.2).

The first model runs with the full domain were initialised with data from a range of sources. Inflow data (Lock 1), used to drive the main river domain, were provided by the MDBA for the three scenarios (Figure 24), i.e. with all environmental water ("***All water***", representing observed conditions), without Commonwealth environmental water ("***no CEW***"), and without any environmental water ("***no eWater***"). These simulations extend previous model runs that started 1 July 2014 (Ye *et al*. 2020), and were run for this assessment for the period between 1 July 2019 and 30 June 2020.

Additional flow specifications for SA Water off-takes were also included. Irrigation return flows were assumed to be negligible over this period and were not included in the model. Similarly, flows from Eastern Mount Lofty Ranges were not included since their contribution to the Lower Lakes was considered to be relatively minor (Cook *et al.* 2010). Meteorological conditions were based on data from Narrung. Between Lake Alexandrina and the Coorong, five barrages were included (Goolwa, Mundoo, Boundary Creek, Ewe Island and Tauwitchere) and set with a spill-over height of 0.72 m AHD. The barrage operation was set to include gate operation based on operational information provided through discussions with representatives of DEW. At the bottom of the domain, two open boundaries were specified, one at the Murray Mouth and one at Salt Creek. Murray Mouth water level was based on Victor Harbor tidal data, which were available at 10 minute resolution. Salt Creek flow data were set based on available flow data from the WaterConnect website (DEW).

Water quality conditions for both boundary points were set based on a linear interpolation of the measured nutrient and salinity data used for this study. Water quality conditions for the river inflow at Lock 1 were determined based on interpolation of available data from Lock 1 or Morgan. For water quality properties for the without environmental water scenarios, rating curves were developed for flow and concentration. Based on the daily flow difference, a scaled concentration was estimated for water quality parameters including salinity, phosphate, ammonium, nitrate, total nitrogen and silica. The physico-chemical information at other sites was used to validate the model.

The influence of environmental water on the concentrations of matter was assessed through a comparison of modelled concentrations for the various scenarios for the Barrages and Coorong. Modelled concentrations are presented as medians of modelled cells within areas surrounding sampling sites (Figure 25). A range in concentrations within those cells is also presented for the 'All water' scenario.

The transport of matter was assessed through modelled exports from the Murray River Channel (Wellington), Lower Lakes (Barrages) and Coorong (Murray Mouth). Findings are presented for salinity, ammonium, phosphate, dissolved silica, organic nitrogen, organic phosphorus and chlorophyll *a*. Salinity is presented as practical salinity units (PSU), a measurement of the measured conductivity to standard potassium chloride (KCl) conductivity. PSU was used for validating model outputs as it overcomes observed differences in electrical conductivity caused by changes in water temperature. One PSU is approximately equal to one part per thousand.

The inflow data that were used to drive the main river domain are treated as indicative only as they do not account for all complexities associated with water accounting, water attenuation through the system and different management decisions that may have been made if the volume of environmental water provided had not been available (Neville Garland, MDBA, pers. comm.). Assumptions made to address these complexities result in uncertainty in the model outputs and so outputs are not to be treated as absolute values (refer to Aldridge *et al.* 2013 for more detail). When assessing the relative differences between scenarios, the uncertainties are considered to influence the accuracy of each scenario equally and so the model outputs are used to assess the general response to environmental water delivery.

Given the increasing efforts to improve and restore the Coorong, a high-resolution model of the Coorong was developed, which is considered to be more accurate in resolving the salt, nutrients and habitat across the system. The Coorong only domain (Figure 26) was run under the same scenarios above, but due to the long residence time, these simulations were run from 1 July 2017 to 30 June 2020, i.e., a period of three years; this window was chosen as it began after the high flow event in 2016 which "reset" salinity levels in the Coorong. The simulation included gauged Salt Creek inputs and measured ocean water levels based on the Barker Knoll telemetered site. Weather data were also used as a boundary condition, in order to predict the effect of wind and evapo-concentration effects. Because of the importance of salinity movement in this system, a detailed salt and nutrient flux analysis was undertaken to understand the rate of salt and nutrient accumulation in both the North and South of the Coorong.

Rather than predicting barrage flows based on water levels either side of the structures, as done in the full domain model, this simulation specifies the daily barrage flows based on flow volumes obtained from SA Water (Figure D1). This figure gives context to the period of focus (1 July 2017 to 30 June 2020), highlighting that it was a period of low flow to the Coorong overall.

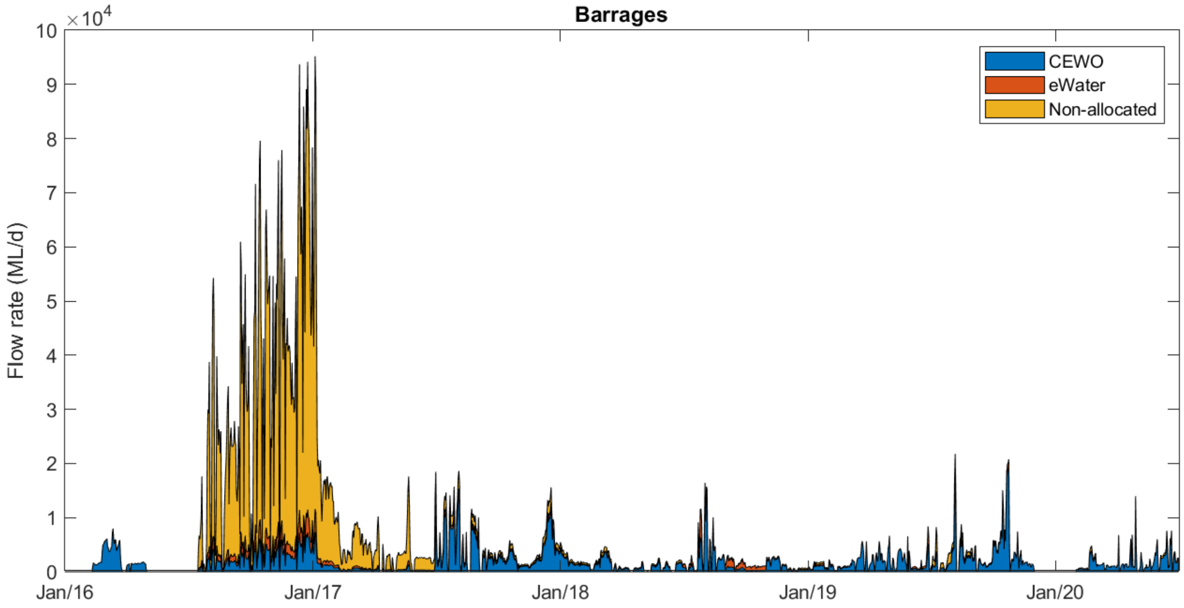

Figure D1. Summary of history of flow over the barrages, from 2016 until mid-2020, spanning the large 2016 flow event, categorised based on water source.

Analysis of the flow over the barrages for the three-year simulation period is shown in more detail in Figure D2. As the data provided for the environmental water accounting do not resolve details of specific barrages and are based on estimates, we had to: (1) understand how to distribute the water through the different barrage reaches, and (2) ensure the predicted amounts from the Commonwealth environmental water water balance modelling were consistent with local estimates of barrage flow. Actual estimates of daily barrage flow from each barrage were obtained from SA Water and DEW, and show the dominance of flows through Goolwa and Tauwitchere over the period of interest, with some minor pulse through Mundoo (Figure D2a).

We then compared the actual total discharge through the five barrages with the estimate from Commonwealth environmental water water balance models (Figure D2b), and for the former we assigned the fractions of environmental water contributions as specified in the Lock 1 flow data. Whilst there was a simulative difference (bias) between the Commonwealth environmental water data and the SA Government data, the difference was relatively minor. This is further summarised into monthly flow totals (Figure D2c), indicating the flow categories and the nominal percentage of Commonwealth environmental water contribution to the barrage flow for each method.

This provides an independent check on the two sources and methods of flow data estimation, and allows us to combine the data about water source contribution and which specific barrages were active. The analysis highlights that Commonwealth environmental water dominated the barrage flows for this period, in many cases making up 100% of the specified flux into the Murray estuary and Coorong. For the final model simulations of the Coorong, we used the actual SA Government flow data over each of the barrages (called the "Base-case" simulation), and then undertook the following: Scenario 1: No environmental water across the barrages (essentially equating to no barrage flow); and Scenario 2: Observed conditions for 2017-18 and 2018-19 (as the Base-case), and no environmental water for the 2019-20 water year.

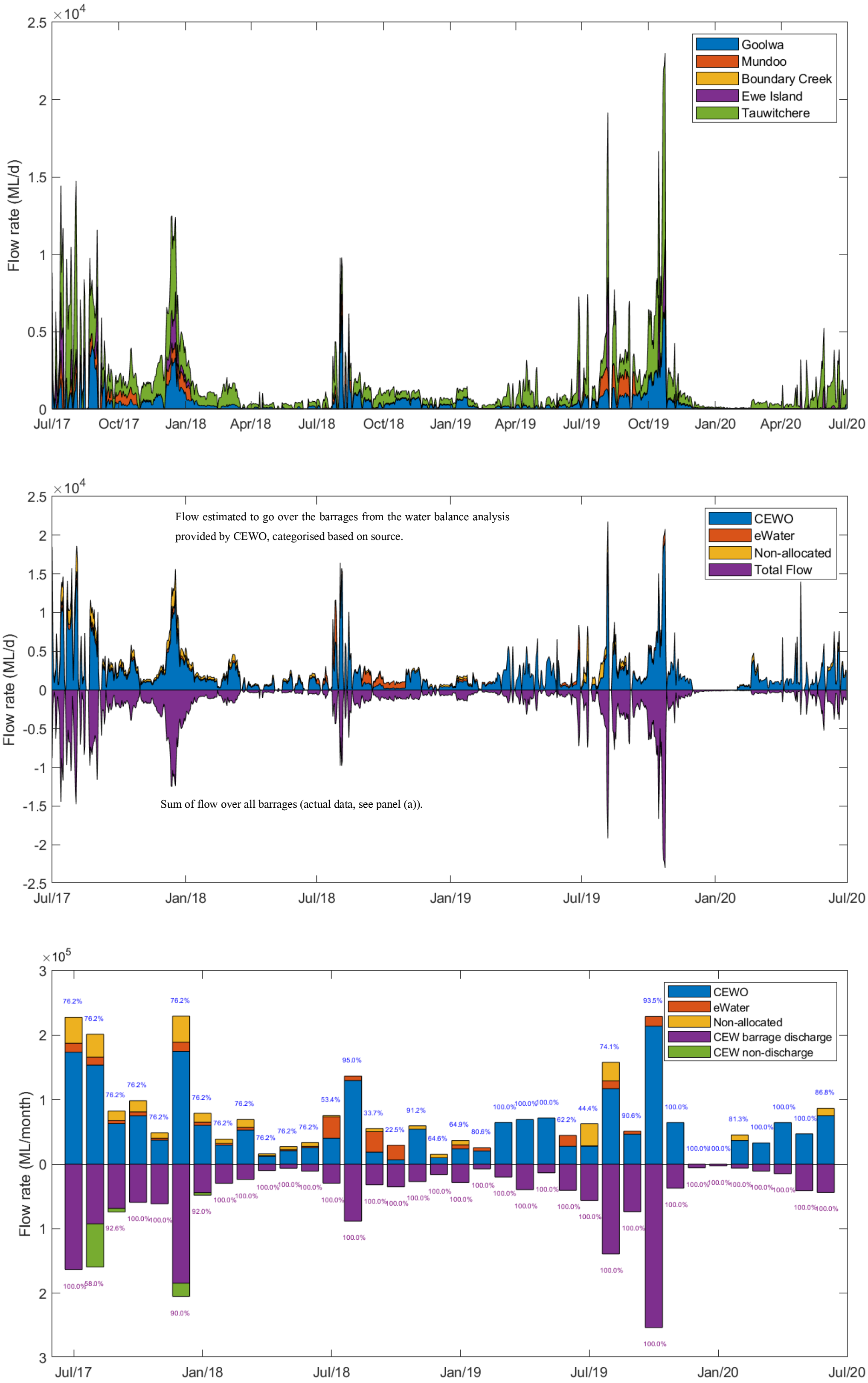

Figure D2. Summary of barrage flow volumes, categorised based on flow per gate (a, top) and based on water source (b, middle). The monthly summary of e-water fractions flowing over the barrages is shown in (c, bottom).

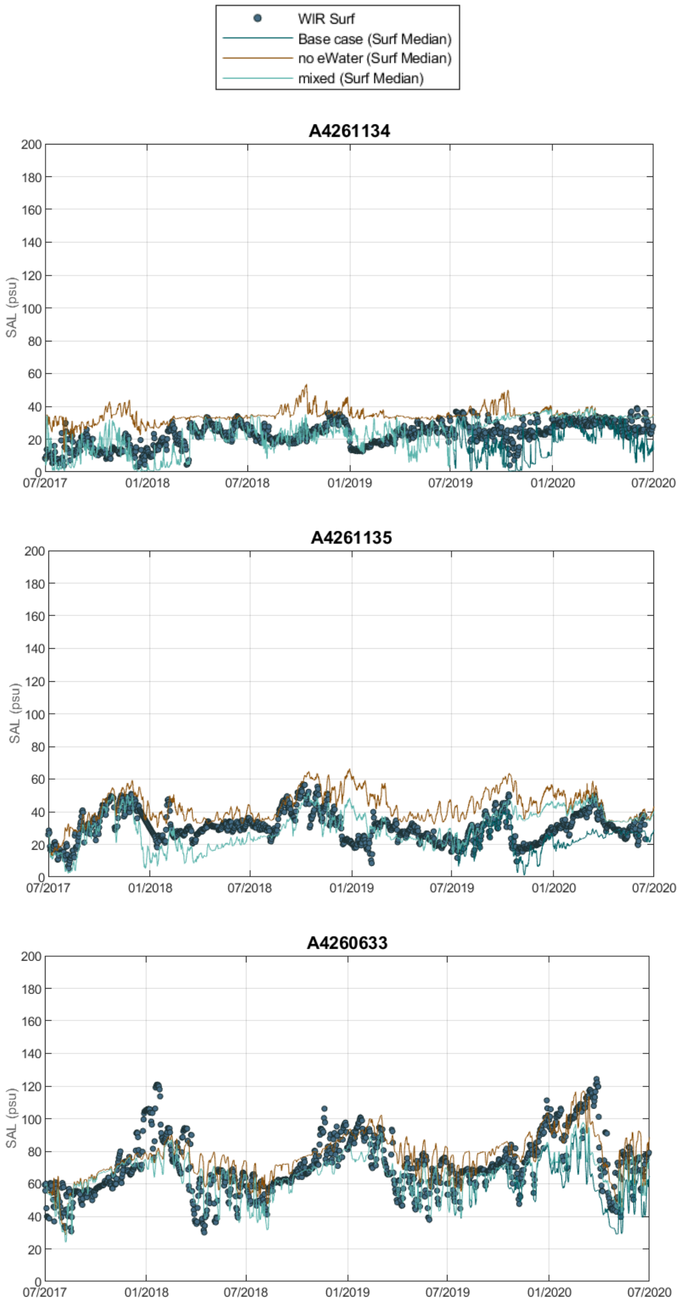

Figure D3. Comparison of measured and simulated salinity at key monitoring points within the Coorong lagoon, moving from the Murray Mouth into the South Lagoon. Model simulations for the Base-case (observed conditions), and Scenario 1 (no eWater) and Scenario 2 (mixed) are shown. A4261134 = Pelican Point, A4261135 = Long Point, A2460633 = Parnka Point.

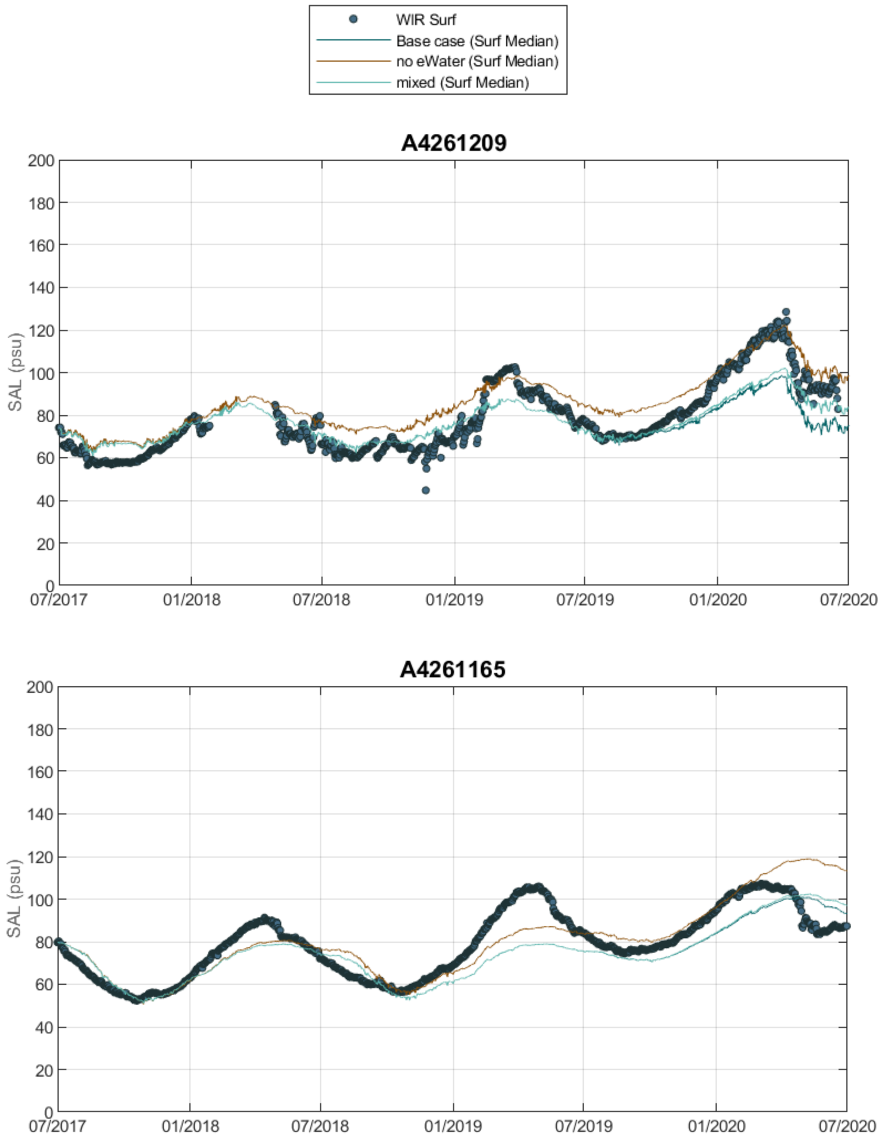

Figure D4. Comparison of measured and simulated salinity at key monitoring points within the Coorong lagoon, moving from the Murray Mouth into the South Lagoon. Model simulations for the Base-case (observed conditions), and Scenario 1 (no eWater) and Scenario 2 (mixed) are shown. A4261209 = Cattle Island, A4261165 = Snipe Island.

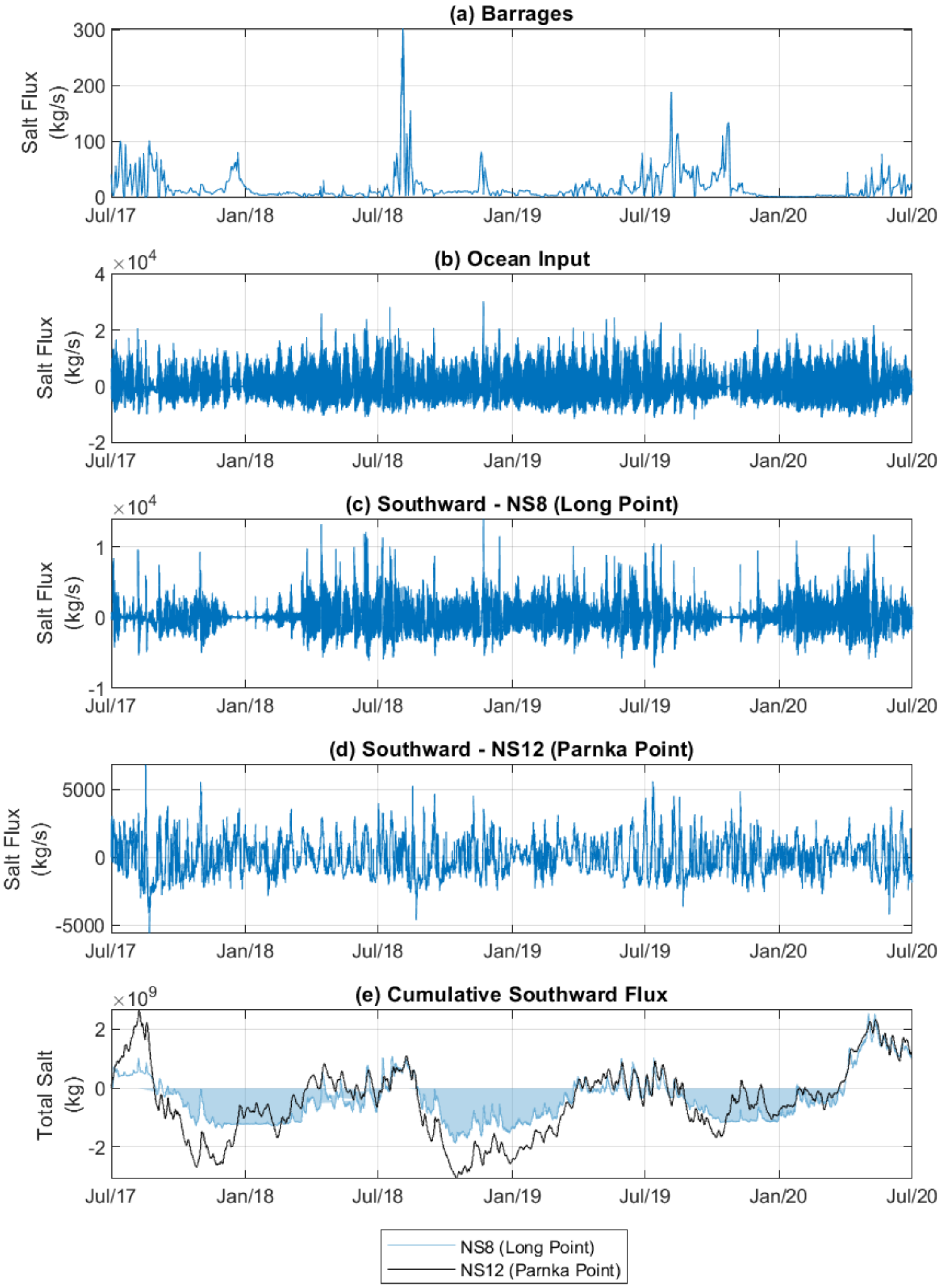

Figure D5. Detailed salt flux analysis at five locations in the Coorong (a-d) in the base-case scenario, which has environmental water flow over the barrages each year (see Figure D2). Panel (e) shows the cumulative salt flux into the North Lagoon (past Long Point) and into the South Lagoon (past Parnka Point); the shaded blue area shows salt export from the Coorong towards the river mouth.

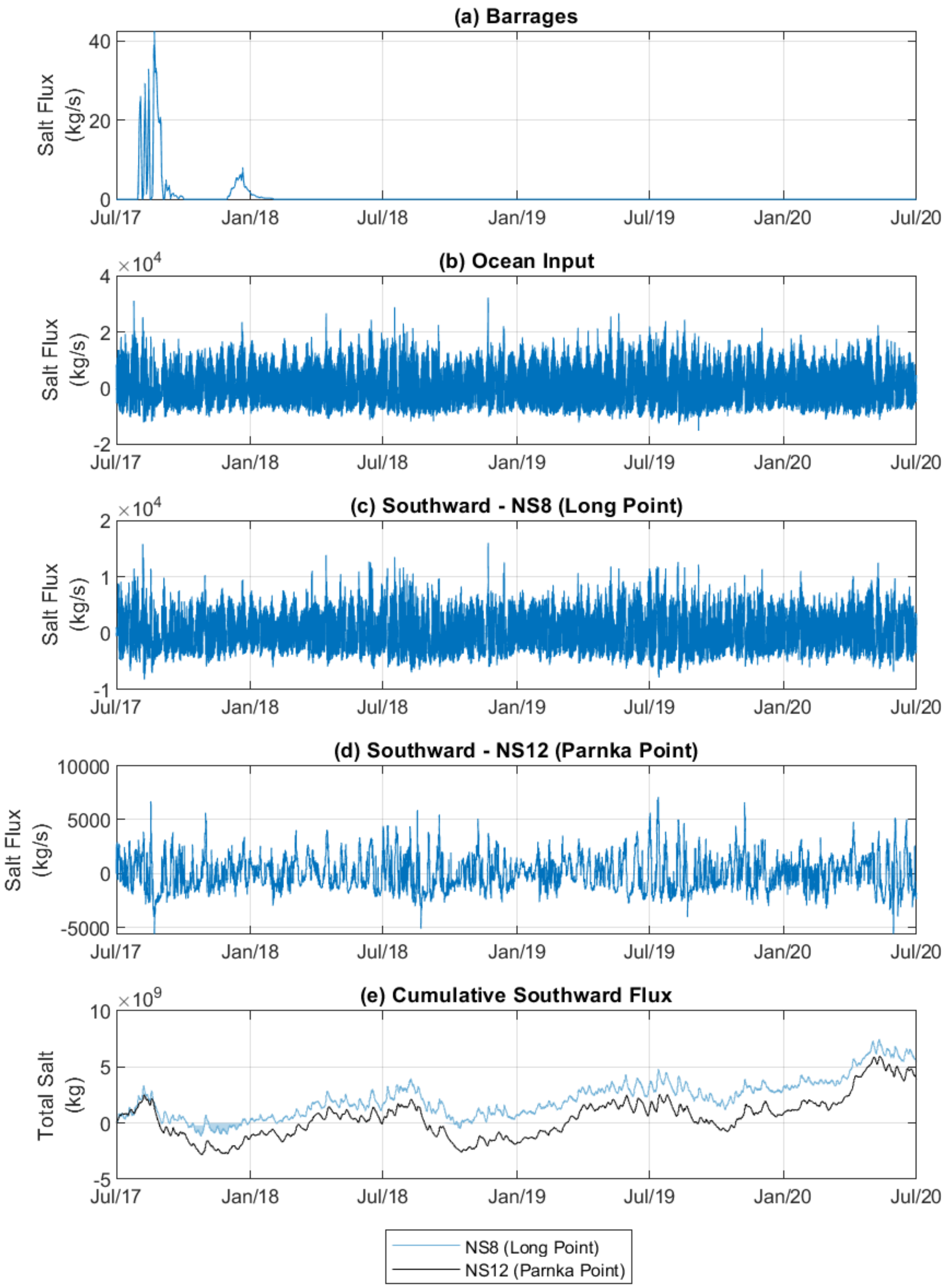

Figure D6. Detailed salt flux analysis at five locations in the Coorong (a-d) for Scenario 1, which has no environmental water flow over the barrages each year. Panel (e) shows the cumulative salt flux into the North Lagoon (past Long Point) and into the South Lagoon (past Parnka Point); the minimal shaded blue area shows salt is always moving into the Coorong towards the South Lagoon, and is much larger than in the base-case (Figures D3 and D4).

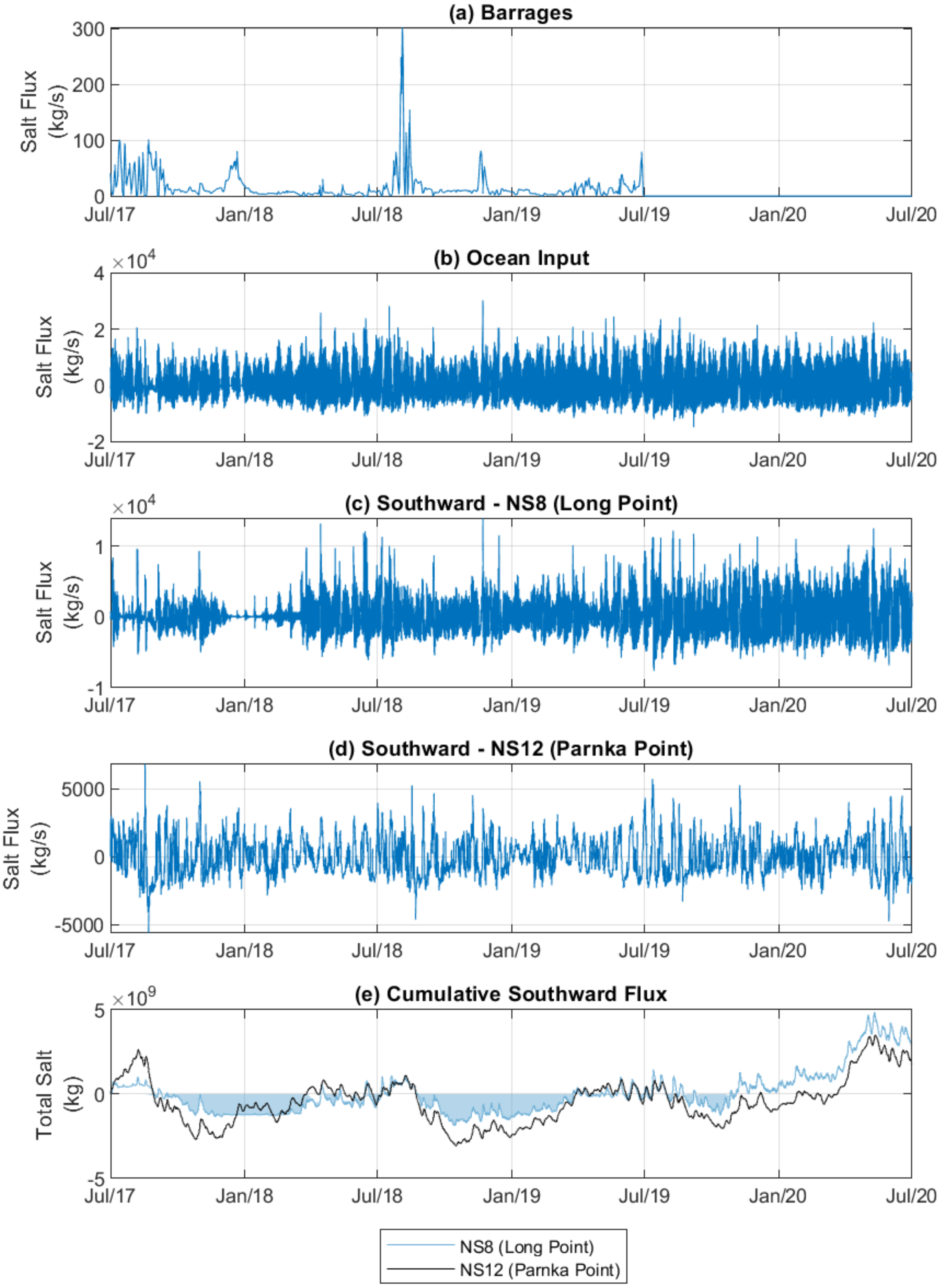

Figure D7. Detailed salt flux analysis at five locations in the Coorong (a-d) for Scenario 2, which has environmental water flow for 2 years, and then no flow over the barrages in 2019-20. Panel (e) shows the cumulative salt flux into the North Lagoon (past Long Point) and into the South Lagoon (past Parnka Point); the minimal shaded blue area in 2019-20 shows salt begins to move towards the South Lagoon.

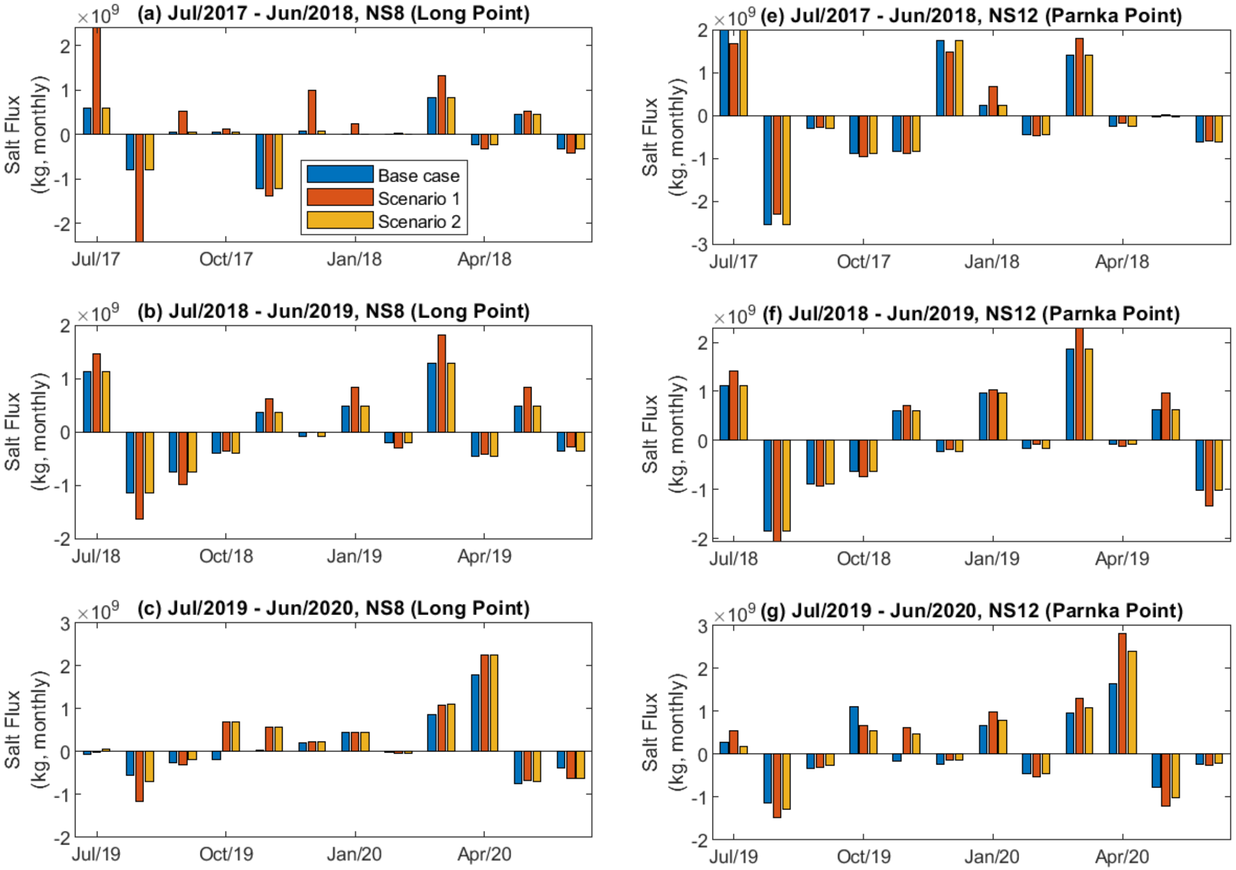

Figure D8. Monthly salt exports with and without environmental water delivery for July 2017–July 2020. Scenarios include with all water, without Commonwealth environmental water (no CEW) and without any environmental water (no eWater). eWater delivery maintained this flux to be close to zero over the period of interest; even one year of no ewater over the barrages contributes to salt accumulation in the North and South Lagoon.

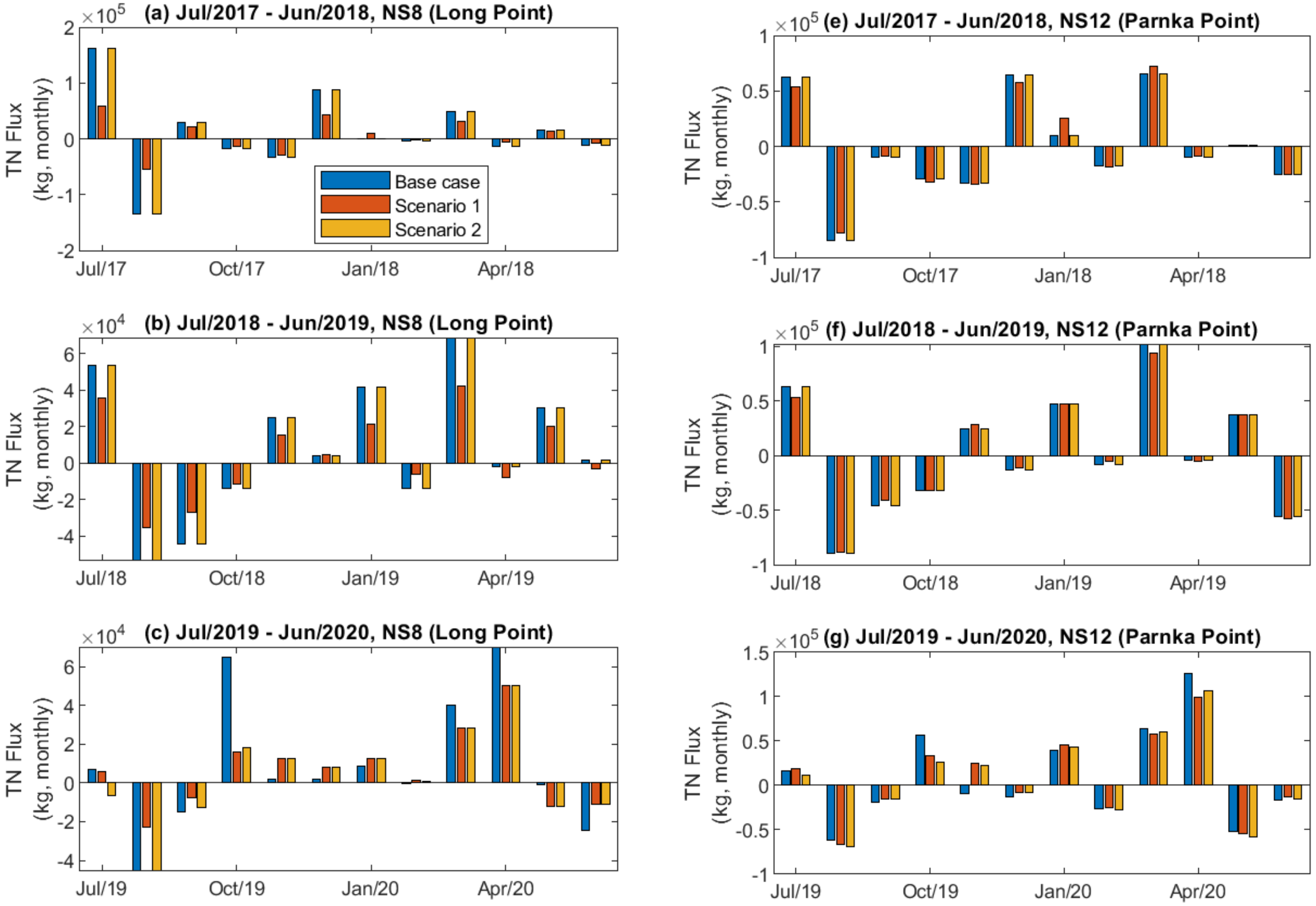

Figure D9. Nitrogen flux in the Coorong with and without environmental water delivery for July 2017–July 2020. Scenarios include with all water (Base-case), without Commonwealth environmental water (no eWater; Scenario 1) and without any environmental water for 2017-18 and 2018-19 water years (Scenario 2).

Littoral Vegetation Diversity and Productivity

Table D1. GPS coordinates for the lowest elevation of each transect.

Reach	Transect	Latitude	Longitude
Lock 1	1	-34.479019	139.596311
Lock 1	2	-34.477575	139.598798
Lock 1	3	-34.446681	139.609714
Lock 1	4	-34.419635	139.613482
Lock 1	5	-34.41855	139.61395
Lock 1	6	-34.391968	139.619028
Lock 4	1	-34.361848	140.565262
Lock 4	2	-34.355854	140.576385
Lock 4	3	-34.355763	140.568069
Lock 4	4	-34.350657	140.562584
Lock 4	5	-34.343043	140.553056
Lock 4	6	-34.342492	140.554519
Lock 6	1	-34.021533	140.867416
Lock 6	2	-34.01918	140.876627
Lock 6	3	-34.016288	140.88713
Lock 6	4	-33.997601	140.879789
Lock 6	5	-33.995581	140.880611
Lock 6	6	-33.994623	140.882183

Table D2. Species list, functional classification, life history strategy, conservation status (state conservation status from listings in Barker *et al.* (2005) (*denotes exotic species, **denotes proclaimed pest plant in South Australia, # denotes listed as rare in South Australia).

Species	**Family**	**Status**	**Life history strategy/growth form**	**Functional Group**
Acacia stenophylla	Fabaceae	Native	Perennial tree	Amphibious
Alternanthera denticulata	Amaranthaceae	Native	Annual herb	Flood dependent
Ammannia multiflora	Lythraceae	Native	Annual herb	Flood dependent
*Apium graveolens**	Apiaceae	Exotic, Naturalised	Annual herb	Terrestrial
Atriplex suberecta	Chenopodiaceae	Native	Annual herb	Flood dependent
Bolboschoenus caldwellii	Cyperaceae	Native	Perennial sedge	Emergent
Brachyscome paludicola	Asteraceae	Native	Annual herb	Flood dependent
Callistemon brachyandrus#	Myrtaceae	Native, Rare in South Australia	Perennial shrub	Amphibious
*Centaurea calcitrapa**	Asteraceae	Exotic, Naturalised	Annual herb	Terrestrial
Centipeda minima	Asteraceae	Native	Annual herb	Flood dependent
Cyperus gymnocaulos	Cyperaceae	Native	Perennial sedge	Amphibious
*Dittrichia graveolens**	Asteraceae	Exotic, Naturalised	Annual herb	Terrestrial
Duma florulenta	Polygonaceae	Native	Perennial shrub	Amphibious
Einadia nutans	Chenopodiaceae	Native	Perennial sub-shrub	Terrestrial
Eleocharis acuta	Cyperaceae	Native	Perennial sedge	Emergent
Enchylaena tomentosa	Chenopodiaceae	Native	Perennial sub-shrub	Terrestrial
*Erigeron bonariensis**	Asteraceae	Exotic, Naturalised	Annual herb	Terrestrial
Eucalyptus camaldulensis	Myrtaceae	Native	Perennial tree	Amphibious
Euphorbia drummondii	Euphorbiaceae	Native	Annual herb	Flood dependent
*Gazania rigens***	Asteraceae	Exotic, Declared Pest Plant in South Australia	Perennial herb	Terrestrial
Glinus lotoides	Aizoaceae	Native	Annual herb	Flood dependent
Glycyrrhiza acanthocarpa	Fabaceae	Native	Annual herb	Flood dependent
Haloragis aspera	Haloragaceae	Native	Perennial herb	Flood dependent
*Heliotropium curassavicum**	Boraginaceae	Exotic, Naturalised	Annual herb	Terrestrial

Species	**Family**	**Status**	**Life history strategy/growth form**	**Functional Group**
*Heliotropium europaeum**	Boraginaceae	Exotic, Naturalised	Annual herb	Terrestrial
Isoetopsis graminifolia	Asteraceae	Native	Annual herb	Flood dependent
Isolepis australiensis	Cyperaceae	Native	Perennial herb	Amphibious
Juncus usitatus	Juncaeae	Native	Perennial rush	Amphibious
Lachnagrostis filiformis	Poaceae	Native	Annual grass	Flood dependent
Limosella australis	Scrophulariaceae	Native	Perennial herb	Flood dependent
Ludwigia peploides	Onagraceae	Native	Perennial herb	Amphibious
Lythrum hyssopifolia	Lythraceae	Native	Annual herb	Flood dependent
Maireana	Chenopodiaceae	Native	Perennial sub-shrub	Terrestrial
*Medicago**	Fabaceae	Exotic, Naturalised	Annual herb	Terrestrial
Melaleuca lanceolata	Myrtaceae	Native	Perennial tree	Terrestrial
*Melilotus albus**	Fabaceae	Exotic, Naturalised	Annual herb	Terrestrial
*Melilotus indicus**	Fabaceae	Exotic, Naturalised	Annual herb	Terrestrial
Myoporum montanum	Myoporaceae	Native	Perennial tree	Terrestrial
Myriophyllum papillosum#	Haloragaceae	Native, Rare in South Australia	Perennial herb	Amphibious
*Oxalis**	Oxalidaceae	Exotic, Naturalised	Perennial herb	Terrestrial
Paspalidium jubiflorum	Poaceae	Native	Perennial grass	Flood dependent
Paspalum distichum	Poaceae	Native	Perennial grass	Amphibious
Persicaria lapathifolia	Polygonaceae	Native	Perennial herb	Amphibious
Phragmites australis	Poaceae	Native	Perennial grass	Emergent
Phyllanthus lacunarius	Euphorbiaceae	Native	Annual herb	Flood dependent
Picris angustifolia	Asteraceae	Native	Annual herb	Terrestrial
Polygonum plebeium	Polygonaceae	Native	Annual herb	Flood dependent
Pseudognaphalium luteoalbum	Asteraceae	Native	Annual herb	Flood dependent
Rhodanthe pygmaea	Asteraceae	Native	Annual herb	Flood dependent
*Rorippa palustris**	Brassicaeae	Exotic, Naturalised	Annual herb	Flood dependent
Schoenoplectus pungens	Cyperaceae	Native	Perennial sedge	Amphibious
Schoenoplectus tabernaemontani	Cyperaceae	Native	Perennial sedge	Emergent
Sclerolaena divaricata	Chenopodiaceae	Native	Perennial sub-shrub	Terrestrial
Senecio cunninghamii	Asteraceae	Native	Perennial shrub	Flood dependent
Senecio runcinifolius	Asteraceae	Native	Perennial herb	Flood dependent

Species	**Family**	**Status**	**Life history strategy/growth form**	**Functional Group**
*Silene nocturna**	Caryophyllaceae	Exotic, Naturalised	Annual herb	Terrestrial
Sphaeromorphaea australis	Asteraceae	Native	Annual herb	Flood dependent
Sporobolus mitchellii	Poaceae	Native	Perennial grass	Flood dependent
Stemodia florulenta	Scrophulariaceae	Native	Perennial herb	Flood dependent
*Symphyotrichum subulatum**	Asteraceae	Exotic, Naturalised	Annual herb	Flood dependent
Teucrium racemosum	Lamiaceae	Native	Perennial herb	Flood dependent
Typha domingensis	Typhaceae	Native	Perennial sedge	Emergent
Wahlenbergia fluminalis	Campanulaceae	Native	Annual herb	Flood dependent
*Xanthium occidentale***	Asteraceae	Exotic, Declared Pest Plant in South Australia	Annual herb	Amphibious

Table D3. Indicator Species Analysis results comparing plant community in each inundation zone at a. Lock 1, b. Lock 4 and c. Lock 6 in December 2019, * denotes exotic species, ** denotes declared pest plant in South Australia, # denotes listed as rare in South Australia; yellow highlighting denotes significant indicator; blue highlighting denotes not significant but exclusive to the Pool level and Inundated zones.

a.

Species	Zone	P
Acacia stenophylla	*Not inundated*	0.6593
Alternanthera denticulata	*Inundated*	0.3179
Ammannia multiflora	*Inundated*	1
*Apium graveolens**	*Inundation extent*	0.3377
Bolboschoenus caldwellii	*Pool level*	0.3379
Brachyscome paludicola	*Not inundated*	0.8874
Callistemon brachyandrus#	*Not inundated*	0.6583
Centaurea calcitrapa	*Not inundated*	0.2791
Centipeda minima	*Pool level*	0.0158
Cyperus gymnocaulos	*Pool level*	0.8368
Duma florulenta	*Inundated*	0.8222
Einadia nutans	*Inundation extent*	0.8868
Eleocharis acuta	*Pool level*	0.001
*Erigeron bonariensis**	*Inundated*	1
Eucalyptus camaldulensis	*Not inundated*	0.6807
Euphorbia drummondii	*Not inundated*	0.6029
*Heliotropium curassavicum**	*Inundation extent*	0.4005
Juncus usitatus	*Pool level*	0.0076
Lachnagrostis filiformis	*Pool level*	0.4945
Limosella australis	*Pool level*	0.3707
Ludwigia peploides	*Pool level*	0.3379
Lythrum hyssopifolia	*Pool level*	0.0416
Maireana	*Inundated*	1
*Medicago**	*Inundated*	1
*Melilotus albus**	*Not inundated*	0.6575
*Melilotus indicus**	*Inundation extent*	0.7459
Myoporum montanum	*Not inundated*	0.6331
Paspalum distichum	*Pool level*	0.002
Phragmites australis	*Pool level*	0.4627
Picris angustifolia	*Inundation extent*	0.7179
Rhodanthe pygmaea	*Not inundated*	0.3529
Schoenoplectus pungens	*Pool level*	0.3317
Schoenoplectus tabernaemontani	*Inundated*	1
Senecio cunninghamii	*Inundation extent*	0.1756
Senecio runcinifolius	*Inundated*	0.3519
Sphaeromorphaea australis	*Pool level*	0.22
Sporobolus mitchellii	*Not inundated*	0.2557

Species	**Zone**	**P**
Stemodia florulenta	*Inundation extent*	0.7782
*Symphyotrichum subulatum**	*Pool level*	0.0024
Teucrium racemosum	*Not inundated*	0.5425
Wahlenbergia fluminalis	*Not inundated*	0.6753
*Xanthium occidentale***	*Inundated*	0.0352

b.

Species	**Zone**	**P**
Alternanthera denticulata	*Inundated*	0.0974
Atriplex suberecta	*Not inundated*	0.1386
Bolboschoenus caldwellii	*Pool level*	0.6365
Brachyscome paludicola	*Not inundated*	0.0686
Centipeda minima	*Inundated*	0.1578
Cyperus gymnocaulos	*Inundation extent*	0.1354
*Erigeron bonariensis**	*Not inundated*	0.6229
Eucalyptus camaldulensis	*Inundation extent*	0.3811
*Gazania rigens***	*Inundation extent*	0.1476
Haloragis aspera	*Inundated*	1
*Heliotropium curassavicum**	*Not inundated*	0.6725
Isoetopsis graminifolia	*Inundated*	0.2623
Isolepis australiensis	*Inundated*	1
Juncus usitatus	*Pool level*	0.5887
Lachnagrostis filiformis	*Inundation extent*	0.1518
Ludwigia peploides	*Inundated*	0.1008
Lythrum hyssopifolia	*Inundated*	0.2543
Melaleuca lanceolata	*Inundation extent*	0.1518
Myriophyllum papillosum#	*Pool level*	0.1672
Paspalidium jubiflorum	*Inundation extent*	0.1476
Persicaria lapathifolia	*Inundated*	0.3951
Picris angustifolia	*Inundation extent*	0.4649
*Rorippa palustris**	*Not inundated*	0.6309
Sclerolaena divaricata	*Not inundated*	0.6459
Senecio cunninghamii	*Inundated*	1
*Silene nocturna**	*Not inundated*	0.6459
Sphaeromorphaea australis	*Inundated*	0.0562
Sporobolus mitchellii	*Inundation extent*	0.3689
Stemodia florulenta	*Inundated*	0.2392
*Symphyotrichum subulatum**	*Inundated*	0.2494
*Xanthium occidentale***	*Inundated*	0.88

c.

Species	Zone	P
Acacia stenophylla	*Inundation extent*	0.7712
Alternanthera denticulata	*Pool level*	0.1862
Ammannia multiflora	*Inundated*	1
Atriplex suberecta	*Not inundated*	0.2575
Bare soil	*Not inundated*	1
Bolboschoenus caldwellii	*Pool level*	0.0056
Brachyscome paludicola	*Not inundated*	1
Centipeda minima	*Inundated*	0.2811
Cyperus gymnocaulos	*Inundated*	0.8232
*Dittrichia graveolens**	*Inundated*	0.244
Enchylaena tomentosa	*Not inundated*	0.1474
*Erigeron bonariensis**	*Inundated*	1
Eucalyptus camaldulensis	*Pool level*	0.3665
Euphorbia drummondii	*Not inundated*	0.5427
Glinus lotoides	*Inundated*	1
Glycyrrhiza acanthocarpa	*Not inundated*	1
*Heliotropium curassavicum**	*Inundation extent*	0.7644
*Heliotropium europaeum**	*Inundation extent*	0.4877
Isoetopsis graminifolia	*Pool level*	0.4827
Isolepis australiensis	*Pool level*	0.7804
Ludwigia peploides	*Pool level*	0.003
*Oxalis**	*Inundation extent*	0.3277
Paspalidium jubiflorum	*Not inundated*	1
Paspalum distichum	*Pool level*	0.3215
Persicaria lapathifolia	*Pool level*	0.0294
Phragmites australis	*Pool level*	0.3295
Phyllanthus lacunarius	*Inundated*	1
Polygonum plebeium	*Pool level*	0.3247
Pseudognaphalium luteoalbum	*Inundated*	0.2665
Senecio cunninghamii	*Not inundated*	0.7508
Sphaeromorphaea australis	*Pool level*	0.003
Sporobolus mitchellii	*Inundation extent*	0.5865
Stemodia florulenta	*Inundated*	0.1098
*Symphyotrichum subulatum**	*Pool level*	0.0166
Typha domingensis	*Pool level*	0.7698
*Xanthium occidentale***	*Inundated*	0.4111

Microinvertebrate assemblage

To estimate the contribution of Commonwealth environmental water to microinvertebrate taxonomic richness (quantified as the integer number of distinct taxa with individuals identified to the lowest possible taxonomic level (e.g., family, genus, species)) and density (quantified as the number of individuals per litre (i.e., volumetric density)), we ignored the potential variation in the relative contribution of longitudinal and lateral sources. We adopt a hypothesis that the additional water volumes contributed by Commonwealth environmental water increased the overall abundance and taxonomic richness of microinvertebrate assemblages at all locations in the Lower Murray, without consideration of the relative contribution of different habitats or functional groups.

Because we have only one river, we are unable to use an experimental approach to answer the evaluation questions. These must necessarily be addressed using a modelling approach, to estimate the difference in the microinvertebrate assemblages under a scenario where the environmental water was not provided. To achieve this, the observed microinvertebrate data was used to build a model based on the observed flow data. Then the modelled flow data, removing the volumes due to environmental water, can be used to predict microinvertebrate taxonomic richness and density if this were the amount of water that had been available. While this comes with considerable uncertainty, it is not possible to answer the questions in any other way with available data. To guide the modelling process, each evaluation question was turned into a hypothesis with specific predictions.

Additional modelling and statistical analyses

Microinvertebrate taxonomic richness of LTIM and MER samples

There are three important elements to biodiversity: total density, taxonomic richness, and evenness (Magurran and McGill 2011). Microinvertebrate density (individuals per litre) and taxonomic richness (number of distinct taxa per litre) were quantified as the mean and 95% confidence limits in all samples in each water year. To quantify evenness, Pielou's measure (Shannon diversity divided by the log number of taxa) was used. Pielou's evenness ranges from 0 to 1, with a value of 1 indicating all taxa had the same number of individuals. Generally, a higher evenness means that the number of individuals is similar for each taxa, and fora given taxonomic richness, a more even assemblage is considered more diverse. Evenness is an important diversity measure as it influences community stability, resistance and resilience and tends to be more responsive to human perturbations than richness (Hillebrand *et al.* 2008).

Functional group abundance

LTIM microinvertebrate samples have previously been compared according to the proportional density of different microinvertebrate functional groups (e.g., pelagic vs. littoral rotifers; Ye *et al.* 2020). Here we have repeated this analysis for MER samples. We also classified the MER and LTIM samples into a minimum set of groups based on the proportion of each functional group in the samples, to determine the main influences of difference in composition. For each sampling event (trip), we calculated proportional functional group representation in pooled samples at the lock scale (i.e., proportional density of each functional group in the three sites each below locks 1 and 6 for the LTIM samples and of the three replicates at the single site below locks 1, 4 and 6 for the MER samples, total number of samples = 79). We classified samples using Ward's method to identify clusters of samples with relatively similar relative proportions of functional groups quantified using the Hellinger distance matrix and used indicator species analysis (Dufrene and Legendre 1997) to determine the optimal number of groups to split samples among (*n* = 3) and to identify representative taxa within each of these groups to aid interpretation. For each taxon, its indicator value for a group of samples is the product of its specificity (mean abundance in samples from that group relative to the other groups) and its affinity (proportion of samples within that group the taxa is observed). This product is multiplied by 100 to give a percentage. An indicator value of 100 for a given group would mean the taxa was only found in samples within that group and occurred in every sample. The significance of the indicator value is tested by randomization.

We then built a binary classification tree to distinguish the conditions most likely to produce the observed similarity between samples within the functional group classes. Binary classification trees use both categorical and continuous predictors to create binary (yes/no) splits among samples, seeking to identify the predictors (and values of these) most associated with the grouping of samples into classes. The classification tree was built using R package 'rpart' (Therneau *et al.* 2019).

Predictors used in modelling

Both categorical and continuous predictors were used to model microinvertebrate flow response. Categorical predictors were month, water year, lock and site. Continuous predictors quantified: (1) flow, (2) flow variability, (3) water quality and (4) inundated floodplain area. Short-term and long-term flow predictors were calculated, with short-term flow quantified as the mean flow on the day of sampling at the nearest lock for each site. Long term flow was quantified as the mean daily flow over the preceding 12-week periods (shorter periods resulted in predictors with a Pearson correlation with daily flow exceeding 0.95). Flow variability was quantified as the trend in 10-day flow volume (denoted 'dQ10'), estimated as the mean daily change in flow volume over the preceding 10-day period (5, 7 and 14-d trends were also calculated, and tested in modelling. However, 10-day trends tended to have the most explanatory power). Water quality parameters available for modelled were daily temperature and electrical conductivity. Floodplain inundated area was quantified using the DEW MIKE model outputs (M. Gibbs, pers comm). These outputs provide a flow-area response curve where the area of floodplain in each weir pool can be estimated based on the daily flow. A generalised additive model (a type of smoothing function) was fit to the discrete MIKE outputs (area of floodplain inundated in each weir pool at a given daily flow) to allow an estimate of inundated area for any flow volume. These were summed over 28- and 60-day periods to estimate inundated floodplain area corresponding to a minimum period required for taxa to emerge from the egg bank and complete one or two life cycles. Models were fit using only one of the estimates, the predictor giving the better explanatory power was adopted for the final model.

Model construction and selection process

As the response variables of interest (i.e., density and taxonomic richness) were count data, each response was first modelled using a generalised linear model with a Poisson error structure and log link function. Where over-dispersion was evident, models were refit using negative binomial error structures. Because the data represent repeated measures, model residuals were assessed for temporal dependence (i.e., autocorrelation) using Pearson's correlation for a lag of one sampling event. However, no evidence of autocorrelation was found in any of the models (all $p > 0.05$). Spatial autocorrelation was assessed using Mantel test of model residuals and Euclidean distances between sites, and again no spatial autocorrelation was evident (all $p > 0.05$).

In each case, model selection was based on fitting a global model that included all predictors: month, site, lock and water year as factor variables, continuous predictors of short- and long-term flow, and all two-way interactions for temperature, electrical conductivity, area of inundated floodplain and 10-day flow trend. An optimal model structure was determined using an automated stepwise procedure that minimised Akaike's information criterion by iteratively fitting different combinations of predictors (function 'stepAIC'). Model predictive performance was assessed using two pseudo-R^2 measures: the amount of explained deviance; and the squared correlation between observed and fitted data. All modelling was done using R V4.0.1, using custom package MASS (Venables and Ripley 2002).

Results – inter-annual comparisons

Diversity (taxonomic richness and evenness) patterns

Across all sampling years, mean taxonomic richness per replicate was lowest in 2017-18 and highest in 2019-20 (Table D4). As is typical of ecological data, density and taxonomic richness were highly positively correlated (Pearson's $r = 0.84$) and mean density per replicate across all sampling years followed the same rank order as that of taxonomic richness (Table D4). The current sampling year (2019-20) stands out as having higher sampling variability, with a standard error of around 10% of the mean density. However, this is difficult to interpret due to the contrasting sampling design between LTIM and MER, where the latter samples were collected at three locks but only one location in each (Table D4). Samples collected in 2019-20 were distinguished from the LTIM samples by the higher relative density of littoral/pelagic rotifers, which are primarily taxa from the *Trichocerca pusilla* complex along with *T. similis* and *T. similis grandis*. This was also evident in the high proportion of samples from Group 2 (see Table D5).

In contrast with taxonomic richness, Pielou's evenness was highest in 2016-17, a year of relatively low mean taxonomic richness and density. Higher evenness values suggest more stable assemblages, as greater evenness in density means more individuals can be lost without any change in the number of taxa. Years of lower evenness in density such as 2014-15 and 2019-20 are consistent with a more skewed density distribution, with many rare taxa, each represented by relatively few individuals.

Table D4. Taxonomic richness, density and evenness of microinvertebrates calculated from raw sub-samples. LTIM samples (years 2014-15 to 2017-18) were based on three replicate sub-samples at three locations downstream of Locks 1 and 6, while MER samples (2019-20) reflect three replicate sub-samples at one location downstream of Locks 1, 4 and 6.

	2014-15 (*n* =108)	**2015-16 (*n* =144)**	**2016-17 (*n* =144)**	**2017-18 (*n* =126)**	**2019-20 (*n* =63)**
Taxonomic richness	17.9 ± 0.48	16.5 ± 0.52	16.6 ± 0.60	13.3 ± 0.56	18.3 ± 0.59
Density (ind./L)	1027 ± 53.4	686 ± 42.5	713 ± 54.6	610 ± 44.3	1058 ± 103.2
Pielou's evenness	0.66 ± 0.02	0.77 ± 0.01	0.80 ± 0.01	0.75 ± 0.01	0.69 ± 0.02

Functional group density

Classification of pooled (at lock scale) LTIM and MER samples into functional group composition and density at the different sampling dates yielded three classes of approximately equal frequency (Figure D10).

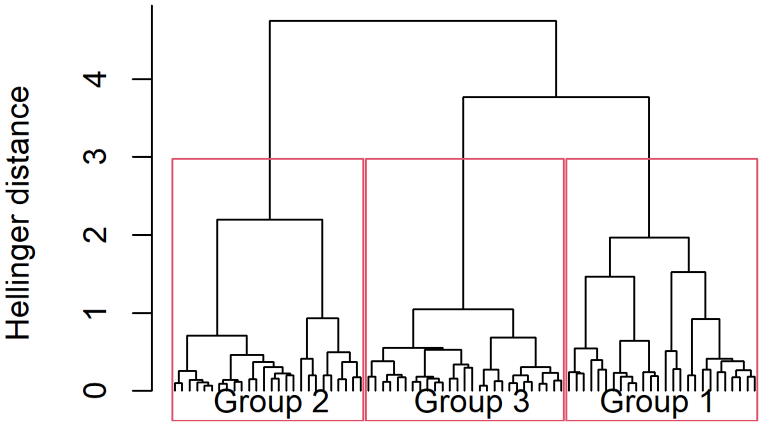

Figure D10. Classification of pooled (within lock) microinvertebrate samples from 2014–2020 according to functional group composition and density using Hellinger distance. Rectangles show the most ecologically informative division into separate functional group classes, each having an indicative functional group.

Group 1 samples were characterised by high density of pelagic cladocerans (indicator value = 0.60, p = 0.001) and pelagic calanoid copepods (IV = 0.36, p = 0.04), Group 2 by littoral/pelagic rotifers (IV = 0.49, p = 0.003) and Group 3 by pelagic rotifers (IV = 0.77, p = 0.001). Across all years of sampling, every group was observed at least once at each site, although there were clear patterns in the relative dominance (Table D5).

Table D5. Proportion of samples from each water year falling within the three functional group classes.

Sampling year	Group 1	Group 2	Group 3
2014-15	0.50	0.33	0.17
2015-16	0.38	0.06	0.56
2016-17	0.12	0.38	0.50
2017-18	0.43	0.29	0.29
2019-20	0.29	0.52	0.19

The question of what determines the number of samples falling into the different groups from year-year was investigated using a classification tree (Figure D11). This method tries to separate the samples into their compositional groups using a series of yes/no decisions. Each decision is based on a different threshold (e.g., flow volume) or condition (e.g., month) and samples are separated according to the conditions at the time of sampling. Each decision is represented by a numbered 'node', which shows the decision. Samples meeting the condition follow the 'yes' (left) path, the others follow the 'no' path to the next decision point (node).

Group 1 samples (which had a high density of pelagic cladocerans and calanoid copepods) were most commonly observed when flow increased rapidly in the 10 days prior to sampling (hereafter 10-day flow trend; dQ10). For example, node 2 (coloured green) shows 80% of all samples taken when the 10-day flow trend was in the top 25% observed were Group 1, while Group 3 samples were never found under these conditions (Figure D11). Group 1 samples were also favoured under extended periods of stable low daily flow below approximately 5.4 GL/d (note 8.6 in the figure refers to the logarithmic scale, node 12; Figure D11) and to a lesser extent where the extent of floodplain inundation was declining (i.e., a negative value for fpcon, node 30). This is consistent with longitudinal transport during the rising limb of flood pulses providing the most likely source of Group 1 taxa, but with some increased probability of observing these taxa from the entrainment of microinvertebrates as floodwaters recede.

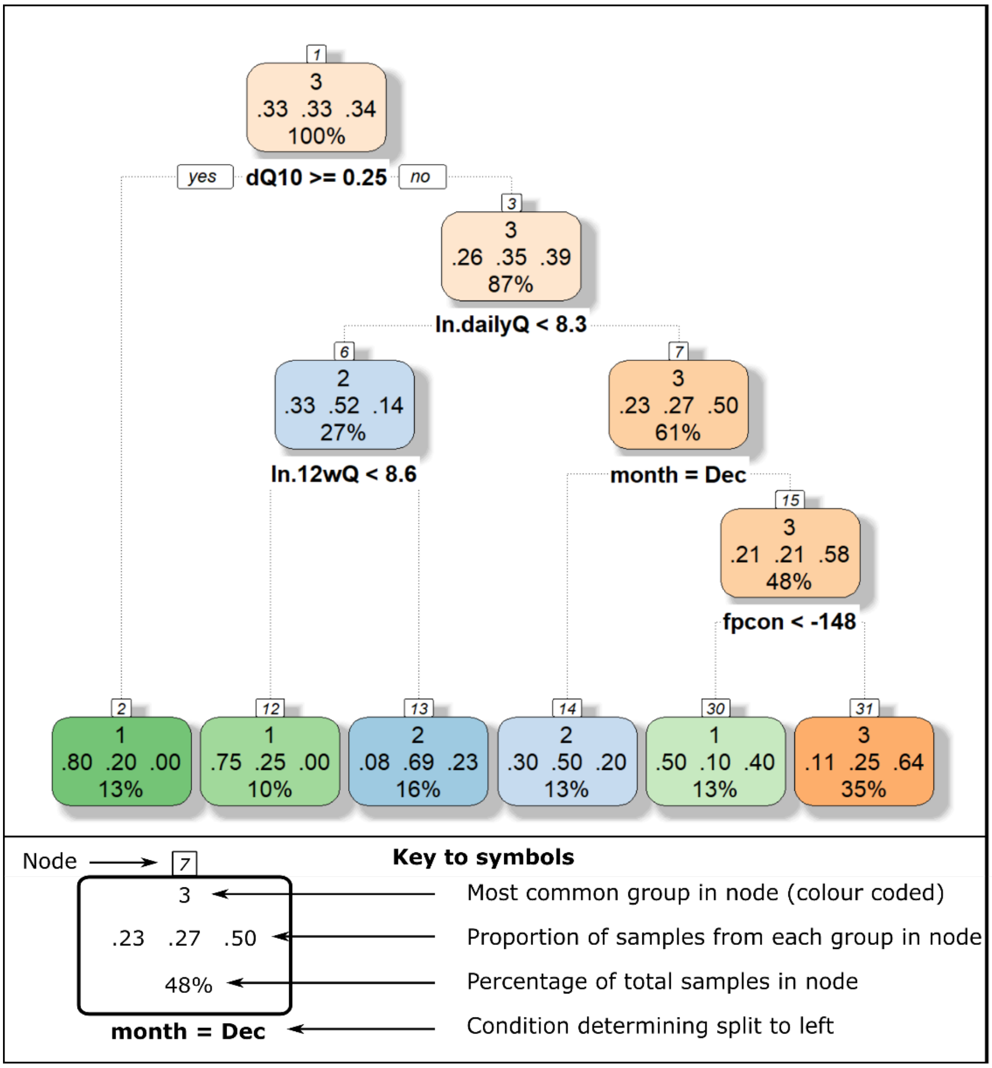

Figure D11. Classification tree for the functional group classes shown in Figure D10. Nodes show the distribution of samples among functional groups. Nodes with conditions specified below them divide samples according to that condition. Nodes in the bottom row (terminal nodes) represent the distribution of samples among groups after following all binary conditions leading to that node.

Group 2 samples (high density of littoral-pelagic rotifers) were present under all sets of conditions, but most favoured during long-term flows exceeding 5.4 GL/d but with relatively constant (or slowly decreasing) flow, or during the month of December. Group 3 samples (pelagic rotifers) were absent during rapidly increasing flow but were favoured at higher (constant or decreasing) daily flows and with higher floodplain connectivity.

Modelled responses to environmental flows

H1: Density and taxonomic richness

Across all years of available data (incorporating water years 2014-15 to 2017-18 and the new data for 2019-20), approximately 18% of observed density was estimated to be attributable to environmental water (Figure 44) with the mean difference ±[95% CI] being 144.7 [112.2, 177.1] ind/L (*t* = 8.80, *df* = 194, *p* < 0.001). Additional Commonwealth environmental water was predicted to have contributed approximately 64% of this increase (mean observed-modelled difference ± [95% CI] = 93.2 [66.7, 119.6] ind/L; t = 6.96, df = 194, $p < 0.001$).

Selected predictors of density (see Table D6) with a positive effect included mean daily flow, temperature, floodplain area inundated over the last 60 days (FPA60) and the 10-day flow trend, while electrical conductivity was involved in negative interaction terms with the latter two predictors (indicating decreasing benefits of both with increasing conductivity). Temperature also had a negative interaction with the 10-day flow trend, perhaps reflecting the transport of individuals from the floodplain on the receding limb i.e., a negative coefficient means that a falling (negative) trend in flow was predicted to result in increased individuals. After accounting for all continuous variables, density tended to increase continually over the sampling season (relative to September values), while water years 2015-16 and 2019-20 were higher than the baseline (water year 2014-15) expectation.

The negative binomial density model had acceptable overall performance, with explained deviance of 0.61 and the squared correlation between predicted and observed density equal to 0.46 (both are measures of explanatory power, a form of pseudo R^2). With this said, there remain considerable uncertainties. For example, the model did not predict universally lower density in the absence of any environmental water (e.g., water year 2016-17 had higher median density without environmental water; Figure 44a). In some instances, this could be a true pattern, for example due to greater local flushing from higher flow velocities. However, across all years, models suggest there are clear benefits from the delivery of environmental water by increasing microinvertebrate density within the channel environment.

Table D6. Results of negative binomial generalised linear model for density (individuals per litre) showing selected predictors, estimated coefficients (Estimate), standard errors in estimation (std.Error) and Type I Error probability (*p*-value). Flow data were log transformed prior to modelling. FPA60 quantifies the area of floodplain inundated over the last 60 days (see Methods for full description of predictor calculation). Negative binomial distribution dispersion parameter (theta) = 4.14, standard error = 0.41.

Predictor	**Estimate**	**std. Error**	***p*-value**
(Intercept)	-3.29	1.17	0.005
Month [Oct]	0.30	0.26	0.247
Month [Nov]	0.71	0.31	0.022
Month [Dec]	0.72	0.34	0.035
Month [Jan]	1.17	0.39	0.003
mean daily flow (ML/d)	0.61	0.11	<0.001
electrical conductivity (EC)	3.3e-03	2.8e-03	0.228
temperature	0.15	0.03	<0.001
water year [2015]	0.14	0.12	0.246
water year [2016]	-0.59	0.23	0.010
water year [2017]	-0.11	0.14	0.452
water year [2019]	0.65	0.20	0.001
10-day flow trend (dQ10)	4.20	1.38	0.002
FPA60	5.9e-04	1.7e-04	0.001
temperature:dQ10	-0.12	0.05	0.015
EC:dQ10	-0.01	2.8e-03	0.008
EC:FPA60	-4.2e-06	-9.0e-07	<0.001

For taxonomic richness (Figure 44b), across all years, environmental water was predicted to account for 9% of observed taxa (mean difference ±[95% CI] between observed and modelled no environmental water scenarios = 2.3 [1.6, 2.9] taxa per sampling event; t = 7.04, df = 194, p < 0.001). Commonwealth environmental water alone was predicted to account for ~64% of this increase (mean difference ±[95% CI] = 1.5 [0.95, 1.96] taxa per sampling event; *t* = 5.65, *df* = 194, *p* < 0.001).

As discussed in Inter-annual comparisons, the most important predictor of taxonomic richness was the density of individuals, which has a strong positive influence on the number of taxa observed (Table D7). Other predictors with a positive influence on richness included the mean long-term (12 week) daily flow, water temperature, and the area of floodplain inundated over the last 28 days (FPA28). As with the density model, electrical conductivity was most influential via a negative interaction with floodplain area but for taxonomic richness, the interaction between temperature and the 10-day flow trend was positive. One possible explanation could be increased transport efficiency from upstream source areas as seasonal temperatures rising. Consistent with this interpretation, taxa richness was highest in late spring and early summer, although month did not have a large effect on taxonomic richness.

The Poisson generalised linear model had similar performance to the density model, with an identical explained deviance of 0.61 but slightly higher squared correlation between predicted and observed density (0.57). Interestingly, there was no overdispersion evident in the model that would warrant the use of a negative binomial error structure (as was required for the density model). However, it is important to note that the uncertainty in the performance of the density model is compounded in the richness model because of the need to use the output from the former model as the density predictor in the latter. To obtain an estimate of the benefits of environmental water, this is essentially unavoidable, because of the high dependence of taxonomic richness on density. However, it does increase model uncertainty and provides an additional complexity to modelling taxonomic richness as opposed to density. Nonetheless, as with the density model, across all years there appears to be clear benefits for microinvertebrate taxonomic richness from environmental water delivery.

Table D7. Results of taxonomic richness Poisson generalised linear model, showing selected predictors, estimated coefficients (Estimate), standard error in the coefficient estimate (std. Error) and Type I error probability (p-value). Explained deviance for the model was 0.61, residual standard error: 0.007. Abundance was predicted for environmental flow scenarios using the model in Table D6.

Predictors	Estimate	std. Error	*p*
(Intercept)	1.00	0.35	0.004
Month [Oct]	0.01	0.11	0.937
Month [Nov]	0.16	0.13	0.203
Month [Dec]	0.16	0.14	0.263
Month [Jan]	0.11	0.16	0.466
abundance	2.1e-04	2.9e-05	<0.001
temperature	0.04	0.01	0.002
electrical conductivity (EC)	7.5e-04	7.3e-04	0.302
10-day flow trend (dQ10)	-0.35	0.37	0.355
mean long-term daily flow	0.10	0.02	<0.001
FPA28	4.2e-04	1.3e-04	0.001
temp:dQ10	0.03	0.02	0.127
EC:FPA28	-2.3e-06	6.8e-07	0.001

H2: Lateral and longitudinal connectivity

Across all years, environmental water delivery accounted for ~18% of the observed density of microinvertebrates dependent on lateral connectivity (mean difference ±[95% CI] between observed and modelled no environmental water scenarios = 8.9 [6.4, 11.4] ind/L; t = 7.06, df = 194, p < 0.001). Commonwealth environmental water alone accounted for ~86% of this (mean difference ±[95% CI] = 7.6 [5.6, 9.6] ind/L; t= 7.53, df = 194, p <0.001).

Across all years, environmental water delivery accounted for ~16.5% of the observed density of microinvertebrates dependent on longitudinal connectivity (mean difference ±[95% CI] between observed and modelled no environmental water scenarios = 83.2 [66.0, 100.4] ind/L; t = 9.56, df = 194, p < 0.001). Commonwealth environmental water alone accounted for ~65% of this increase (mean difference ±[95% CI] = 54.1 [41.2, 66.9] ind/L; t = 8.29, df = 194, p < 0.001).

Table D8. Results of negative binomial generalised linear model for abundance (individuals per litre) of taxa dependent upon lateral connectivity showing selected predictors, estimated coefficients (Estimate), standard errors in estimation (std.Error) and Type I Error probability (p-value). Flow data were log transformed prior to modelling. FPA60 quantifies the area of floodplain inundated over the last 60 days (see Methods for full description of predictor calculation). Negative binomial distribution dispersion parameter (theta) = 2.41, standard error = 0.26.

Predictor	Estimate	std. Error	*p*-value
(Intercept)	-4.41	3.79	0.245
Month [Oct]	-0.46	0.38	0.216
Month [Nov]	-2.04	0.45	<0.001
Month [Dec]	-1.91	0.51	<0.001
Month [Jan]	-2.35	0.56	<0.001
mean daily flow (ML/d)	-0.67	0.15	<0.001
mean long-term daily flow	0.82	0.31	0.007
electrical conductivity (EC)	3.3e-03	2.8e-03	0.228
water year [2015]	-0.69	0.18	<0.001
water year [2016]	-1.47	0.57	0.010
water year [2017]	-0.43	0.19	0.028
water year [2019]	-0.47	0.28	0.095
temperature	0.36	0.14	0.010
electrical conductivity	0.03	0.02	0.027
10-day flow trend (dQ10)	1.77	0.74	0.017
FPA60	4.0e-04	2.3e-04	0.086
temperature:EC	-1.3e-03	7.0e-04	0.061
EC:dQ10	-5.6e-03	3.5e-03	0.104
EC:FPA60	-2.0e-06	1.2e-07	0.106

Table D9. Results of negative binomial generalised linear model for abundance (individuals per litre) of taxa dependent upon longitudinal connectivity showing selected predictors, estimated coefficients (Estimate), standard errors in estimation (std.Error) and Type I Error probability (p-value). Flow data were log transformed prior to modelling. FPA60 quantifies the area of floodplain inundated over the last 60 days (see Methods for full description of predictor calculation). Negative binomial distribution dispersion parameter (theta) = 2.41, standard error = 0.26.

Predictor	Estimate	std. Error	*p*-value
(Intercept)	4.51	2.96	0.128
Month [Oct]	0.75	0.33	0.022
Month [Nov]	1.35	0.39	0.001
Month [Dec]	1.31	0.44	0.003
Month [Jan]	1.78	0.49	<0.001
mean daily flow (ML/d)	0.58	0.13	<0.001
mean long-term daily flow	0.82	0.31	0.007
electrical conductivity (EC)	3.3e-03	2.8e-03	0.228
water year [2015]	-0.09	0.15	0.532
water year [2016]	-0.84	0.28	0.003
water year [2017]	-0.36	0.17	0.033
water year [2019]	0.19	0.25	0.427
temperature	-0.20	0.13	0.109
electrical conductivity	-0.04	0.01	0.003
10-day flow trend (dQ10)	2.76	1.07	0.010
FPA60	4.4e-04	2.1e-04	0.033
temperature:EC	-1.9e-03	6.8e-04	0.003
temperature:dQ10	-1.1e-01	5.1e-02	0.027
EC:FPA60	-3.6e-06	1.1e-06	0.001

H3: Spring microinvertebrate prey species density

Across all years, observed microinvertebrate fish prey species density (i.e., the total density of all taxa in Table 17) during spring (October and November) was lowest in 2019-20 (mean [95% CI] = 73.9 ind/L, [48.3, 99.8] and highest in 2016-17 (202 ind/L, [93.1, 312.6]). However, differences in density between years were not statistically significant (Kruskal Wallis X^2 = 5.84, df = 4, p = 0.21), due to the high variability within sites in all sampling years, particularly 2016-17 (Figure 46).

Modelled spring microinvertebrate prey species density in the absence of any environmental water was estimated at ~37% lower (mean difference ±[95% CI] between observed and modelled no environmental water scenarios = 44.0 [36.2, 51.9] ind/L; t = 11.16, df = 101, p = <0.001). Commonwealth environmental water alone was estimated to account for ~86% of the density increase (37.7 [30.4, 45.1] ind/L per sampling event; t = 4.12, df = 101, p < 0.001).

A clear uncertainty in the building of the spring fish prey species model lies in the selection of suitable taxa. In this analysis, a broad interpretation of prey species was adopted, but future work should seek to identify the most important taxa, allowing a more sophisticated modelling approach. The spring fish prey species density model also had additional statistical uncertainties, with a low squared correlation between predicted and observed data (0.31). However, the amount of explained deviance was comparable to the other models (0.46) and the most influential predictors (Table D10) were largely in accord with the other density models (e.g. Table D6).

Table D10. Results of negative binomial generalised linear model for spring microinvertebrate prey-species density (individuals per litre) showing selected predictors, estimated coefficients (Estimate), standard errors in estimation (std.Error) and Type I Error probability (*p*-value). Flow data were log transformed prior to modelling. FPA28 quantifies the area of floodplain inundated over the last 28 days (see Methods for full description of predictor calculation). Negative binomial distribution dispersion parameter (theta) = 3.07, standard error = 0.30.

Predictor	Estimate	std. Error	*p*-value
(Intercept)	19.18	3.19	<0.001
Month [Oct]	-0.30	0.33	0.363
Month [Nov]	-0.82	0.39	0.036
Month [Dec]	-0.65	0.44	0.137
Month [Jan]	-0.51	0.48	0.283
mean long term flow	1.19	0.26	<0.001
electrical conductivity (EC)	0.06	0.01	<0.001
temperature	0.62	0.12	<0.001
water year [2015]	-0.01	0.16	0.955
water year [2016]	-1.24	0.50	0.014
water year [2017]	0.01	0.16	0.951
water year [2019]	-0.03	0.24	0.889
10-day flow trend (dQ10)	1.71	0.75	0.023
FPA28	1.5e-03	4.0e-04	<0.001
temperature:EC	-2.4e-03	6.0e-04	<0.001
EC:dQ10	-7.1e-03	3.3e-03	0.030
EC:FPA28	-1.1e-05	2.1e-06	<0.001
dQ10:FPA28	6.4e-04	2.7e-04	0.018

Discussion

Functional group density

Group 1 (high density of pelagic cladocerans and pelagic calanoid copepods) and Group 3 (pelagic rotifers) were most dominant in low flow years (2014-15 and 2015-16, respectively), however were favoured under different conditions. Group 1 is dominated by pelagic cladocerans and pelagic calanoid copepods, important food resources for fish such as juvenile golden and silver perch and Murray cod larvae (e.g. Humphries 2005; Kaminskas and Humphries 2009; King *et al.* 2009; Puckridge and Walker 1990; Rowland 1998; Shiel *et al.* 1982; Zampatti and Leigh 2013b). This group of organisms generally require still open water for reproduction and long water residence times to develop into dense populations (e.g. months) (Obertegger *et al.* 2007). Consequently, these groups are commonly generated within habitats such as permanent/semi-permanent backwaters, wetlands and lakes and at times transferred to the main river channel (Furst *et al.* 2020; Gigney *et al.* 2006). Group 1 was found to be strongly favoured when the 10-day flow trends were increasing rapidly and likely due to already established communities being flushed from permanent backwater habitats, low-lying wetlands and from within weir pools. This group was also favoured under extended periods of less variable mean daily flow below approximately 5.4 GL/d. Under these circumstances, it is likely that lake-like conditions, often created above weirs in the main river channel during periods of extended low flow, were facilitating production of large populations of pelagic copepods (e.g. *Boeckella triarticulata*) (Hynes and Hynes 1970). The other conditions where Group 1 samples were favoured were when flows were declining with limited areas of floodplain inundation. This pattern is consistent with flow pulses that flush backwater and low-lying wetland habitats, providing the most likely source of Group 1 taxa, while increasing the probability of observing these taxa from the entrainment of microinvertebrates when flows recede. In comparison, Group 3 samples (pelagic rotifers) were favoured at higher (constant or decreasing) daily flows and with higher floodplain connectivity. Pelagic rotifers are the most abundant group of zooplankton throughout the river. Despite still needing slow or still water to reproduce, they have high reproduction rates and thus do not require water residence times as long as cladocerans and copepods (e.g. Nandini and Rao 1997). Furthermore, they are generally weaker swimmers and consequently are more prone to entrainment than larger zooplankton. Therefore, floodplain inundation creates expansive areas of slow/non-flowing water for rapid reproduction and population growth while the higher flows/connectivity transfer them to the main channel. A better understanding of what drives surges in populations and communities of pelagic rotifers may provide useful in is the future as they are food resources for other rotifers, copepods, cladocerans and small fish such as Australian smelt and rainbowfish larvae (Conde-Porcuna and Sarma 1995; Gilbert and Williamson 1978; King 2005; Stemberger 1985).

Diversity (taxonomic richness and evenness) patterns

Both richness and evenness can indicate changes in resistance and resilience, the two components of community stability (Hillebrand *et al.* 2008). Increasing richness tends to result in increased resilience, as it increases the probability that those taxa which possess life history traits that allow them to rapidly recover from disturbance are present in the community. Community resistance is thought to depend more on evenness (or its opposite, dominance), with the likely outcome dependent on the traits of the dominant taxa; if the dominant taxa are resistant to disturbance, increasing evenness decreases resistance (Hillebrand *et al*. 2008). If dominant taxa possess traits enabling them to resist disturbance, for example flooding, the two sampling years with the lowest evenness and highest richness could represent the most stable microinvertebrate communities across the sampling record, although this would require knowledge of the trait distribution to confirm.

Modelled effects of environmental flows for microinvertebrates

Overall, modelling demonstrated that environmental water delivery has consistently provided benefits for the microinvertebrate community in the LMR. While varying across years, on average, microinvertebrate density would have been 17–37% lower than observed in the absence of any environmental water, with Commonwealth environmental water accounting for 64–86% of the increase due to environmental water. Importantly, most of these benefits were delivered during low flow years, helping to maintain higher productivity and ecosystem function. Environmental water appeared to proportionally benefit taxa dependant on lateral connectivity more so than longitudinal connectivity/transport. The influence of environmental water was clearly greatest on microinvertebrate prey species density in spring, with environmental water accounting for an average of 37% of observed values and Commonwealth environmental water contributing 40–100% of this.

Each of the predictors identified provide a valuable starting point for conceptualising and modelling to inform more effective environmental water delivery to benefit the microinvertebrate community in the LMR. Research to refine these relationships and test a range of predictors will be highly beneficial. For example, the main uncertainty in the calculation of predictors is determining the area of floodplain inundated that contributes to local scale richness at a given sampling point in the channel. In future, it might be possible to calculate two predictors for each site, one giving the area of floodplain that has been inundated in the local environment, the other accounting for upstream contributions. Conceptually the two predictors would represent local and regional floodplain contributions and including one or the other in models might afford an alternative way to quantify the importance of lateral (local) and longitudinal transport of microinvertebrates. Another possible predictor to calculate could be the area of connected wetland habitat (as opposed to floodplain area). There are also uncertainties in selecting suitable response predictors to test hypotheses. A major contributor is the lack of a robust definition of the taxa that are dependent on longitudinal vs lateral connectivity and of those that provide high quality food resources. To provide more detailed guidance to fine tune environmental water delivery for specific outcomes, it is important to continually improve these definitions and refine model predictors.

Fish indicators – statistical outputs

Table D11. PERMANOVA pairwise comparison test results for golden perch electrofishing abundance (individuals per 90 second shot) in the gorge geomorphic zone (10 sites) of the Lower Murray River in autumn from 2015–2020. *P*-values presented in bold are significant comparisons, using Bonferroni corrected α = 0.0033 (Narum 2006) for comparisons between years (fifteen comparisons).

Comparison	**Large-bodied**	
	t	*P (perm)*
2015 vs. 2016	0.2726	0.8134
2015 vs. 2017	1.4563	0.1831
2015 vs. 2018	0.8253	0.4354
2015 vs. 2019	3.6323	**0.0015**
2015 vs. 2020	3.5222	**0.0018**
2016 vs. 2017	1.6998	0.1248
2016 vs. 2018	0.6930	0.5271
2016 vs. 2019	4.7217	**0.0001**
2016 vs. 2020	4.3351	**0.0004**
2017 vs. 2018	0.8086	0.4573
2017 vs. 2019	2.1526	0.0637
2017 vs. 2020	1.8277	0.1044
2018 vs. 2019	2.8114	0.0145
2018 vs. 2020	2.7290	0.0185
2019 vs. 2020	0.2371	0.8786

Table D12. Results of the model selection procedure for linear regression models fitted to Murray cod length–weight baseline data (2004–2020).

Model	**AICc**	**Npar**	**ΔAICc**	**Likelihood**
Exponential	54.9896	2	438.1684	<0.001
Linear	1265.7601	3	1648.9389	0
Cubic	-383.1788	5	0	1
Qudratic	-23.3887	6	359.7901	<0.001

Table D13. PERMANOVA pairwise comparisons of the relative condition index (K*rel*) of YOY Murray cod between years (weir pools pooled). P-values presented in bold are significant comparisons (α = 0.05).

Comparison	*t*	*p* value
2015, 2016	0.617	0.543
2015, 2017	2.849	**0.016**
2015, 2018	0.452	0.656
2015, 2019	1.909	0.067
2015, 2020	0.492	0.623
2016, 2017	2.615	**0.028**
2016, 2018	1.213	0.244
2016, 2019	0.838	0.410
2016, 2020	0.084	0.933
2017, 2018	3.613	**0.004**
2017, 2019	1.713	0.098
2017, 2020	2.255	**0.028**
2018, 2019	2.762	**0.008**
2018, 2020	1.029	0.315
2019, 2020	1.763	0.082

Table D14. Results of the model selection procedure for linear regression models fitted to 2019-20 Murray cod age–length data.

Model	AICc	Npar	∆AICc	Likelihood
Linear	283.977	1	6.0812	0.044
Gompertz	277.896	3	0	0.919
Von Bertalanffy	284.346	2	6.4498	0.037

Table D15. PERMANOVA main test comparisons of Murray cod catch-per-unit-effort (CPUE) between years and weir pools for non-targeted electrofishing.

Factor	df	*Pseudo-F*	*p* value
Year	5	2.229	0.066
Weir Pool	1	2.014	0.164
Year x Weir Pool	5	1.042	0.387
Residuals	36		

Table D16. PERMANOVA main test and pairwise comparisons of Murray cod catch-per-unit-effort (CPUE) between years and weir pools for targeted electrofishing. P-values presented in bold are significant comparisons (α = 0.05).

Factor	**df**	***Pseudo-F***	***p* value**
Year	6	3.973	**0.006**
Weir Pool	1	4.453	**0.042**
Year x Weir Pool	5	0.442	0.810
Residuals	36		

Pairwise comparisons (between years)	***t***	***p* value**
2013, 2014	0.334	0.744
2013, 2015	1.401	0.193
2013, 2016	1.925	0.097
2013, 2017	0.450	0.657
2013, 2019	1.624	**0.037**
2013, 2020	1.901	0.081
2014, 2015	2.023	0.059
2014, 2016	2.939	**0.011**
2014, 2017	0.346	0.738
2014, 2019	2.893	**0.020**
2014, 2020	3.522	**0.005**
2015, 2016	0.718	0.466
2015, 2017	1.666	0.124
2015, 2019	0.533	0.566
2015, 2020	1.429	0.176
2016, 2017	2.594	**0.030**
2016, 2019	0.303	0.867
2016, 2020	0.830	0.435
2017, 2019	2.695	**0.029**
2017, 2020	3.139	**0.011**
2019, 2020	1.129	0.318

Table D17. PERMANOVA pairwise comparison test results for large- and small-bodied fish assemblages in the gorge geomorphic zone of the Lower Murray River from autumn 2015–2020. *P*-values presented in bold are significant comparisons, using Bonferroni corrected α = 0.0033 (Narum 2006) for comparisons between years (fifteen comparisons).

Comparison	**Large-bodied**		**Small-bodied**	
	t	*P (perm)*	*t*	*P (perm)*
2015 vs. 2016	2.0305	0.0065	1.1526	0.2541
2015 vs. 2017	3.5839	**0.0006**	2.0765	0.0125
2015 vs. 2018	1.7638	0.0400	1.2070	0.2185
2015 vs. 2019	1.2874	0.1767	1.0302	0.3419
2015 vs. 2020	1.3092	0.1679	1.2146	0.2132
2016 vs. 2017	2.2942	0.0036	3.7883	**0.0002**
2016 vs. 2018	1.7945	0.0289	0.8127	0.5881
2016 vs. 2019	1.6992	0.0421	1.7025	0.0483
2016 vs. 2020	2.4776	0.0043	2.2132	0.0100
2017 vs. 2018	2.6821	**0.0015**	3.1583	**0.0007**
2017 vs. 2019	3.1843	**0.0006**	1.9555	0.0170
2017 vs. 2020	3.0791	**0.0004**	2.8131	**0.0002**
2018 vs. 2019	1.7619	0.0417	1.4932	0.1031
2018 vs. 2020	1.7910	0.0387	2.1635	0.0127
2019 vs. 2020	1.4898	0.1118	2.0557	0.0129

ACRONYMS

AHD	Australian Height Datum
CEW	Commonwealth environmental water
CEWO	Commonwealth Environmental Water Office
CLLMM	Coorong, Lower Lakes and Murray Mouth
DEW	Department for Environment and Water
DOC	Dissolved organic carbon
ENP	Ecosystem net production
ER	Ecosystem respiration
GPP	Gross primary production
LMR	Lower Murray River (South Australian section of the Murray River).
LTIM	Long-Term Intervention Monitoring
M&E	Monitoring and Evaluation
MDB	Murray–Darling Basin
MDBA	Murray–Darling Basin Authority
NPL	Normal pool level
NSW DPIE	New South Wales Department of Planning, Industry and Environment
PSU	Practical salinity units
RMIF	River Murray Increased Flows
TL	Total length
TLM	The Living Murray
VEWH	Victorian Environmental Water Holder
YOY	Young-of-year

GLOSSARY

Allochthonous	Refers to foreign or outside sources. For example, organic matter of an allochthonous source is that which has been produced outside of the river channel, e.g. terrestrial or floodplain material.
Autochthonous	Refers to local sources. For example, organic matter of an autochthonous source is that which has been produced within the river channel.
Base flow	Flows that are confined to the low flow part within the river channel.
Biofilm	A collection of microorganisms (e.g. bacteria) attached as a 'film' on living (e.g. tree root) and non-living (e.g. wooden pylon) surfaces.
Direct trade	"Direct trade" refers to an order for a specified volume of environmental water to be delivered at the South Australian border. Typically a timing and profile for the delivery is specified and river operators can meet the order by providing water from any available source.
Flood or flooding	Refers to flows that are overbank. In South Australia, this is deemed to be above bankfull flow (45,000 ML/d).
Freshes (flow)	Flows greater than base flow but below bank level.
Epibenthic	Organisms living on the surface of sediment.
Epiphytic	Organisms that are attached to plants.
Hatch date	The date at which fish emerge from their eggs, which is one day prior to a fish laying its first daily growth increment on an otolith.
Heleoplankton	Plankton derived from billabongs and other floodplain still, generally-vegetated, waters.
In situ	Used to describe monitoring *in* the field.
Lentic	Refers to slower water velocities associated with 'pool water' habitat in highly regulated systems, typically median velocities of approximately ≤0.3 m/s.
Littoral	The margin along the bank of the river.
Lower Murray	Term used to describe the Selected Area of monitoring, which includes the Lower Murray River (see below) and the Coorong, Lower Lakes and Murray Mouth (CLLMM).
Lower Murray River (LMR)	Defined as the main channel of the Murray River between Wellington and the South Australian border.
Lotic	Refers to flowing water, typically with median velocities of approximately >0.3 m/s.
Pulse (flow)	A description given to the shape of a hydrograph that is characterised by an increase in discharge, followed by a decrease in discharge, often of similar slope.
Recruitment (reproduction)	Refers to individuals passing the critical stages of early life (e.g. larval) and becoming juveniles in a population, described here as age 0+ years.
Respiration (ecosystem)	Ecosystem respiration is the measure of oxygen depletion in water by respiring animals.
RMIF	River Murray Increased Flows: a type of environmental water. Water entitlements recovered under the Snowy Water Initiative (established in 2002) via infrastructure upgrades and water purchase, which receive annual allocations and are used to supply environmental water to the Snowy River (Snowy River Increased Flows, SRIF) and River Murray (RMIF).
Primary productivity	The rate at which energy is converted to organic substances by autotrophs (e.g. algae and plants) during photosynthesis.
Salt flux	The measure (mass/time) of the movement of salt over a given area or point.

Southern connected Basin	The southern connected Basin is a network of the Murray River and all tributaries that flow into it between the Hume Dam and the sea. The Lower Darling (below Menindee Lakes) is considered part of the Southern Connected Basin, whilst all rivers upstream of Menindee Lakes are considered as the Northern Basin.
QSA	Murray River discharge (Q) to South Australia at the SA-NSW border.
Unregulated flows	Unregulated flows occur when water in the system exceeds demands and are declared to be unregulated by the appropriate authority (source: http://www.bom.gov.au/water/awid/id-1026.shtml). They can be driven by substantial rainfall from upper tributaries, spills from headwork storages and rainfall rejection events.
Weir pool	Stretch of river between two weirs.