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River Murray Channel Productivity Monitoring of the Southern Spring Flow 2020

Gavin Rees[1], Tapas K Biswas[1], Darren Giling[2], Robyn J. Watts[3], Xiaoying Liu[3], Rod Oliver[4], John Pengelly[1], Zygmunt Lorenz[4], Qifeng Ye[5], Paul McInerney[1], Ross Thompson[2], Tim Malthus[1], Klaus Joehnk[1]

[1]CSIRO Land & Water

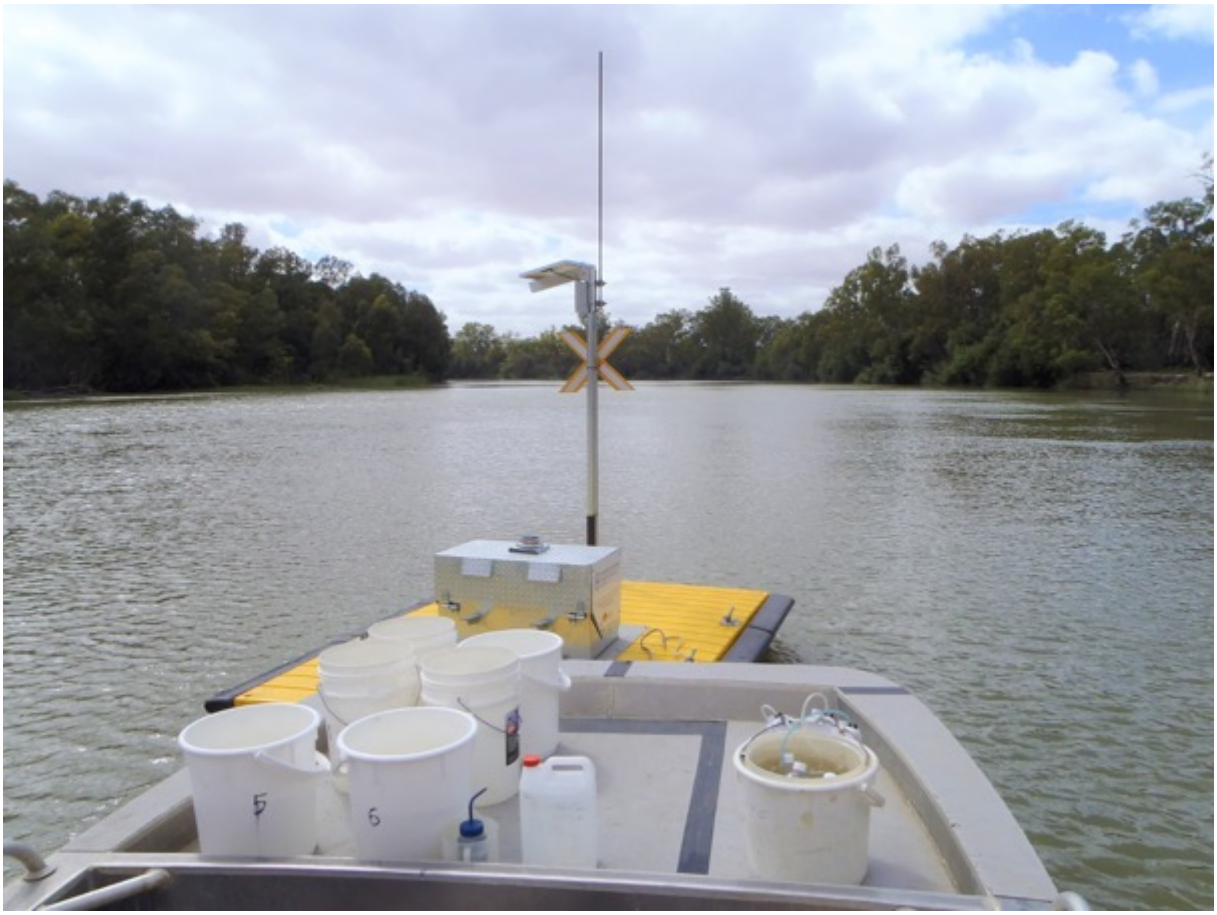
[2]University of Canberra

[3]Institute for Land, Water and Society, Charles Sturt University

[4]School of Biological Sciences, University of Adelaide

[5]South Australian Research and Development Institute

A technical report to the Murray-Darling Basin Authority for the project MD005528



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This report was written by Gavin Rees, Tapas Biswas and Darren Giling with support from others for the Murray-Darling Basin Authority.

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Front page photo: Lower Murray monitoring station downstream Chowilla (credit: University of Adelaide)

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Executive summary

Managed flows for the environment are a key mechanism that can be used by water managers to maintain or improve river and floodplain condition. In Spring 2020, New South Wales, Victorian, and South Australian agencies, the MDBA and CEWO coordinated a system scale environmental flow event, also known as the 2020 Southern Spring Flow that mimicked some of the spatial and temporal aspects of the natural flow regimes in the River Murray system.

Commencing in October 2020, the productivity response from this coordinated environmental flow event was measured along the River Murray. The elevated water levels from these flows inundated around 25% of Barmah-Millewa Forest before returning to the River Murray. These flows then combined with flows from the Goulburn, Murrumbidgee and Lower Baaka/Darling Rivers to create a flow pulse down the length of the River Murray from Yarrawonga the Coorong in South Australia.

As water for the environment delivery in the Southern Murray-Darling Basin continues to evolve, there is a growing need to better understand outcomes and learn from coordinated flow delivery at a system scale. The overall objectives of the project, by monitoring the productivity response from the 2020 Southern Spring Flow were:

1. To test the suitability of implementing the CEWO Flow-MER stream metabolism indicators along the River Murray channel.
2. To test suitable pilot indicators for in-stream productivity for a longer-term surveillance monitoring program that aims to capture productivity responses to a range of flow events and help understand the effect of different flow event timings.

To date, stream metabolism measurements in the River Murray channel have only been undertaken in the Lower Murray and as part of some targeted analysis of flooding associated with the Barmah-Millewa Forest. To address objectives, we measured stream metabolism and concentrations of key constituents (carbon, nutrients and Chlorophyll-*a*) at seven selected sites along the River Murray channel and one site on the Edward/Kolety River. We also deployed an emerging digital remote sensing device (HydraSpectra camera) at two sites to generate high frequency chlorophyll-*a* measurement (a surrogate for productivity) to test its potential to capture productivity responses.

Similar to the 2019 Southern Spring Flow event, the 2020 flow event mobilised considerable amount of carbon and nutrients from the Barmah-Millewa Forest (BMF) into the main river channel. This pulse of carbon and nutrients supported in-channel production though increasing basal food resources such as biofilms and phytoplankton and therefore increasing food resources available for higher trophic levels in the foodweb.

The stream metabolism measurements showed that gross primary production (GPP) measured upstream of the BMF was highest among all the sites due to instream production being dominated by autotropic organisms (such as algae, cyanobacteria and plants). Water for the environment delivered to the BMF mobilised dissolved organic carbon from the floodplain and the receiving River Murray channel downstream of the forest was found to be strongly heterotrophic, demonstrated by high ecosystem respiration (ER), consistent with a boost in in-stream production.

Increased heterotrophic production was detected as far as Boundary Bend, although GPP and ER became balanced towards the end of the monitoring program. Both the Lower Murray sites in South Australia showed an increase in the rate of GPP from late October through to mid-November 2020 and as the flow pulses reached these sites in mid-November 2020, overall response was often balanced.

Key Outcomes

Stream metabolism measurements provided the following important insights into the River Murray stream productivity responses associated with the 2020 Southern Spring Flow event.

- There was a significant increase in productivity response downstream of the Barmah-Millewa Forest, consistent with the mobilisation of carbon and nutrients from the floodplain into the River Murray and Edward-Kolety River.
- A clear positive productivity response was detected at Barham and Boundary Bend, lasting for up to 4 weeks. A strong heterotrophic response was also detected at Torrumbarry, showing overall increased metabolism at this site, but there was greater variability over time at Torrumbarry.
- Continuous stream metabolism measurements demonstrated increased productivity occurred at the Barham and Boundary Bend sites (longitudinal response), whereas monitoring of constituents alone (dissolved organic carbon, nutrients and chlorophyll-*a*) provides equivocal evidence of productivity responses
- The Lower Murray sites showed a small increase in volumetric gross primary production in early November 2020, but for much of the monitoring period, gross primary production and ecosystem respiration were generally balanced.

While stream metabolism indicators are well developed and provide daily measures of productivity response, the approach continues to be refined.

- A 'metabolic fingerprint' approach currently being developed aggregates all data from a given sites and provides a clear graphical representation of productivity responses at each of the sites.
- Stream metabolism measurements require dissolved oxygen logging probes to be maintained at approximately fortnightly intervals, that can be incorporated into existing monitoring programs and provide a cost effective way to continuously measure productivity changes.
- Additional measurements such as cross section and water depth will allow area-based measurements of productivity (production per unit length of river channel), giving better comparison or response between sites.
- For a longer-term surveillance, emerging digital remote sensing measurement of productivity indicators/surrogates (DOC, Chlorophyll-a) by above water low maintenance HydraSpectra camera can provide high frequency (15 min interval) data from any such monitoring program.

1 Introduction

1.1 Aquatic ecosystem metabolism

Productivity in aquatic ecosystems refers to the rate of generation of biomass by different organisms. Rather than being a measure of how much of a given material or biomass of a given organisms is present, it is a measure of the rate of change associated with organisms, thus it is a measure of the functioning of components of a system, rather than simply the structure of different components of a system. Productivity of autotrophs such as plants is called primary productivity, whereas production by heterotrophic organisms such as bacteria and animals are termed secondary productivity.

Gross Primary Productivity (GPP) is the conversion of energy from sunlight into organic molecules (e.g. sugars) during photosynthesis and is carried out by all micro autotrophs including cyanobacteria, algae and other microphytes in the system (Figure 1). Ecosystem Respiration (ER) is the collective respiration of all the aquatic organisms present (both autotrophs and heterotrophs) and is carried out as organisms obtain energy through oxidising carbon compounds (Figure 1). Aquatic ecosystem metabolism, often simply referred to as stream metabolism, characterises the collective production and oxidation of organic carbon by all the organisms at a given site (Figure 1) (Odum 1956; Young and Huryn 1996; Oliver and Merrick 2006; Bernhardt et al. 2018).

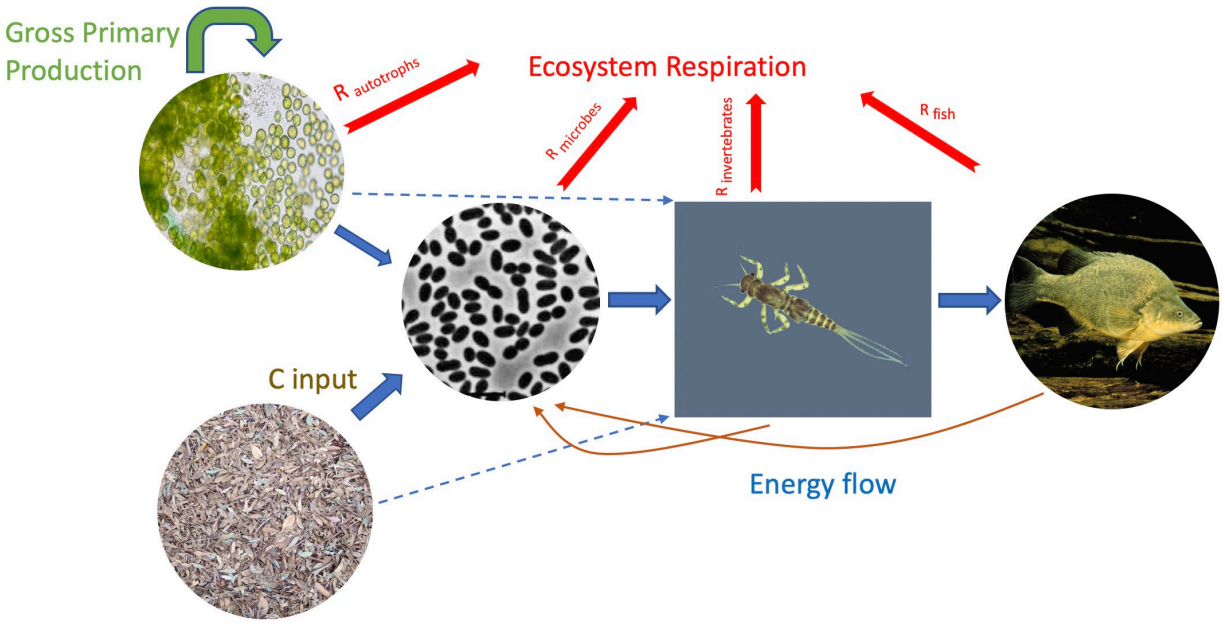

Figure 1. A simplified representation of gross primary production (GPP), ecosystem respiration (ER) and energy flow in a stream food web. GPP is carried out by algae and plants (autotrophs) and ER is carried out by all other trophic levels of the food web. Litter is a source of cabon to the system, metabolised by microbes and consumed directly by invertebrates. Higher trophic levels also feed carbon back though the food web as waste and decaying material. Modfied from(Rüegg et al. (2020).

The rates of GPP and ER provide direct measures of secondary productivity, however may not be able to be correlated with the productivity of organisms higher in the food chain (e.g. fish) (Ruegg et 2020) This is because there can also be factors other than food resources that may limit the growth of higher trophic organisms. High GPP represents the production of more biomass at the

base of the food web and more basal food resource is required to support higher trophic levels of the foodweb. High ER indicates that a lot of organic carbon is being respired, suggesting a high biomass and/or activity of heterotrophic organisms in the ecosystem, meaning more food resource is available to support the higher trophic levels of the food web.

The process of photosynthesis produces oxygen (O_2), whilst respiration consumes O_2. The rate of change of dissolved oxygen (dDO/dt) can thus be described by the formula:

$$d\text{DO}/dt = \text{GPP-ER} + K(DO_{sat}\text{-}DO)$$

where K, the oxygen reaeration coefficient, is a value that reflects the rate of transfer of oxygen across the surface of the water, DO_{sat} is the concentration of oxygen at saturation, and DO is the observed dissolved oxygen. Highly turbulent streams have high values of reaeration, whereas slow-moving lowland rivers such as the River Murray have low reaeration rates.

This means that by measuring the rate of change of DO over 24 hours, it is possible to estimate daily GPP and ER at a given time when the reaeration rate is known or can be calculated.

The availability of sensitive, stable and comparatively inexpensive DO logging devices means that it is relatively easy to obtain the data to calculate GPP and ER. As a consequence, ecosystem metabolism has emerged as a useful measure of ecosystem function and has been employed to measure productivity responses from managed flows in a number of studies within the Murray-Darling Basin (Vink et al. 2005; Oliver and Merrick 2006; Cook et al. 2015) and overseas (Appling et al. 2018; Jankowski et al. 2021). Since 2015, GPP and ER measures have been employed widely as part of the CEWO's Long-Term Intervention Monitoring Program (LTIM) (Hale et al. 2014) and continues at selected areas in the MDB as part of the CEWO Flow Monitoring, Evaluation and Research (Flow-MER) projects (Watts et al. 2018; Dyer et al. 2019; Webb et al. 2019; Ye et al. 2020). Measurements of GPP and ER in the River Murray have generally been limited to few sites within main channel as part of the Lower Murray Selected Area of the Flow-MER project (Ye et al. 2020) or at other sites where targeted research questions have been addressed (Cook et al. 2015), therefore an objective of this project was to also determine whether further stream metabolism measurements would complement existing data-collection programs.

1.2 2020 Southern Spring Flow

Improved instream productivity was one of the key ecological objectives of the 2020 Southern Spring Flow event, where managed flows for the environment were coordinated with other flows to create a pulse along the length of the River Murray with the objective of increasing productivity.

The 2020 Southern Spring Flow event involved co-ordinated management of flows from the Murray, Goulburn, Murrumbidgee and Baaka/Lower Darling rivers (Figure 2) from September to December 2020. At its peak, flows reached 15,000 ML/d downstream of Yarrawonga from mid-October to mid-November 2020, and close to 18,000 ML/d at the South Australian border in the Lower Murray in late November 2020.

These flows are estimated to have inundated approximately 25% of the BMF (Figure 3) with the return waters from the forest transporting constituents such as dissolved organic carbon (DOC) and nutrients into the river channel, which is hypothesised to stimulate instream productivity. Further downstream, water level rises would inundate small areas, and we hypothesised this

inundation would also release carbon and nutrients to the river channel, albeit less than the inundation of the BMF, thereby stimulating instream and downstream production leading to increased GPP and ER.

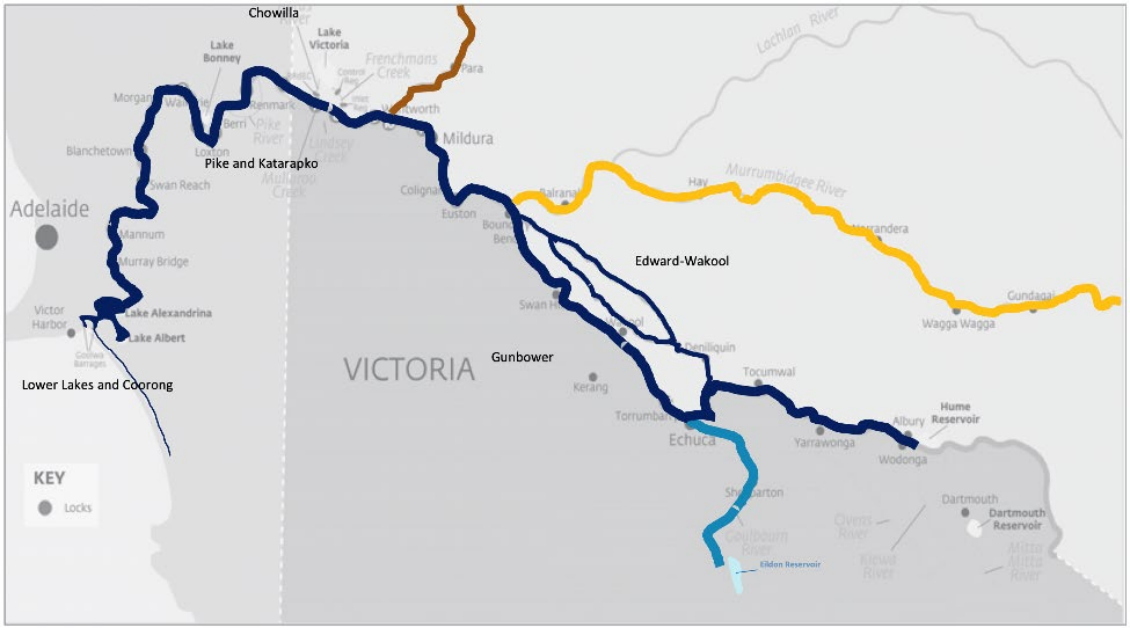

Figure 2. Map showing flow coordination between the major southern basin rivers - The River Murray (dark blue), Goulburn (light blue), Murrumbidgee (yellow) and Lower Baaka/Darling Rivers (brown). Map supplied by MDBA.

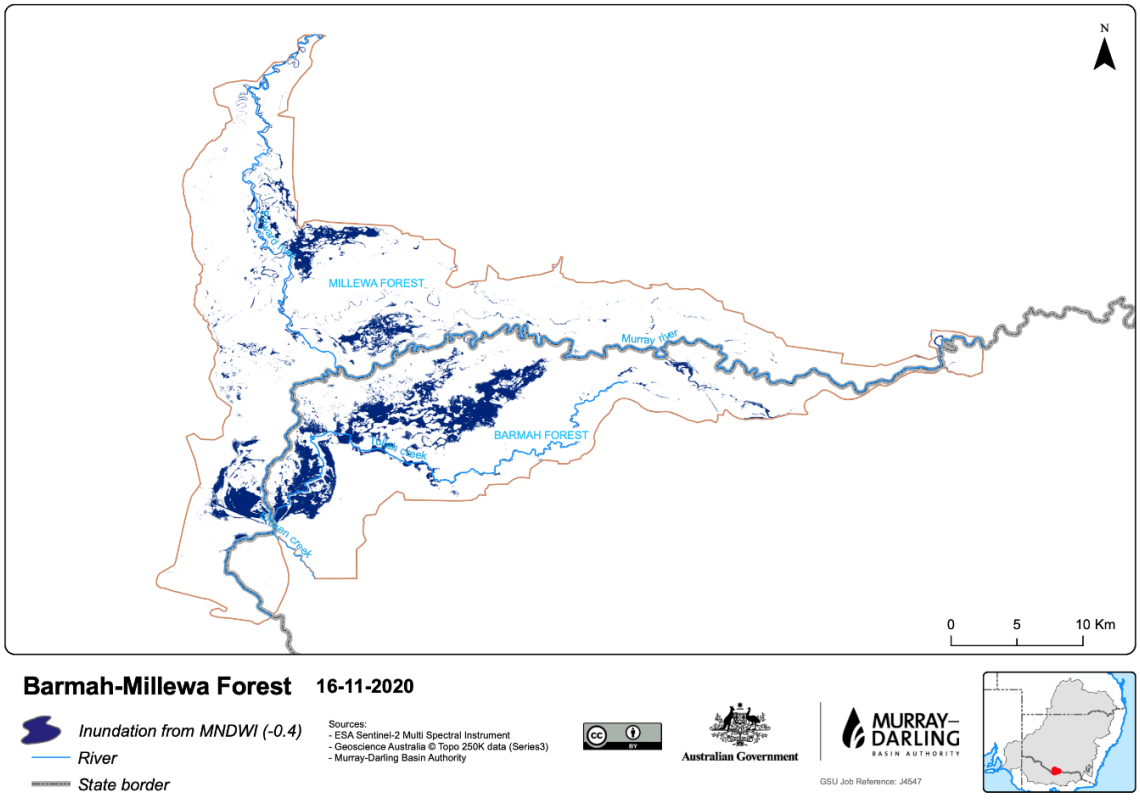

Figure 3. Map showing extent of inundation of the Barmah-Millewa Forest during the 2020 Southern Spring Flow. Map supplied by MDBA.

Although stream metabolism analyses have been carried out across a range of river systems, application to date in the River Murray Channel has only been in the Lower Murray in South

Australia. Given this, the approach of this investigation used was to monitor the metabolic outcomes of the 2020 Southern Spring Flow in other parts of the River Murray Channel. This was to determine its potential as a standardised method for monitoring productivity responses to a range of flow events including coordinated water for the environment delivery and natural flooding in the River Murray Channel. The specific questions addressed in this study are:

1. Can the CEWO Flow MER productivity monitoring indicators (stream metabolism) be applied across the River Murray Channel, and if it is considered suitable, which areas should be targeted for future monitoring?
2. For the pilot indicators– recommendations regarding the most reliable and cost-effective indicators/surrogates for a longer-term surveillance monitoring program for productivity in the River Murray that aims to capture productivity responses to a range of flow events.

2 Methods

2.1 Field sites

Field sampling was carried out at seven sites on the River Murray and one site on the Edward/Kolety River (Figure 4, Table 1). The sampling sites were chosen to determine how different sections of the river would respond to the 2020 Southern Spring Flow. The approach allowed for interrogation of question 1 '*Can the CEWO Flow MER productivity monitoring be applied to the River Murray Channel, and if it is considered suitable, which areas should be targeted for potential future monitoring*'

COVID-19 border restrictions played a role in the selection of the Lower Murray sites as the original intent was to have one site closer to the confluence of the Lower Baaka/Darling River and the River Murray. At the time of planning and into the start of the sampling program, border closures made it impossible for the South Australian team members to cross the border into New South Wales. As a result, sites for the Lower Murray were selected within the South Australian border.

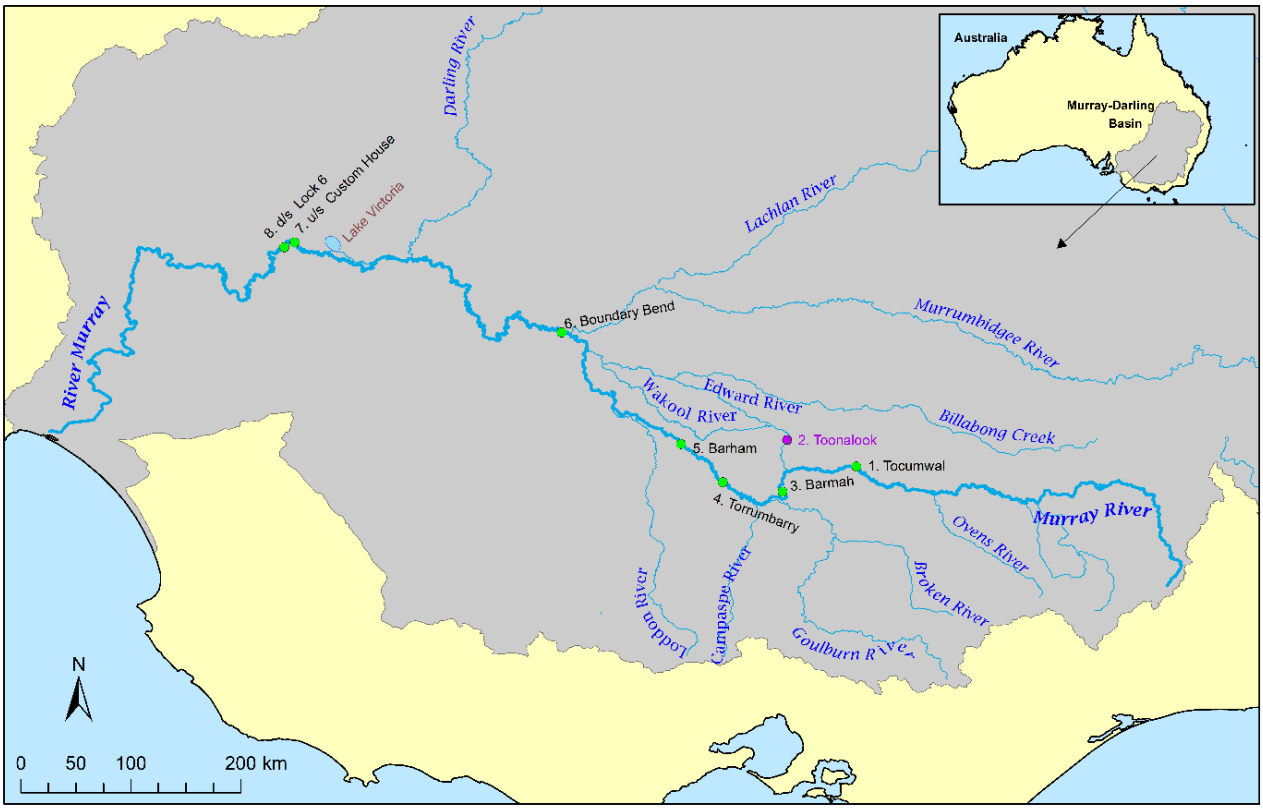

Figure 4. Map of study sites corresponding to Table 1, showing locations for monitoring of Southern Spring Flow 2020. Green circles represent main Murray channel sites and purple show the site on the Edward/Kolety river.

Table 1 Details of sites for monitoring of Southern Spring Flow 2020

Site		Response	Location
1.	River Murray @ Tocumwal	Upstream Barmah-Millewa Forest	-35.813, 145.559
2.	Edward/Kolety River @ Toonalook	Millewa Forest flooding input	-35.596, 144.991
3.	River Murray @ Barmah Township	Barmah and Millewa forests flooding	-36.019, 144.955
4.	River Murray @ Torrumbarry	Main channel processing	-35.942, 144.465
5.	River Murray @ Barham	Main channel processing	-35.629, 144.123
6.	Murray R @ Boundary Bend Township	Main channel processing. Downstream of Murrumbidgee confluence	-34.715, 143.147
7.	River Murray @ upstream of Custom House	Main Channel downstream of Lower Baaka/Darling and Lake Victoria	-33.979, 140.961
8.	River Murray @ downstream of lock 6-Chowilla floodplain	Main channel downstream of Chowilla floodplain	-34.026, 140.840

For the remainder of this report, we used shortened names for each site, simply reflecting the site location, e.g. Tocumwal, Custom House, etc. (Figure 4).

2.2 Water quality monitoring

Fortnightly grab water samples and spot measurements using multi-probe sondes were collected for key water quality parameters. Water quality measurements included dissolved organic carbon (DOC), total nitrogen (TN), total phosphorus (TP), ammonium (NH_4-N), oxides of N (NOx -nitrates and nitrites), filterable reactive P (FRP) and Chlorophyll-*a* (Chl-*a*). For each sampling site, three replicate grab samples for each metric were collected from the flowing part of the river or within weir pools, as close to the centre of the weir pool as practicable.

All analyses were carried out in the CSIRO analytical laboratory, Thurgoona, NSW, which operates under National Association Testing Authority (NATA) accreditation. Detailed sampling and analysis protocols are given at Appendices A2 and A3.

Discharge (or flow rate) data, provided by MDBA were used to calculate daily loads of carbon, nutrients and Chlorophyll-*a* from daily concentrations. Loads were calculated by multiplying the concentration of materials by discharge, to give an amount per time. An increase in load can result from the generation of new carbon or nutrients which can be used by biota.

2.3 Gross primary production and ecosystem respiration

2.3.1 Logger deployment and data collection

Gross primary production (GPP) and ecosystem respiration (ER) were measured as described by (Hale et al. 2014). Field crews deployed Zebra-Tech D-Opto continuous monitoring dissolved oxygen (DO) loggers at Tocumwal, Barmah, Torrumbarry, Custom House and downstream of Chowilla. For the Tocumwal, Barmah and Torrumbarry sites, loggers were manually deployed between 30 and 50cm depth in flowing water, attached to a chain and float mechanism that was

tethered to pontoons or instream structures. Loggers were retrieved every two weeks and replaced with clean, calibrated DO loggers, to provide near continuous logging over the period of the project. All new DO loggers specific to this project were programmed to measure DO at 10-minute intervals.

In situ DO probes that are part of State government agency monitoring programs are present on the Edward/Kolety river at Toonalook, River Murray at Barham and Boundary Bend. These probes are cleaned and maintained fortnightly. Dissolved oxygen readings were being recorded at 15-minute intervals by these three in-situ probes and DO data were obtained directly from on-line data warehouses.

Light sensors that continuously log photosynthetically active radiation (PAR) were deployed in open spaces on properties adjoining the sites where DO loggers were deployed, or on infrastructure within the river (e.g. pontoons or within fenced areas) to improve security and avoid shading during the day. Light loggers were set to integrate light over 10- or 15-minute intervals, consistent with their respective DO logger.

2.3.2 Additional data collection

Atmospheric pressure values were obtained from the nearest Bureau of Meteorology monitoring sites. Discharge data for each site was obtained from the MDBA.

2.3.3 Data analysis

Raw data from DO and PAR loggers, and atmospheric pressure readings were combined and aligned according to their time stamps for further analysis. All data were quality checked for aberrant values. A logger temporarily removed from the water during deployment, as can happen in public places, led to occasional single point spikes in DO and these were either removed, or time was manually replaced with an average of the previous and subsequent DO (note: DO generally varied no more than 0.01 mg over a 10 min interval and the averaging approach therefore was valid). On one occasion, the PAR logger was turned upside down by a stranger, resulting in very low light readings. This erroneous data was not used for GPP and ER calculations.

Quality-checked data from successive loggers for each site were aggregated into single data files for final analysis. Rates of GPP and ER were estimated using the R package BASEmetab (Grace et al. 2015; Giling et al. 2018). The models were provided with prior information on the reaeration coefficient K, which was set to be approximately 2 day^{-1} for all sites based on typical observed values for lowland rivers. This initial estimate was updated for each day as the modelling procedure sought the combination of parameters pertaining to GPP, ER, and reaeration that produce the best fit to the data. Final mean estimates of K ranged from 1.05 to 2.47 day^{-1}. Estimates of daily GPP and ER were only retained for further interpretation if there was a good agreement between observed and predicted DO time series (i.e., the model converged and had a $R^2 > 0.75$). Note that the LTIM program generally used a R^2 cut-off of 0.9, but this was reduced to 0.75 in some cases, and we used the more lenient criteria given the short duration of the 2020-21 monitoring. We added an additional constraint that ER must be at least 20% of GPP. Extremely

high GPP relative to ER is not biologically feasible because the autotrophic organisms must also perform respiration, so such a case is likely a spurious model fit and was discarded.

2.4 HydraSpectra camera

As part of this project we also tested an emerging technology by deploying HydraSpectra cameras that continuous monitor a range of water quality parameters such as chlorophyll-a. Cameras were installed from 9 November 2020 at two sites; one upstream of the BMF floodplain at Tocumwal and one downstream at Barmah (see Appendix B1). While deploying equipment, we received excellent assistance from local businesses in getting them installed on a jetty and on the bank of a resort, both privately owned. The COVID-19 pandemic severely limited our efforts to deploy them on time and to all three locations, which we initially planned for. Since this device is an addendum to test an emerging technology (HydraSpectra) for monitoring river productivity, a high frequency Chl-a measurement at Tocumwal site for a short period is provided in the Appendix B1 (see Figure B4).

2.5 Monitoring sites data analysis

2.5.1 Grouping of sites

For final analysis, the results from eight sites were aggregated into three groups based on key hydrological and biogeochemical responses that were likely to occur due to the 2020 Sothern Spring Flow watering event. The three groups are: 1) those sites that were directly relevant to understand the role of inundation of BMF, and return waters from the floodplain in stimulating in-stream productivity, 2) sites in the middle Murray to examine changes in production and water quality downstream of BMF, and 3) response of the Lower Murray Chowilla and weir pools. The categories are:

1. Barmah-Millewa sites: Murray @Tocumwal, Edward/Kolety @Toonalook and Murray @Barmah.
2. Mid – Murray sites: Murray @Torrumbarry, Murray @Barham and Murray @Boundary Bend.
3. Lower Murray sites: Murray @upstream of Custom House and Murray @downstream of lock 6-Chowilla floodplain.

3 Results

3.1 Hydrology of the 2020 Southern Spring Flow

The overall discharge associated with the 2020 Southern Spring Flow occurred through flows from the Murray, Goulburn, Murrumbidgee and Lower Baaka/Darling Rivers. Releases from Hume Dam targeted flows of up to 15,000 ML/d downstream of Yarrawonga. Regulators within the BMF were opened to maintain river level and distribute water across the Barmah-Millewa Forest Floodplain. Discharge at Boundary Bend was considerably higher than the sites immediately upstream which can be attributed to minor inflows from Loddon (<500 ML/d) and Wakool (~1,000 ML/d) rivers and mainly due to major inflows from the Murrumbidgee River, which started at 1,104 ML/d on 28 October 2020 and peaked at 4,268 ML/d on 15 November 2020 (see Appendix A6). Hydrographs for all sites are shown in figures describing productivity changes measured by water quality change parameters as well as in metabolism measurement figures.

3.2 Water quality changes

Increased flows associated with the 2020 Southern Spring Flow mobilised organic materials from the BMF. The DOC increased from nearly 3 to 6 or 7 mg/L immediately downstream of the BMF while the concentration upstream at Tocumwal remained at approximately 3 mg/L throughout the monitoring period (Figure 5). For both the upstream and downstream of the forest, DOC readings were more or less same by the beginning of January 2021. A small peak in DOC concentrations was measured in late October 2020 at Torrumbarry and Barham, before showing a steady decline for the remainder of the monitoring period. DOC concentration at Boundary Bend remained around 5 mg/L and did not show any significant change during the monitoring period. The absence of any rise in DOC could be explained by dilution with cumulative flows from tributaries especially Murrumbidgee River between Barham and Boundary Bend. For Lower Murray sites, DOC readings were between 4 and 5 mg/L and showed no noticeable change over time.

Chlorophyll-a (Chl-*a*) concentrations were higher at Tocumwal most of the time than both the sites immediately downstream of the BMF (Figure 6). For both the downstream sites, Chl-*a* showed little change over time until the later part of the study, when a general increase occurred, consistent with the general increase in temperature that was detected across all sites (Figure 12; for water temperature see Appendix A5). The Chl-*a* concentrations at Torrumbarry and Barham in the mid-Murray varied over time, but the pattern didn't always reflect changes to discharge. Chl-*a* concentration at these sites increased towards the end of the monitoring period again consistent with increased water temperatures. In contrast, for the Lower Murray sites, Chl-*a* increased markedly over the period from October to December 2020 before showing a small decline, followed by another increase to approximately 25 µg/L. The increase towards the end of the monitoring period is consistent with increasing water temperature, as seen at other sites.

The total nitrogen (TN) concentration at Tocumwal increased steadily from 282 µg N/L to 352 µg N/L over the monitoring period, irrespective of any change in discharge (Figure 7). In contrast, TN concentrations downstream of the BMF increased to approximately 430 µg N/L in response to discharge. TN showed a second increase over time (most notably at Barmah), however, it is not

clear as to the driver for this. TN concentration at Torrumbarry and Barham peaked at 610 and 500 µg N/L respectively, which coincided with the initial increase in flow. TN concentration then declined, followed by minor fluctuations throughout the monitoring period. No significant increase in TN concentration was detected at Boundary Bend (consistent with DOC, noted above), and the TN remained relatively consistent throughout the remainder of the monitoring period, with an apparent rise in early January. TN concentrations at Custom House increased from approximately 460 to 570 µg N/L in late October, followed by a small decline then remained consistent. TN increased from approximately 420 µg N/L to 550 µg N/L downstream of Chowilla and remained at the concentration until late November before decreasing to approximately 420 µg N/L.

The total phosphorus (TP) concentrations were lowest at Tocumwal, increasing from approximately 20 µg P/L to approximately 34 µg P/L at the end of the Spring Flow period, then remained steady throughout the monitoring period (Figure 8). TP concentrations were elevated at Barmah and Toonalook, varying between sampling times, but showed a general increase throughout the study. TP showed little consistent and appreciable change at either Torrumbarry, Barham or Boundary Bend over time. TP was highest at both Custom House and Chowilla, with an initial peak of approximately 60 µg P/L coinciding with initial flow increases. TP decreased before a further rise, peaking in mid-December 2020.

Other than one occasion, ammonium and oxides of nitrogen (NO_x) concentrations showed little change over time at all the mid-Murray sites, and on occasions, many of the analyses for both ammonium and NO_x were below the limits of detection (see Appendix A4). NO_x concentration was particularly high on one occasion at Torrumbarry and Barham, but we are unable to attribute any clear reason for this outlier. Ammonium and NO_x were elevated at the Lower Murray sites. Ammonium varied considerably over time but also between replicates at Custom House, as indicated by large error bars. Two significant peaks occurred with NO_x concentration, which both occurred against frequent values at the limit of detection. We are not able to attribute reasonable causes for these unusually high values.

Filtrable reactive phosphorus (FRP), a measure of bio-available phosphorus, in the mid-Murray sites ranged from below levels of detection to the peak at approximately 6-8 µg P/L during mid-November 2020 at the BMF sites (see Appendix A4). There was no significant difference in concentrations between the sites upstream and downstream of the BMF. FRP was often at limits of detection at Torrumbarry, Barham and Boundary Bend, but increased to approximately 5 µg P/L over October and November 2020, before returning to limits of detection. FRP at Custom House was 5 µg P/L throughout the monitoring period and ranged from limit of detection to 5 µg P/L downstream of Chowilla.

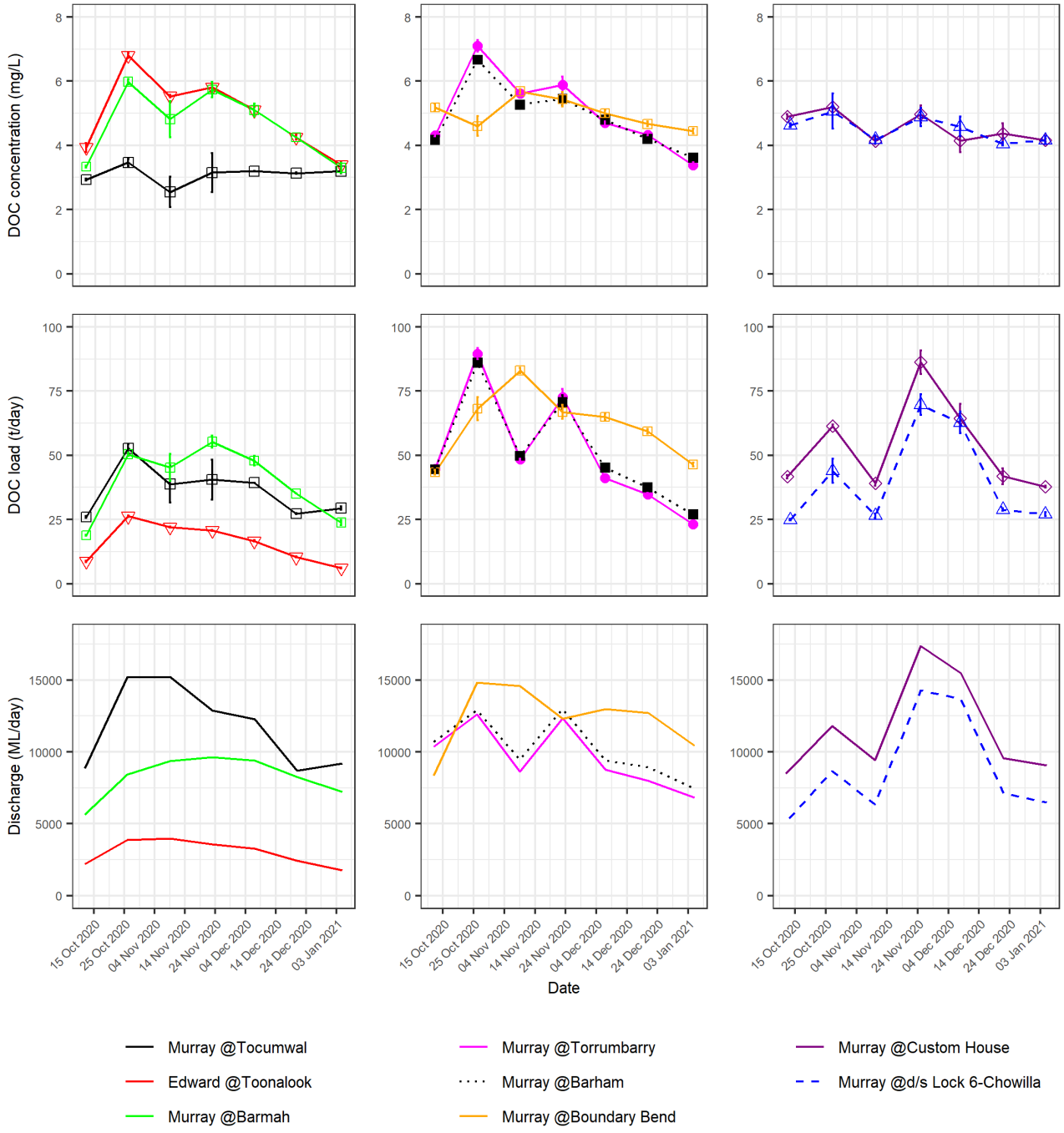

Figure 5. Dissolved organic carbon concentrations, (top graphs), load (middle graphs) and relevant river discharges (bottom graphs). Left column shows Barmah-Millewa sites, middle column are mid- Murray sites and right column shows Lower Murray -South Australian sites.

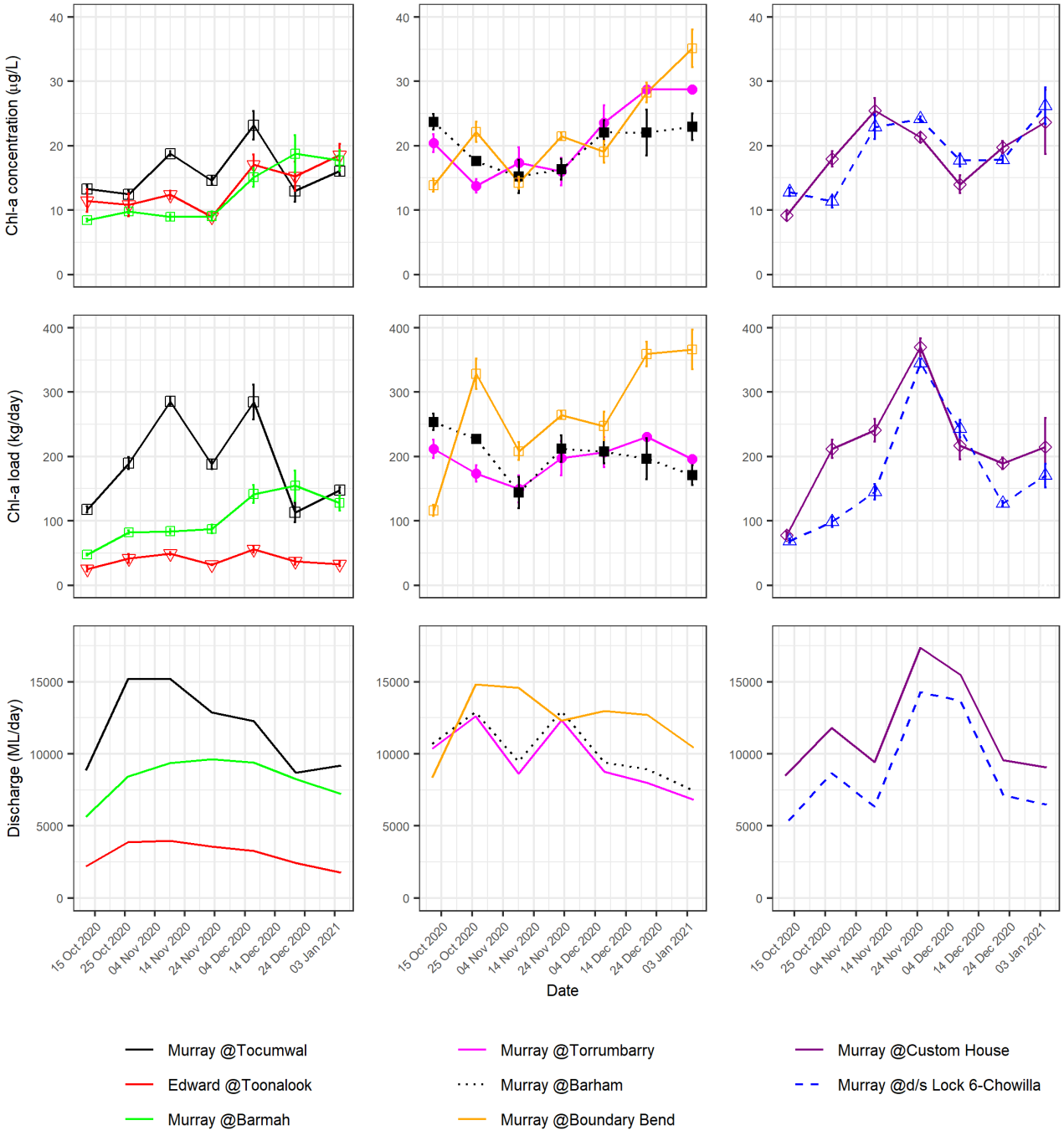

Figure 6. Chlorophyll-a concentrations, (top graphs), load (middle graphs) and relevant river discharges (bottom graphs). Left column shows Barmah-Millewa sites, middle column shows mid Murray sites and right column shows Lower Murray -South Australian sites.

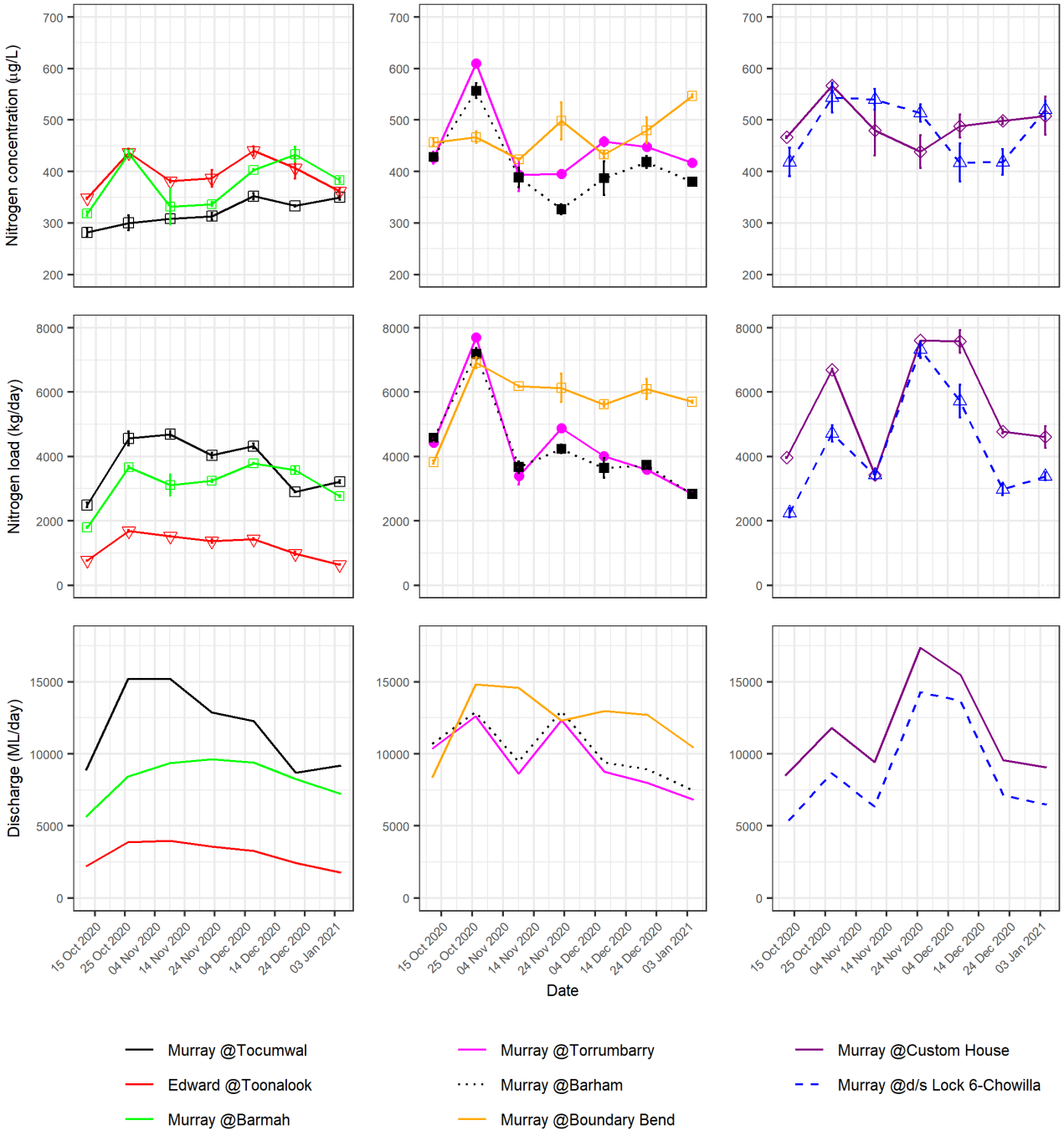

Figure 7. Total nitrogen concentrations, (top graphs), load (middle graphs) and relevant river discharges (bottom graphs). Left column shows Barmah-Millewa sites, middle column are middle Murray sites and right column shows Lower Murray -South Australian sites.

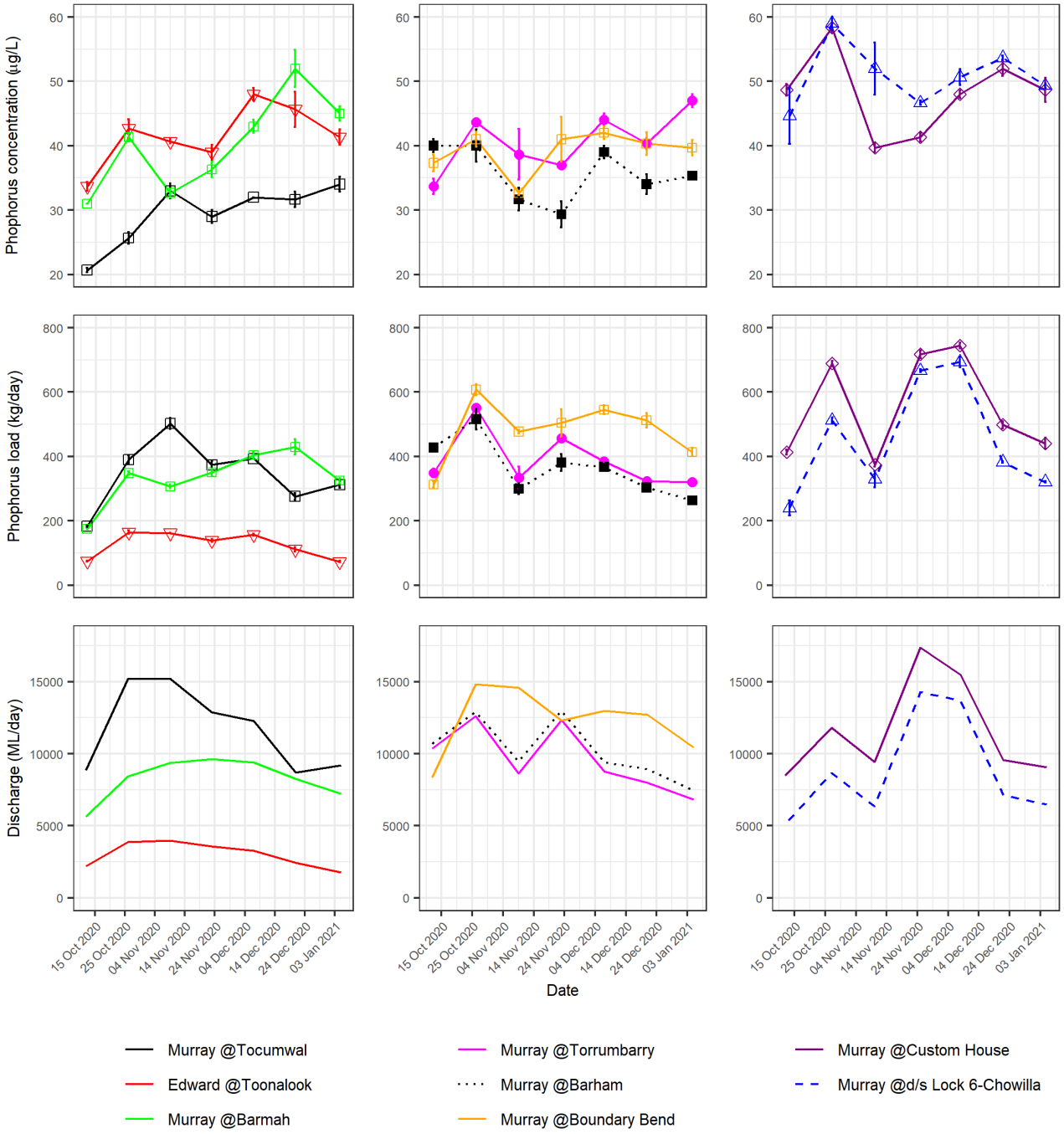

Figure 8. Total phosphorus concentrations, (top graphs), load (middle graphs) and relevant river discharges (bottom graphs). Left column shows Barmah-Millewa sites, middle column are mid Murray sites and right column shows Lower Murray - South Australian sites.

3.3 Stream Metabolism

GPP and ER values were of the same order as previously published results from studies on the River Murray and more widely across the CEWO Flow MER sites (Cook et al. 2015; Watts et al. 2017; Dyer et al. 2019)

There was clear longitudinal variation in rates of GPP and ER over the monitoring period (Figure 9). The highest mean rates of GPP were observed upstream of BMF at Tocumwal (2.27 mg O_2 L^{-1} day^{-1}), which was almost double the mean ER rate at that site (1.19 mg O_2 L^{-1} day^{-1}). This indicates the site was autotrophic (net accumulator of carbon) and suggests a strong reliance of the river food

web on energy produced in-situ by aquatic algae and plants. It is difficult to ascertain whether the Southern Spring Flow contributed to this high productivity at Tocumwal because the monitoring period did not begin until after the flow pulse commenced. However, following the conclusion of the environmental flows, GPP rates at Tocumwal decreased. This may indicate the flow pulse stimulated GPP, perhaps through mobilising nutrients.

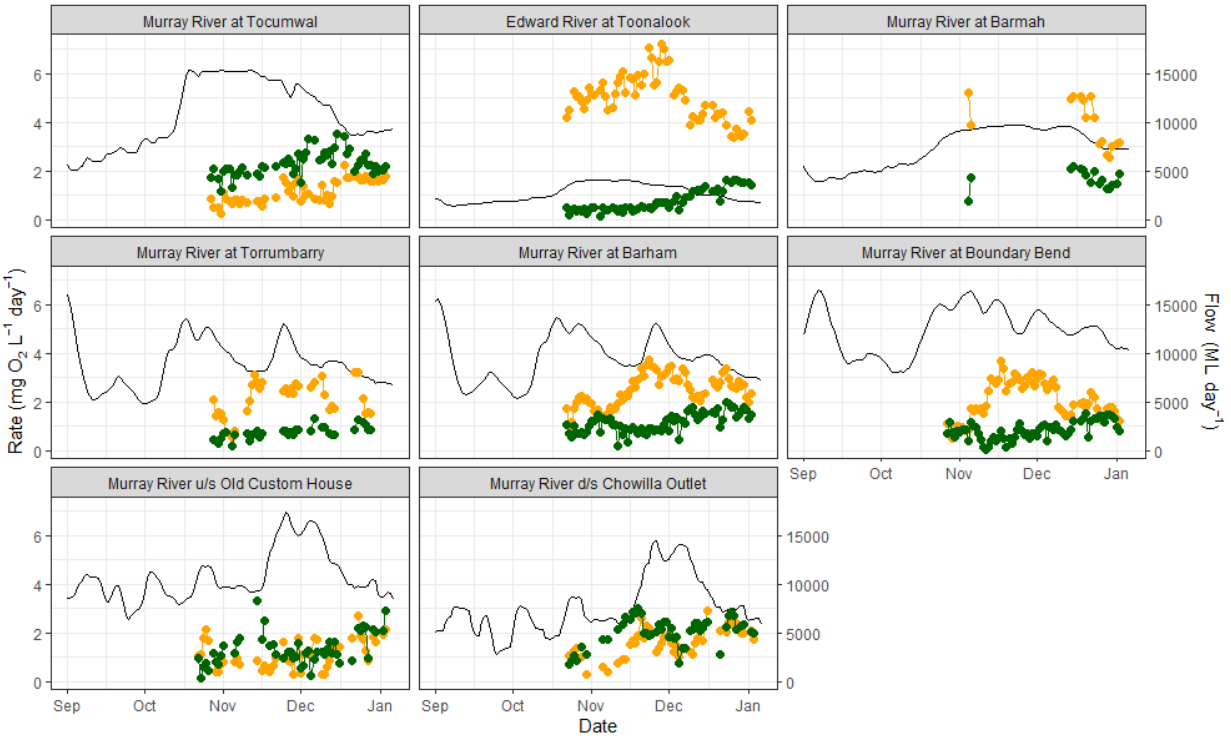

Figure 9. Temporal variation in GPP (dark green points), ER (orange points), and discharge (black line) at the eight monitoring sites. Metabolic results are expressed volumetrically (i.e. mg O_2 L^{-1} day^{-1}). Days with poor model fits or missing data (e.g. due to logger failure) are not shown.

The pattern was reversed in the water entering the smaller Edward/Kolety River from the Millewa Forest, and immediately downstream of the forest at Barmah. At these sites, ER outstripped GPP, demonstrating net heterotrophy (net consumption of carbon). This is consistent with substantial amount of organic carbon inputs from the forest or upstream being respired by heterotrophs, potentially driving high ecosystem secondary productivity. This also indicates that secondary productivity is higher in Edward/Kolety system, for a lot less water requirement. Similar patterns were observed at Torrumbarry, Barham, and Boundary Bend, with rates of GPP similarly low and increasing slightly over the monitoring period, likely due to warmer temperatures (Figure 12). ER at these sites was elevated between mid-November and mid-December 2020.

At the Lower Murray sites in South Australia (Custom House and downstream of Chowilla outlet), GPP and ER were generally more balanced than at the upstream sites. Both sites showed an increase in the rate of GPP from late October through to mid-November and as the flow pulses reached these sites in mid-November 2020, there was a reduction in the rate of GPP per litre of water. This reduction is hypothesised to be caused by a dilution of the organisms living in the water column and potential changes in the underwater light conditions. The increased flows were generally associated with increased productivity in most sites, particularly at Chowilla outlet (Figure 10).

A marked drop in temperature of between 2 and 5 degrees occurred across all sites at the end of November to beginning of December 2020 (Figure 12). It is probable that some of the short-term variation that occurred in ER could be due to the sudden temperature changes.

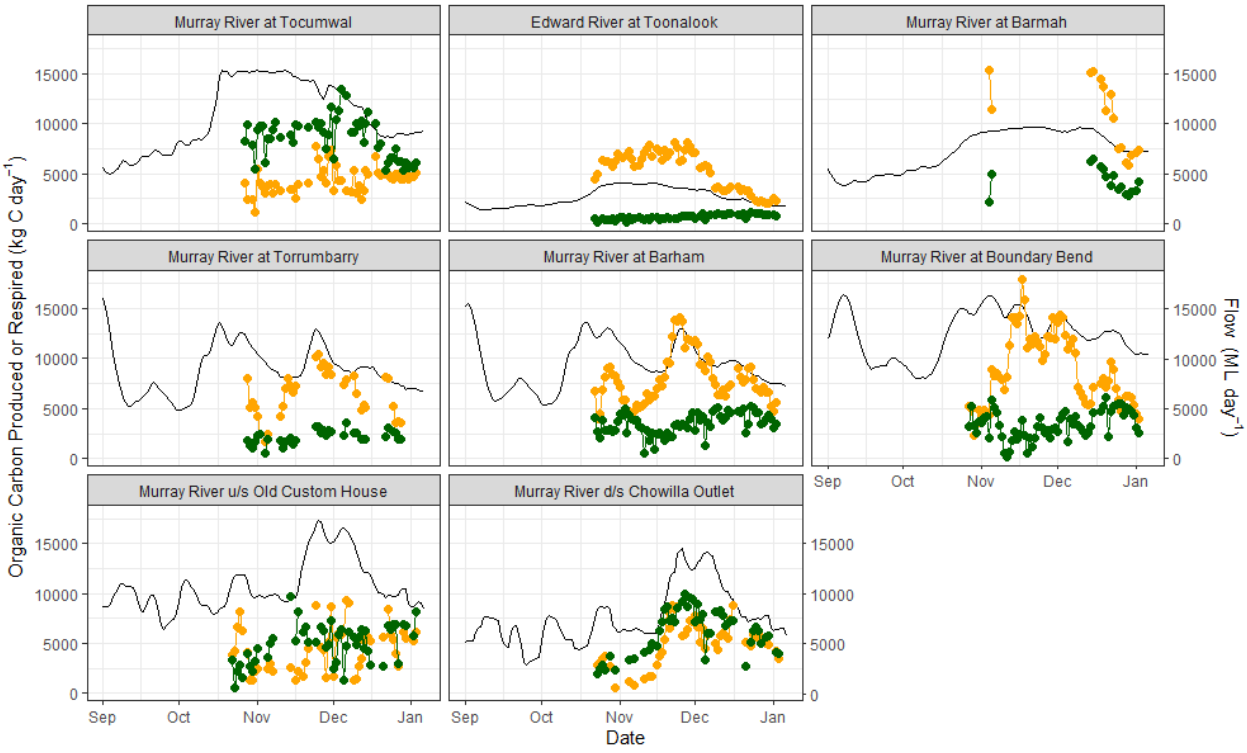

Figure 10. Temporal variation in GPP (dark green points), ER (orange points), and discharge (black line) at the eight monitoring sites. Metabolic results are expressed as a system total (i.e. kg C day^{-1}), calculated by multiplying the volumetric rate by the daily flow. Days with poor model fits or missing data (e.g. due to logger failure) are not shown.

Initial GPP and ER estimates at the Barmah site gave poor fits to the metabolism model, resulting in few points in time where GPP and ER at the Barmah were considered reliable (Figure 9). Close exploration of the raw data showed that maximum DO for many days occurred some 7-10 hours later than the time when maximum light irradiation occurred (data not presented). Maximum DO and light being this far out of step is not realistic, particularly given maximum DO (i.e. photosynthetic activity) occurred in darkness. Transport of oxygen rich water down the river, from a site where high photosynthetic activity occurred represents a reasonable explanation for the time shift (noting technical aspects of logger integrity were examined but no logger failure was found). DO and PAR data were manually realigned to levels that were consistent with other sites, reinterrogated within the BASE analysis and produced a greater number of estimates that fitted the model (Figure 11). The reinterrogated data showing an extensive rise in ER throughout November 2020.

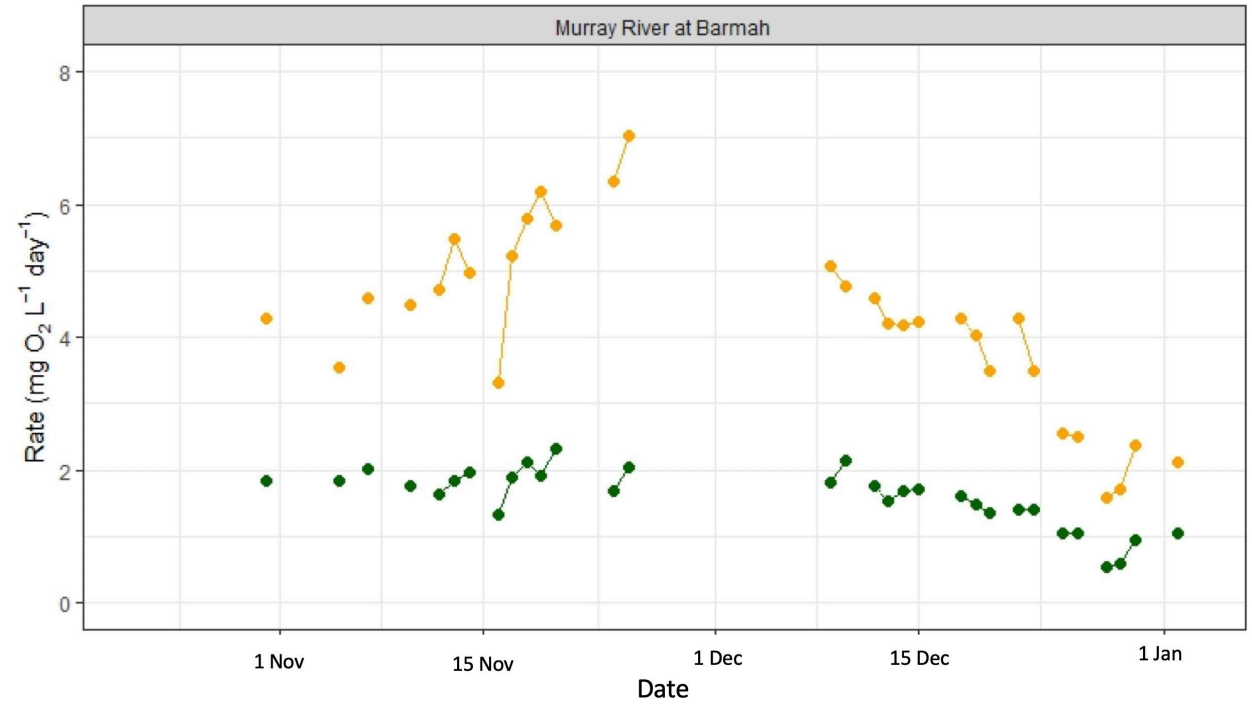

Figure 11. Temporal variation in GPP (dark green points), ER (orange points) for the River Murray at Barmah (mg O_2 L^{-1} day^{-1}) following manual time series correction.

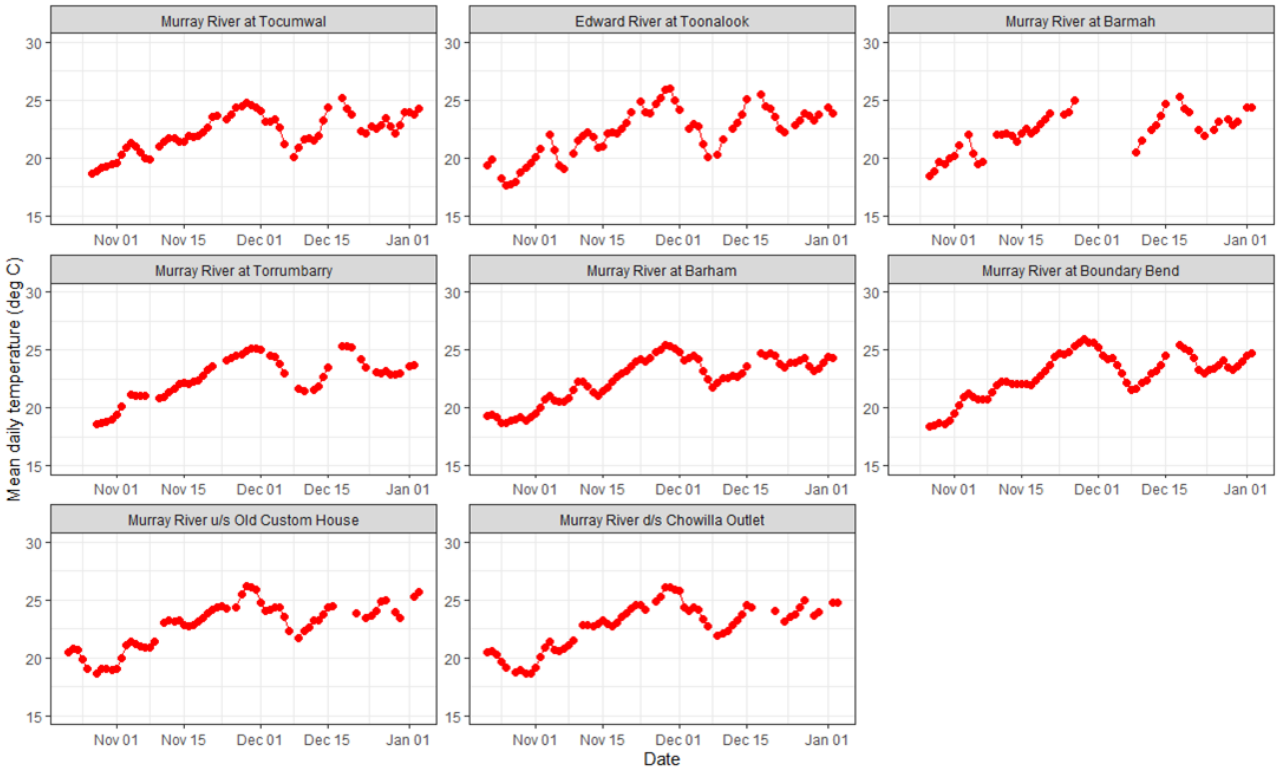

Figure 12. Mean daily temperature at the eight monitoring sites.

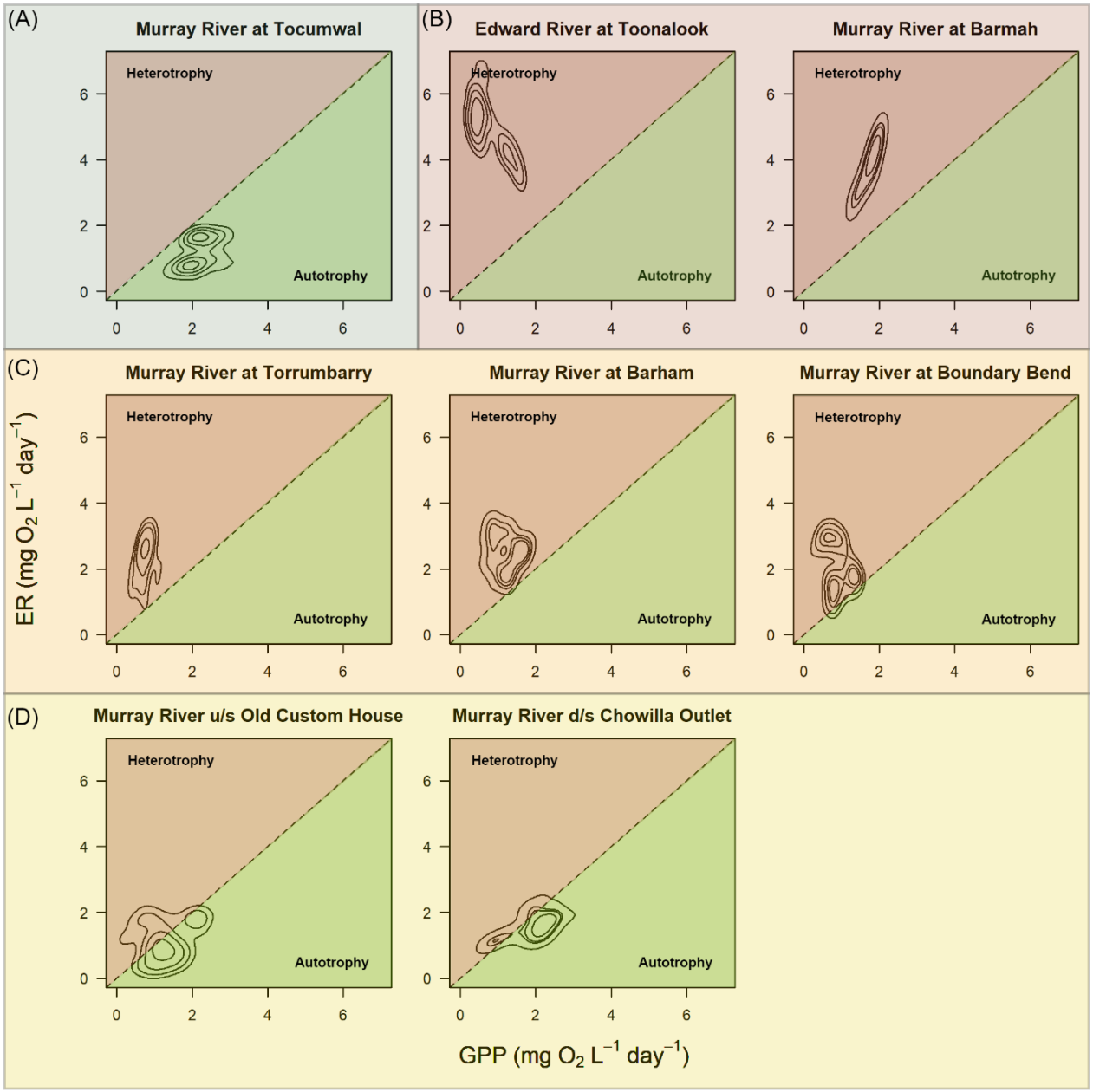

Figure 13. Metabolic fingerprints generated from the 2019-20 River Murray Spring Flow. The diagrams show GPP and ER displayed with a kernel density plot. The contour lines indicate the areas estimated to contain the top 25, 50, 75, and 90% of the GPP and ER points (from the inner to outermost contour lines, respectively). The dashed line indicates when GPP and ER are equal (i.e., net ecosystem productivity is 0).

Metabolic fingerprints are an emerging tool for visualising and diagnosing changes in ecosystem metabolism across time or space and in response to drivers (Bernhardt *et al.* 2018), and we derived fingerprints for the 2020 Southern Spring Flow data (Figure 13). Metabolism was dominated by autotrophic metabolism at the upstream site at Tocumwal (Figure 13A). As the pulse moved downstream and inundated the BMF, terrestrial carbon was liberated and washed into the river channel, increasing in-channel DOC and shifting the metabolic fingerprint towards heterotrophy downstream of the BMF (Figure 13B). The metabolic fingerprints show heterotrophy still dominated from Torrumbarry to Boundary Bend, but started to trend towards autotrophy, brought about by carbon from upstream being incorporated into foodwebs, combined with limited generation of additional carbon within the river channel (Figure 13C). By the time the flow reached the Lower-Murray, the metabolic fingerprints had returned to a balanced state.

4 Discussion/Recommendations

DOC, TN and TP concentrations and loads associated with the BMF the 2020 Southern Spring Flow broadly support the results from the 2019 Southern Spring Flow event (Rees et al. 2020) where elevated concentrations downstream of the forest were greater than upstream. Carbon and nutrients transferred from the forest will support in-channel production though biofilms and planktonic algae. For the mid Murray sites, other than one observation where increased concentrations were detected, there was little change in concentrations at Barham and Torrumbarry for much of the sampling period. High variability in concentration between sample times and the limited number of samples limits the capacity to make strong inference about any effect of flow on nutrient concentrations in the middle and lower sections of the River Murray.

The stream metabolism measurement methods based on continuous and high-resolution records of DO generated reliable estimates of ecosystem production and provided important insights into River Murray stream responses associated with the 2020 Southern Spring Flow event. Key insights include:

- Demonstrating a significant increase in productivity downstream of the Barmah-Millewa Forest.
- Clear demonstration of a productivity response to flows at mid-Murray sites (Barham and Boundary Bend) occurred for up to 4 weeks. A strong heterotrophic response was also detected at Torrumbarry, but with a greater variability over time.
- The Lower Murray sites showed a small increase in GPP in early November 2020, but for much of the monitoring period, GPP and ER were generally balanced.

It is important to note that strong inferences on the responses of GPP and ER to the 2020 Southern Spring Flow are difficult across all sites as there was often little or no data for any extended period prior to the increase of flows thus baseline conditions are unknown.

DOC concentrations were lowest and the river channel was strongly autotrophic upstream of the BMF, but elevated DOC drove the system to be heterotrophic downstream of the forest, consistent with previous reports (Cook et al. 2015). Productivity measures at Barmah gave poor fit to the models used in this study to calculate GPP and ER and consequently we have limited data for the River Murray channel immediately downstream of the BMF. The limited data that was obtained at Barmah site indicated a very high ER relative to GPP. Increased ER was clearly notable in the Edward/Kolety River at Toonalook and this finding supports other observations of increased productivity in this system following instream pulses (Watts et al 2019). The increased ER could be detected as far downstream as Boundary Bend, demonstrating a significant longitudinal effect that was not as apparent during the 2019 Spring Flow pulse (Rees et al. 2020). Increased ER demonstrates mobilisation of carbon and nutrients, which provides a strong supply of carbon and energy for instream foodwebs. GPP and ER at Boundary Bend returned to a more balanced state towards the end of the monitoring period and were trending in the same fashion at Torrumbarry and Barham.

Rates of GPP exceeded ER at the Lower Murray sites from early to mid-November 2020 but were reasonably balanced for the remainder of the monitoring period. In general, DOC changed little

over time in the Lower Murray, however, peaks in Chl-*a* generally reflected the increase in GPP. The Lower Murray LTIM/MER stream metabolism results have shown that water for the environment (i.e., more flow) can produce a small boost to overall productivity, although the level of effect has been quite small and potentially restricted mainly by the limited water volumes available to deliver and the largely stable water levels regulated by weirs in this region (Ye et al. 2021). An apparent limited response in GPP and ER at the Lower Murray sites provides valuable contrast to other sampling sites. The results here support the work of Ye et al (2021) who suggest that with the current possible flow rates and delivery constraints, flows will only generate small increases in GPP and ER at the Lower Murray sites. This observation provides valuable insights into factors that are limiting productivity and sustained ecological outcomes in the Lower Murray. Either greater flows, or additional measures are required to occur with small flows if sustained productivity increases are desirable. This data also suggests that water managers should consider appropriate ecological targets associated with environmental flows. For example, if increased fish biomass is considered an important target, then achieving that target will require appropriate habitat, hydraulics and sufficient food resources. Food availability (timing, amount, appropriate type), should be examined in concert with physical/hydraulic parameters to understand how they influence the fish outcomes. This will inform targeted management/interventions, including the delivery of water for the environment, to provide favourable conditions and promote productivity to support riverine food web.

It is important to recognise that stream metabolism measurements, (GPP and ER) as a single, or standalone metric cannot currently provide a target value to assess the success of productivity response to flows or if the response is ecologically significant In other words, it is not yet appropriate to make a single GPP value a target across river systems until more information is available to understand how values exist under a baseline condition. Current understanding of river function describes in general terms how production and respiration ratios change along the length of large rivers (Vannote et al. 1980). However those ratios are affected by different flows and structural aspects of river channels and attempts have been made to integrates the models that consider how rivers function (Humphries et al. 2014). The metabolic fingerprints we present here (Figure 13) are emerging as a useful tool to summarise data that has been collected over wider temporal and spatial scales, allowing a rapid inspection of the most typical rates of GPP and ER (the inner most contour of plots), the balance between GPP and ER (the position of the fingerprint relative to dashed line), the total variability in GPP and ER (the size of the outermost contour), and the correlation between GPP and ER. The availability of long-term baseline data will allow investigation of temporal trends, by plotting multiple fingerprints for each site. As the information behind the fingerprints grows, they could be used to show the stream metabolism at specific times of interest and allow an assessment of whether metabolism during a particular period falls outside the typical window of rates at that site, thus enabling it as a tool to monitor and predict productivity outcomes from managed flows. It is useful to note that the fingerprints do not show the temporal trends in rates (these are shown in Figure 9).

Stream metabolism revealed responses that may not previously have been detected simply by measuring concentrations of constituents. For example, measuring DOC mobilisation has been used as a surrogate for indicating increased production, based on the argument that an increase in DOC will increase ER. However, as discussed elsewhere, simply measuring the concentration of DOC does not indicate the rate that the material is being generated or transformed, therefore

potentially miss detecting the extent of the response (Rees et al. 2020). For example, DOC concentration at Boundary Bend increased by approximately 1mg/L in early November 2020, followed by a steady decrease, yet ER increased almost four-fold in mid-November 2020, followed by a steady decline. It is important to note that, in this study, carbon and nutrient measurements were only taken fortnightly, unlike metabolism measurement, which were calculated at 24-hour intervals. Constituents such as DOC, Chl-*a* and nutrients are potentially highly dynamic and important changes in concentrations may not be detected by the time interval between sampling periods.

Linkages between GPP and ER and secondary production by higher trophic organisms have not yet been clearly demonstrated, and the underlying measurements of GPP and ER cannot yet be used to predict how much energy is ultimately transferred to higher trophic levels (e.g. fish). This knowledge gap is in part due to the lack of integrated research specifically designed to answer such question. The use of metabolism measurements and their capacity to predict secondary production is recognised as a an active area of research (Rüegg et al. 2020). While currently GPP and ER cannot yet be used to predict biomass increases of fish, an appropriate level of GPP must be present with suitable habitat and instream conditions (e.g. hydraulics) to support fish biomass in riverine systems. The delivery of water for the environment could not only influence the GPP (overall energy supply), but also mediate energy pathway through the food webs, thus potentially leading to better higher trophic outcomes.

The key questions addressed in this report are elaborated below:

1. Can the CEWO Flow MER stream metabolism monitoring be applied to the River Murray Channel and if it is considered suitable, which areas should be targeted for future monitoring.

2. For the pilot indicators– recommendations about what are the most reliable and cost-effective indicators/surrogates for a longer-term surveillance monitoring program for productivity in the River Murray that aims to capture productivity responses to a range of flow events.

4.1 Stream metabolism monitoring: suitability for the River Murray

Productivity monitoring using the stream metabolism method can be applied to the River Murray channel. Metabolism methods have been developed over several years and have been used to demonstrate increases in river productivity in response to flow manipulations. Importantly, the stream metabolism approach generates high frequency data and so it is able to detect daily changes in instream function, which can subsequently be combined to provide estimates over any time frame that is of interest (e.g. weekly, monthly, whole of event). Single time point analysis of dynamic parameters, such as chl-*a* are prone to high temporal variability, which can obscure any real concentration changes that may have occurred, so sole reliance on those forms of measurement can have limited capacity to detect responses to variations in flow.

Programs such as the CEWO Flow-MER has generated large data sets over long periods of time, and in different rivers in the Murray-Darling Basin. Interrogation of data from the selected area sites of the CEWO's Flow-MER is ongoing and its Basin-scale assessments are being used to

demonstrate Basin-scale outcomes of Commonwealth environmental water; support adaptive management; and fulfil CEWH legislative requirements under the Basin Plan.

In situ DO probes that are currently part of existing State Government water quality monitoring programs (in our study, sites at Toonalook, Barham and Boundary Bend), provided high quality data that could be used for GPP and ER estimates. In addition to providing quality data, this approach attaches monitoring equipment within in channel infrastructure and is less likely to suffer from theft or tampering by the public.

Integration of GPP and ER over distance

Metabolism methods derive measures of productivity at single points in the main channels and consequently, measurements reflect an integration of productivity that has occurred upstream. Results from this project indicate that productivity derived on the Barmah-Millewa Forest floodplain, or potential areas up stream where high productivity may have occurred (e.g. Barmah and Moira Lakes) could potentially be detected downstream at Barmah township. Downstream transport and integration of data from upstream sources of productivity have not been widely considered in the past but warrant further examination in the future.

Appropriate estimates of scalable GPP and ER

The current Flow-MER method for analysis of GPP and ER over scales (and comparison between sites) adopts an approach similar to that where 'loads' are calculated, based on volumetric rates of production (C produced/L/day). This approach is not widely used among researchers who measure instream production and some caution is required in interpretation of this data. We recommend that any future analyses should investigate in detail which approaches are defensible. To this end, we propose that production estimates be carried out on areal measurement measurements.

Baseline data

There is very little baseline data available for the River Murray, other than long-term data collected at the Lower Murray CEWO Flow MER sites, largely during the Spring–Summer period. A robust baseline data consisting of measurements before and after flow events will value add to assess comparative effectiveness and benefits of size, timing and duration of any future flow event.

River geomorphology and instream debris

River geomorphology together with instream woody debris plays an important part in determining the overall productivity outcome of flow events. Stream metabolism is a collective measure of respiration and production at a site and large wide rivers will be dominated by the metabolism in the water column. In smaller rivers with woody debris and more frequent shallow benches and backwaters, high surface to volume ratio, and habitat complexity will make a greater contribution to overall productivity than the water column alone. This is evident when small volumes of water are delivered to smaller systems in the Edward/Kolety Wakool system, resulting in significant increase in DOC relative to the small volume of environmental water delivered. It is reasonable to expect that responses from delivered volumes of water will vary across different river systems, or parts of a river.

4.2 Pilot indicators - recommendations

Cost effective indicators

Determining cost effectiveness of any indicator/surrogate for long term monitoring programs is ultimately driven by the monitoring objectives that are set for any given monitoring program. In this report we have shown ecological value in measuring stream metabolism, which can be done in a cost-effective way through continuous logging DO and light loggers. By way of comparison, an alternative monitoring objective could be to demonstrate the quality of food resources in response to a managed flow. Such an approach remains an expensive analytical method, but it can potentially provide a valuable measure as to the effectiveness of a managed flow.

Stream metabolism

In situ DO probes are an effective monitoring tool providing high frequency data that cannot be gained from spot measurements. DO probes do require cleaning and calibration at approximately 2 to 3 week intervals during summer e.g., at mid or Lower Murray sites. Maintenance periods are likely to be extended during winter periods, reducing the frequency of field trips.

Potential site locations for stream metabolism

- Site selection should balance between those that provide data to inform ecological outcomes targeted by water and the cost to implement measurements at those sites. For example, probes potentially could be positioned at a series of sites along the Murray that represent the influence of major river inflows (e.g., Goulburn, Murrumbidgee, Darling, Edward/Kolety) and major floodplain connections (e.g., Barmah, Koondrook-Perricoota, Hattah, Chowilla, Pike and Katarapko) to provide data about the influence of the tributaries and floodplains on stream metabolism in the River Murray. It is important that a monitoring site upstream of Barmah-Millewa forest be considered. Long-term deployment of a DO logger at Tocumwal (or equivalent site between Hume Dam and Barmah-Millewa Forest) should be considered.
- Consideration should be given to probes security and the balance between scientifically chosen sites vs leveraging off sampling sites of existing programs. Logger security is an issue with manually deployed loggers, particularly as highest use of the river by the public tends to occur at the time when logger deployment usually occurs. For example, it could be better in the long term if DO loggers were installed and maintained by WaterNSW, VICDPIE, DEW or MDBA at key hydrometric stations along the main channel, where other relevant supporting data is currently being collected. DO loggers as part of an existing hydrographic station network could be a cost-effective approach and once connected to telemetry, can trigger immediate action in case of logger failure. We recommend investment to install DO probes to existing hydrological gauges.
- Future use of DO loggers could also include targeted deployment of loggers where investigation might be warranted. For example, deployment could occur along with other monitoring programs that may be targeting fish responses associated with floodplain inundation and the interaction with the river channel. This additional

deployment could assist in addressing the knowledge gaps associated with metabolism measurements.

Additional measures

Other parameters should be added into the overall metabolism monitoring approach to provide increased capacity to interpret metabolism measurements. We propose incident irradiance and vertical attenuation coefficients and temperature profiles be determined at the monitoring sites. These measurements will be improved assessment of how production occurs throughout the water column. Temperature profiles will also allow testing of the assumption that water columns are mixed. In addition, major features of river metabolism are influenced by the characteristics of the wetted channel including average water depth and average cross-sectional area. These can be obtained from bathymetric maps or transects of monitoring sites through the association of water level and flow. Applying these additional measurements enables greater understanding of the mechanics and drivers of primary production, and the response to flow, thus improving capacity to predict responses in the future.

Further thought can be given regarding the most expedient mathematical routines for fitting the data to the dissolved oxygen model (this project used the BASE model approach). Not all data can be fitted to the model as there are several implicit assumptions, including that the water column is completely mixed. There are a range of approaches available to researchers to derive GPP, ER and reaeration coefficients from raw data and consideration needs to be given to alternative approaches.

HydraSpectra as a remote reconnaissance monitoring tool for river productivity

We have tested an emerging remote sensing productivity measurement tool (HydraSpectra) at Tocumwal and Barmah sites for Chl-*a* concentrations upstream and downstream of BMF (see section 2.4). This approach provided high frequency data that could be applied to a monitoring program, particularly demonstrating the high variability that may occur in Chl-*a* concentrations in response to flows which are not detected by weekly sampling programs.

HydraSpectra is a relatively low-cost (in comparison to water sample analysis), compact, reliable device, which exploits the fundamental physics-based principle that spectral reflectance signals emanating from algal-dominated inland waters contain information which can be related to important water quality indicators e.g., Chl-*a*, turbidity, CDOM. Hydraspectra cameras give instant Chl-*a*, CDOM and turbidity measurements. Once installed this equipment requires minimal maintenance and provides high frequency (15 min interval) remote sensing data. While this technique is only just beginning to be used in monitoring programs, we showed that this could become one of the most reliable and cost-effective ways to monitor indicators/surrogates for a longer-term surveillance program. (Appendix B1).

5 Glossary and abbreviations

Allochthonous: The term is also applied to production and is referring to production in the river that has been derived from materials external to the river, such as terrestrial vegetation and floodplains.

Autochthonous: In river ecology, the term is applied to production (namely, autochthonous production) and is referring to production that has occurred within the river channel, such as that from phytoplankton, within biofilms on woody debris and hard surfaces, and bottom sediments.

Barmah-Millewa Forest (BMF) covers approximately 650 square kilometres between Tocumwal, Deniliquin and Echuca, the BMF is Australia's largest river red gum forest. The forest is adjacent to the main channel of the River Murray and experiences relatively frequent flooding

CEWH: Commonwealth Environmental Water Holder (CEWH) is a statutory position established under the Water Act 2007 (Water Act) responsible for managing the Commonwealth environmental water holdings.

Chl-*a*: Chlorophyll-a (Chl-*a*) is the photosynthetic pigment in algae. Chl-*a* concentration in water samples is used as an indicator of phytoplankton biomass.

CDOM: Coloured dissolved organic matter (CDOM) is the optically measurable component of dissolved organic matter in water. Also known as *chromophoric* dissolved organic matter.

DOC: Dissolved organic carbon (DOC) is the total organic carbon in a sample that has passed through a membrane filter that has a pore size of 0.45 μm.

Flow-MER: The Commonwealth Environmental Water Office (CEWO) Monitoring, Evaluation and Research (MER) Program (Flow-MER) integrates and replaces monitoring and research activities under the Long-Term Intervention Monitoring (LTIM) and Environmental Water Knowledge and Research (EWKR) projects**.** The Flow-MER Program consists of evaluation, research and engagement at the Murray-Darling Basin-scale and on ground monitoring, evaluation, research and engagement across seven Selected Areas.

FRP: Filterable reactive phosphorus (FRP) is often equated to phosphate (sometimes also referred to as orthophosphate). Since the analysis is derived from a filtered sample, the reaction to detect the phosphate can also detect organic phosphorus, that may have passed through the filter. Thus, the sample is more strictly referred to as FRP, rather than phosphate.

Productivity: In ecology, productivity refers to the rate of generation of biomass in an ecosystem. Productivity of autotrophs such as plants and algae are called primary productivity, whereas secondary productivity is production by heterotrophic organisims, from bacteria to animals.

NOx: Oxides of nitrogen (NOx) is a generic term for the mixture of nitrogen oxides.

TN: Total nitrogen (TN) is the sum of nitrate/nitrite, ammonium, dissolved organic nitrogen and particulate organic nitrogen.

TP: Total phosphorus (TP) is the sum of orthophosphate, dissolved organic phosphorus and particulate organic phosphorus.

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7 Appendices

A.1 Stream metabolism

Detailed descriptions of the approach to making stream metabolism measurements are described in Hale et al (2014). Single station continuous logging dissolved oxygen (DO) probes were deployed at sites within the Murray channel, along with light (PAR) loggers, which will log measurements at 10 minute intervals. GPP, ER and reaeration rates were estimated from the diel oxygen curves for each site. The BASE program (Grace et al 2015) was used to analyse the data, noting that the program has been updated to BASEv2 to included recommended improvements (Song et al. 2016).

Regular maintenance, data download and calibrations were carried out at regular intervals throughout the monitoring program. Fortnightly maintenance was carried out for all the sites except those data obtained from already existing probes.

A.2 Water sampling, processing and transport

All samples were collected from the flowing part of the river, or within weir pools, as close to the centre of the weir pool as practicable. In addition, spot measurement of standard physico-chemical parameters (temperature, pH, turbidity, dissolved oxygen and conductivity) was undertaken, recorded. Samples were transported to the CSIRO Albury NATA accredited laboratory for measuring following constituents:

a. Dissolved organic carbon (DOC).
b. Dissolved nutrients – (NH_4, NOx, FRP)
c. Total nitrogen (TN)
d. Total phosphorus (TP)
e. Chlorophyll-a (Chl-*a*)

Sampling Methods, processing and transportation

Three replicates of water samples were collected from each site at each fortnightly trip in a 500 mL bottle which was rinsed with river water three time before sample collection. For DOC, 30 to 40 ml of water sample was collected by passing through 0.45-µm pore-size membrane syringe filters into sterile 70 mL jars with proper labelling. For dissolved nutrients, 10 mL filtered sample was collected into 15 mL vials with proper labelling and leaving air space to allow for expansion during freezing. For TN and TP, a further 30-40 ml unfiltered sample of water was collected in pre-rinsed 70 mL jars and labelled.

For Chlorophyll-a, the water sample was filtered immediately after collection as chlorophyll pigments react with oxygen and light. A volume of 200 ml of sample water was vacuum filtered through GF/C filter papers (Whatman®) and then the filter paper was wrapped immediately in aluminium foil, labelled and frozen.

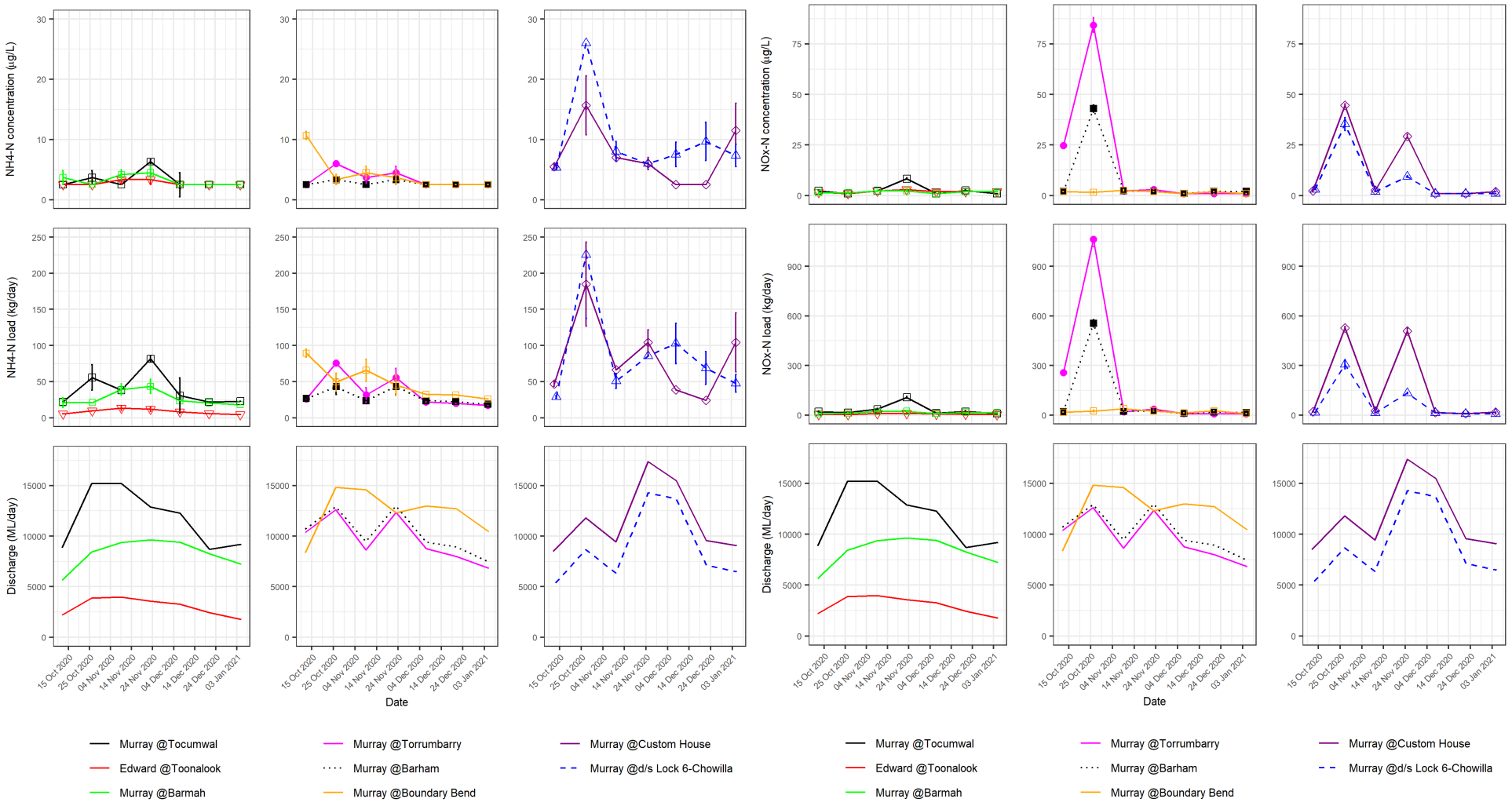
Sample blanks (Milli-Q water) were collected fortnightly and included in sample analysis. The blank was used to estimate the amount of contamination introduced during the sample collection procedure.
All water samples and filter papers were properly labelled, stored on ice whilst in the field and then kept frozen until analysis.

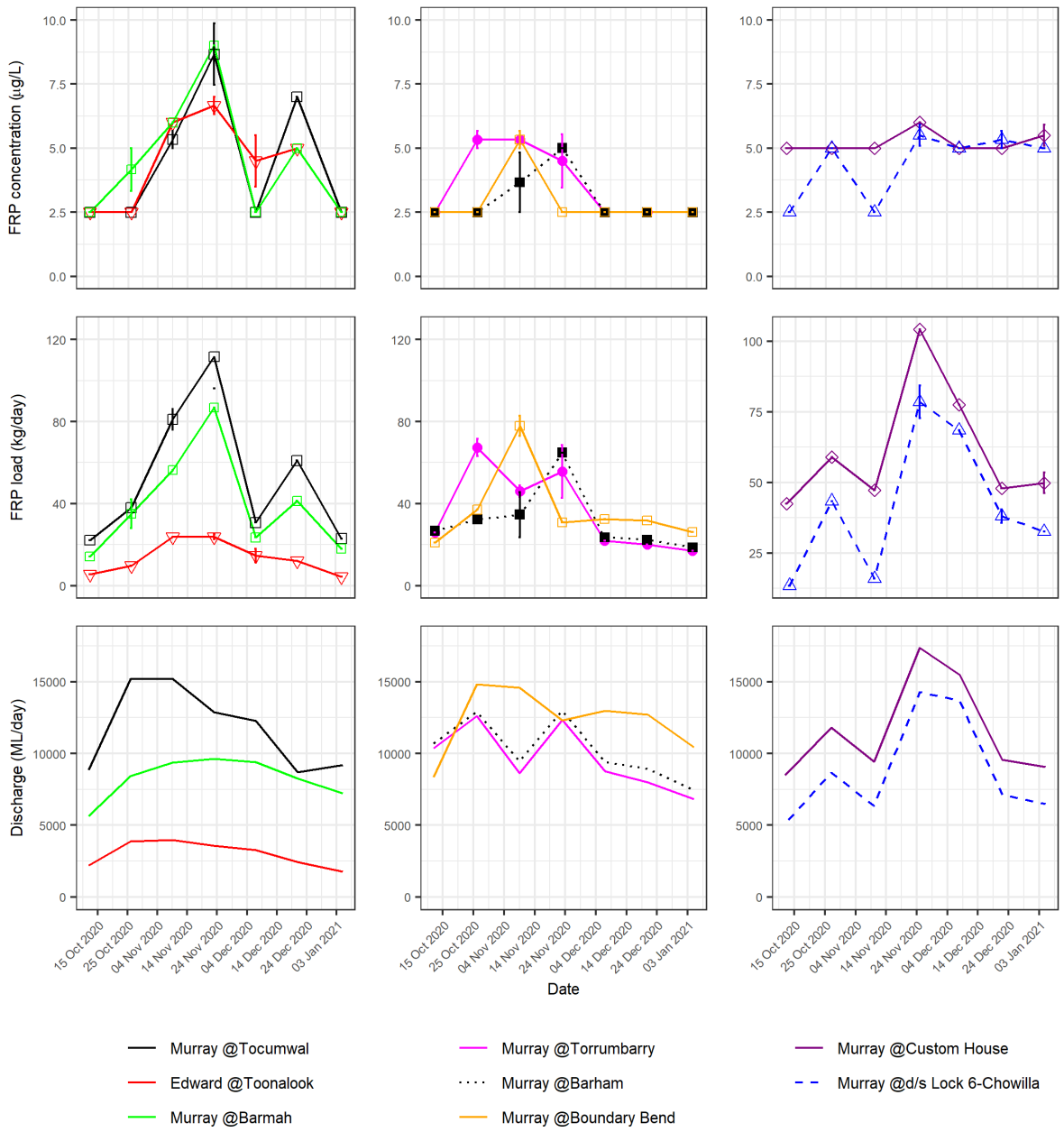
A.3 Detailed chemical analyses

All analyses were carried out in the CSIRO analytical laboratory, Thurgoona, NSW. The laboratory operates under National Association Testing Authority accreditation. The following standard methods were used to examine the relevant carbon and nitrogen analytes:

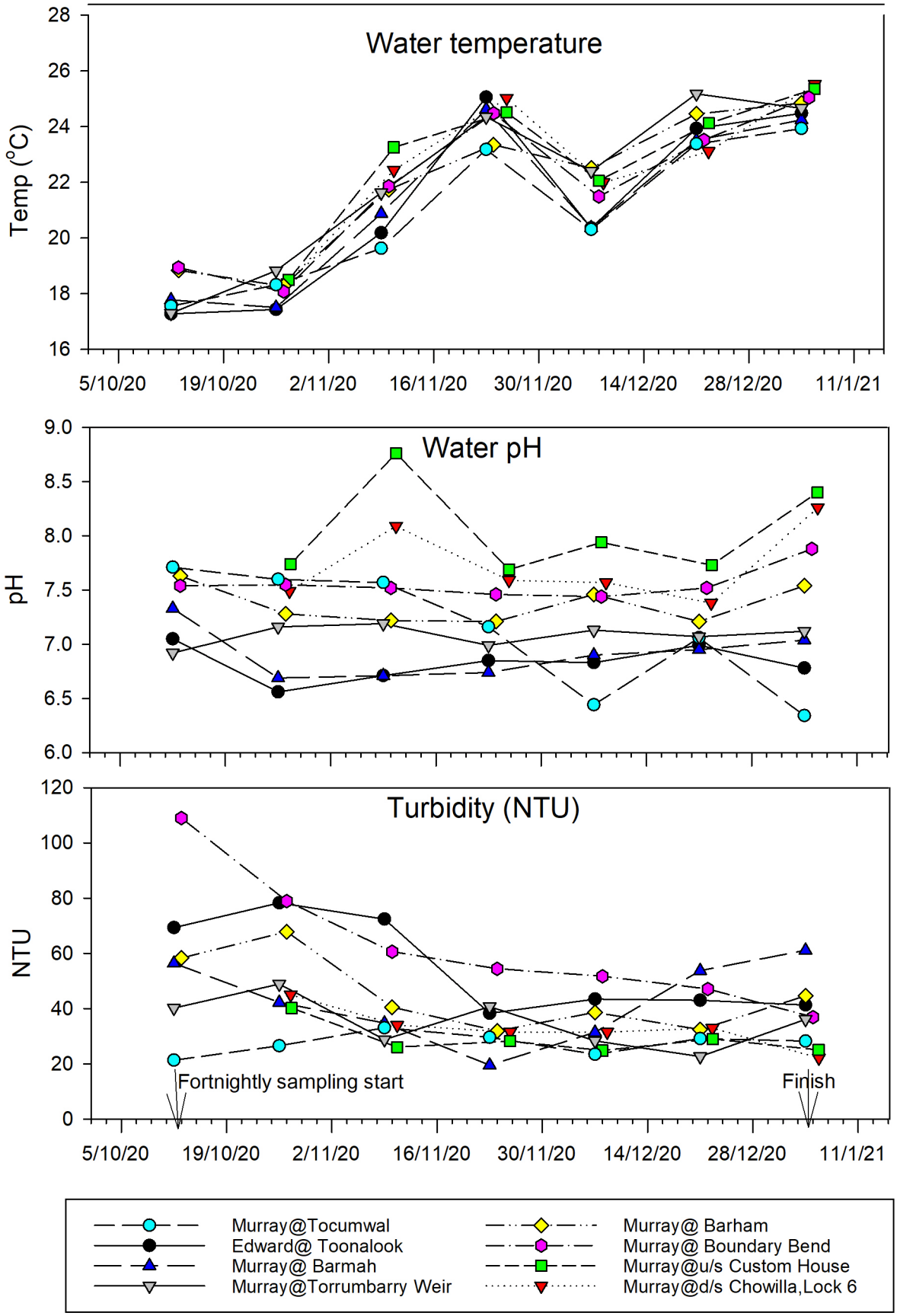
1. NO_3^- was converted to NO_2^- by passing a buffered sample through a column of Cu-coated Cd. Total NO_2^- was then converted to the diazonium salt by reacting with sulfanilamide. The 4-sulfanilamide benzenediazonium chloride is then coupled with N-(1-naphthyl) ethylenediamine dihydrochloride to form a pink dye. Its absorbance was measured colorimetrically at a wavelength of 520 nm.
2. Orthophosphate present in the sample was reacted with ammonium molybdate and potassium antimony tartrate in an acidic medium to form molybdo-phosphoric acid. This was then reduced by ascorbic acid to give a molybdo-phosphoric blue complex, the absorbance of which was measured spectrophotometrically at 880 nm.
3. Organic forms of N and NH_3 present in the sample were digested in an alkaline solution of NaOH-$K_2S_2O_8$ and oxidized to form NO_3^-. NO_3^- in the digestion sample was then reduced to NO_2^- by passing a buffered sample through a column of Cu-coated Cd. Total NO_2^- was then converted to the diazonium salt by reacting with sulfanilamide. The 4-sulfanilamide benzenediazonium chloride was then coupled with N-(1-naphthyl) ethylenediamine dihydrochloride to form a pink dye. Its absorbance is then measured colorimetrically at a wavelength of 520 nm.
4. Organic forms of P present in the sample were digested in an alkaline solution of NaOH-$K_2S_2O_8$ and oxidized to form PO_4^{3-}. PO_4^{3-} present in the sample then reacts with ammonium molybdate and potassium antimony tartrate in an acidic medium to form molybdo-phosphoric acid. This was reduced by ascorbic acid to give a molybdo-phosphoric blue complex, the absorbance of which was measured spectrophotometrically at 880 nm.
5. Chlorophyll pigments were extracted in 90% filtered Ethanol (AR100) and placed in a water bath for 5 min at 75°C. Chlorophyll-*a* was measured by spectrophotometric absorption without acid correction and concentrations calculated as µg/L .
6. Dissolved organic C (DOC) analysis was performed by high temperature combustion (680°C) on a catalyst bed using a TOC-L analyzer by Shimadzu (Kyoto, Japan). Total Inorganic C (TIC) was removed by purging an acidified sample with ultra-pure air. Dissolved organic C in the sample was then injected directly onto the catalyst bed and converted to CO_2. The CO_2 generated was carried by ultra-pure air and detected by NDIR. The resulting mass of CO_2 is proportional to the mass of DOC in the sample.

A.4 Concentrations and loads of NH_4-N (Left), (NOx (Right) and FRP (next page)

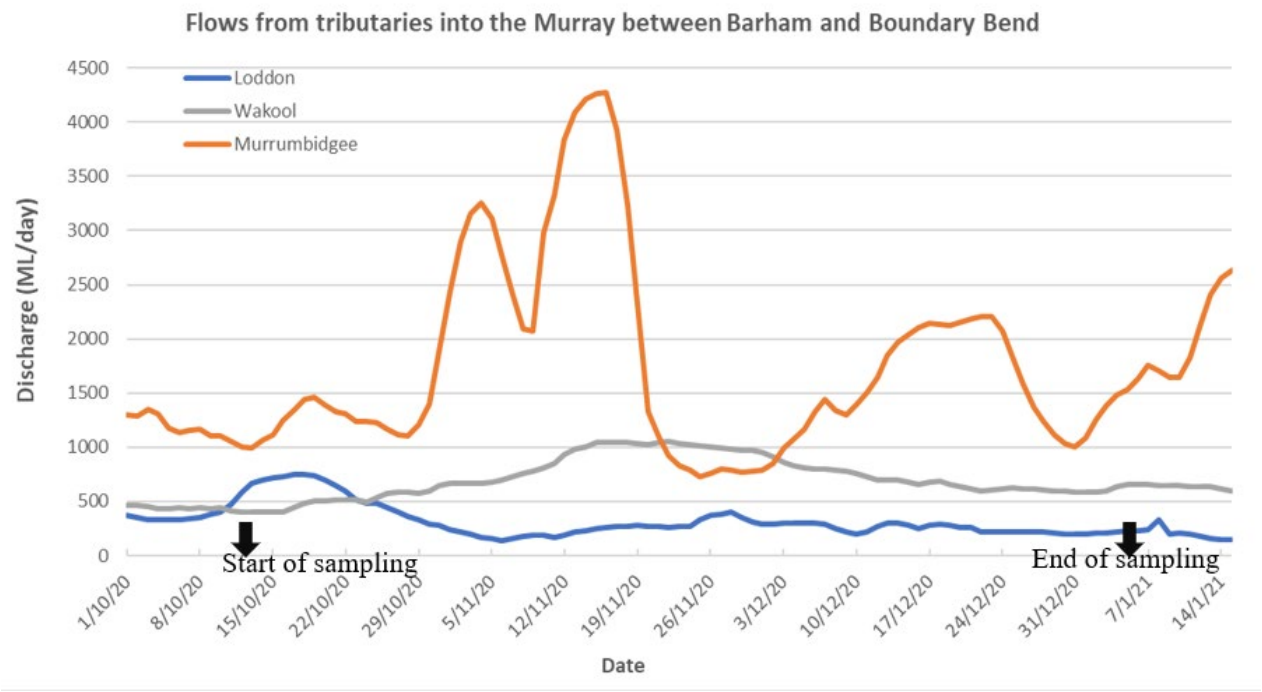



FRP concentration (µg/L)
FRP load (kg/day)
Discharge (ML/day)
Date
15 Oct 2020
25 Oct 2020
04 Nov 2020
14 Nov 2020
24 Nov 2020
04 Dec 2020
14 Dec 2020
24 Dec 2020
03 Jan 2021
Murray @Tocumwal
Edward @Toonalook
Murray @Barmah
Murray @Torrumbarry
Murray @Barham
Murray @Boundary Bend
Murray @Custom House
Murray @d/s Lock 6-Chowilla

A.5 Spot measurements of turbidity, pH and water temperature.



A.6 Flows from Loddon, Wakool, and Murrumbidgee into the Murray



B.1 HydraSpectra for long-term, regular maintenance-free surveillance

Two HydraSpectra devices were installed at Tocumwal upstream of the Barmah-Millewa floodplain (site 1, HS15) and at Barmah (site 3, HS16) on the Murray (Figures B1, B2). A third installation in SA was not possible because of COVID-19 state border restrictions. The devices were installed with optimal azimuthal view angles of 146° and 248°, respectively, to measure the reflectance of the water's surface every 15 minutes. Shortly after installation, HS16 was vandalised and although it continued to function, it was viewing the water incorrectly from which to derive accurate reflectance data. The camera was redeployed on 21 March 2021 at a viewing azimuth of 252°.

Figure B1. The installations of HydraSpectra at Tocumwal Site 1 (left, HS15) and Barmah Site 3 (right, HS16). HS15 and HS16 are serial number for the equipment.

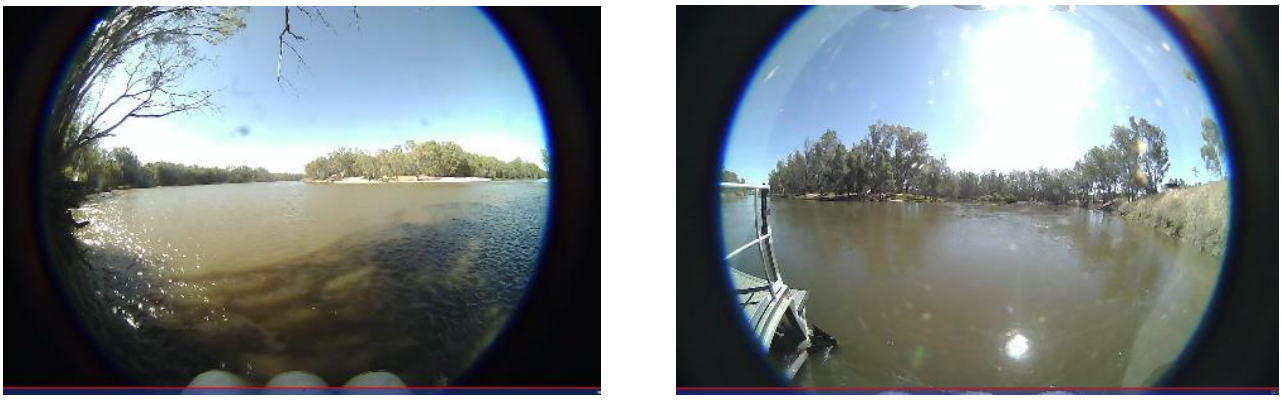


Figure B2. Representative views obtained from the horizontal cameras on the HydraSpectra devices to indicate their outlook.

HydraSpectra measures inputs from different sensors measuring ambient irradiance, diffuse radiation from the sky and water-leaving radiance across the 400-850 nm wavelength range. From these measures an accurate estimate of surface reflectance is made from which water quality parameters such as chlorophyll concentrations can be derived.

The default chlorophyll algorithm applied to HydraSpectra is a three waveband index mostly designed to monitor high algal concentrations typical of algal bloom concentrations (> 20 mg m^{-3}). Calculated chlorophyll concentrations applied to the Tocumwal data suggested that the device was underestimating chlorophyll in comparison to those measured on water samples using laboratory pigment extraction (Figure 5, main report). This suggests the algorithm needs calibration for the River Murray water conditions, but there were insufficient laboratory samples to do this accurately. Instead, an approximate calibration was undertaken.

Figure B3 shows the full time sequence of estimated chlorophyll concentrations determined by the HydraSpectra at Tocumwal between November 2020 and March 2021 period. The data show chlorophyll peaks (~20-25 mg m^{-3}) in mid-November and early-mid December 2020; these peaks are evident in the water sampling data (Figure 5 main report). Since then chlorophyll concentrations have remained relatively low (< 15 mg m^{-3}).

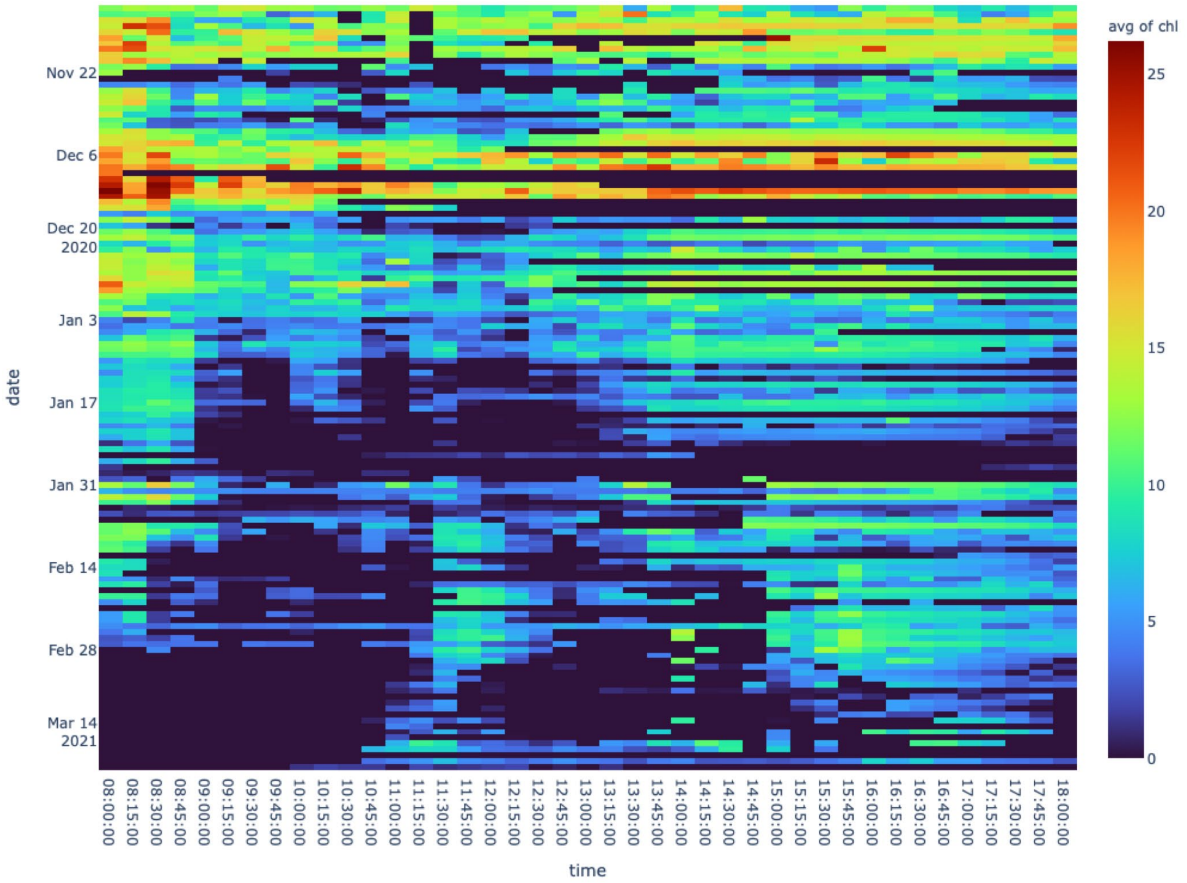


Figure B3. Full time sequence of estimated chlorophyll concentrations estimated by HydraSpectra at the Tocumwal site between November 2020 to March 2021 and over time of day.

These results obtained from the time sequence of daily maximum Chl-a concentrations at the Tocumwal site are shown against the daily air temperature and rainfall data in FigureB4. This shows high frequency Chl-a concentrations fluctuating but generally sustained around 10 mg m^{-3} from late December to March. Fluctuations may be due to errors in estimation related to algorithm assumptions, and to possible variations in flow and weather patterns. This data

highlights the highly variable dynamic and temporal nature of the data and of the need to more frequently detect variations in water constituents compared to conventional water sampling methods which potentially missing short-term fluctuations.

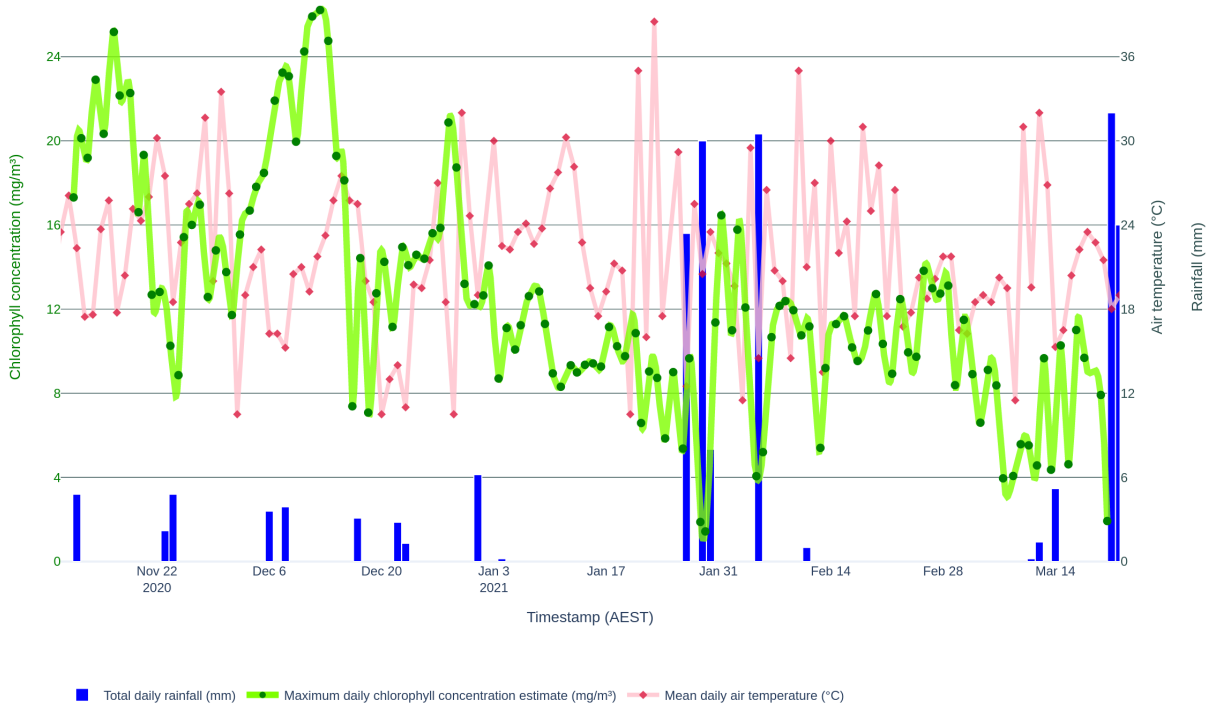


Figure B4. Maximum estimated daily Chl-a concentrations measured by Hydraspectra in relation to rainfall and average air temperature at Tocumwal between November 2020 and March 2021.

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Contact us

1300 363 400
+61 3 9545 2176
csiroenquiries@csiro.au
csiro.au

For further information

CSIRO Land & Water
Gavin Rees
+61 2 6**0519381**
Gavin.rees@csiro.au

Tapas Biswas
+61 2 62464438
tapas.biswas@csiro.au
csiro.au/L**and & Water**