

Addendum to the Long-Term Intervention Monitoring Program for the lower Goulburn River

Winter Monitoring 2018-19

prepared for the

Commonwealth Environmental Water Office

Submitted by:

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1. Preamble

The Commonwealth Environmental Water Office, in partnership with the Goulburn-Broken Catchment Management Authority, Goulburn-Murray Water, and the Murray-Darling Basin Authority is moving to provide greater environmental flows during winter in the Lower Goulburn River.

Under the original Monitoring and Evaluation Plan (MEP) for the LTIM Project in the Lower Goulburn River, little monitoring was planned for the winter period, as it was less of a focus for environmental water delivery, and to improve efficiencies for the overall project.

In May 2018, the CEWO invited the Lower Goulburn River LTIM Project team to develop proposals and rationales for additional monitoring to be conducted during the final 12 months of the five-year LTIM Project, with a focus on identifying the effects of winter environmental flows.

Two types of flow actions were proposed (Figure 1). A winter fresh, delivered over the June-July period had been delivered before, but not specifically monitored. New for 2018-19 would be the provision of high variable baseflows over the period between the end of the winter fresh and the beginning of the spring fresh.

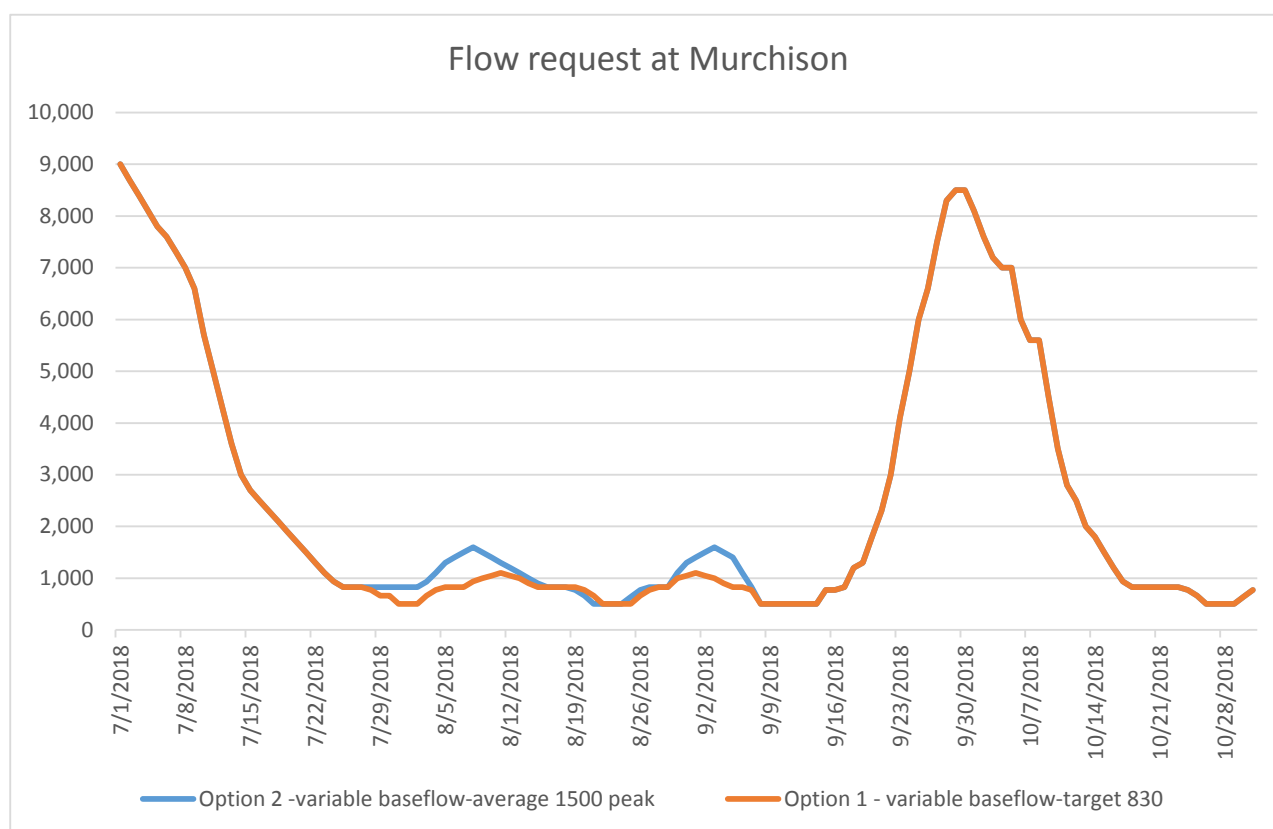


Figure 1. Potential hydrographs for flows in the Lower Goulburn River over winter 2018-19 (GBCMA, unpubl.). The second half of the proposed winter fresh is shown, along with two possibilities for high variable base flows, dependent upon water availability. The heavily monitored spring fresh is also shown, beginning mid-September.

The CEWO and the Lower Goulburn LTIM Project team undertook a prioritization process to identify those proposed monitoring activities that would be taken forward to winter monitoring in 2018-19. Below, the each of the final preferred monitoring activities is described, along with the ecological rationale for undertaking it.

2. Summary of proposed winter monitoring

Table 1 below summarizes the proposed monitoring activities to be undertaken in the lower Goulburn River for winter 2018-19. This includes extensions of existing activities (Ecosystem metabolism), application of existing approaches to new areas (Lamprey movement), and completely new activities (Sediment and plant propagule transport and deposition, Crustacean habitat use, Algal biofilm production). Further details for each proposed activity are included in the sections below, noting that far more detail is provided for the new monitoring

activities compared to those that build upon existing approaches described in the main MEP. Note that some of these activities rely on data to be collected during the following summer to infer the effects of winter flows.

Table 1. Summary of proposed monitoring activities, following the summary table included in the original Monitoring and Evaluation Plan. Abbreviations: CEW – Commonwealth environmental water, LGR – lower Goulburn River.

Monitoring activity	Cate-gory	Zones	No. sites per zone	Rationale for inclusion	Evaluation questions
Sediment and plant propagule transport and deposition	III	1&2	1 + 2	Plant propagule dispersal by flow (hydrochory) is an important process supporting vegetation recruitment and ecosystem diversity of riparian zones, and may be the most important source of propagules to riparian zones and wetlands. Hydrochory can be associated with sediment movement (non-buoyant propagules), or independent of sediment (buoyant propagules). Flow regulation affects both longitudinal connectivity of sediment and propagules (by physical barriers such as dams) and flow patterns which drive transport and deposition. The extent to which environmental water can restore hydrochory needs to be determined.	What does CEW delivered in winter contribute to sediment transport and deposition in the LGR? What does CEW delivered in winter contribute to plant propagule transport and subsequent deposition in the LGR?
Ecosystem metabolism	I	1&2	2	THE LTIM program has been showing some very strong seasonal effects on rates of metabolism, yet wintertime measurements have been thus far constrained to some of winter at one site. A full wintertime data set will enable determination of annual metabolism budgets for the Goulburn River and test the question as to whether wintertime metabolism follows the prediction of low rates due to cooler temperatures and shorter days. Watering actions frequently occur during wintertime, so establishment of wintertime 'normal' metabolic rates can be contrasted with rates under higher flow regimes.	What does CEW delivered in winter contribute to ecosystem metabolism in the LGR? Is ecosystem metabolism in winter in the LGR a substantial component of the full-year ecosystem metabolism?
Crustacean habitat use	III	1,2 +	4 + 5 + 3	Crustacean monitoring will be conducted at multiple sites in both zones to characterize crustacean abundance, biomass and habitat use under variable flows, with an additional site upstream of the lower Goulburn River to provide a point of comparison in the same river system that will not be affected by the same variable flow regime, and another 2 sites in the Broken River to provide another point of comparison with a river system that is unlikely to be experiencing variable flows during the monitoring period (and if it does, these flows will be natural). These results will help determine any beneficial impacts of variable flows on crustaceans and provide information on short-term crustacean habitat use changes under variable flows, helping to interpret data from routine LTIM monitoring.	Does CEW contribute the exploitation of novel habitats by large-bodied crustaceans in the LGR?
Algal biofilm production	III	1, 2 +	1 + 1 + 1*	Algal biofilm monitoring will be conducted at two sites during winter/spring and summer/autumn to characterize changes in biomass and community structure temporally and under variable flows. An additional site in the Broken River will be assessed to provide a point of comparison with a river system that is unlikely to be experiencing variable flows during the monitoring period (and if it does, these flows will be natural). These results will help determine any beneficial impacts of variable flows on algal biofilms and provide information to assist in interpretation of macroinvertebrate and stream metabolism data already included in routine LTIM monitoring.	What does CEW contribute to algal biofilm production in the LGR? Do rates of algal productivity in the LGR differ between summer and winter?
Lamprey movement)	II	1, 2 +	NA	Understanding lamprey spawning migrations in the River Murray system and the influence of flow on migration is important for system scale management of flows and connectivity for highly mobile species.	What does CEW delivered in the LGR contribute to Lamprey movement in the Lower Murray River and connected southern basin?

3. Sediment and plant propagule transport and deposition

3.1 Ecological Rationale

Propagule dispersal by flow (hydrochory) is an important process supporting vegetation recruitment and ecosystem diversity of riparian zones (Nilsson, Gardfjell & Grelsson, 1991), and may be the most important source of propagules to riparian zones and wetlands. In rivers propagules are dispersed by water in two ways:

1. Dispersal of non-buoyant propagules entrained with sediment movement.
2. Dispersal of buoyant propagules. These may be more likely to be deposited at the water/land boundary as flows recede.

Hydrochorous dispersal is supported by hydrological and sediment connectivity in rivers, and influenced by flow patterns and seasonality, landscape position and geomorphology. Dispersal associated with sediment movement may be influenced by the properties of the sediment (amount, organic content and particle size). In addition, propagules that are deposited with sediment may have better recruitment success.

Flow regulation affects both longitudinal connectivity of sediment/propagules (by physical barriers such as dams) and flow patterns which drive transport and deposition. The extent to which environmental flows can restore these processes depends on both appropriate flow regimes (both magnitude and seasonality) being delivered, and hydrochorous propagule sources being available. The links between flow magnitude (elevation of inundation), seasonality and sediment/propagule deposition on different types of in-channel features (bars, benches, and banks) needs to be better quantified. Additionally, the link between sediment properties and propagule assemblages needs to be determined.

An assessment of sediment and propagule deposition will enable 1) biodiversity objectives to be better understood, 2) links between flow magnitude and timing, properties of transported sediments and plant propagule abundance and assemblages to be elucidated, and 3) better informed management of the pattern and timing of delivery of environmental flows to achieve ecological objectives.

3.2 Evaluation questions

- What does Commonwealth environmental water delivered in winter contribute to sediment transport and deposition in the lower Goulburn River?
- What does Commonwealth environmental water delivered in winter contribute to plant propagule transport and subsequent deposition in the lower Goulburn River?

3.3 Complementary monitoring and data

Water level gauging at sites and hydraulic models developed for other monitoring programs are valuable tools to demonstrate levels of inundation arising from certain flows (e.g. models developed for the LTIM Physical Habitat Monitoring and Victorian Environmental Flow Monitoring and Assessment Program).

3.4 Monitoring locations and timing

3.4.1 Locations

Locations for turf mats should be based on: sites directly influenced by Commonwealth environmental water flow deliveries, existing transects so that cross sectional data can be incorporated into the assessment, sites where nearby gauging stations exist, sites with appropriate access, but limited public access

The following three sites have been selected for monitoring in winter-spring 2018:

- Darcys Track
- Loch Garry
- McCoys Bridge

3.4.2 Timing

Sediment and propagule deposition assessments should be undertaken at least four times over winter/spring targeting different flow types (winter fresh, spring fresh, variable baseflow, IVT). Deployment, removal and replacement of sampling equipment (turf mats) should be undertaken at low flows.

3.5 Monitoring protocol

3.5.1 Equipment

Equipment required includes:

- Field equipment:
 - Synthetic turf mats (360 mm x 240 mm, 19 mm pile)
 - Pegs (heavy-duty tent pegs or similar)
 - Survey equipment (total station) for survey of mat elevations
 - Large resealable bags
- Laboratory equipment (sediment):
 - Drying oven (55°C)
 - Foil trays
 - Balance
 - Furnace (550°C)
 - Crucibles
 - Sieves – including, at a minimum, 2 mm and 0.5 mm aperture
 - Laser diffraction particle sizer, or alternative equipment for sizing of fine (<0.5 mm) particles
 - Reagents
 - Sodium hexametaphosphate (Calgon)
- Nursery equipment:
 - Drying oven (40°C)
 - 125 µm sieve
 - Standard seedling trays (1 per sample)
 - Sterile seed raising media
 - Temperature controlled glasshouse (with TCs maintained between 15-25C) with automatic misting system.

3.5.2 Protocol

Based on the method of (Goodson *et al.*, 2003) the following will be undertaken:

Deployment:

- Paired turf mats (for sediment and propagule monitoring) are deployed in groups of six replicates per feature with four features per site
- Mats are secured at four corners with pegs
- Feature selection is based on site geomorphology but aims to capture the diversity of features, particularly depositional features, on site. Ideally, features should include a bar, bench and bank.
- One additional set of six mats (unpaired, for propagule monitoring only) is deployed above the expected top water level for monitoring of aerial seed dispersal
- Mats are deployed at levels to target different flow types
 - Bar mats are inundated in low flows
 - Bench and bank mats are inundated in freshes and possibly IVTs
- Mat locations and levels are surveyed by total station

Retrieval:

- Turf mats are retrieved at low flow
- The bottom surface of each turf mats is wiped clean of any adhering soil or organic material before being placed in large resealable bags

- Leaves and small twigs (but not logs) on the surface of the turf mats are collected for processing
- Large litter that extends outside the mats should be broken off and discarded
- Lost mats are recorded
- Mats are replaced in same location by new mats

Laboratory analysis of sediment:

- Mats are dried for 48 hours at 55°C
- Sediment is brushed/tapped off each mat into a tray and weighed
- Large leaves and twigs are removed and weighed
- A subsample of known dry mass (20-50 g) is taken and placed in furnace at 550°C for 3 hours, then reweighed to obtain loss-on-ignition estimate of organic content
- A further subsample of 300 g (or remainder of sample if less than 300 g) is taken for particle size analysis:
 - Sample is dispersed by addition of water and sodium hexametaphosphate (around 10 ml of 25% solution per 100 ml of water)
 - Sample is washed through a 0.5 mm sieve with a low-pressure water jet. The effluent is reserved for laser particle sizing. The captured material is dried, weighed, and then dry-sieved through a 2 mm sieve (and other sieves if further resolution of particle size distribution is required). The retained and passing fractions are weighed.
 - The material finer than 0.5 mm is analysed using a laser diffraction particle sizer using standard procedure.

Propagule germination:

Processing of turf mats

- Large leaves and twigs are removed and rinsed thoroughly over a 125 µm sieve to collect attached seeds before being discarded.
- Mats are agitated in buckets of tap water to dislodge propagules. The wash water is then filtered through 125 µm sieve to recover propagules. This mesh size ensures very small seeds (e.g. *Juncus* spp) are collected.
- The presence of the floating plants *Lemna* spp and *Azolla* spp should be recorded and removed. Counts of *Lemna* and *Azolla* are unreliable as they form colonies of vegetatively produced individuals which are prone to fragmenting during sampling and multiply rapidly.
- Viable vegetative fragments (e.g. green or turgid buds) are recorded and placed in separate trays if they cannot be identified immediately.
- All other material retained by the sieve is placed in paper bags and dried at 40°C for 1 week.
- The dried material is then spread evenly over soil trays. If there is a large amount of material it should be spread over several trays.

Preparation of soil trays and set up

- Germinations trays are filled to a depth of c. 5 cm with soil
- Seeds are germinated under moist soil conditions (Casanova & Brock, 2000; Williams *et al.*, 2008).
- Germination trays are kept moist using an automatic misting system operated 3-4 times per day
- Trays are placed in a glasshouse to provide a favourable temperature range for germination and to avoid contamination with wind dispersed propagules.
- For each sampling event ten “blank” soil trays should be distributed among the samples to detect contamination of trays.

Monitoring germination

- Species identification and counts of germinants are carried out weekly progressing to fortnightly as germination slows.
- Germinants are monitored for 4-6 months depending on the amount of continuing germination
- Seedlings are removed once identified and once they flower.

- If new germinants are observed at 4 months monitoring may need to continue for longer.

Plants that cannot be identified at the end of the germination period are transplanted and grown on.

4. Ecosystem metabolism

4.1 Ecological rationale

Winter is generally thought to be a time of low activity in stream metabolism, because both production and respiration rates are dependent upon temperature. However, counterexamples to this prevailing wisdom exist. A six-year continuous data set found flow but not temperature was found to have profound effects on the magnitude of metabolism (Marcarelli, Van Kirk & Baxter, 2010). Additionally, higher winter baseflows provide the opportunity for greater entrainment of terrestrial carbon into the river channel (through inundation of low-level bars). This would prevent organic matter accumulating (Acuna *et al.*, 2004), potentially reducing the frequency and severity of blackwater events following runoff in warmer months.

4.2 Evaluation questions

- What does Commonwealth environmental water delivered in winter contribute to ecosystem metabolism in the lower Goulburn River?
- Is ecosystem metabolism in winter in the lower Goulburn River a substantial component of the full-year ecosystem metabolism?

4.3 Monitoring protocol

Loggers are already in place with arrangements made for regular data downloading and maintenance at the existing 4 LTIM logger sites. No new sites will be required.

Data collection, site visitation and logger maintenance will all be performed exactly as per the established sampling protocol. Data downloads will be performed every 4-6 weeks depending on weather conditions and river height - safe access to the 4 sites and safe (and achievable) downloading.

5. Crustacean habitat use

5.1 Ecological rationale

Alternating periods of low and high flows are critically important in regulating life cycles and species interactions in the food web; the lengths of functionally important food chains (number of trophic levels) were found to increase with natural regimes of flow variation (Power, Dietrich & Finlay, 1996). Variable winter flows could then be expected to be of benefit for Goulburn River macroinvertebrates.

Previous results from the Goulburn LTIM macroinvertebrate monitoring program have shown that crustaceans seem to be particularly responsive to flows in the lower Goulburn River; one of the most significant responses observed was an increase in shrimp and prawn biomass in response to large, natural flood events in spring, 2016. Possible reasons for this increase could involve a change in food availability and stream productivity under different flow scenarios, as well as changing availability of particular habitats, such as snag availability, bank vegetation inundation and macrophyte bed maintenance, in response to these flows. These habitats can provide important refuges for crustaceans at different life stages, sources of food (including through deposition or biofilm growth), shelter from predators or increased flows, territories and breeding refuges. Variable flows in winter could be beneficial to crustaceans over the short term by changing the availability of habitats, and in the long term by sustaining important crustacean habitats such as bank vegetation and macrophyte beds. The purpose of this winter monitoring is to evaluate how variable flows affect crustacean habitat use, which will answer whether these variable winter flows are beneficial to crustaceans over the short term as well as provide information that will help interpret crustacean responses to other flow events, including spring freshes, that have been previously observed.

5.2 Evaluation questions

- Does Commonwealth environmental water contribute the exploitation of novel habitats by large-bodied crustaceans in the lower Goulburn River?

5.3 Monitoring protocol

5.3.1 Locations for monitoring

Monitoring will occur at a maximum of 12 sites in the Goulburn river catchment to characterise crustacean habitat use under variable flow conditions (Table 2). Site 1 is upstream of Zones 1 and 2 in Lake Nagambie to provide a site within the same river system that will not be experiencing variable winter flows. Sites 2 to 5 are along the Goulburn River in Zone 1, while Sites 6 to 10 are along the Goulburn River in Zone 2. Monitoring at these latter sites will provide multiple data points about crustacean responses to environmental flows. Sites 11 and 12 are on the Broken River, and will not be impacted by variable flows, again allowing another point of comparison.

Table 2. Monitoring sites for crustacean habitat assessment.

Site	LTIM Zone	Latitude	Longitude
Goulburn River at Kirwans Bridge Rd	Upstream	36.747037°S	145.140155°E
Goulburn River at Moss Road	1	36.707142°S	145.179229°E
Goulburn River at Salas Road, Murchison	1	36.624335°S	145.230148°E
Goulburn River at Bridge Road, Toolamba	1	36.497908°S	145.369435°E
Goulburn River at Watt Road, Shepparton	1	36.394851°S	145.363927°E
Goulburn River off Reedy Swamp Rd, Shepparton	2	36.325138°S	145.357739°E
Goulburn River at Loch Garry	2	36.241955°S	145.285934°E
Goulburn River at McCoys Bridge	2	36.176646°S	145.117990°E
Goulburn River at Yambuna Bridge	2	36.130839°S	145.003214°E
Goulburn River at Stewarts Bridge Road	2	36.103056°S	144.854380°E
Broken River at Central Avenue	Tributary	36.433041°S	145.461783°E
Broken River at Gowangardie Weir, Nalinga	Tributary	36.430595°S	145.697075°E

At each site, 6 bait traps will be deployed and divided among two main habitat types (depending on habitat availability): complex habitats (macrophytes, snags) and less complex habitats (bare). These bait traps will be deployed in the afternoon and retrieved the following morning, where all crustaceans within them will be removed and preserved in 100% ethanol (with the exception of yabbies, which will be weighed wet, have their carapaces measured and will then be returned alive to the river). The intention is to visit all 12 sites early in the week (Monday and Tuesday nights) and then a return visit will be made later in the week (Wednesday and Thursday nights) so that crustaceans will be assessed twice at each site under different flow conditions. However, the actual number of sites visited will be dependent upon the amount of time actually required to visit each site, as well as travel time between sites and early sunset times during winter. If fewer sites are visited, those that were visited earlier in the week will be prioritised for a second visit to ensure temporal replication at these sites.

5.3.2 Methods

If time permits, sweep samples will be taken at each site using a kick net, targeting different habitats for a set period of time (two minutes) to determine what crustaceans are present in these habitats. Sweep samples will be preserved in 100% ethanol and all crustaceans will be removed from the samples in the laboratory.

Samples will be processed in the laboratory by identifying crustaceans, counting their abundance, measuring their carapace lengths and drying these for dry weights. Dry weight measurements involve air drying animals for 24 hours, followed by oven drying at 60°C for a further 24 hours prior to weighing.

Data analyses will involve comparing changes in crustacean presence, abundance and dry weights among different habitat types over different flow periods.

5.4 Schedule of monitoring

Monitoring will take place over a period of five consecutive days in mid-August, 2018, when variable flows are being delivered in the lower Goulburn River and it is predicted that river heights will change over those days. Bait traps will be deployed overnight twice at each site during in the week, with the intention of monitoring crustaceans at each at two different river heights. An approximation of the monitoring schedule is given in Table 3.

Table 3. Approximate monitoring schedule.

	Day				
Activities	Monday	Tuesday	Wednesday	Thursday	Friday
Morning		Bait traps retrieved from sites 1-6	Bait traps retrieved from sites 7-12	Bait traps retrieved from sites 1-6	Bait traps retrieved from sites 7-12
Afternoon	Bait traps deployed at sites 1-6	Bait traps deployed sites 7-12	Bait traps deployed at sites 1-6	Bait traps deployed sites 7-12	

6. Algal biofilm production

Algae are dominant primary producers and an important foundation of many riverine food webs. They are one of the main sources of energy for higher trophic levels. As such, algal succession and total biomass may be a driving or explanatory variable in determining the structure of higher trophic levels, such as benthic macroinvertebrates. Disturbances, such as regulation of flows in riverine systems, are an important regulator of structure and biomass of algal assemblages. Increased flows or flooding in river systems can temporarily alter the composition of algal assemblages and result in both increases and decreases in algal biofilm abundance. In turn, alterations to algal biomass and assemblage structure have been correlated with changes in taxonomic richness, biomass and density of invertebrate communities. The current research being undertaken as part of the LTIM assesses the impacts of environmental water on macroinvertebrate communities and stream metabolism; however, there is a lack of information on how algal communities are impacted, which could be an important link between these other two trophic levels.

The resilience and susceptibility of algal assemblages to changes in flow are likely to be dependent on the timing of the flow releases, i.e.: whether they occur in summer/autumn or winter/spring.

The purpose of benthic algal monitoring is to evaluate how variable flows affect algal biofilm production over a seasonal scale. Such measurements will be important to better understand whether increased flows alter biomass and composition of algal biofilm communities and how this may impact on the macroinvertebrate and stream metabolism measures already being investigated. Monitoring sites and the rationale for inclusion are given in Table 1.

6.1 Evaluation questions

- What does Commonwealth environmental water contribute to algal biofilm production in the lower Goulburn River?
- Do rates of algal productivity in the lower Goulburn River differ between summer and winter?

6.2 Monitoring protocol

6.2.1 Locations for monitoring

Monitoring will occur at two sites in the Goulburn river catchment and one site on the Broken River (Table 4). Site 1 will be situated in the Goulburn River in Zone 1 at Darcy's Track, while Site 2 will be in the Goulburn River in Zone 2 at McCoys Bridge. If Site 1 is not suitable, Loch Garry (also Zone 2) will be considered instead. The Broken River site will be used as a reference site that will not be impacted by variable flows.

Table 4. Monitoring sites for crustacean habitat assessment.

Site	LTIM Zone	Latitude	Longitude
Goulburn River at Darcy's Track*	1	36.452839°S	145.359563°E
Goulburn River at Loch Garry*	2	36.241955°S	145.285934°E
Goulburn River at McCoys Bridge	2	36.176646°S	145.117990°E
Broken River at Central Avenue	Tributary	36.433041°S	145.461783°E

*If Darcy's Track is unsuitable, Loch Garry will be used instead.

6.2.2 Methods

The impacts of variable flows on benthic algal biofilms will be assessed through evaluating the structure and biomass of algal biofilms colonised on artificial substrates.

At each site, artificial substrate samplers will be deployed over two 4-week and a one 8-week period during each of winter and summer. Artificial substrate samplers will consist of plastic disks 11cm in diameter, held on a rope. Each rope will have two disks separated to allow one disk to be situated in the photic zone (where light is likely to be optimal for growth) and the second disk in the non-photoc zone (where light is likely to be limiting for growth) when deployed. The top of the rope will be attached to a float, allowing the artificial substrate sampler to maintain the same depth in the water column during the deployment period, as water levels change. The bottom end of the rope will be tethered to a weight, which will hold the sampler vertical in the water column. The float will be tethered to the bank, ensuring the sampler is not washed downstream (Figure 2). At each site, 6 artificial substrate samplers will be deployed on day 0. After 4 weeks, three will be removed and another 3 deployed, while 3 will remain. The newly deployed samplers and those remaining will be left for a further 4 weeks, after which they will be retrieved.

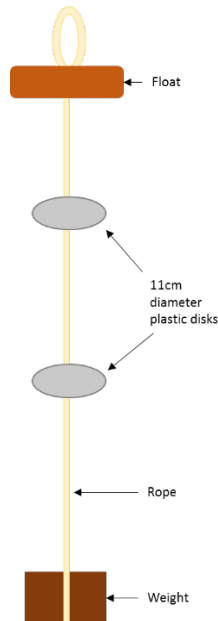


Figure 2. Diagram of the proposed artificial substrate sampler unit. This unit would be deployed by tethering the top rope loop to a line of rope tethered to the bank.

Upon retrieval, artificial substrate samples will be placed in bags on ice in the dark and immediately returned to the laboratory for processing. Disks will be processed for algae within 24hrs of collection.

The biofilm will be scrubbed from each disk into distilled water using a soft brush. The sample will be thoroughly homogenised and then divided evenly for analyses of community structure and biomass determination. Biomass will be determined through determination of chlorophyll a content and ash free dry mass (AFDM). For chlorophyll a, a subsample of scrubbed biofilm will be filtered through GF/C filters. Filters will then be extracted in acetone in the dark at 4°C overnight. The extract will be centrifuged and then chlorophyll a measured spectrophotometrically using standard methods. For AFDM, a subsample of scrubbed biofilm will be filtered through pre-ashed and pre-weighed GF/C filter papers. The filter paper will then be dried at 80°C for 24hrs, weighed, combusted for 4 hours at 500°C and re-weighed. Filters will be weighed to 4 decimal places and converted to dry weight and AFDW. The percentage of organic matter will be calculated as the proportion of AFDW to dry weight and converted to a percentage to standardise across sample dates and sites.

Community structure will be determined using multi-wavelength-excitation PAM fluorometry. This method assesses community structure based on spectral properties of different groups of algae. Specifically, three groups of algae can be differentiated – the diatoms, cyanobacteria and chlorophytes. A subsample of scrubbed biofilm will be dark-adapted for 20 mins. Afterwards, minimum chlorophyll fluorescence (f_0) will be measured using a four-wavelength-excitation PAM fluorometer. Low-intensity modulated measuring light will be provided by four different LEDs peaking at 470, 520, 645 and 665 nm and the minimum fluorescence measured and recorded at each wavelength.

Data analyses will involve comparing changes in algal biofilm biomass and structure among different sites and deployment periods.

6.2.3 Schedule of monitoring

Monitoring will take place over two seasons, Winter and Summer. Winter sampling will begin in mid-August 2018, when variable flows are being delivered in the lower Goulburn River. Artificial substrate samplers will be deployed at sites for 4 and 8 weeks. At 4 weeks, a set of samplers will be retrieved, while a set will remain and a new set will also be deployed. After 8 weeks all samplers will be retrieved. An approximation of the monitoring schedule is given in Table 5.

Table 5. Approximate monitoring schedule.

Activity	Season					
	Winter/Spring 2018			Summer/Autumn 2019		
	August	September	October	January	February	March
Algal biofilms	Deploy artificial substrate samplers	Retrieve a set of artificial substrate samplers and replace with a new set.	Retrieve all deployed artificial substrate samplers	Deploy artificial substrate samplers	Retrieve set of artificial substrate samplers and replace with a new set.	Retrieve all remaining artificial substrate samplers

7. Lamprey movement

Winter flows in the Goulburn River are at least partly designed to stimulate upstream migration of diadromous adult lampreys from the Lower Murray River to spawn upstream (Koehn & O'Connor, 1990; McDowall, 2000). Juveniles then gradually migrate downstream and return to sea the following winter and remain there for 3-4 years (McDowall, 2000). There are historical reports of lamprey being found in the Goulburn, but no recent records.

The aim of this component is to elucidate the timing and spatial extent of upstream migrations of lamprey, identify potential spawning regions/sites (e.g. the Goulburn River), and the importance of increased winter flows for their upstream migration. Ultimately this data would contribute to an understanding of how freshwater flow and connectivity influence lamprey life-history and population dynamics in the MDB, and in turn inform environmental water delivery for CEWO.

At this stage, potential benefits of Goulburn River winter flows to lamprey migration is conceptualised as contributing to basin-scale ecological outcomes, but there remains the possibility of re-establishment of the local spawning grounds for lamprey.

7.1 Evaluation questions

- What does Commonwealth environmental water delivered in the lower Goulburn River contribute to Lamprey movement in the Lower Murray River and connected southern basin?

7.2 Monitoring protocol

Procedures will follow those outlined in the current SOP for Fish Movement.

7.2.1 Schedule of monitoring.

Lamprey (up to 30 individuals) will be collected in the lower Murray River from fishways on the Murray Barrages by SARDI as part of fishway trapping scheduled for winter 2018. Lamprey collected will be tagged with VEMCO V7 acoustic transmitters and a passive integrated transponder tag.

A large-scale acoustic receiver array has already been established from the Murray barrages to Yarrawonga, including major tributaries of the River Murray such as the Goulburn, to determine movement patterns.

Data from the receiver array will be downloaded approximately in early-mid 2019. Results will be incorporated into the LTIM annual report for 2018/19.

8. Data analysis and reporting

As with the existing LTIM Project monitoring in the Lower Goulburn River, the main analysis of data collected for the winter monitoring will be undertaken at the University of Melbourne to ensure greater consistency in methods used for analysis and reporting of results. Processed monitoring data will be uploaded to the Monitoring Data Management System in accordance with data management protocols. All new results will be presented in the annual report as per other monitoring matters.

It is impossible to be completely prescriptive of the specific model structures that will be used to analyze the winter monitoring data at this stage, but the programs for sediment deposition, propagule transport, and algal biomass will use comparisons among different periods of the year (winter to warmer season), but over similar flow regimes (e.g. winter fresh compared to autumn fresh; winter variable flows compared to summer IVTs) to make inferences about the effect of winter flow regimes. Any Goulburn specific data that arises from the lamprey tagging at the Murray mouth will be included in the annual area-level evaluation report, potentially including an extension of the golden perch movement analysis. Metabolism analyses will continue to use existing methods, with the data set for the final year covering a full 12 month period, allowing us to infer the effects of winter flows (as well as warmer season flows) on metabolic parameters. Finally, the crustacean habitat use surveys are intended to provide an additional line of evidence to explain crustacean biomass responses to elevated flows after the 2016 floods. Analyses of these data will focus on identifying a change in habitat use with different flow conditions over the intensive survey period.

9. References

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