

# Benthic Macroinvertebrate Response Monitoring in the Coorong and Murray Mouth, 2012/13

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**Final Report  
for the Department of Environment and Natural Resources**

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## 1. Executive Summary

- This report presents findings from monitoring of macroinvertebrate response to continued freshwater inflows into the Murray Mouth and Coorong. The monitoring period from September 2012 to March 2013 covers the third year of water release following the drought breaking rainfalls in 2010, which allowed environmental flow into the lower reaches of the Murray-Darling Basin. The objectives for this monitoring centred on a series of key questions linked to the environmental management of the system (Table 1). By addressing these questions, data from this monitoring will support understanding of the resilience of macroinvertebrates, which are a key component to the functioning of estuarine and lagoon ecosystems, and can guide future management of water releases and restoration measures.
- Sampling was carried out at nine sites located between Monument Road near the Goolwa Barrage and Villa de Yumpa at the northern end of the South Lagoon. As in previous monitoring, samples were taken from exposed mudflats or close inshore (subject to water levels) and in deeper permanently submerged locations. In addition, mudflats on the Younghusband peninsula were sampled at two sites and occasions (Pelican Point in September 2012, February and March 2013; and Long Point in September 2012 and March 2013). Sampling took place in September and December 2012, and February and March 2013. Besides assessing the diversity and abundance of macroinvertebrates, size measurements of organisms were taken to obtain size frequency distributions for assessments of recruitment events. An approach to monitor settlement was trialled and while some data were obtained, the design required refinement. To link macroinvertebrate records with environmental conditions, sediment and water characteristics were obtained from each sampling site and time. Further observations and settlement plates targeted the occurrence and spread of tubeworms (*Ficopomatus enigmaticus*).
- The monitoring showed continued recovery of macroinvertebrate communities, with diversity and abundances increasing throughout the Murray Mouth and North Lagoon in comparison to the previous monitoring periods, and further expansion of distribution ranges towards the South Lagoon. The continued water release enables long-lasting improvements in environmental conditions, especially a reduction of area affected by hypersalinity, with larger regions of the Coorong thus becoming habitable again for macroinvertebrates. Several bivalve species rarely seen in recent years (for example *Arthritica helmsi*, *Spisula trigonella*) are becoming established and a sabellid polychaete, not found in our previous monitoring of the Coorong, was recorded. Yet, overall abundances remained dominated by amphipods and chironomid larvae.
- Based on size-frequency distributions, recruitment occurred for most of the polychaete species in the Murray Mouth and Coorong, as well as for several bivalve species. Most of the recruitment occurred around December and February. Capture of small-sized polychaetes and bivalves in sediment trays indicated their dispersal capability and potential to recolonise mudflats or sediments further south in the Coorong. In this monitoring period, abundances in mudflats were similar or higher than those in permanently submerged sediments, indicating the recovery after years of exposure during the drought.
- Macroinvertebrate communities were distinct from those occurring during the drought. Over various time frames, communities showed strong differences based on flow scenarios, but in the Coorong, recovery in the southern reaches of the North Lagoon and at Villa de Yumpa is taking longer. Salinity and, to a lesser extent, sediment characteristics were influencing the distinction of communities in the regions of the Murray Mouth and Coorong.

- The presence of living tubeworms (*Ficopomatus enigmaticus*) was recorded from most sites in the Murray Mouth and northern North Lagoon, with settlement on exposed frames occurring at Monument Road, Mundoo Channel, and to a lesser extent at Pelican Point and Long Point.
- As macroinvertebrates are a prime food supply for overwintering shorebirds, bird counts were carried out in association with the benthic studies. Eleven shorebird species were recorded and Red-necked Stints, Sharp-tailed sandpiper and Red-capped plover were numerically most abundant, especially at Ewe Island and Villa de Yumpa. Birds were observed to be foraging at all sites at each of the four surveys in this monitoring period. The abundance of shorebirds at particular sites did not match the highest abundances of macroinvertebrates, and attraction of birds to sites with high food supply *versus* depletion of macroinvertebrate prey from foraging birds could not be differentiated from the data. Experimental approaches are needed to fully understand predator-prey relationships. Some of the variation in the distribution and abundance of shorebirds was due to sediment characteristics.
- This monitoring has documented the further improvement in macroinvertebrate communities in the Murray Mouth and North Lagoon, and the persistence of the pattern of continued colonisation of the Coorong remains to be recorded in coming years. Following the changes after the water release commenced in late 2010 for several years has elucidated different stages of recovery and the longer time needed for a stressed system to restore key ecosystem components. These findings are valuable information for assessments of resilience in the Murray Mouth and Coorong that can guide future planning for water release scenarios.

**Table 1. Summary table of key questions and summary of findings for the 2012/2013 macroinvertebrate surveys, including page references for results**

#	Monitoring Objective	Key Questions	Summary of Findings	Ref.
1.	To assess the response of:	1. Are there indications of continued system recovery in 2012/2013 following the significant flows of 2010-11 and further flows in 2011-12, when a recovery was first documented?	<ul style="list-style-type: none"> <li>• Yes, this monitoring revealed further improvements in environmental conditions and macroinvertebrate communities and thus documented the beneficial effects of continued flows to enable a recovery of the system.</li> </ul>	
	Benthic macro-invertebrates to the continued water availability following the recent drought	a) Will environmental conditions within the Murray Mouth reflect true estuarine conditions and have they further improved in the North and South Lagoons since flows recommenced in 2010?	<ul style="list-style-type: none"> <li>• Salinity in the Murray Mouth region changed from brackish to marine over this survey period, following a reduction of river inflow in late spring/early summer. This seasonal change reflects estuarine conditions.</li> <li>• Salinity in the Coorong was marine to hypersaline and also showed a seasonal increase, yet remained consistently lower since flow commenced in late 2012 compared to previous drought periods.</li> <li>• The continued water release has long-lived benefits for the restoration of salinities in the Murray Mouth and Coorong.</li> </ul>	pg. 15
		b) Has recruitment continued for macroinvertebrate species which underwent recruitment in 2011-12 and is recruitment occurring for other species in 2012-13?	<ul style="list-style-type: none"> <li>• Continued recruitment was observed of <i>Simplisetia aequisetis</i>, <i>Capitella sp.</i>, <i>Nephtys australiensis</i> and <i>Australonereis ehlersi</i> in 2012/2013. Yet, apart from <i>S. aequisetis</i>, this recruitment was often detected at single sites only.</li> <li>• Juveniles were also found for the polychaetes <i>Boccardiella limnicola</i>, <i>Ficopomatus enigmaticus</i>, <i>Euchone cf. variabilis</i>, and the bivalves <i>Spisula trigonella</i>, <i>Soletellina alba</i> and <i>Arthritica helmsi</i>, yet mostly at particular sites only for each species.</li> </ul>	pg.18
		c) Can the recruitment of species in 2012/2013 be linked with the timing of differing flow scenarios to identify drivers of macroinvertebrate recruitment?	<ul style="list-style-type: none"> <li>• Recruitment of <i>Simplisetia aequisetis</i> may be related to salinity changes generated by freshwater inflows over the barrages into the Coorong.</li> <li>• Recruitment of most macroinvertebrates occurred around December and February, and may be induced by flow-related salinity changes or rising temperature over summer.</li> </ul>	pg.23

#	Monitoring Objective	Key Questions	Summary of Findings	Ref.
		d) Will there be any further recolonisation of mudflats by macroinvertebrates absent from areas in previous monitoring 2010-12?	<ul style="list-style-type: none"> <li>• At mudflats from all sites further arrivals of species, not previously encountered at the site, occurred during the 2012/2013 surveys.</li> <li>• A polychaete species (<i>Euchone</i> cf. <i>variabilis</i>, Sabellidae) not previously recorded in this water release study in the Coorong, was common in a mudflat from the peninsula site. This may be the same as species as recorded by Geddes from the North Lagoon in the 1980s.</li> <li>• Macroinvertebrate abundances in mudflats were similar or higher than in subtidal sediments, driven by the highly abundant and widespread amphipods and chironomid larvae.</li> <li>• Recolonisation of mudflats was particularly pronounced at Ewe Island and Pelican Point over summer 2012/2013.</li> <li>• Yet, bivalves not or rarely found in previous monitoring were more common in subtidal sediments.</li> </ul>	pg.24
		e) Will recolonisation of macroinvertebrate species occur in the South Lagoon if salinities remain lowered in this region?	<ul style="list-style-type: none"> <li>• Salinities in the South Lagoon have remained lowered during 2012/2013 since flow commenced. Species recorded here last year were still present, and recolonisation by the polychaetes <i>Simplisetia aequisetis</i> and <i>Capitella</i> sp. was evident from occasional collections of a few individuals at Villa de Yumpa. These species were not previously collected from this site.</li> </ul>	pg.28
		f) Have species maintained or further increased their distribution range in comparison to 2011-12?	<ul style="list-style-type: none"> <li>• During the 2012/2013 monitoring, an expansion of macroinvertebrate distributions into the Coorong was recorded, evidenced by higher species numbers and abundances compared to the previous year.</li> <li>• <i>Simplisetia aequisetis</i> expanded its distribution range to all sites except Parnka Point. <i>Arthritica helmsi</i>, <i>Soletellina alba</i> and hydrobiid snails were collected from all sites in the Murray Mouth and also appeared in the northern end of the North Lagoon.</li> <li>• While <i>Capitella</i> sp. still dominated in sediments at Long Point and Noonameena, it was spreading into adjacent sites, including the northern end of the South Lagoon.</li> <li>• Some species (<i>Australonereis ehlersi</i> and <i>Nephtys australiensis</i>)</li> </ul>	pg.30



#	Monitoring Objective	Key Questions	Summary of Findings	Ref.
			remained confined in their distributions to a few sites.	
1.	To assess the response of:  Benthic macro-invertebrates to the continued water availability following the recent drought	g) Has the macroinvertebrate community been restored to pre-drought conditions following the 2011-12 barrage flows into the Coorong; and are they comparable to values observed prior to drought conditions?	<ul style="list-style-type: none"> <li>• Macroinvertebrate communities in each region are becoming increasingly distinct from those prevailing during the drought and first flood response, indicating recovery.</li> <li>• Distributions of macroinvertebrates in relation to salinity ranges are becoming more similar to records by Geddes (1987) following barrage flows between 1983-1985.</li> </ul>	pg.32
		2. Which environmental conditions influence the distribution, abundance and community structure of macroinvertebrates?	<ul style="list-style-type: none"> <li>• Salinity continued to be the main environmental factor that influenced the distribution, abundance and community structure of macroinvertebrates throughout the very distinct regions of the Murray Mouth and Coorong.</li> <li>• Sediment characteristics (% organic matter, % silt and clay and % coarse sand) were secondary in influencing community structure.</li> </ul>	pg.35
		3. Are there similarities or differences in the community structure of macroinvertebrates across differing flow scenarios (drought/flood)?	<ul style="list-style-type: none"> <li>• Communities in the Murray Mouth and North Lagoon region showed strong differences based on flow scenarios, but differences were not apparent at the South Lagoon site (Villa de Yumpa), where recovery is taking longer.</li> <li>• Different species characterise the communities at most sites during the drought and different stages of response to water release.</li> <li>• Variability in the community compositions decreased, possibly indicating a reduced stress.</li> </ul>	pg.40
		4. Do <i>Ficopomatus enigmaticus</i> reefs in the Murray Mouth and North Lagoon region exhibit new growth?	<ul style="list-style-type: none"> <li>• New growth and living tubeworms (<i>F. enigmaticus</i>) were observed at Monument Road.</li> <li>• Living tubeworms were occurred also at Hunters Creek, Pelican Point, Mark Point, Long Point and reef structures were observed at Noonameena and Villa de Yumpa.</li> <li>• Settlement plates and frames showed colonisation by tubeworm larvae at Monument Road and Mundoo Channel in February and March 2013, and to a lesser extend at Pelican Point and Long Point in February 2013.</li> </ul>	pg.43

#	Monitoring Objective	Key Questions	Summary of Findings	Ref.
		<p>5. Is there a relationship between diversity, abundance and habitat use of shorebirds and the availability of their macroinvertebrate food source at study sites?</p> <ul style="list-style-type: none"> <li>- What is the prey availability (based on the depth distribution and size) for shorebirds at study sites?</li> <li>- Does the prey available in mudflats at study sites determine the bird's spatial distribution across sites?</li> <li>- Which environmental factors and microhabitats are important for birds at study sites?</li> </ul>	<ul style="list-style-type: none"> <li>• Eleven species of shorebirds were recorded foraging on intertidal mudflats at the study sites during the 2012/13 survey.</li> <li>• Red-necked Stint were by far the most abundant, with Sharp-tailed Sandpiper and Red-capped Plover also being common.</li> <li>• Prey availability at study sites in the Coorong was determined by macroinvertebrate species distribution, with prey for short-billed species, like Red-capped Plover, being fairly widely distributed, while larger polychaetes favoured by longer-billed species, occurred at fewer sites.</li> <li>• There were only weak relationships between shorebird distribution and prey availability or environmental conditions, possibly because of the snap-shot nature of sampling highly mobile foraging birds.</li> </ul>	pg.44

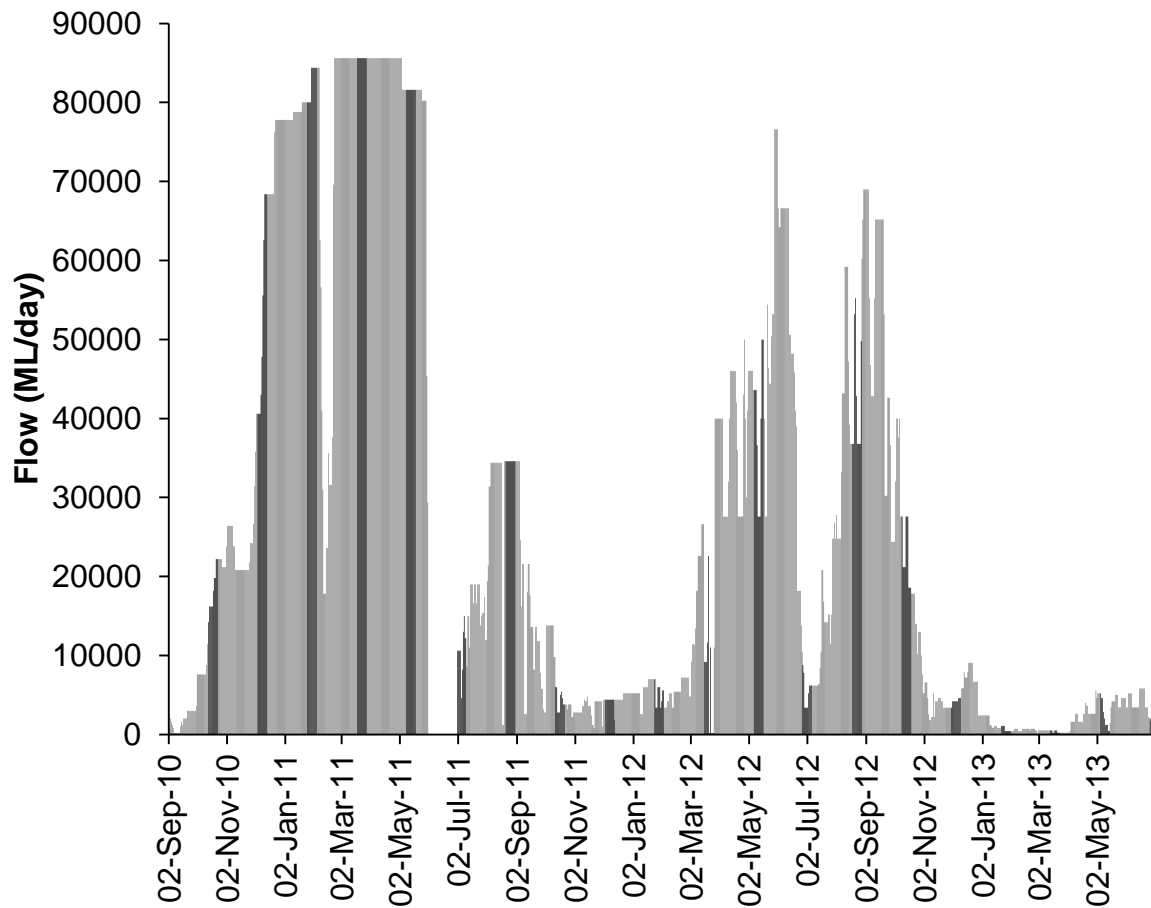
## 2. Introduction

Connectivity between the major regions of the Coorong, Lower Lakes and Murray Mouth has been restored since spring 2010, when substantial water flow from the Murray-Darling Basin reached the estuary and was released over the barrages. Prior to this flow restoration, the Murray Mouth and Coorong were affected by drought induced environmental degradation (Lester & Fairweather 2009), which had led to a decline in macroinvertebrate communities (Dittmann *et al.* 2010a). Freshwater flows are important drivers for estuarine habitat characteristics and if flows are inhibited, environmental degradation has to be managed (Elliott *et al.* 2007; Elliott & Whitfield 2011). Monitoring informs management on the effectiveness of restoration and mitigation measures, as recovery pathways can be unpredictable (Gaertner-Mazouni & de Wit 2012; Verissimo *et al.* 2012; Marques *et al.* 2013). This report presents findings from continued monitoring of the macroinvertebrate response to continued freshwater inflows (Figure 1) following the Millennium Drought (Leblanc *et al.* 2012).

Macroinvertebrate monitoring of the water release that commenced in spring 2010 (Figure 1) has so far revealed a series of responses, which included changes in benthic abundances in sub- and intertidal sediments, extension of distribution patterns and re-occurrence of species not recorded during the drought (Keuning *et al.* 2012). While some of the observed responses resembled changes recorded after water release in the 1980's (Geddes 1987), the benthic communities still deviated from community characteristics found at the start of the Millennium Drought (Dittmann *et al.* 2012). Further monitoring of macroinvertebrate responses will gain insight on the resilience of benthic organisms under changing environmental conditions and the flow-on effects for higher trophic levels. Environmental changes expected with continued flow over the barrages were reduced salinity and improved water quality in the Coorong, continued high water level, localised restoration of true estuarine character, and intensified connectivity between adjacent ecosystems.

Ecological response monitoring of macroinvertebrates to continued water release was thus undertaken to test a series of key questions linked to environmental management of the system (Table 1). These questions required assessments of diversity and abundance of macroinvertebrates in the Murray Mouth and Coorong and the extent of their distribution ranges in relation to water and sediment quality characteristics, analyses of recruitment processes and recolonisation of key invertebrate species, as well as links with higher trophic levels through shorebird surveys. The monitoring was conducted at nine sites across the Murray Mouth and Coorong, including locations with different water depths and some on the peninsula side of the Coorong. Sampling was carried out at four occasions over spring and summer 2012/2013. In addition, targeted monitoring was conducted to investigate the recruitment of larvae from the water column.

This report is structured around the key questions outlined in Table 1, and additional results are presented in an appendix. Answering the key questions is based on data collected in the 2012/2013 survey period as well as previously collected data from the same sampling sites to investigate any changes across time. These investigations will evaluate the benefit of the continued flow for macroinvertebrates, and ultimately the food webs in the Coorong and Murray Mouth.



**Figure 1: Daily barrage flow from the Lower Lakes into the Murray Mouth and Coorong from September 2010 up until May 2013 during the third consecutive year of recommenced flow. Based on data from the River Murray Operations Group.**

## 3. Materials and Methods

### 3.1 Sampling sites and dates

Benthic macroinvertebrates were sampled in September and December 2012, and February and March 2013 at nine sites between the Murray Mouth region and northern end of the South Lagoon (Figure 2, Table 2). Most of these sites were surveyed in previous studies (Dittmann *et al.* 2011a, Dittmann *et al.* 2011b, Keuning *et al.* 2012). Two sites (Sugars Beach and Tauwitcherie) previously included in macroinvertebrate response monitoring (see Keuning *et al.* 2012) were omitted for the 2012/2013 monitoring to allow more regular sampling of sites further south (i.e. Parnka Point and Villa de Yumpa) and addition of sampling locations on the Youngusband Peninsula. The December 2012 survey overlapped with the annual "The Living Murray" (TLM) monitoring, and only supplementing sites (Mark Point, Long Point), subtidal and Youngusband Peninsula locations which are not part of the TLM, were sampled for this project.

To obtain a transect across the Coorong, additional samples were collected from the Young Husband Peninsula side at Long Point and Pelican Point in September 2012, February (Pelican Point only) and March 2013. Sampling of both peninsula locations was intended in February, but as the Long Point peninsula site could not be accessed in February 2013, both peninsula sites were sampled in March 2013 to obtain a complete data set. At Parnka Point, both the northern and southern sides were sampled during February and March 2013 to compare conditions across the narrow channel (Hells Gate) between the North and South lagoons.

Samples were taken at two water depths, from the near-shore intertidal<sup>1</sup> (0-0.5 m water depth) and subtidal. Subtidal locations were up to 150 m towards the deeper channel from the intertidal location. Due to the changing water levels over time, the water depths at subtidal locations varied between 0.5 m to over 1.5 m. Water levels throughout the Coorong and Murray Mouth region were high during September 2012 sampling, reaching the shoreline or exposing only a narrow strip (< 1 m) of sediment. By December 2012, water levels had dropped, exposing mudflats up to 200 m wide (Ewe Island) in the Murray Mouth, and 300 m at Villa de Yumpa in the South Lagoon. At the North Lagoon sites, mudflats were between 1 m (Noonameena) and up to 10 m wide (Parnka Point). Water levels dropped further over summer and in February and especially March 2013, wider areas of mudflats were exposed at most sites. Samples were taken from unvegetated sediments, but the presence of *Ruppia* sp. was noted at several sites in the Coorong (Mark Point, Parnka Point and Villa de Yumpa).

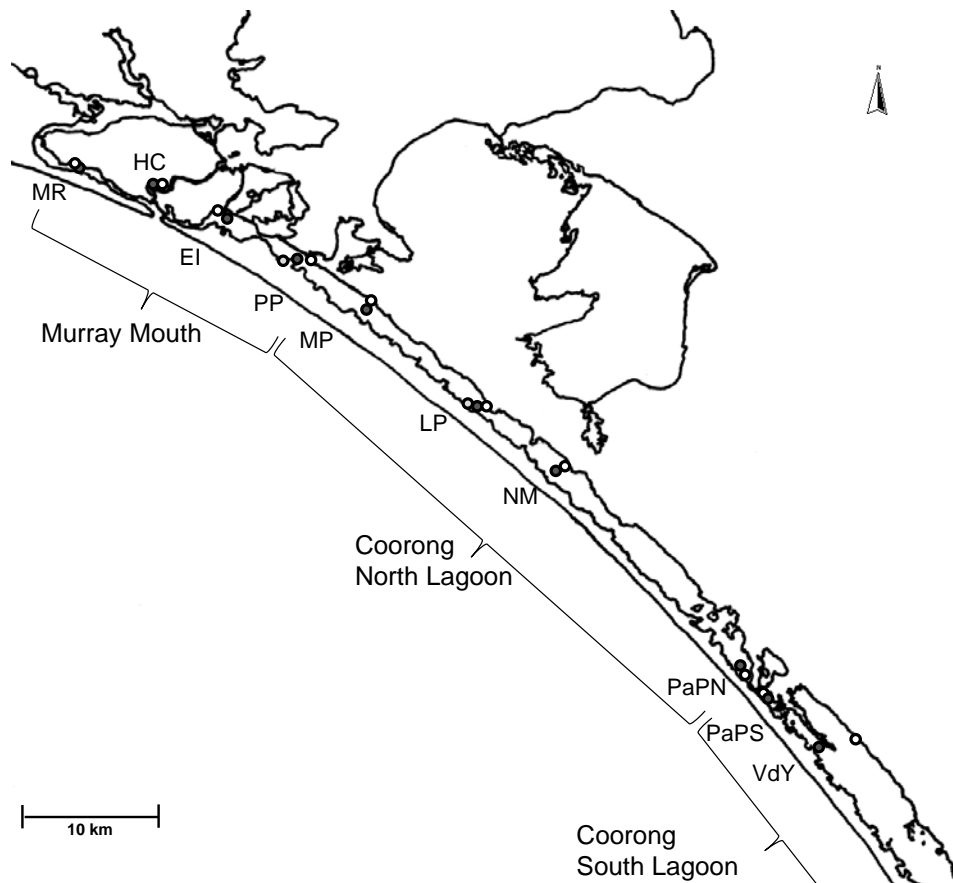
### 3.2 Environmental parameters

At each site, water quality parameters including temperature (° C), salinity (ppt), oxygen concentration (O<sub>2</sub> mg/l) and saturation (O<sub>2</sub> %) and pH of the water overlying the mudflats (intertidal) or subtidal sediments were measured using either a TPS WP-81 (for pH, temperature and salinity) and WP-82Y (for dissolved oxygen) electrode, and/or a YSI 85 multi-parameter electrode. If possible, measurements were taken with two electrodes as malfunctions can occur, which happened during March 2013, when no measurements could be taken for water quality at sites south of Pelican Point. Missing data was supplemented for analysis where possible with data collected by the EPA for Pelican Point, Mark Point, Long Point and Villa de Yumpa.

To characterise the sedimentary environment for benthic organisms, three replicate sediment samples were taken each for grain size, organic matter and chlorophyll-a. Sediment samples for grain size and organic matter were taken with a 60 ml cut-off syringe (surface area 6.6 cm<sup>2</sup> to a depth of

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<sup>1</sup>Note the terminology is used although tides are only affecting the Murray Mouth region.



**Figure 2. Study sites in the Murray Mouth and Coorong during the September 2012 – March 2013 monitoring period, see Table 2 for site acronyms. Sampling locations per site are indicated as open circles for ‘intertidal’, closed circles for submerged locations, and grey circles for peninsula locations (at Pelican Point and Long Point only). Parnka Point was sampled on the northern and southern side of Hells Gate on some occasions. Hunters Creek is located on Mundoo Channel, where additional settlement plates for tubeworms were deployed.**

approximately 10 cm). Samples of the sediment parameters were stored on ice in the field and frozen on return to the lab until further analysis. Grain size was determined by laser diffraction using a particle size analyser (Malvern Mastersizer 2000). Sediment grain size samples were thawed and the fraction >1 mm sieved off manually to avoid blockage in the machine. The weights of this fraction and of the remaining sediment were determined for later normalisation of the data to correct for this procedure. Median and quartiles, as well as percentage of various particle sizes were obtained from the Mastersizer output. Sediment sorting ( $S_o$ ) was calculated from the ratio of the quartiles ( $P_{25}$  and  $P_{75}$ )  $S_o = (P_{25}/P_{75})^{1/2}$ , based on the metric scale. The three sediment samples taken for sediment organic matter were analysed separately to account for spatial variation and to calculate means and standard error. To obtain a bulk parameter of organic matter as % dry weight (d.w.), sediment samples were dried to constant weight (for 24 – 36 hours) at 80 °C and then burnt in a muffle furnace at 450 °C for 5 hours.

For sediment chlorophyll-a, three replicate samples were taken per site by inserting a 5 ml vial about 1 cm into the sediment, 5 ml of methanol was added to extract the chlorophyll, and the vial was heavily shaken before being wrapped in aluminium foil (Seuront and Leterme 2006) and frozen for later analysis with a fluorometer (Turner 450). After the initial reading for total chlorophyll, drops of 0.1 M hydrochloric acid (HCl) were added to the samples to correct for phaeophorbides.

**Table 2. Overview of the macroinvertebrate sampling design in the Murray Mouth and Coorong, showing the dates and months of various sampling activities. Acronyms for months are S (September 2012), D (December 2012), F (February 2012) and M (March 2013). See Figure 2 for site locations. n/s = not sampled.**

Site	Sampling Date				Water Quality, Sediment Characteristics, Macrobenthic Community and Size Frequency Distribution	Recruitment trays deployed	Ficopomatus Frames Collected	Bird Surveys
	September '12	December '12	February '13	March '13				
Monument Road (MR) - Intertidal	19/09/2012	11/12/2012	5/02/2013	13/03/2013	S D F M	S F	F M	S D F M
Subtidal	19/09/2012	11/12/2012	5/02/2013	13/03/2013	S D F M			
Mundoo Channel	(Ficopomatus settlement frames only)						F M	
Hunters Creek (HC) - Intertidal	19/09/2012	11/12/2012	5/02/2013	12/03/2013	S D F M			S D F M
Subtidal	19/09/2012	11/12/2012	5/02/2013	12/03/2013	S D F M			
Ewe Island (EI) - Intertidal	18/09/2012	11/12/2012	5/02/2013	12/03/2013	S D F M	S F		S D F M
Subtidal	18/09/2012	11/12/2012	5/02/2013	12/03/2013	S D F M			
Pelican Point (PP) - Intertidal	18/09/2012	3/12/2012	12/02/2013	19/03/2013	S D F M		D F M	S D F M
Subtidal	18/09/2012	18/12/2012	12/02/2013	19/03/2013	S D F M			
Peninsula	18/09/2012	n/s	12/02/2013	19/03/2013	S F M			
Mark Point (MP) - Intertidal	18/09/2012	4/12/2012	12/02/2013	19/03/2013	S D F M	S F		S D F M
Subtidal	18/09/2012	4/12/2012	12/02/2013	19/03/2013	S D F M			
Long Point (LP) - Intertidal	17/09/2012	3/12/2012	11/02/2013	18/03/2013	S D F M		D F M	S D F M
Subtidal	17/09/2012	4/12/2012	12/02/2013	18/03/2013	S D F M			
Peninsula	17/09/2012	n/s	n/s	19/03/2013	S M			
Noonameena (NM) - Intertidal	17/09/2012	4/12/2012	11/02/2013	18/03/2013	S D F M	S F		S D F M
Subtidal	17/09/2012	4/12/2012	11/02/2013	18/03/2013	S D F M			
Parnka Point (PaPN) - Intertidal	17/09/2012	4/12/2012	11/02/2013	18/03/2013	S D F M			S D F M
Subtidal	17/09/2012	3/12/2012	11/02/2013	18/03/2013	S D F M			
Parnka Point (PaPS) - Intertidal	n/s	4/12/2012	11/02/2013	18/03/2013	D* F M			F M
Subtidal	n/s	n/s	11/02/2013	18/03/2013	F M			
Villa de Yumpa (VdY) - Intertidal	17/09/2012	3/12/2012	11/02/2013	18/03/2013	S D F M			S D F M
Subtidal	17/09/2012	3/12/2012	11/02/2013	18/03/2013	S D F M			

\* Macrobenthic community sampled only

### 3.3 Macrofauna

To investigate macroinvertebrate species composition and abundance within sediments, handheld PVC corers (83.32 cm<sup>2</sup> surface area) were used for intertidal sediments at each site. For each of the land-based sites that were intertidal or under shallow water, ten haphazardly placed replicate samples were taken. Each replicate core was inserted into the sediment to approximately 15 cm depth, sealed with a stopper to avoid disturbance of the sample and dug out with a shovel or by hand. Submerged sites were sampled with a benthic Ekman grab (225 cm<sup>2</sup> surface area), deployed from a small boat, when the overlying water was deeper than 1 m. The grab penetrated approximately 10 cm into the sediment. The grab was only used at some subtidal sites in September and December 2012, and in March 2013, as water levels receded over summer. When water levels were <1 m, subtidal locations were sampled using the PVC corers (83.32 cm<sup>2</sup> surface area) in the vicinity of the boat.

All samples were sieved through a 500 µm mesh size *in situ*. Sorting of live samples was carried out within several days and organisms were identified to the lowest possible taxonomic level and individual numbers for each species counted. Amphipods were not differentiated into family or species. For insects, larval and pupae life stages were distinguished during sorting. When samples could not be processed live within a few days, due to the unexpected high number of macroinvertebrates encountered in the samples, they were preserved in 70 % ethanol (February 2013).

To investigate whether recruitment of the four most abundant polychaete species (*Capitella* sp., *Australonereis ehlersi*, *Simplisetia aequisetis* and *Nephtys australiensis*<sup>2</sup>) had occurred, the length of complete specimens of these four polychaete species were measured. Specimens were laid out on graph paper to determine their length. Thirty individuals per species per sample were measured for size-frequency distribution. When there were fewer than 30 individuals of a species, all individuals of that species were measured. Measurements were also taken of additional macroinvertebrates (e.g. bivalves) to obtain a range of their sizes.

### 3.4 Larval recruitment

To investigate recolonisation potential of benthic macroinvertebrates, a targeted monitoring approach was included in the 2012/13 survey period. Settlement of pelagic larvae from the water column into the sediment was to be assessed with larval recruitment trays at four sites; Monument Road and Ewe Island in the Murray Mouth region, and at Mark Point and Noonameena in the North Lagoon. At each site, three types of trays were used. These consisted of (i) defaunated sediments, (ii) ambient sediment transferred into trays, and (iii) undisturbed ambient sediment. Defaunated sediment was obtained by using sediment from each respective site which was frozen at -20°C for 24 hours and checked to ensure it contained no living fauna before being deployed in trays in the field.

Trays for holding sediments were constructed from PVC pipe (110 mm diameter) cut into 20 cm lengths. For easier insertion of these pipes into the sediment, two holes (approximately 1 cm diameter) were drilled into the lower sides, so they remained below the surface of the sediment. A level plastic barrier was fixed inside the PVC pipe about 3 cm beneath the top opening to create a tray that contained the sediments. When deployed in the field, the plastic barrier sat on the sediment surface, and the tray section of the PVC pipes was thus elevated 3 cm above the ambient sediment. This design was chosen to inhibit intrusion of benthic fauna from sediment below the trays, and reduce colonisation by bed load transport.

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<sup>2</sup>Note that *Nephtys australiensis* is now recognised as *Aglaophamus australiensis* but *N. australiensis* is retained throughout this report for ease of comparison to earlier reports and data sets.



For each of the treatments, five replicate trays were deployed in a fully randomised grid at each of the four sites, in shallow water depths. The trays were inserted with about 30 cm interspace. Samples of all three treatments were taken at the time of deployment, using a cut-of syringe (1.5 cm<sup>2</sup> surface area) inserted to the depth of the tray, or 3 cm sediment for the ambient controls. Three replicate samples were taken per unit and pooled. This sacrificial pseudoreplication was done to capture any small-scale patchiness of larval spatfall, and to reduce the amount of samples to be processed. Trays were re-sampled a fortnight later, but changes in water level or windy condition caused loss or inaccessibility for sampling at several sites and sampling occasions. The number of true replicate samples for treatments (i) and (ii) was therefore low and only used for observational records together with data from the undisturbed control treatment (iii).

As the depth of samples taken varied between trays subject to erosion, data were calculated to the volume of sediment per sample instead of the surface area. In the lab, samples were processed by repeated decantation with seawater through a series of sieves with 500 µm (to separate macro-invertebrates) and 52 µm mesh size (to concentrate larvae and meiofauna).

### **3.5 *Ficopomatus enigmaticus* settlement frames**

Four *Ficopomatus enigmaticus* settlement frames were deployed to test for the presence and distribution range of tubeworm larvae, one each at Monument Road, Mundoo Channel, Pelican Point and Long Point. Mundoo Channel was added as a site as tubeworm reefs were observed here in the past. The top of each frame was positioned approximately 30 cm under the surface of the water. The frame design followed the experiments by Rolston (see Dittmann *et al.* 2009) which consisted of a 81 cm x 58 cm PVC frame with eight ceramic tiles attached (see Dittmann *et al.* 2011 for diagram of frame). Tiles (rough side facing out) were deployed during September and December 2012 and in February 2013 and collected on the next sampling occasion (i.e. December 2012, February and March 2013, respectively). Deployment times thus varied from one to three months. The individual tiles were inspected for settlement of *F. enigmaticus*.

### **3.6 Shorebird surveys**

Shorebirds were surveyed four times at each of the nine intertidal sites between September 2012 and March 2013, during the non-breeding overwintering season of migratory shorebirds. Identification and counts of shorebirds occurred on the same day as macroinvertebrate sampling in September and December 2012, but during February and March 2013 shorebird observations were made two days after the macroinvertebrate sampling due to logistical reasons. Shorebirds were identified and counted using either a Leica Televid telescope with a 20-60x eyepiece or Bauch and Lomb Elite 10x42 binoculars. Observations of behaviour and substrate use were also taken. Behaviour was either recorded as flying, roosting or foraging, while substrate was classified as dry mud, wet mud, water's edge and shallow water.

### **3.7 Data analysis**

To determine if there have been any improvements in environmental conditions within the Murray Mouth and North and South Lagoons in 2012/2013 (key question 1a) the current conditions measured in 2012/2013 (this report) were graphically compared to historical data (dating back to December 2004 where possible) (Dittmann *et al.* 2012).

To investigate recruitment of macroinvertebrate species and recolonisation of mudflats (key questions 1b-g), current (2012/2013) distribution ranges, abundances of key species (e.g. polychaetes) and taxonomic groups (e.g. amphipods) as well as size frequency distributions were determined and compared to previous known distributions (including those measured prior to the Millennium Drought).

Recolonisation of mudflats by species from subtidal populations was investigated by comparing subtidal and intertidal species distributions over the survey period. Size-frequency distributions for key species were investigated to determine recruitment of juveniles into populations in 2012/2013. When recruitment was observed in 2012/2013, occurrence of juveniles was related to environmental flows and salinities. SIMPER was used to compile lists of characteristic species for each site that were compared to previous surveys (Dittmann *et al.* 2010, Dittmann *et al.* 2011, Keuning *et al.* 2012) and historical information (Geddes & Butler 1984; Geddes 1987). Principle Coordinate Plots (PCO) were created for comparisons over longer term using TLM monitoring data (Dittmann *et al.* 2012), based on fourth root transformed data and Bray-Curtis similarity with a dummy value due to many zero values in the data.

Distance-based linear models (DistLM) and distance-based redundancy analysis (dbRDA) plots were used to determine if environmental factors measured at the time of sampling, specifically sediment characteristics and water quality, were important for determining the distribution, abundance and community structure of macroinvertebrates in the Coorong (key question 2).

Macroinvertebrate communities within the Coorong and Murray Mouth region have been sampled by Dittmann and others since June 2004 using the same methods employed in the current survey 2012/2013 (Dittmann *et al.* 2006a, Dittmann *et al.* 2006b, Dittmann & Nelson 2007, Dittmann *et al.* 2008, Barring *et al.* 2009, Dittmann *et al.* 2010, Dittmann *et al.* 2011, Keuning *et al.* 2012). This period has been divided into two flow scenarios based on drought and flood conditions, with little, infrequent or no flows over the barrages during drought years (2004 – 2009) and continued flows over the barrages during flood years (2010 – current). To determine if macroinvertebrate communities were similar or different across drought and flood conditions (key question 3), SIMPER was used to calculate average similarities among samples and compile lists of characteristic species for samples collected during drought and flood years, separately for each sampling location. Mark Point was omitted from analysis as there were too few sampling events ( $n = 5$ ) at this site between June 2004 and May 2012. Canonical analysis of principle coordinates (CAP) discriminate analysis based on drought (Jun 2004 to December 2009) and flood periods up to the current survey (December 2010 to May 2012) was then used to determine if distinct communities existed during these two periods ('drought' versus 'flow') at each sampling location. Samples collected during the 2012/2013 survey were then fitted to the CAP model generated for each location and assigned to a community type based on the two flow regimes to investigate the current state of macroinvertebrate communities within the Coorong.

Observations of *Ficopomatus enigmaticus* reefs, settlement frames and living specimens collected in macrobenthic samples were combined to assess the distribution of this species and the status of populations at each sample site (key question 4).

To determine the prey availability for shorebirds and investigate whether there were any relationships between shorebird distribution and prey availability (key question 5) a number of taxa were identified from previous reports (Dittmann *et al.* 2006) as potential prey items and their spatial and temporal distribution was compared to that of shorebirds. Taxa identified as potential prey items were polychaetes, especially *Simplisetia aequisetis* (known to be preferred prey for waders, especially Curlew Sandpiper) and *Capitella sp.* (a potential prey item for short-billed waders), the mollusc *Arthritica helmsi* (a preferred prey item of sandpipers), Amphipoda and insect larvae of Chironomidae (both being potential prey items for short-billed waders). The distribution of shorebirds was also related to environmental factors using DistLM. All analyses were carried out using the software PRIMER v6 with PERMANOVA add-on (multivariate techniques).

## 4. Results and Discussion

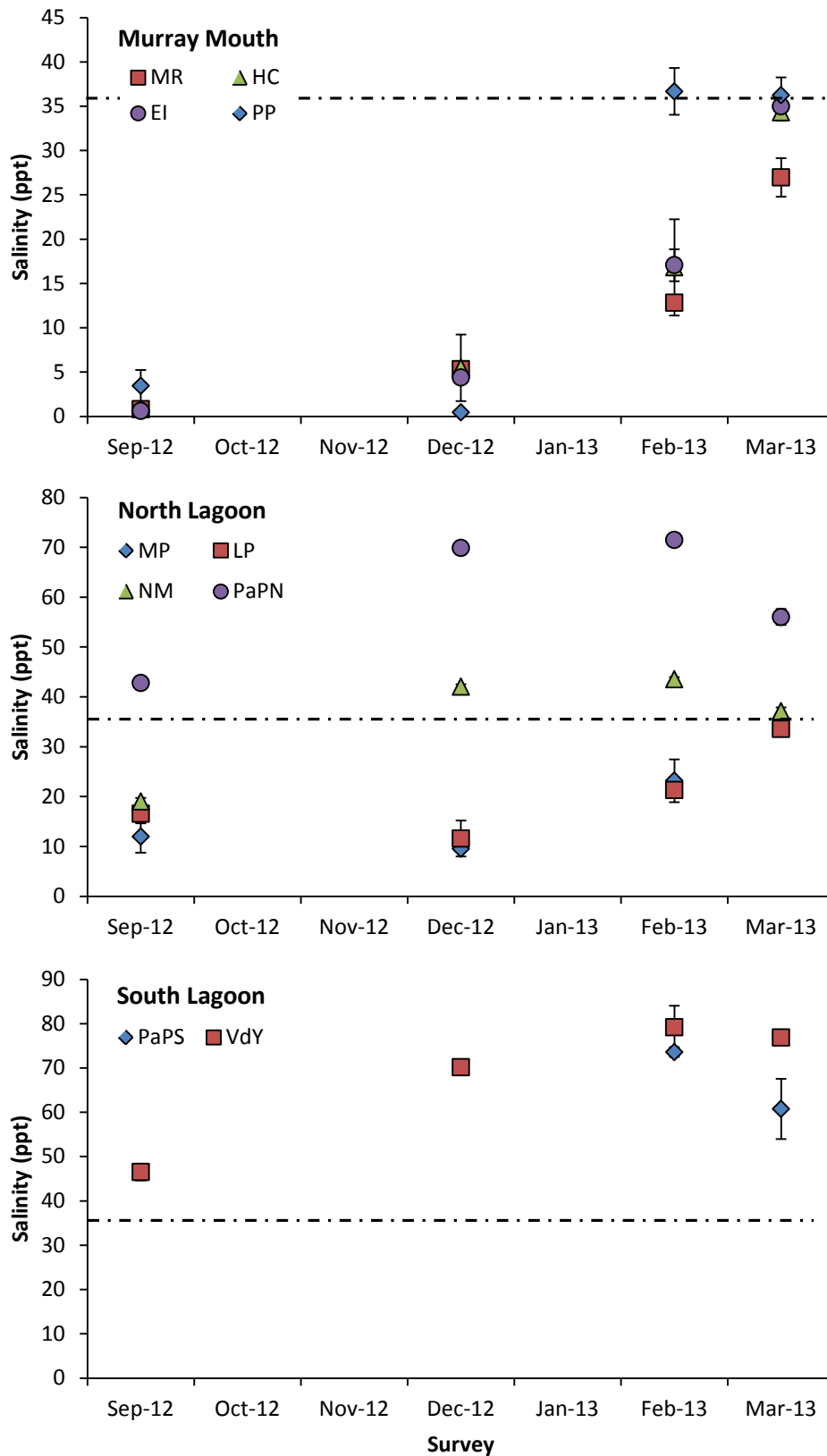
### 4.1 Environmental Conditions

***Key Question 1a) Will environmental conditions within the Murray Mouth reflect true estuarine conditions and have they further improved in the North and South Lagoons since flows recommenced in 2010?***

Salinity in the Murray Mouth varied at each study site across the four sampling occasions (Figure 3) for the 2012/2013 survey. Salinities were oligohaline at all four sites in the Murray Mouth in September and December 2012, ranging from 0.5 ppt at Pelican Point to 5.5 ppt at Hunters Creek (Figure 3). These salinities were similar to those observed during 2010/2011. Salinity increased in the Murray mouth over summer and was meso-euhaline, between 12 ppt at Monument Road and 36 ppt at Pelican Point (Figure 3). On average across the four sampling occasions in 2012/2013, salinity in the Murray Mouth region was estuarine, between 10 to 20 ppt. Compared to previous years, the average salinity in the Murray Mouth was lower than in the drought (data from December 2007 to February 2010), but higher than during the peak water release in 2010/2011 (Figure 4a).

In the North Lagoon, a salinity gradient occurred from the northern end (Mark Point) to the southern end (Parnka Point) and salinity was also rising within each site over the four sampling events. In the northern end (Mark Point and Long Point), salinities increased only later in summer, whereas salinity increased already by December in the southern end of the North Lagoon (Noonameena and Parnka Point), which was hypersaline over most of summer (Figure 3). There was only a small increase in salinity across Hells Gate on the two sampling occasions when measurements were taken from both the north and south side of Parnka Point (February and March 2013) (Figure 3). On average, across the four sampling occasions in 2012/2013, salinity in the North Lagoon was estuarine to hypersaline (between 20 to 60 ppt). This was a substantial reduction (~50%) in salinity compared to the drought period between December 2007 and February 2010, and within the range of reduced salinities recorded since the water release commenced in late 2010 (Figure 4b). The current water release has achieved a more long-lived reduction in salinities in the North Lagoon than a small water release in 2005 (Figure 4b).

Salinity at the South Lagoon site, Villa de Yumpa, was hypersaline, and also increased from 47 ppt in September 2012 to nearly 80 ppt for the remaining sampling occasions (Figure 3). Yet, even with these hypersaline conditions, salinity was lower than between November 2006 and February 2010 (Figure 4c). While the salinity in this South Lagoon site has not further improved since the previous year, it has remained consistently lower over several years since the water release commenced in late 2010 (Figure 4c).



**Figure 3. Average salinity at each study site for each sampling occasion during the 2012/2013 macroinvertebrate survey. Sites are labelled as in Table 2. Note different scales of the y-axis. The dashed line shows the salinity of seawater (36 ppt) for comparison among panels. Error bars indicate standard deviation.**

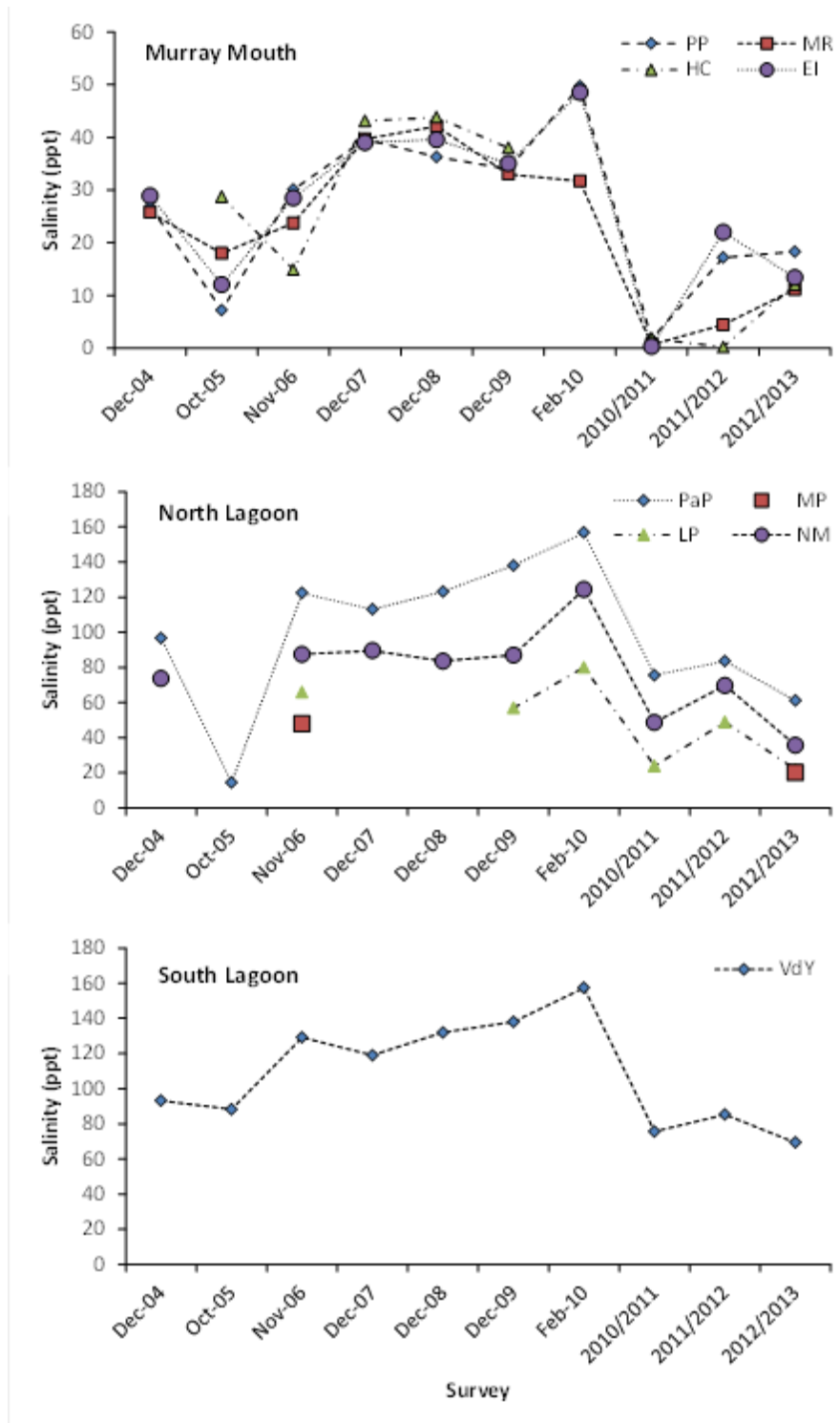


Figure 4. Average salinity at those study sites in the Murray Mouth and Coorong surveyed for this water release project and The Living Murray Icon Site condition monitoring (Dittmann *et al.* 2012) since December 2004, capturing also a small water release in late 2005 (Dittmann *et al.* 2006). Sites are labelled as in Table 2. Note different scales of the y-axis. The dashed line shows the salinity of seawater (36 ppt) for comparison among panels. Lines are only provided for guidance between sites, but further fluctuation could have occurred between the surveys.

## 4.2 Macroinvertebrate Recruitment

### **Key Question 1b) Has recruitment continued for macroinvertebrate species which underwent recruitment in 2011/2012 and is recruitment occurring for other species in 2012/2013?**

In the previous survey period (2011/2012), the timing of recruitment events for macroinvertebrates in the Murray Mouth and Coorong was species-specific (Keuning *et al.* 2012). Small individuals of the polychaetes *Capitella sp.* and *Simplisetia aequisetis* occurred continuously throughout the survey period, while those of *Nephtys australiensis* and *Australonereis ehlersi* were more abundant in spring/summer and summer/autumn respectively (Keuning *et al.* 2012). Juveniles of these species were also recorded throughout or at some time in the current survey period in 2012/2013, and their recruitment in the Murray Mouth and Coorong thus continued.

During the 2012/2013 survey, recruitment of the polychaete, *Simplisetia aequisetis* was investigated in more detail. Worms of this species ranged in size from 1 to 84 mm and the range of sizes was similar over most sites and times (Figure 5). At Long Point, larger worms only started to occur later in summer, a possible indication of the juvenile recolonisation of sediments at this site. High frequencies of small-sized *S. aequisetis* occurred at Mark Point and Long Point in December 2012 and at Pelican Point in February 2013, but were not as evident during the following sampling occasion (Figure 5). During March 2013, a high proportion of small-sized *S. aequisetis* occurred at all sites, yet not at all locations. At Monument Road, Hunters Creek and Ewe Island, more small worms occurred in intertidal than subtidal sediments in March 2013. Size-frequency distributions of *S. aequisetis* were largely similar in intertidal, subtidal and peninsula locations, yet larger worms at the Murray Mouth sites were mostly from subtidal sediments (Figure 5). Females with visible ovaries and egg-lined tubes of this species were observed in samples during March 2013 and reproduction of this species thus continued well into late summer. This last observation also indicates the occurrence of benthic development stages, and estuarine nereidids are known to have flexibility in their reproductive mode (Sato 1999). The 2012/13 survey results for *S. aequisetis* indicate that although small individuals of this species were observed at most sites throughout they survey, as during the 2011/12 survey, the recruitment of small individuals of this species into populations may be triggered by salinity (see also key question 1c).

The opportunistic polychaete *Capitella sp.* is rather small-sized, ranging from 2 to 31 mm in lengths (Figure 6). It was most prominent in the North Lagoon at Long Point and Noonameena. Size-frequency distributions of *Capitella sp.* between these sites showed some similarity, such as with the occurrence of smaller worms (< 4 mm in length) during December 2012 and February 2013 (Figure 6). However, no specific peaks in the proportion of small *Capitella sp.* were observed (Figure 6), indicating that recruitment of *Capitella sp.* could be continuous during the summer months. No larger capitellids were found in the September survey, and not until March 2013 in intertidal sediments at Noonameena, yet larger worms were present in subtidal samples from this site in December 2012. Size-frequency distributions also differed between locations at Long Point, where larger capitellids were found in the intertidal and a higher proportion of small worms was observed in the subtidal in March 2013 (Figure 6).

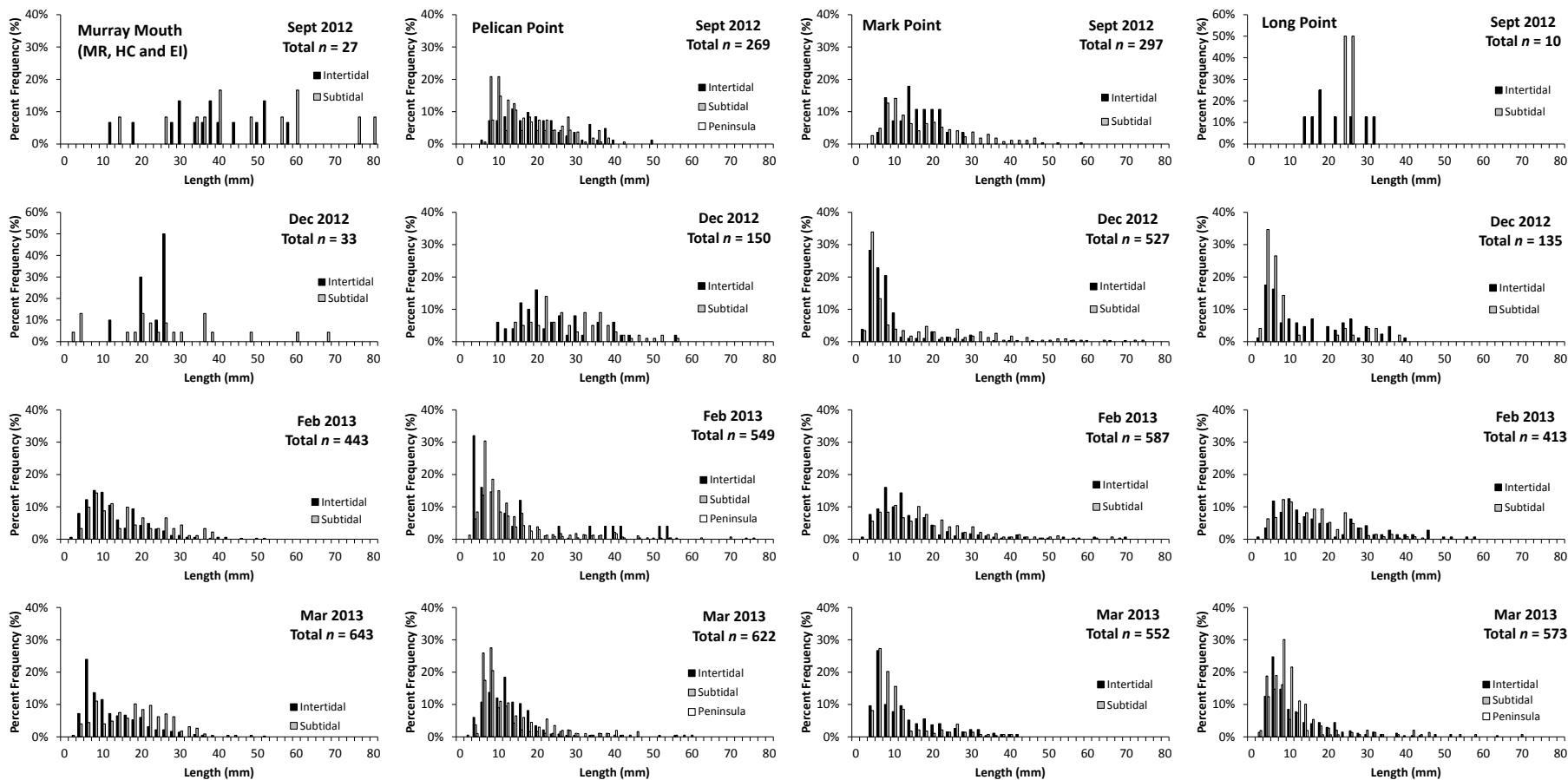
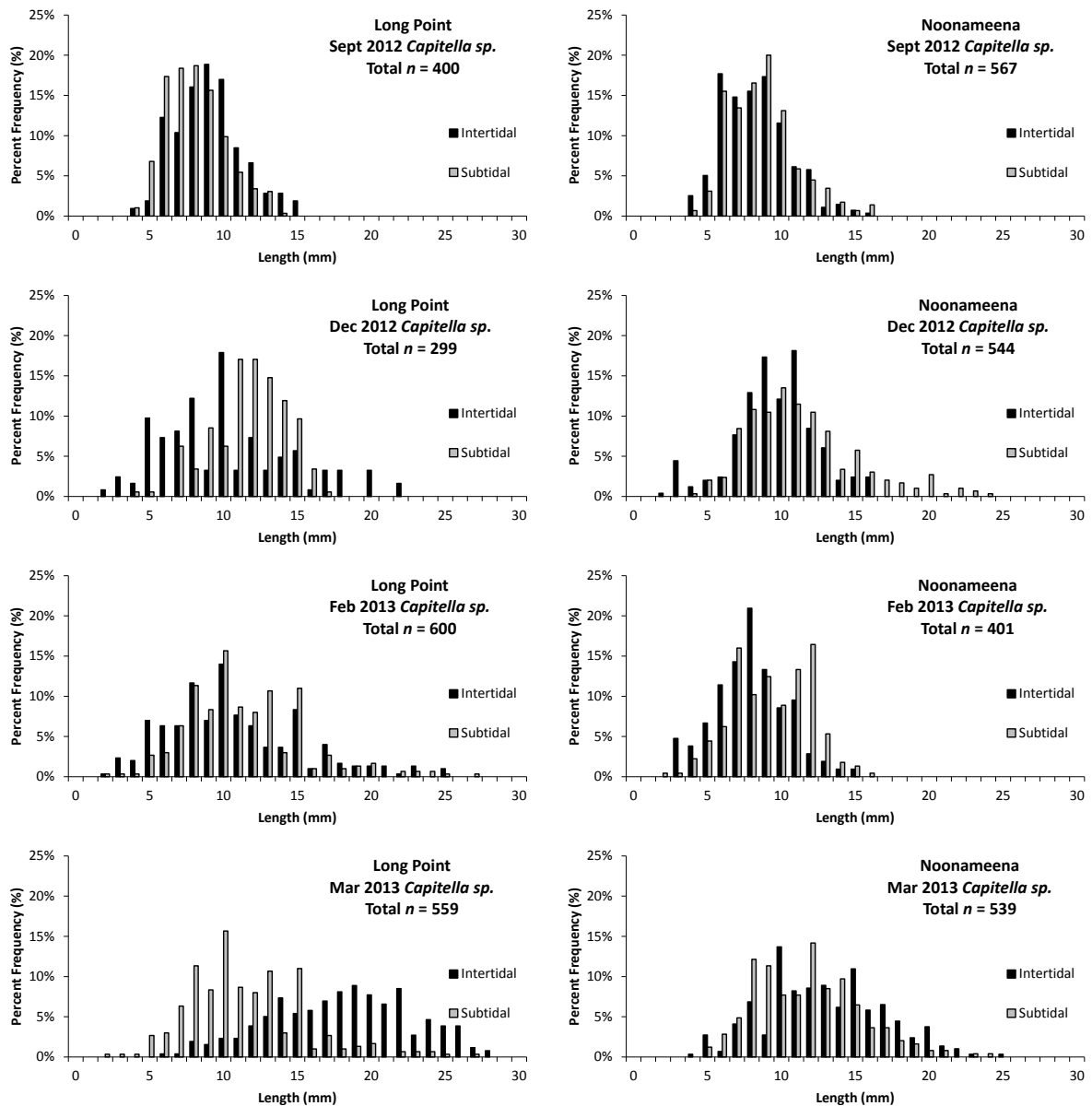


Figure 5. Size-frequency distribution histograms for *Simplicisetia aequisetis* for each sampling occasion (rows) and specific sites (columns). Murray Mouth sites (excluding Pelican Point) are grouped because size-frequency patterns for Monument Road (MR), Hunters Creek (HC) and Ewe Island (EI) were the same. Note not all plots have the same scale for per cent frequency (%).

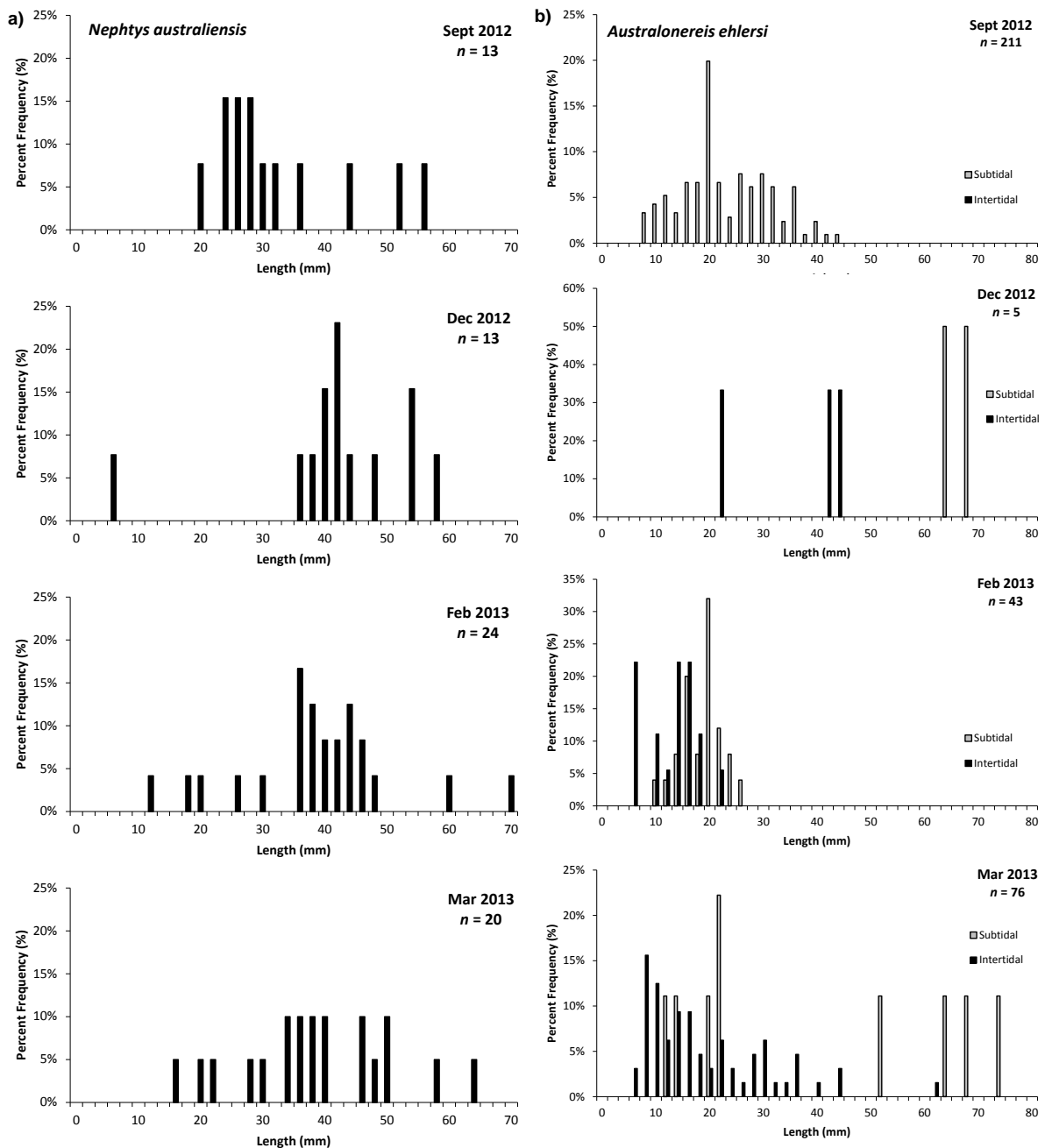


**Figure 6. Size-frequency distribution histograms for *Capitella sp.* for each sampling occasion at Long Point and Noonameena in the North Lagoon.**

The predatory species, *Nephtys australiensis*, occurred in much lower abundances than the previous two polychaete species, and thus size-frequency distributions of *N. australiensis* are based on fewer measurements. In addition, with the exception of Ewe Island, the occurrence of this species was limited to a few individuals per site, and size-frequency plots have been generated based on individuals from samples at Ewe Island only (combined intertidal and subtidal, where it was more common). No specific peaks in the proportion of small-sized individuals were observed for this species, although some smaller individuals occurred during December 2012, February and March 2013 (Figure 7a). Larger specimens of this species were regularly encountered, and *N. australiensis* was observed to reach lengths up to 7 cm at sites in the Murray Mouth. The size ranges of *N. australiensis* were similar to those recorded in the 2011/2012 survey, yet frequencies of small specimens were higher during the previous summer surveys (Keuning *et al.* 2012).



*Australonereis ehlersi* occurred almost exclusively at Noonameena in the 2012/2013 survey, where it was most abundant in September 2012. Over 200 individuals were measured with a modal length of 20 mm (average  $23.4 \pm 9.5$  mm) (Figure 7b). The size range for this nereidid polychaete was 5-74 mm. A higher proportion of small worms occurred in March 2013 when more large individuals were recorded as well (Figure 7b). The smaller specimens found in February and March 2013 came mainly from intertidal sediments, whereas larger *A. ehlersi* were from subtidal sediments. In December 2012, only five individuals could be measured, which were of larger sizes. This is similar to the previous year, which also showed more juveniles in March 2012 (Keuning *et al.* 2012).



**Figure 7.** Size-frequency distribution histograms for a) *Nephtys australiensis* at Ewe Island and b) *Australonereis ehlersi* at Noonameena for each sampling occasion in the North Lagoon. Measurements from worms from inter- and subtidal locations were combined for *N. australiensis*, but kept separate for *A. ehlersi*.

Based on size measurements taken from further macroinvertebrate species found in the 2012/2013 survey, recruitment occurred for several other species. Juveniles were often recorded at particular sites only and more often at submerged sampling locations. Egg carrying amphipods were also noted throughout the study.

*Boccardiella limnicola* (Polychaeta, Spionidae) occurred at all sites in the Murray Mouth, especially at Monument Road. These small tube dwelling worms reached up to 25 mm in length. In September, they were only found in subtidal sediments and of medium sizes. A recruitment event occurred in late spring, as a high frequency of small sized worms was recorded in December. The frequency of smaller worms was still apparent in February, but a shift towards larger sizes was evident, which continued in March. Size frequencies of worms from inter- and subtidal locations were similar in December and February, and the larger worms found in March were collected in the intertidal.

Some samples were taken from tubeworm reefs (*Ficopomatus enigmaticus*), at Mark Point in September and February. The lengths of these worms ranged from 3 – 20 mm, with a modal length of 8 and 10 mm in the two months respectively. A few tubeworms were collected from Monument Road in the Murray mouth as well, but these were smaller in size (5 mm). (See also key question 4)

A previously unrecorded sabellid polychaete (*Euchone cf. variabilis*) was found in sediments on the peninsula side at Long Point in March. These are small sized polychaetes, ranging in size from 2-5 mm, with a modal length of 2 mm, which could indicate recent recruitment.

Several bivalves were recorded and measured as well. *Spisula trigonella* occurred in subtidal sediments at Long Point and Noonameena, with similar size frequencies noted for each month (modal length 12-14 mm in September, December and February). Their size ranged from 2-14 mm and no distinct recruitment was evident in our sampling period. *Soletellina alba* was initially recorded in subtidal sediments at Long Point in the North Lagoon, but occurred throughout the Murray Mouth by March 2013 (Appendix, Figure A8). These bivalves ranged in size from 1–17 mm, with a higher frequency of juveniles (modal length 1.25 mm) in December 2012, indicating that recruitment occurred. *Arthritica helmsi* also re-occurred in the study area. This micro-mollusc occurred in size ranges from 0.6-2.3 mm, and presence of juveniles is impossible to differentiate with this narrow size range. In December and February shells from 0.6-1.8 mm in size were found in subtidal sediments at Mark Point and Long Point. Specimens of 1-2 mm in size were also found in intertidal sediments at Pelican Point in February.

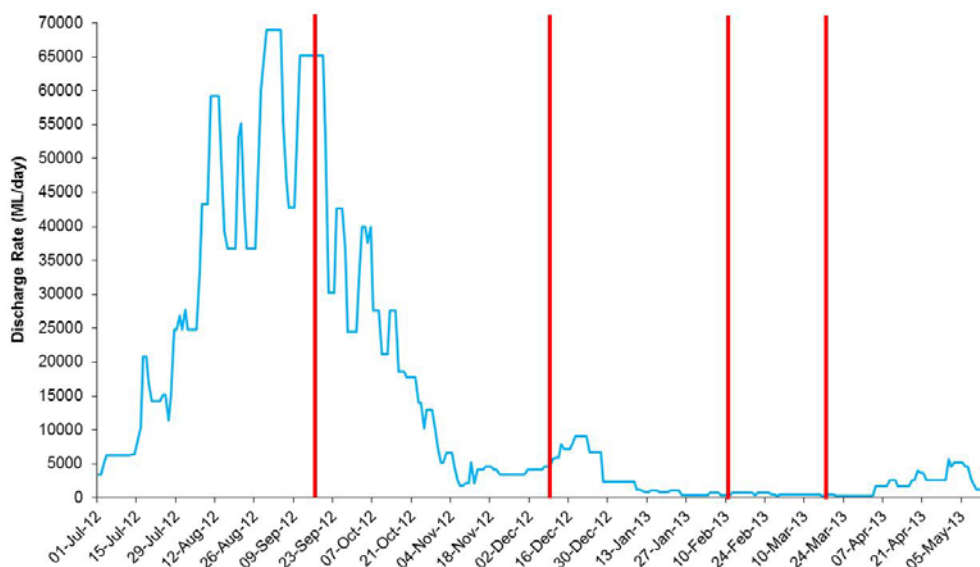
Overall, there was not only continued recruitment of those polychaetes already recorded in the previous summer survey period, but also recruitment of several other macroinvertebrate species (Table 3). Yet, recruitment was patchy, and the wider distribution of macroinvertebrates in the Murray Mouth and Coorong will be determined by a combination of pre- and post-settlement processes (Olafsson *et al.* 1994).

**Table 3. Occurrence of small individuals of polychaete species observed in benthic samples on each sampling occasion.**

Species	Location	Small individuals observed			
		Sep-12	Dec-12	Feb-13	Mar-13
<i>Simplisetia aequisetis</i>	Murray Mouth		✓	✓	✓
	Pelican Point			✓	✓
	North Lagoon		✓		✓
<i>Capitella spp.</i>	North Lagoon		✓	✓	
<i>Nephtys australiensis</i>	Ewe Island		✓	✓	✓
<i>Australonereis ehlersi</i>	Noonameena				✓

**Key Question 1c) Can the recruitment of species in 2012/2013 be linked with the timing of differing flow scenarios to identify drivers of macroinvertebrate recruitment?**

Prior to the water release in 2010, the occurrence of juvenile polychaetes and molluscs was confined to the northern end of the North Lagoon between Pelican Point and Mark Point (Dittmann *et al.* 2010b), whereas recruitment of macrobenthos occurs more wide spread now (this report). Flow affects macroinvertebrates through changes in water level and salinity, and the flow volume is taken as a proxy for these associated changes in environmental conditions. In the survey period 2012/2013, flows over the barrages were greatest in September 2012, and then declined into summer, with a slight increase again in December 2012 (Figure 8). By January 2013 flows had reduced substantially and remained very low for the rest of the sampling period (Figure 8). Corresponding to these flows, salinity in the Murray Mouth region and in the northern end of the North Lagoon (Mark Point and Long Point) remained brackish until December 2012, but then increased over the remainder of the sampling period (Figure 3).



**Figure 8. Total barrage flows (per day) into the Coorong from the Lower Lakes between July 2012 and May 2013 (data from River Murray Operations Group). Red lines indicate approximate timing of sampling during the 2012/2013 survey.**

Recruitment of Nereidids, such as *Simplisetia aequisetis*, is determined by a number of environmental factors, including salinity (Wilson 2000). Small individuals of *S. aequisetis* were first recorded in samples when salinities at the sites had reached around 10 ppt. This occurred at different times for different sampling sites in the Coorong, with Noonameena further south (and further from freshwater inflows over the barrages) exceeding marine salinities by December 2012, and sites around the Murray Mouth (closest to inflows over the barrages) exceeding 10 ppt by February 2013 (Figure 3). These dates correspond to the occurrence of small individuals in samples at Long Point and Mark Point in December 2012 and at Pelican Point in February 2013 (Figure 5). Recruitment of *S. aequisetis* is occurring much more wide spread since flow resumed, as in February 2010, during the drought, juveniles were only picked up at Pelican Point (Dittmann *et al.* 2010b).

The occurrence of small individuals of *Capitella sp.* in samples from Long Point and Noonameena was observed in December 2012 and February 2013 (Figure 6), when salinities at these two sites were very different (Figure 3). *Capitella sp.* is an opportunistic species adapted to highly changeable environments, with a short life-cycle that is completed in a few months and rapid reproduction, and thus may be less sensitive to changes in salinity to trigger reproduction and recruitment (Mendez *et al.* 2000; Ramskov & Forbes 2008). During the drought, juveniles of *Capitella sp.* were found at Pelican

Point and Mark Point in February 2010 (Dittmann *et al.* 2010b), whereas they are now occurring further into the North Lagoon.

Time lags in the response of larvae to freshwater inflow complicate correlations between flow dynamics and recruitment records, which can be further affected by post-settlement dispersal (Poulton *et al.* 2004). Recruitment can also be affected by turbulence (Fuchs *et al.* 2010) and bedload transport (Turner *et al.* 1997; Norkko *et al.* 2002). The loss of sediment from our settlement trays (see below) indicated a high rate of turbulence at the sediment-water interface, driven to different extents by wind, tides and outflowing water at the study sites, which could affect the colonisation of sediments.

### Larval recruitment

The targeted monitoring for larval recruitment was not very successful, as sediments were dislodged from settlement trays at most sites at both deployment occasions. From the first deployment in October 2012, control samples showed presence of juvenile nereidid worms at Mark Point, but only few juvenile polychaetes at the other sites. From trays deployed at the end of January 2013 and collected in February 2013, control samples and treatment trays at Mark Point contained mainly juvenile *Simplisetia aequisetis* (classified as worms < 10 mm in length) and *Arthritica helmsi*, while those at Noonameena contained mainly *Capitella sp.* (juveniles classified as worms < 5 mm in length). Species collected in the trays were similar to those occurring in the ambient sediment, unlike similar studies carried out elsewhere (Lundquist *et al.* 2006).

The sediment in the azoic treatment trays was colonised exclusively by small sized specimens of *S. aequisetis* and *Capitella sp.* respectively, whereas the ambient control samples contained higher frequencies of larger worms. Procedural control sediments were occupied by a wider range of sizes of these polychaetes, yet skewed towards more juveniles. As there was no pronounced recruitment peak of *S. aequisetis* at Mark Point during February 2013 (Figure 5), the targeted monitoring gave valuable information on the occurrence of recruitment at this time.

## 4.3 Macroinvertebrate Recolonisation

### **Key Question 1d) Will there be any further recolonisation of mudflats by macroinvertebrates absent from areas in previous monitoring 2010-2012?**

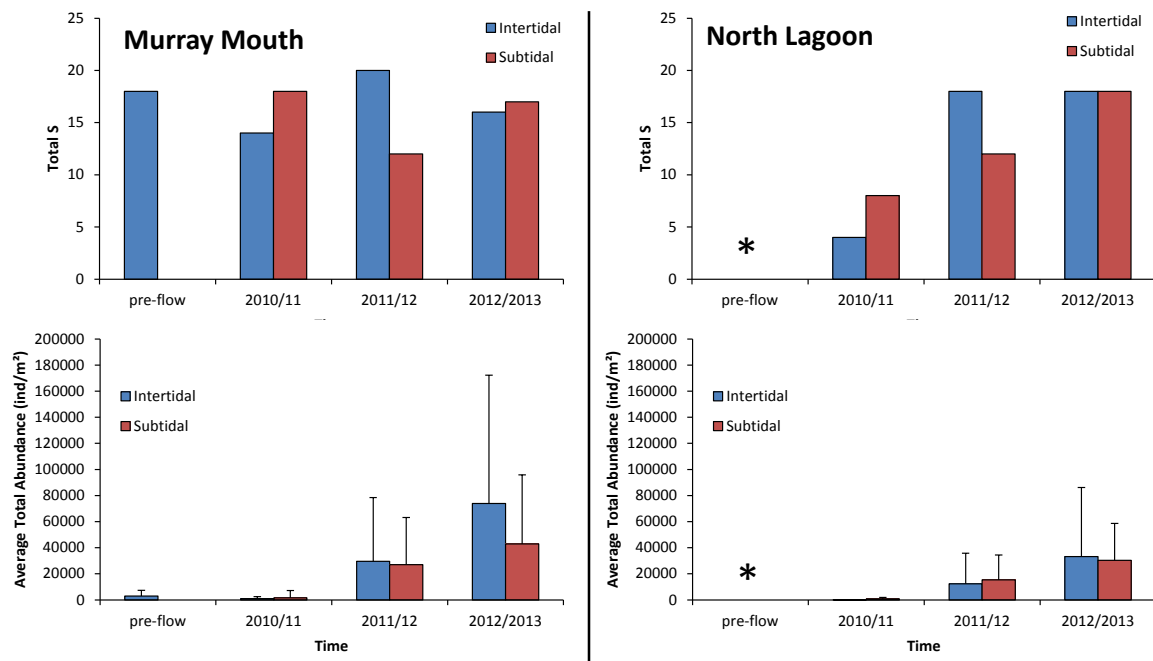
During the survey period in 2012/2013, mudflats were inhabited by key macroinvertebrates in the system as well as colonised by further macroinvertebrate species rarely or not seen in recent years. A species not recorded in the 2010-2012 monitoring, *Euchone cf. variabilis* (Polychaeta, Sabellidae), was very common on the mudflat at the peninsula side of Long Point in March 2013. Compared across the recent years, species numbers in mudflats of the Murray Mouth are comparable to the pre-flow situation, and similar to subtidal sediments (Figure 9). In the North Lagoon, the total number of species found in mudflats did not increase further above the previous monitoring period, yet tripled compared to the time immediately after the flood. Here, species numbers increased in subtidal sediments (Figure 9). Averaged across all sites and months over the respective survey periods, abundances in mudflats in the Murray Mouth and North Lagoon appeared to have increased in this third summer since flow recommenced, and to be also higher in mudflats compared to subtidal sediments in the Murray Mouth (Figure 9).

In detail, benthic abundances were as high or higher in mudflat sediments on the mainland and peninsula sides than at subtidal locations in 2012/2013 (Figure 10). The abundance pattern was driven mainly by amphipods and chironomid larvae (Figure 11), as in the previous year (Keuning *et al.* 2012). Amphipods occurred in high abundances at most sites during the survey period, and made up most of the total benthos in the intertidal mudflat at Pelican Point in September 2012 and March 2013. Chironomids also occurred mainly in mudflats sediments throughout the study area and at all survey

times (Figure 11). Compared to the previous monitoring year (Keuning *et al.* 2012), their abundances increased in mudflats at Ewe Island.

The polychaete *Simplisetia aequisetis*, which began recolonising mudflats in the Murray Mouth and North Lagoon during 2011/2012, further recolonised mudflats in the Murray Mouth in 2012/2013, especially at Monument Road and Pelican Point (Figure 11). The increase in abundances of *S. aequisetis* towards late summer 2012/2013 occurred mainly at intertidal mudflat sites, most pronounced at Mark Point and Long Point, but also at all Murray Mouth sites including Ewe Island.

Abundance of *Capitella sp.*, was always greatest in subtidal sediments at Long Point, where at Noonameena, abundances were more similar between the two depths locations. Abundances of *Capitella sp.* increased over the sampling period in both mudflat and subtidal sediments (Figure 11).

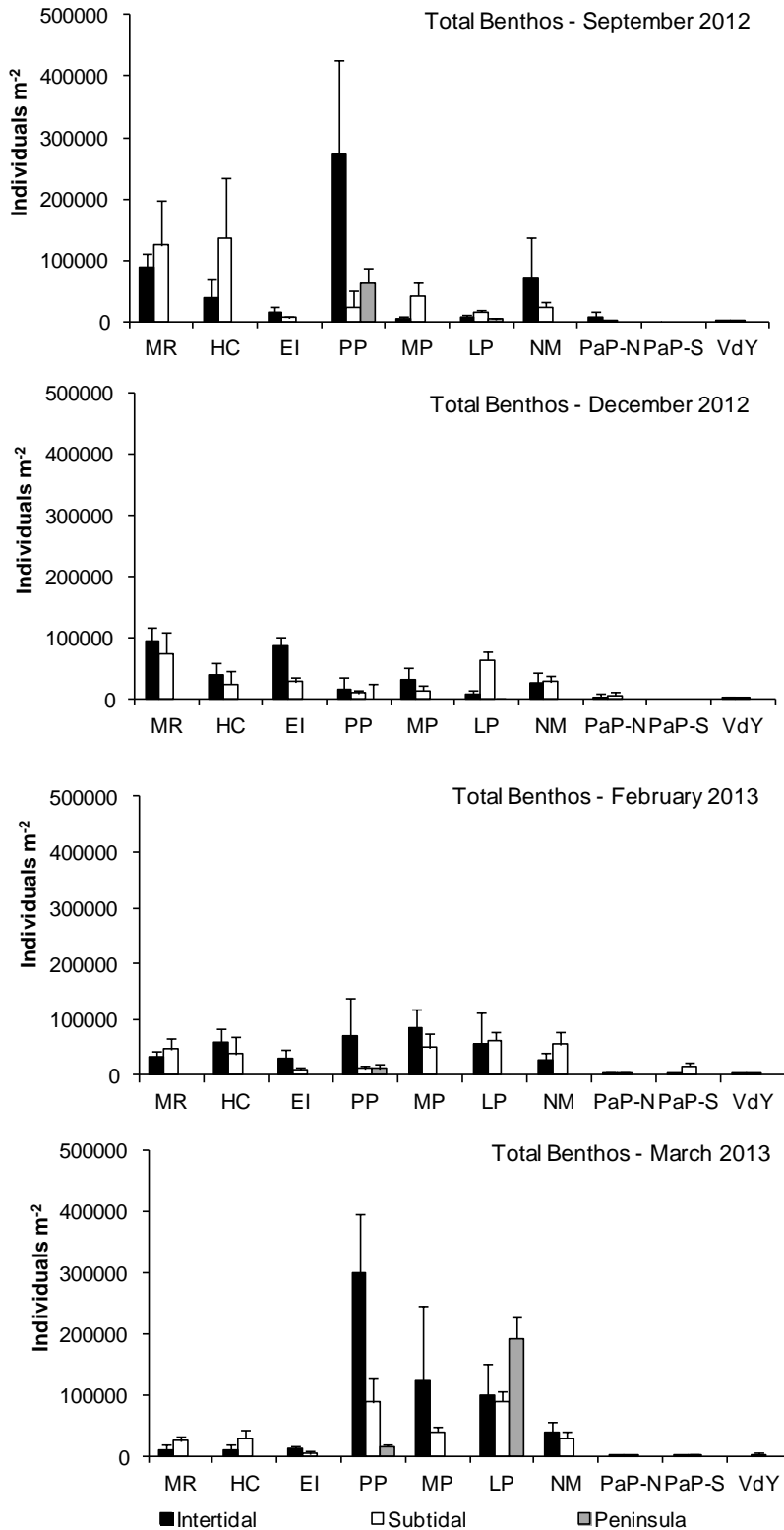


**Figure 9. Total number of species recorded in samples (Total S) and average total abundance (individuals per m<sup>2</sup>; with standard deviation) of macroinvertebrate communities sampled during periods of different flow regimes in the Murray Mouth and North Lagoon regions. Time spans used are covering the drought (pre-flow: August 2009 – August 2010), and sampling periods for the water release monitoring after flows over the barrages commenced (2010/11: December 2010 – April 2011; 2011/12: October 2011 – May 2012; 2012/13: this current report). Asterisk indicates that no sampling occurred during the pre-flow time frame in the North Lagoon (other than a one-off TLM monitoring). Note that different sites and different numbers of sites were included for the Murray Mouth and North Lagoon regions for each monitoring time span.**

Some of the bivalves absent or rare in the previous monitoring periods (*Arthritica helmsi*, *Spisula trigonella*, *Soletellina alba*) were mainly recorded from subtidal sediments (Appendix Figure A8). Only *A. helmsi* started to appear in intertidal samples in December 2012 and became more abundant in mudflat sediments at Mark Point in March 2013. A single specimen of *S. trigonella* was collected from mudflats at Noonameena in September 2012, but this species was not found again in intertidal locations for the rest of the survey period. *Soletellina alba*, however, started to appear in mudflat samples in February and March 2013 at several sites in the Murray Mouth and North Lagoon where it also occurred in higher abundances in adjacent subtidal locations.

Based on the macroinvertebrate distribution patterns and abundances in inter- and subtidal sediments, source populations for the recolonisation of mudflats would have occurred in subtidal sediments of the same region or adjacent areas. Recolonisation patterns in sandflats can be explained by hydro-dynamics, as larvae or dispersing life stages depend on tide or wind driven transport with overlying

water, but volumes of freshwater flow can also affect their transport (Lundquist *et al.* 2004; Lundquist *et al.* 2006).



**Figure 10: Average abundances (individuals per m<sup>-2</sup>; n=10, with standard deviation) of total macroinvertebrates recorded at intertidal, subtidal and peninsula locations during September and December 2012, and February and March 2013. Both peninsula sites were sampled during September 2012 and March 2013, but at Pelican Point peninsula only in February 2013. See Figure 2 for site locations.**

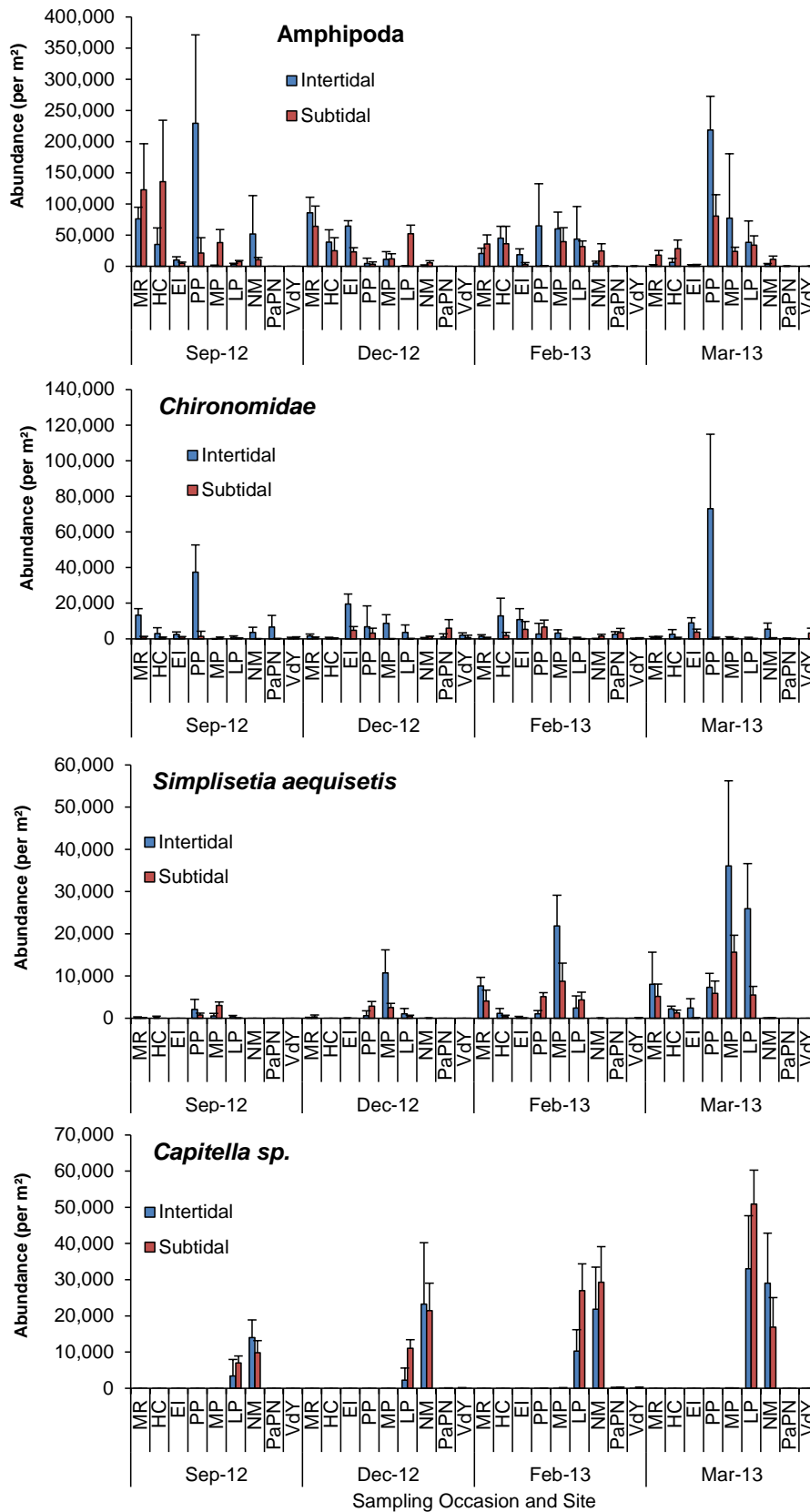


Figure 11: Average abundances (individuals per m<sup>-2</sup>; n=10, with standard deviation) of key macro-invertebrate taxa recorded at intertidal and subtidal locations during September and December 2012, and February and March 2013. Note that peninsula locations are not considered in these graphs.

**Key Question 1e) Will recolonisation of macroinvertebrate species occur in the South Lagoon if salinities remain lowered within this region?**

Previously, macroinvertebrate communities at the South Lagoon site, Villa de Yumpa, have contained only halophylic species, the larvae of salt-tolerant Diptera (Chironomidae and Ephydriidae), Oligochaetes, and species of salt lake crustaceans of the Orders Ostracoda and Amphipoda. During 2012/2013, salinity at Villa de Yumpa was lower than recorded in February 2010 for the third year in a row (Figure 4c). Two polychaete species, *Simplisetia aequisetis* and *Capitella sp.*, which had not previously been recorded from Villa de Yumpa, were found in samples during the summer of 2012/2013 (Table 4). Yet, both species only occurred sporadically in samples. *Capitella sp.* was observed first in intertidal samples during December 2012 (5 individuals), then in subtidal samples in February 2013 (9 individuals). *S. aequisetis* was found in subtidal sediments only, during February 2013 (3 individuals). Other species recorded at Villa de Yumpa in 2012/2013 were found here in the previous survey as well (Oligochaeta, Amphipoda and Ostracoda, Chironomids and other insect larvae). A continued lower salinity will support recolonisation of the South Lagoon, and further sampling is required to determine if other macroinvertebrates will establish in the South Lagoon.



**Table 4. Presence/Absence table of species for each site with average abundances of each species, calculated across both sampling locations (intertidal and subtidal) and all sampling occasions during the 2012/2013 survey period. Note that for Peninsula sites samples were only collected from the intertidal location. Numbers are average abundance (Individuals per m<sup>2</sup>).**

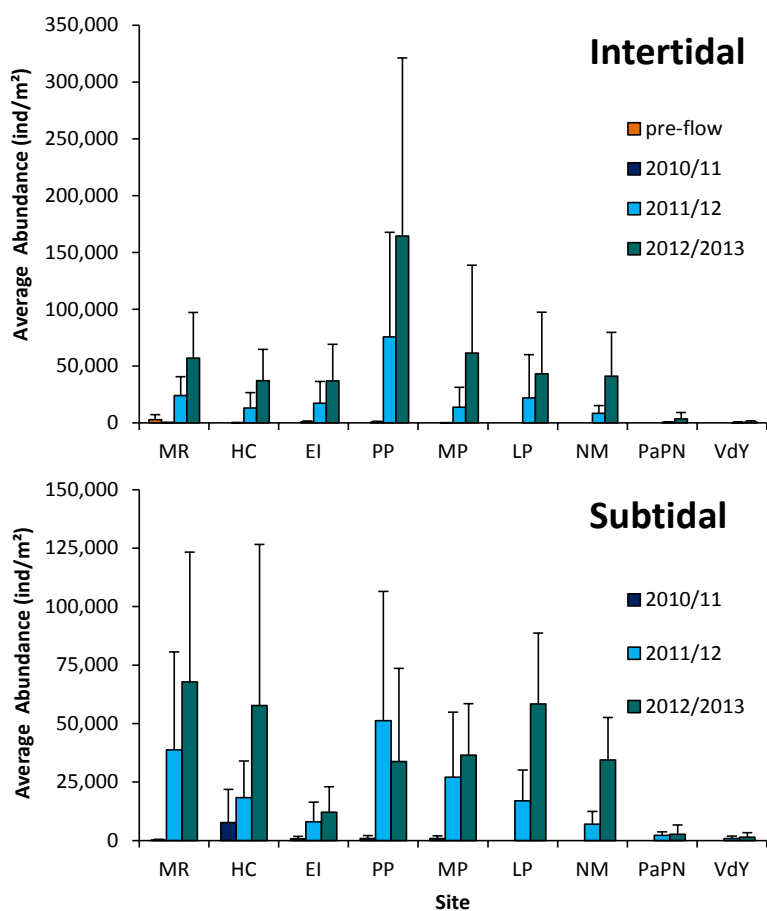
Species List	Murray Mouth					North Lagoon					South Lagoon	
	Monument Road	Hunters Creek	Ewe Island	Pelican Point	Pelican Point Peninsula	Mark Point	Long Point	Long Point Peninsula	Noonameena	Parnka Point North	Parnka Point South	Villa de Yumpa
<i>Oligochaeta</i> sp.	9	106	1043	467	28	373	689	60	261	42	0	2
<i>Capitella</i> sp.	0	0	0	2	8	15	18104	37080	20697	32	177	21
<i>Simplisetia aequisetis</i>	3202	655	332	3217	8801	12384	5003	1446	10	0	0	5
<i>Australonereis ehlersi</i>	0	0	8	0	0	0	22	6	938	2	0	0
<i>Nephtys australiensis</i> <sup>1</sup>	0	4	100	8	4	0	1	0	0	0	0	0
<i>Ficopomatus enigmaticus</i>	3	0	0	0	0	48	0	0	8	0	0	0
<i>Euchone variabilis</i>	0	0	0	0	0	0	0	5803	0	0	0	0
<i>Boccardiella limnicola</i>	3676	67	2	603	472	4	0	0	0	0	0	0
<i>Arthritica helmsi</i>	1	3	20	141	300	1434	6	42	0	0	0	0
<i>Spisula trigonella</i> <sup>2</sup>	0	0	0	0	28	0	3	6	93	0	0	0
<i>Soletellina alba</i>	38	9	15	8	16	0	93	30	3	0	0	0
Uniden. Juv. Bivalve	0	0	0	0	424	0	0	0	0	0	0	0
Hydrobiidae	14	58	31	15	20	18	0	0	0	0	0	0
<i>Salinator fragilis</i>	0	0	2	2	0	0	2	0	0	0	0	0
Ostracoda	0	2	0	0	0	0	0	0	0	325	0	0
Amphipoda	52833	43705	15712	77733	15386	32765	26144	52628	14053	18	21	5
Mysidacea	99	4	14	39	4	121	25	150	31	3	0	0
<i>Paragrapsus gaimardii</i>	50	3	5	2	0	6	4	0	0	0	0	0
<i>Amarinus laevis</i>	2	0	0	0	0	0	0	0	0	0	0	0
Chironomidae larvae	2471	2782	7066	16480	5357	1666	703	1416	1582	2496	4312	941
Chironomidae pupae	31	57	130	411	56	27	6	12	41	195	291	60
Ceratopogonidae larvae	0	2	0	0	0	0	0	0	0	18	3	0
Ephyridae larvae	0	0	0	6	0	1	0	0	6	2	15	0
Ephyridae pupae	0	0	0	0	0	2	63	0	0	2	0	0
Dolichopodidae larvae	15	0	11	50	0	50	6	0	35	0	0	0
Dolichopodidae pupae	2	0	0	2	0	2	0	0	2	0	0	0
Hydrophilidae larvae	0	0	0	0	0	0	0	0	2	0	0	0

<sup>1</sup> now accepted as *Aglaophamus australiensis*

<sup>2</sup> previously *Notospisula trigonella*

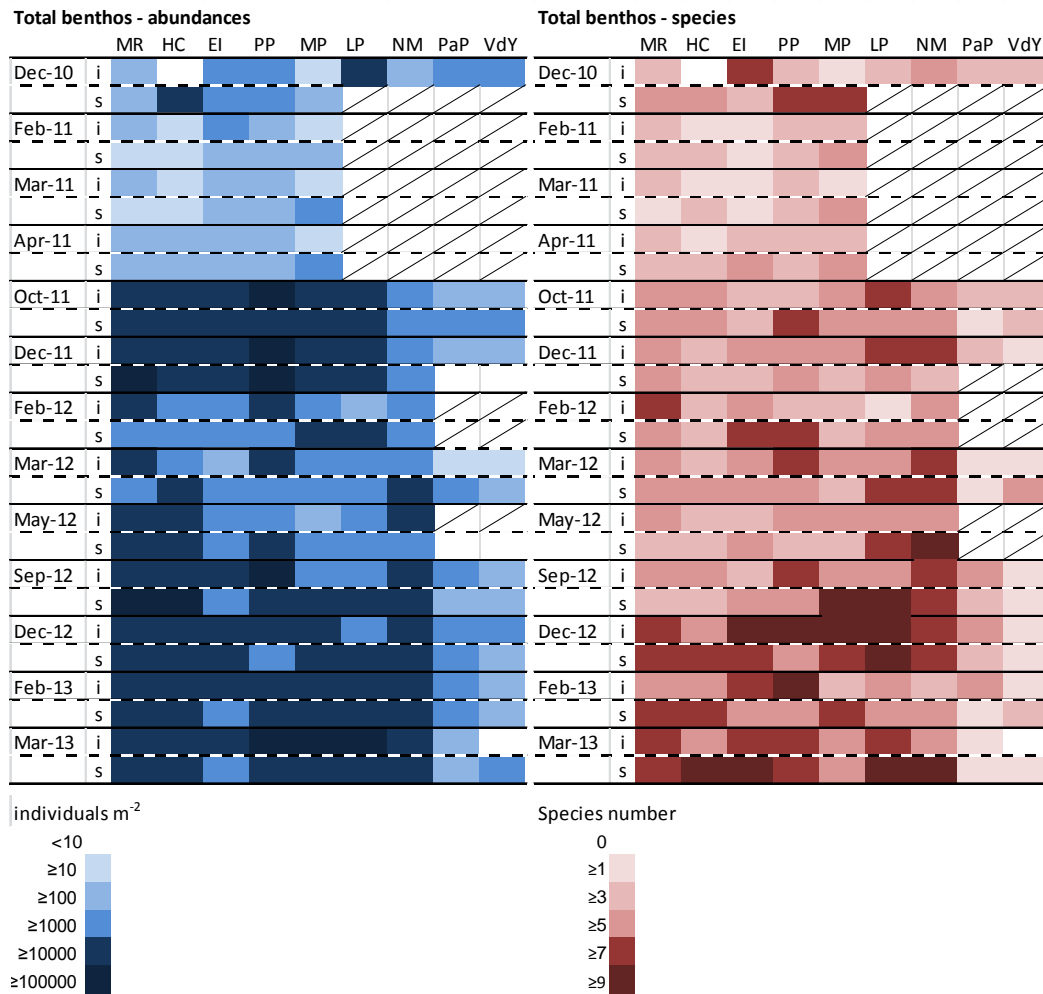
**Key Question 1f) Have species maintained or further increased their distribution range in comparison to 2011/2012?**

During the monitoring period 2012/2013, both species numbers and abundances of macro-invertebrates increased and shifted further south into the North Lagoon, while also maintaining and increasing at sites in the Murray Mouth (Figures 12, 13). In addition, a species of polychaete (*Euchone* cf. *variabilis*, Sabellidae, Fabriciinae) was recorded for the first time in this water release monitoring in samples from the Long Point Peninsula site in March 2013 (Table 4). This may indicate a recurrence of Fabriciinae recorded after water releases in the early 1980s (Geddes & Butler 1984; Geddes 1987).



**Figure 12. Abundance (individuals per m<sup>2</sup>; average with standard deviation) of total macrobenthos during periods of different flow regimes in the Murray Mouth and Coorong. Time spans used are covering the drought (pre-flow: August 2009 – August 2010), and sampling periods for the water release monitoring after flows over the barrages commenced (2010/11: December 2010 – April 2011; 2011/12: October 2011 – May 2012; 2012/13: this current report). Only sites surveyed over several years are included. Note that the time spans comprise at least four survey events each over the respective spring and summer periods, hence the high variability within each time span.**

The expansion of distribution range in 2012/2013 was most evident in *Simplisetia aequisetis*, which was now found at all sites except Parnka Point (Appendix Figure A7). *S. aequisetis* occurred with few specimens at Noonameena in this study period, first in subtidal and by March in intertidal sediments. At sampling sites in the Murray Mouth and northern North Lagoon (especially at Mark Point), abundances of this species generally increased, with peak abundances recorded in intertidal sediments in late summer (Figure 11). This increase included higher abundances at Ewe Island and Hunters Creek, where *S. aequisetis* was rare in the previous monitoring periods after flow commenced.



**Figure 13.** Patterns of change in the distribution of macroinvertebrates since flow commenced, illustrated by average abundances (individuals per m<sup>2</sup>) and number of species found in sediments of the Murray Mouth and Coorong sites during the water release study. Abundances and species numbers are indicated by colour codes (see legends). Sampling locations are differentiated as intertidal (i) and subtidal (s). Peninsula locations are not included in this comparison as first sampled in 2012/2013. Discontinued sampling sites in the Murray Mouth from the 2010/2011 and 2011/2012 monitoring periods are also left out. Sites in the Coorong that are crossed out were not sampled in the earlier monitoring periods yet for Dec-10, data are used from the 2010/2011 TLM monitoring and an additional sampling at Long Point.

Other species who occurred at more sites in the study area in the 2012/2013 monitoring period were *Boccardiella limnicola*, and the bivalve *Arthritica helmsi* (Table 4). *B. limnicola* had started to appear in subtidal locations at some sites in the Murray Mouth during 2011/2012 (Keuning *et al.* 2012), and was now recorded from Monument Road to Mark Point, albeit in low abundances at some of the sites (Table 4). *A. helmsi* was collected at all sites in the Murray Mouth region, and as far south in the North Lagoon as Long Point during the 2012/2013 monitoring (Table 4), whereas it was only collected from Ewe Island during 2011/2012 (Keuning *et al.* 2012). *A. helmsi* thus increased its distribution range and was most abundant at Pelican Point and Mark Point (Table 4). *Soletellina alba*, which was recorded in samples from Ewe Island, Long Point and Noonameena during 2011/2012 (Keuning *et al.* 2012), occurred throughout most of the Murray Mouth and North Lagoon by March 2013. Expansion of the distribution range of these species indicates an increase in suitable habitat as estuarine conditions prevail in the Murray Mouth and North Lagoon of the Coorong.

*Capitella sp.* occurred predominately at Long Point and Noonameena in the North Lagoon, from where it started to extend into neighbouring sites in the Coorong, including the northern reaches of the South Lagoon (Table 4, Appendix Figure A7). This species was first observed at Parnka Point in samples

from the subtidal location during December 2012, then at inter- and subtidal locations here in February 2013, but not in the following month. In the previous monitoring period, this species, which is an indicator for polluted or eutrophic sediments (Tsutsumi 1990; Ramskov & Forbes 2008), was found at several sites in the Murray Mouth and in the North Lagoon, but shifted away from the Murray Mouth sites during 2012/13, further indicating recovery of the Murray Mouth region as estuarine conditions persist.

Other species, however, remained confined to particular sites or contracted in their distribution range. *Nephtys australiensis* was almost exclusively found at Ewe Island and adjacent sites during the 2012/2013 monitoring, whereas it had a wider distribution range from Monument Road and Long Point in the previous monitoring periods (Appendix Figure A7). The distribution range of *Australonereis ehlersi* mainly contracted into the North Lagoon during 2012/2013, whereas this species was previously found at several sites in the Murray Mouth (Appendix Figure A7). *Spisula trigonella* remained confined to mainly Long Point and Noonameena in the North Lagoon, where it was found in the previous year as well, with abundances increasing at Noonameena towards the end of summer 2013 (Appendix Figure A8).

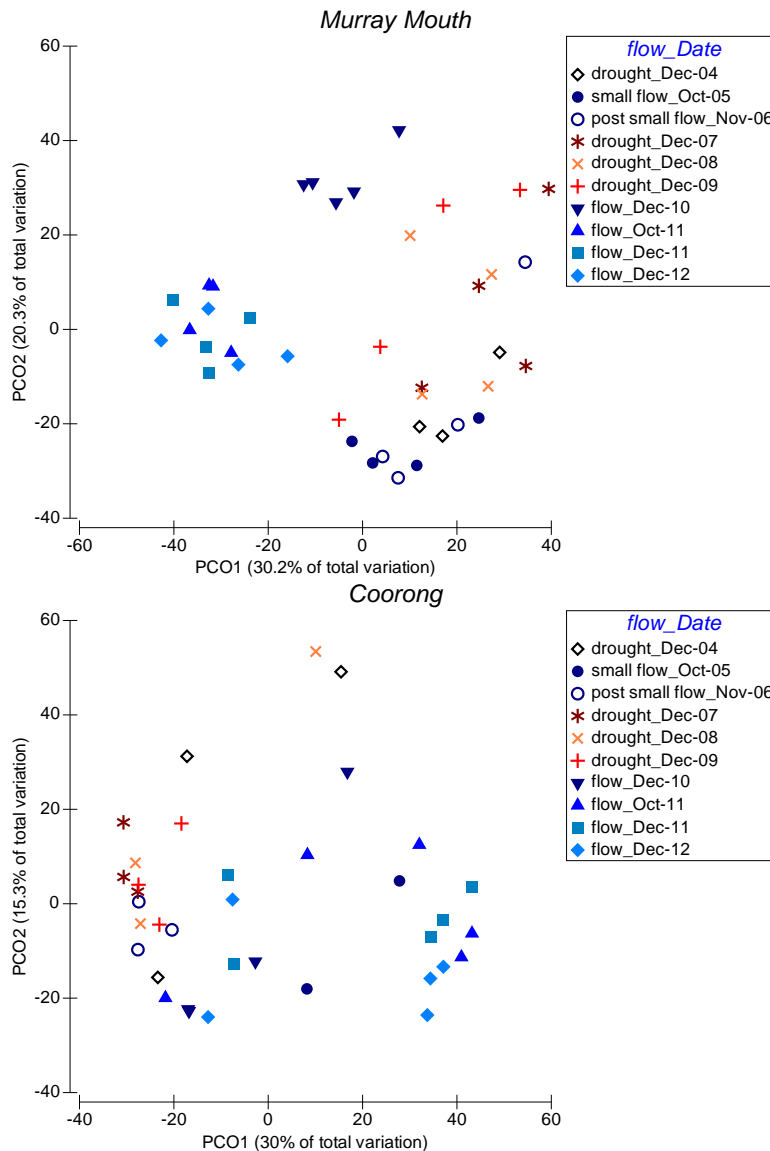
Of the two groups of organisms distributed widely throughout the Murray Mouth and Coorong, amphipods occurred in higher abundances at Noonameena during the 2012/2013 monitoring period, and were found again around Parnka Point in late summer (Appendix, Figure A7). Chironomids were abundant again in the Murray Mouth, but less at Monument Road and more at Ewe Island and pelican point over summer 2012/2013 (Appendix, Figure A7).

**Key Question 1g) Has the macroinvertebrate community been restored to pre-drought conditions following the 2011/2012 barrage flows into the Coorong?**

For both the Murray Mouth and Coorong, a change in benthic communities from those that persisted over the drought years and immediately after flow commenced is evident from multivariate analyses (Figure 14). In the Murray Mouth, the further barrage flows have supported a recovery pathway for macroinvertebrate communities, yet there was little further change between 2011/2012 and 2012/2013. For the Coorong, communities became increasingly distinct from the drought years, but separation of sites was less clear, reflecting the larger gradient of environmental conditions and progressive recovery in this region following restored flows.

The species composition at Murray Mouth sites was fairly similar over the 2012/2013 monitoring (Table 4). The macroinvertebrate community included a number of species that are tolerant of near freshwater and estuarine conditions, including the polychaetes *Simplisetia aequisetis* and *Nephtys australiensis*, Hydrobiid gastropods, the bivalves *Salinator fragilis* and *Arthritica helmsi* and the omnipresent larvae of Chironomidae (Diptera). The macroinvertebrate community in the Murray Mouth region was characterised by high abundances of Amphipoda and Chironomid larvae when conditions were fresh (September and December 2012; Table 5). *Simplisetia aequisetis* increased in abundance as salinities were rising to brackish and marine conditions in the Murray Mouth over summer, and was identified by SIMPER analysis as a further characteristic species in February and March 2013 (Figure 3; Table 5).

For longer term comparisons, only qualitative data on the presence of species in the Coorong are available (Geddes & Butler 1984; Geddes 1987), taken during a period of flow over the barrages between March 1983 and March 1985, but no historic data on macroinvertebrates exist for the Murray Mouth region. During the 2012/2013 monitoring period, similar species as recorded by Geddes and Butler (1984) and Geddes (1987) for fresh and estuarine conditions in the North Lagoon were found in the Murray Mouth region (Table 6).



**Figure 14. PCO plots of macroinvertebrate communities in the Murray Mouth and Coorong, based on sites sampled since December 2004 until December 2012, including Monument Road, Ewe Island and Pelican Point for the Murray Mouth and Long Point, Noonameena, Parnka Point and Villa de Yumpa for the Coorong. Only sampling dates between October and December were considered to avoid any confounding seasonal effects. Data from 2004 to 2010 (Coorong data for 2010 only) are taken from annual TLM monitoring, and from 2010-2012 from water release monitoring.**

Macroinvertebrate species compositions at Mark Point and Long Point during 2012/2013 were most similar to those observed when estuarine/hypersaline conditions persisted in the Coorong, and those observed in the southern North Lagoon during 2012/2013 were most similar to those observed historically in the region when hypersaline conditions persisted (Table 6; Geddes & Butler 1984; Geddes 1987).

Reflecting gradients in salinity observed in the North Lagoon (Figure 3), there was a transition in the abundance of macroinvertebrate species found between sites in the northern (Mark Point and Long Point) and southern (Noonameena and Parnka Point) parts of the North Lagoon. Communities at Mark Point and Long Point in the northern North Lagoon were characterised by Amphipoda, *S. aequisetis*, and in spring also by chironomid larvae and oligochaetes (Table 5). Estuarine species were less abundant south of Long Point (Table 4). *Simplisetia aequisetis* became uncommon in samples, and *Capitella sp.* became the most abundant polychaete in the North Lagoon (Table 4). Their abundances increased towards late summer when salinities were reaching marine conditions (Figures 3 and 11).

*Hydrobiid* gastropods were not recorded from samples south of Mark Point, and *Salinator fragilis*, *Arthritica helmsi* and *Nephtys australiensis* were not found south of Long Point (Table 4).

Communities at Noonameena and Parnka Point were different to sites further north and mainly characterised by chironomid larvae, *Capitella sp.* and amphipods (Table 5). Although *S. aequisetis* was still found, *Capitella sp.* dominated abundances, along with Amphipoda (Table 4). At the South Lagoon site, Villa de Yumpa, the macroinvertebrate community was very distinct in being dominated by halophylic species, especially the Chironomid larvae (Table 4). Unlike 2011/2012, *Simplisetia aequisetis* and *Capitella sp.* were now recorded at Villa de Yumpa. The community was characterised by chironomid larvae in September and December 2012, with *Capitella sp.* also being characteristic during February 2013 and Amphipoda during March 2013 (Table 5). The macroinvertebrate species composition in the South Lagoon during 2012/2013 was most similar to the hypermarine to hypersaline communities observed by Geddes and Butler (1984) and Geddes (1987).

**Table 5. SIMPER results showing average similarities (Ave. Sim.) and characteristic taxa that contributed highly (greater than 10%) to average similarity among samples within each region (divided into northern and southern reaches for the North Lagoon based on observed differences in salinities, see Figure 3), for each sampling occasion. Taxa are listed in decreasing order of contribution to the average similarity (per cent contributions given in brackets next to each species). Site acronyms are as listed in Table 2.**

Region	Site	September 2012		December 2012		February 2013		March 2013	
		Ave. Sim	Characteristic taxa	Ave. Sim.	Characteristic taxa	Ave. Sim.	Characteristic taxa	Ave. Sim.	Characteristic taxa
Murray Mouth	All MR, HC, EI, PP	58.8	Amphipoda (72.0) Chironomid larvae (19.9)	55.7	Amphipoda (68.1) Chironomid larvae (19.2)	55.8	Amphipoda (45.5) Chironomid larvae (26.6) <i>Simplisetia aequisetis</i> (15.9)	47.5	Amphipoda (37.5) <i>Simplisetia aequisetis</i> (29.6) Chironomid larvae (18.3)
North Lagoon	Northern MP, LP	46.1	Amphipoda (44.3) Oligochaeta (16.3) <i>Simplisetia aequisetis</i> (12.6) Chironomid larvae (12.0)	42.4	Amphipoda (45.2) <i>Simplisetia aequisetis</i> (25.5) Chironomid larvae (13.5)	68.0	Amphipoda (67.0) <i>Simplisetia aequisetis</i> (22.0)	63.1	Amphipoda (52.6) <i>Simplisetia aequisetis</i> (32.6)
	Southern NM, PaP	22.6	Chironomid larvae (32.1) Amphipoda (19.6) <i>Capitella sp.</i> (19.2)	32.6	Chironomid larvae (45.5) <i>Capitella sp.</i> (29.9)	40.5	Chironomid larvae (43.8) <i>Capitella sp.</i> (31.1) Amphipoda (13.4)	30.0	Chironomid larvae (59.5) <i>Capitella sp.</i> (21.3) Amphipoda (11.5)
South Lagoon	Villa de Yumpa	81.5	Chironomid larvae (67.4)	69.0	Chironomid larvae (81.8)	37.3	Chironomid larvae (76.9) <i>Capitella sp.</i> (21.6)	36.3	Chironomid larvae (70.7) Amphipoda (11.6)

**Table 6. Presence/absence of taxa from communities within the Coorong from historical sources (Geddes & Butler 1984; Geddes 1987) and the current report.**

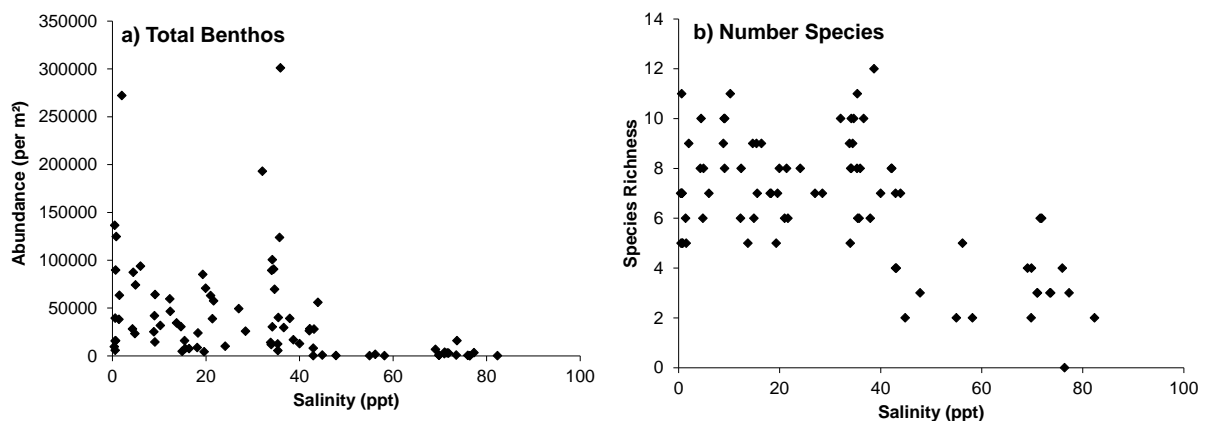
Taxa	Historical communities (Geddes and Butler 1984; Geddes 1987)				Current communities (this report)			
	Freshwater (0 - 2 ppt)	Estuarine (5 - 30 ppt)	Hypermarine (35 - 50 ppt)	Hypersaline (50+ ppt)	Murray Mouth	Northern North Lagoon	Southern North Lagoon	South Lagoon
<i>Simplisetia aequisetis</i>	absent	present			present	present	present	present
<i>Australonereis ehlersi</i>	present	present			present	present	present	absent
<i>Nephtys australiensis</i>	absent	present			present	present	absent	absent
<i>Ficopomatus enigmaticus</i>		present			present	present	present	absent
<i>Capitella sp.</i>	absent		present		present	present	present	present
<i>Arthritica helmsi</i>		present			present	present	absent	absent
<i>Spisula trigonella</i>	present	present			absent	present	present	absent
Hydrobiidae		present	present		present	present	absent	absent
<i>Salinator fragilis</i>	present		present		present	present	absent	absent
Ostracoda				present	present	absent	present	absent
Isopoda				present	absent	absent	absent	absent
Amphipoda		present	present		present	present	present	present
Chironomidae larvae			present	present	present	present	present	present
Ephydriidae Larvae			present	present	present	present	present	present

## 4.4 Relationships between Macroinvertebrate Communities and Environmental Conditions

### **Key Question 2) Which environmental conditions influence the distribution, abundance and community structure of macroinvertebrates?**

Salinity is known to significantly affect the distribution of many species within the Coorong (Kangas & Geddes 1984; Geddes 1987; Dittmann *et al.* 2012; Keuning *et al.* 2012), both along the natural salinity gradients within the Coorong and over time (seasonal and annual variation subject to flows) (Figures 3 and 4). Yet, further environmental factors and life history characteristics of macroinvertebrate species need to be considered for analysing causes of spatial and temporal changes. The distribution ranges for a number of species observed in the Coorong in 2012/2013 (see section 4.3; Key Question 1.f) may be related to salinity gradients.

Salinity alone accounted for 44.8% of the total variation in total abundance of macroinvertebrates across all sites and sampling occasions when tested by DistLM (Pseudo- $F = 49.45$ ;  $p = 0.0001$ ). Peaks in total abundance were observed at low salinities (0-5 ppt; driven by a high abundance of Amphipoda) and marine salinities (35 ppt; driven by a high abundances of Amphipoda, the polychaetes *Simplisetia aequisetis* and *Capitella sp.* and larvae of Chironomidae), with a marked decline in abundances once hypersaline conditions were observed (45+ ppt) (Figure 15a). A similar pattern was observed for the number of species (S) in samples, with a decline in S as salinity increased to hypersaline conditions (Figure 15b). However, because a number of taxa were not identified to species level (e.g. Amphipoda), species numbers indicated here are most likely an underestimate and must be considered with caution.



**Figure 15. Scatter plot of a) total benthos abundance and b) total number of species (macroinvertebrate taxa) versus salinity (x-axis). Each point represents a sampling location and occasion (pooled across replicates) during the 2012/2013 monitoring.**

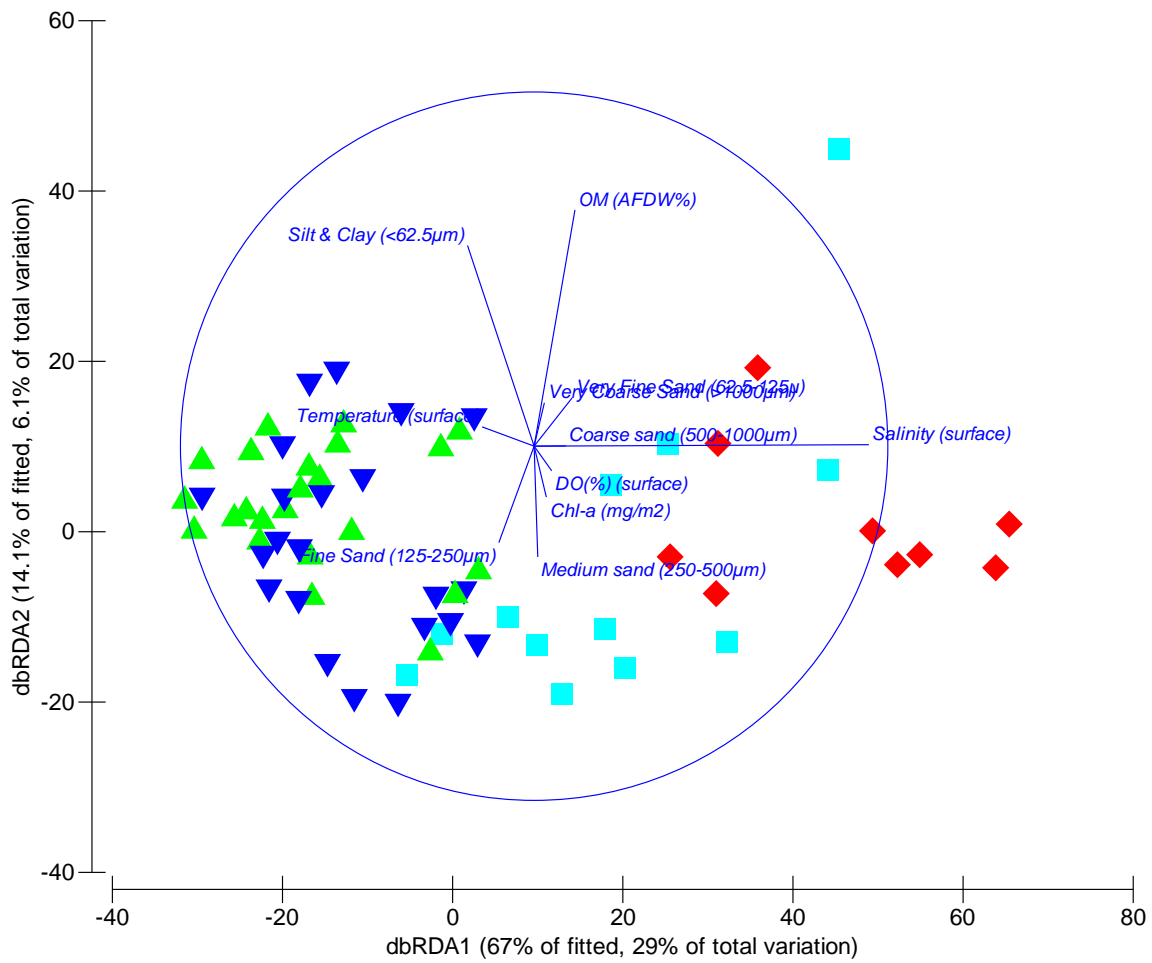
There was a clear separation of macroinvertebrate community structures based on location within the Coorong from the South Lagoon to the Murray Mouth when tested by DistLM (Figure 16). Differences in salinity were significantly correlated with differences in macroinvertebrate community structure (Pseudo- $F = 21.16$ ;  $p = 0.0001$ ), explaining one quarter ( $R^2 = 0.25$ ) of the total variation, and most of the variation in the left-right spread of data points in Figure 16. This indicates that spatial salinity gradients from north to south in the Coorong and Murray Mouth may influence community structure of macroinvertebrates. Vertical spread of these data explained a smaller proportion of the total variation in community structures of macroinvertebrates, and was best explained by sediment characteristics, specifically changes in percentage organic matter (4.4 % of total variation) and the percentage of silt and clay (6.1 % of total variation) (Figure 16). Given the strong natural gradient in the system, the links between environmental conditions and macroinvertebrate communities are further explored per region.

In the Murray Mouth region (Monument Road, Hunters Creek, Ewe Island and Pelican Point) the macroinvertebrate community was significantly influenced by salinity (Pseudo- $F = 4.34$ ;  $p = 0.0053$ ), accounting for 13.8 % of the total variation in community structure within this region. The dbRDA plot shows that the salinity effects occurred on a temporal scale (Figure 17), with salinity changing over subsequent sampling occasions, being highest in this region during March 2013 (Figure 3). This indicates that changes in salinity over the sampling period may influence community structure of macroinvertebrates; however, this effect cannot be separated from changes that may have occurred over time regardless of salinity. The percentage content of organic matter in sediments also explained a further 10.1 % of the total variation in the data, accounting for variation among sites within sampling occasions.

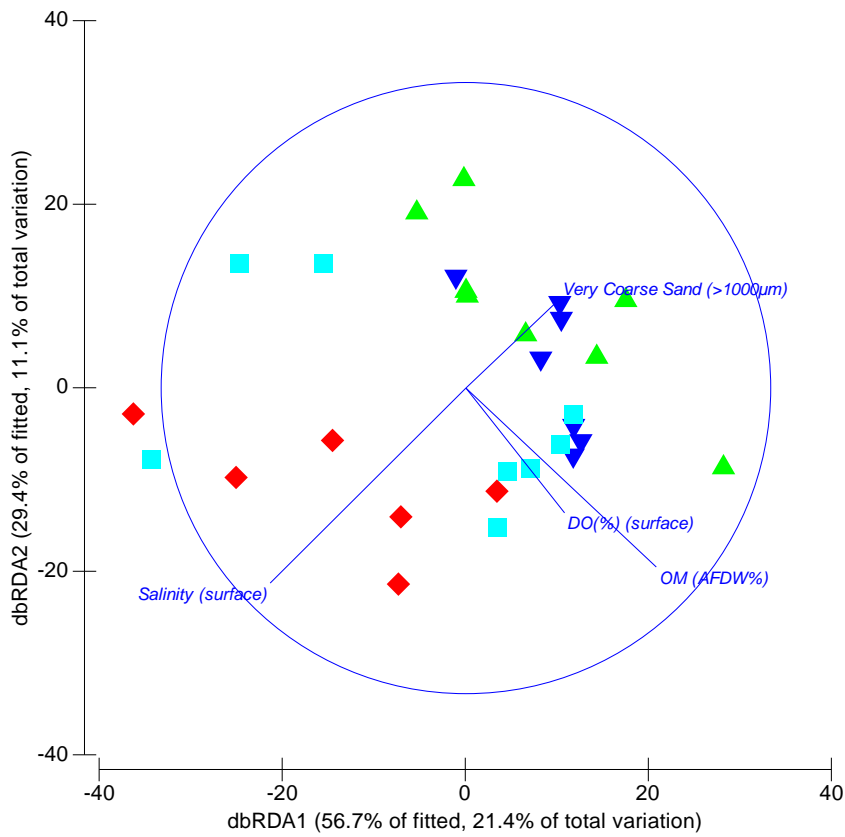
Salinity was also a significant factor for differentiating communities in the North Lagoon (Pseudo- $F = 8.38$ ;  $p = 0.0002$ ), and accounted for over a quarter (26.7%) of the total variation observed in samples. Sampling locations represent a north-south salinity gradient in the North Lagoon from Mark Point to Parnka Point (North) which was reflected by the spread of samples from left to right on the dbRDA plot (Figure 18). A further 17.1% of the variation in the data was accounted for by the percentage content of silt and clay in sediments (9.3%) and the percent content of organic matter in sediments (7.7%), represented by the spread in the data along the y-axis of the dbRDA (Figure 18).

With only two sites sampled in the South Lagoon, salinity differences were less apparent between sampling locations and there was no temporal salinity gradient at Villa de Yumpa (Figure 3). A single sediment characteristic, the percentage of coarse sand in samples, was significantly correlated with macroinvertebrate community structure (Pseudo- $F = 2.75$ ;  $p = 0.032$ ), explaining 28.2% of the total variation observed in the data. The dbRDA plot shows a general separation of samples based on sampling location, with samples from Villa de Yumpa grouping to the top and to the left, and those from Parnka Point south grouping near the bottom and to the right (Figure 19). These sites differed in average value of percentage of coarse sand in sediments, with much lower percentages of coarse sand in sediments at Parnka Point (South) (average = 1.86%) than Villa de Yumpa (average = 5.26%).

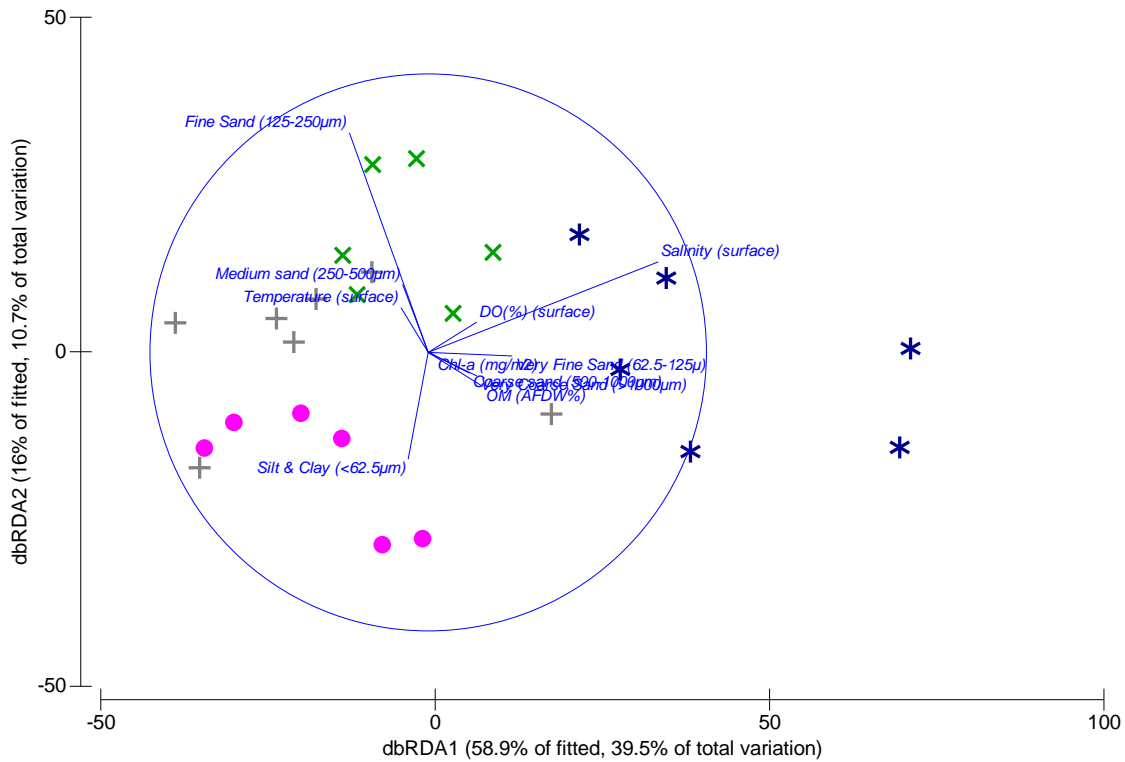




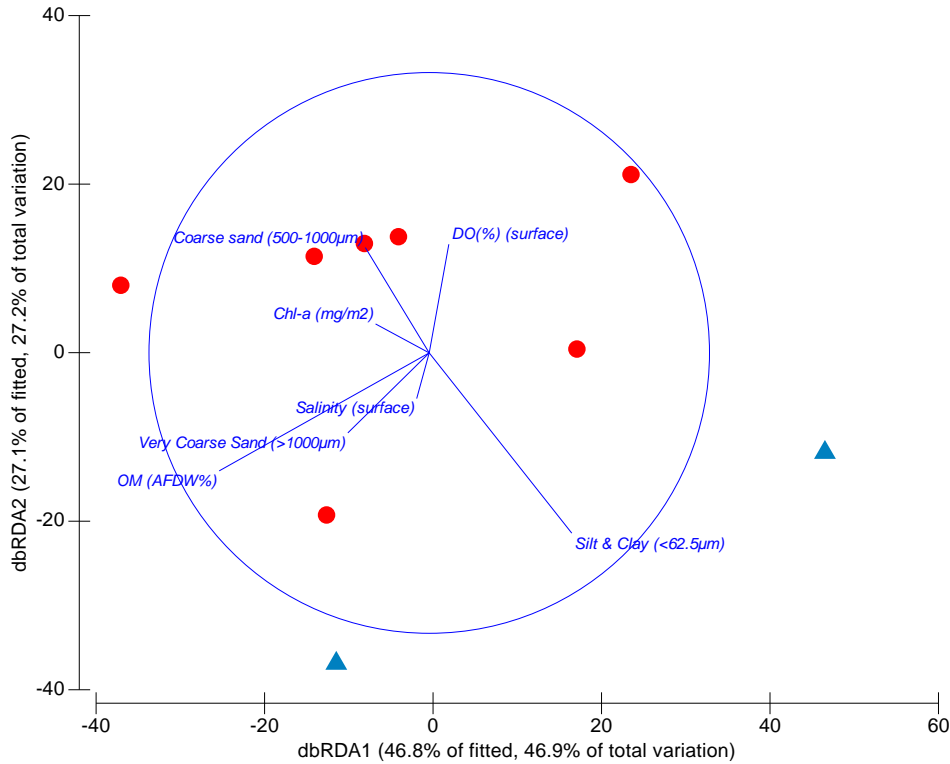
**Figure 16. Distance-based redundancy analysis (dbRDA) plot with vector overlay showing macroinvertebrate community structure for each site and sampling occasion. Each symbol represents the macroinvertebrate community at each site during each sampling occasion (pooled across replicates). Symbols plotted close together represent similar macroinvertebrate communities. Symbols used here represent regions; red diamonds are South Lagoon sites (VdY, PaPS); light blue squares are southern North Lagoon sites (PaPN, NM); blue triangles are northern North Lagoon sites (LP, MP and PP) and green triangles are Murray Mouth sites (MR, EI and HC). Vectors indicate the relative strength and direction of influence of measured environmental variables on macroinvertebrate community structure.**



**Figure 17. dbRDA plot with vector overlay showing macroinvertebrate community structure for the Murray Mouth region across the four sampling occasions. Each symbol represents the macroinvertebrate community at each site during each sampling occasion. Symbols used here represent sampling occasions; green triangles for Sept 2012; blue triangles for Dec 2012; light blue squares for Feb 2013; red diamonds for March 2013.**



**Figure 18.** dbRDA plot with vector overlay showing macroinvertebrate community structure for sites in the North Lagoon for the four sampling occasions. Each symbol represents the macroinvertebrate community at each site during each sampling occasion. Symbols used here represent sampling locations; pink circles for Mark Point; grey crosses for Long Point; green crosses for Noonameena; dark blue asterisk for Parnka Point (North).



**Figure 19.** dbRDA plot with vector overlay showing macroinvertebrate community structure for sites in the South Lagoon for the four sampling occasions. Each symbol represents the macroinvertebrate community at each site during each sampling occasion. Symbols used here represent sampling locations; red circles for Villa de Yumpa; blue triangle for Parnka Point (South).

**Key Question 3) Are there similarities or differences in the community structure of macroinvertebrates across differing flow scenarios?**

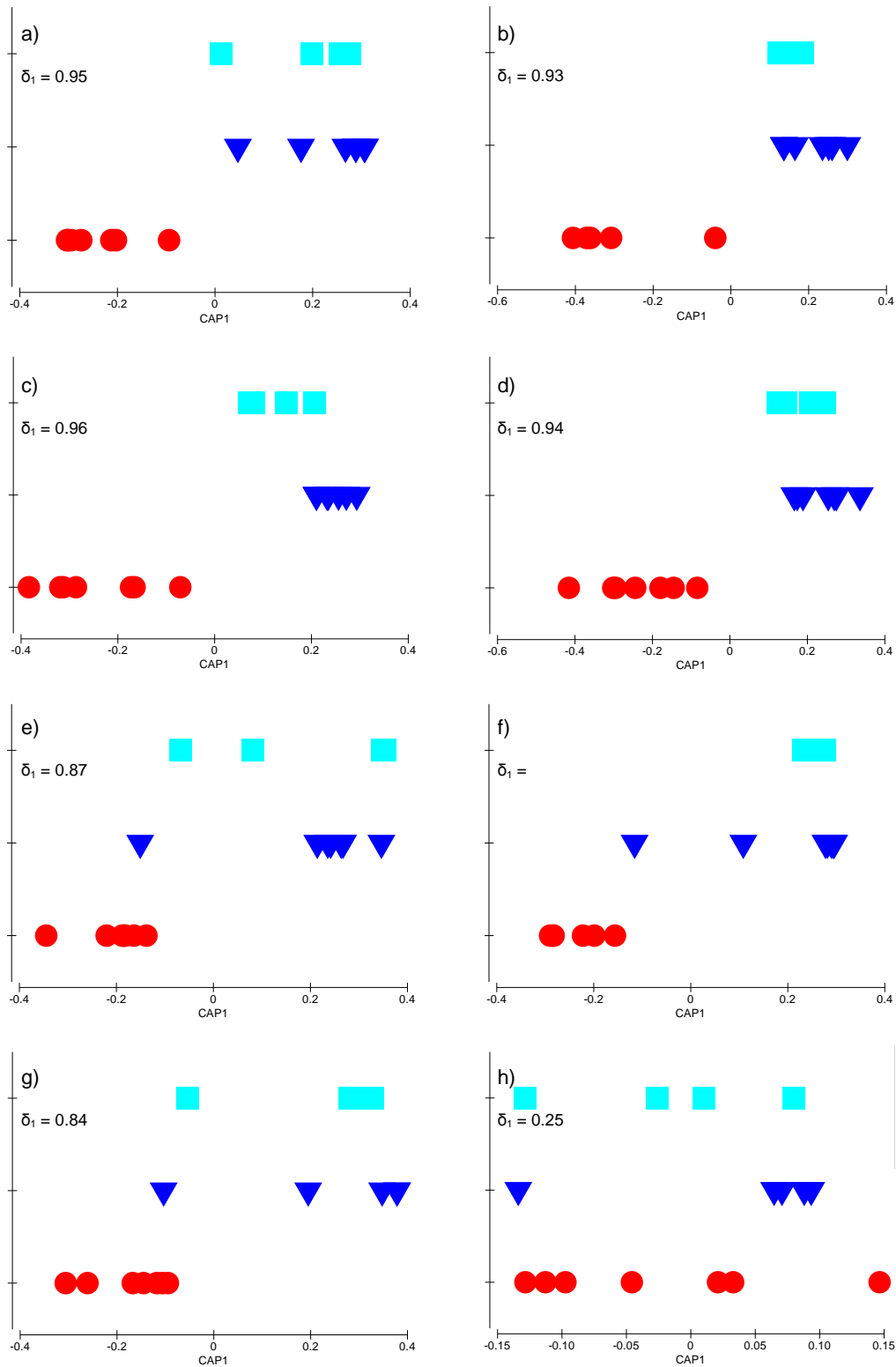
Macroinvertebrate communities at the four sites in the Murray Mouth region exhibited distinct changes between drought and flood scenarios. During drought years, when salinities in the Murray Mouth region were marine or hypersaline (Figure 4), macroinvertebrate communities had a low average similarity of only 35.8%, indicating high variability as a sign of stress or disturbance (Warwick & Clarke 1993). The community was characterised by *Capitella sp.* (an indicator species for disturbance and/or pollution), *Arthritica helmsi* and Amphipoda and *Simplisetia aequisetis* (SIMPER analyses, Table 7). Under flood conditions, when salinities in the Murray Mouth region dropped to fresh or brackish conditions, average similarities were slightly higher, at 45.9%, indicating some reduction in variability. The community was characterised by high abundances of amphipods and chironomid larvae (Table 7). Canonical correlation values ( $\delta_1$ ) from CAP for all sampling locations in the Murray Mouth region are large (Figure 20a-d), indicating a very strong association between macroinvertebrate communities and flow scenarios at these sites. Communities sampled during each sampling event for the 2012/2013 survey were all classified as 'flood' communities, indicating the continued effects of freshwater flows over the barrages generating estuarine conditions in the Murray Mouth region.

In the North Lagoon, differences in macroinvertebrate communities between drought and flood scenarios were also apparent. During drought years, when conditions in the North Lagoon were hypersaline (Figure 4) macroinvertebrate communities in this region showed signs of disturbance, being characterised by *Capitella sp.* and Chironomidae, and a low average similarity among samples of only 19.0%, indicating high variability (Table 7). During flood years, when the North Lagoon returned to marine and estuarine salinities, average similarities within the region increased to 24.8%, but were still low (Table 7). Although communities in the North Lagoon were still characterised by Chironomidae and *Capitella sp.*, Amphipoda also became important (Table 7). Canonical correlation values ( $\delta_1$ ) from CAP were still high for all sampling locations (Figure 20e-g), but not as high as those seen in the Murray Mouth region. This could indicate that although there is still a strong relationship between flow scenarios and macroinvertebrate communities in the North Lagoon, the effects of continued environmental flows on recovery of macroinvertebrate communities may take longer to establish in the North Lagoon, compared to the Murray Mouth region.

Macroinvertebrate communities at the South Lagoon site, Villa de Yumpa, showed only a weak relationship with different flow scenarios. During drought years, when salinities at Villa de Yumpa reached over 150 ppt (Figure 4), average similarity among samples was low (28%) and communities were characterised only by the larvae of the halophylic Chironomidae (Table 7). Under flood conditions, salinities at this site dropped to around 70 ppt (Figure 4), but average similarity increased only slightly to 29.9%, and communities were characterised by Chironomidae and Amphipoda (Table 7). The canonical correlation value ( $\delta_1$ ) from CAP for samples collected at Villa de Yumpa was very low and there was a general overlap of samples collected under different flow scenarios (Figure 20h), indicating that the effects of environmental flows into the Coorong may take longer to manifest at Villa de Yumpa.

**Table 7. SIMPER results showing average similarities (Ave. Sim.) among samples for sites and regions of the Murray Mouth and Coorong over several flow scenarios. 'Drought' samples were collected between June 2004 and December 2009 as part of the annual TLM monitoring (Dittmann *et al.* 2012), 'Flood' refers to samples collected between December 2010 and May 2012, with the current monitoring period starting in September 2012 as a separate column. Macroinvertebrate taxa are listed in a decreasing order of contribution to the average similarity among samples within each site/region for each time period.**

Region	Site	Drought		Flood		2012-2013	
		Ave. Sim.	Characteristic species	Ave. Sim.	Characteristic species	Ave. Sim.	Characteristic species
Murray Mouth	Monument Road	37.9	<i>Simplisetia aequisetis</i> <i>Nephtys australiensis</i> <i>Arthritica helmsi</i>	59.4	Amphipoda Chironomidae	52.4	Amphipoda Chironomidae <i>Simplisetia aequisetis</i> <i>Boccardiella limnicola</i>
	Hunters Creek	36.9	<i>Capitella sp.</i> <i>Arthritica helmsi</i> Amphipoda	35.0	Amphipoda Chironomidae	68.8	Amphipoda Chironomidae
	Ewe Island	38.7	Amphipoda <i>Capitella sp.</i> <i>Arthritica helmsi</i> <i>Simplisetia aequisetis</i>	46.0	Amphipoda Chironomidae	62.1	Amphipoda Chironomid Oligochaeta
	Pelican Point	53	<i>Capitella sp.</i>	45.5	Amphipoda Chironomidae <i>Simplisetia aequisetia</i>	54.1	Amphipoda Chironomidae <i>Simplisetia aequisetis</i>
	OVERALL	35.8	<i>Capitella sp.</i> <i>Arthritica helmsi</i> Amphipoda <i>Simplisetia aequisetis</i>	45.9	Amphipoda Chironomidae	55.6	Amphipoda Chironomidae
North Lagoon	Long Point	55.2	<i>Capitella sp.</i>	26.2	Amphipoda <i>Capitella sp.</i> Chironomidae	49.9	<i>Capitella sp.</i> Amphipoda <i>Simplisetia aequisetis</i> Chironomidae
	Noonameena	21.3	Chironomidae <i>Capitella sp.</i>	45.0	<i>Capitella sp.</i> Chironomidae	66.1	<i>Capitella sp.</i> Amphipoda
	Parnka Point	19.6	Chironomidae	25.7	Chironomidae Ostracoda	37.5	Chironomidae Ostracoda
	OVERALL	19.0	Chironomidae <i>Capitella sp.</i>	24.8	Chironomidae Amphipoda <i>Capitella sp.</i>	33.6	Amphipoda Chironomidae <i>Capitella sp.</i>
South Lagoon	Villa de Yumpa	28.0	Chironomidae	29.9	Chironomidae Amphipoda	24.2	Chironomidae



**Figure 20. Canonical analysis of principal coordinates (CAP) plots showing macroinvertebrate communities when sampled at each site during drought years (red circles), flood years (blue triangles) and during the 2012/2013 survey (light blue squares) for each site a) Monument Road; b) Hunters Creek; c) Ewe Island; d) Pelican Point; e) Long Point; f) Noonameena; g) Parnka Point and h) Villa de Yumpa. Corresponding canonical correlations ( $\delta_1$ ) are given for each CAP.**

## 4.6 Invasive Tubeworms (*Ficopomatus enigmaticus*)

### Key Question 4) Do *Ficopomatus enigmaticus* reefs in the Murray Mouth and North Lagoon region exhibit new growth?

Access to *Ficopomatus enigmaticus* reefs was not possible during most sampling occasions because of high water levels in the Murray Mouth and Coorong, thus reefs marked in 2011 could not be re-measured. Yet, observations on the presence of living *F. enigmaticus* were made whenever possible. *F. enigmaticus* reefs were observed to be alive and growing at Monument Road during all sampling occasions (Table 8). Living *F. enigmaticus* in the Murray Mouth region were also found at Hunters Creek and Pelican Point in late summer 2013 (Table 8). *F. enigmaticus* reefs were also seen in the North Lagoon during most surveys at both Mark Point and Long Point, though reefs at this latter site exhibited no signs of new growth. Reef structures were observed at Noonameena, where live tubeworms were also found in macroinvertebrate samples, and at Villa de Yumpa (Table 8).

**Table 8. Presence of living *Ficopomatus enigmaticus* at study sites throughout the Murray Mouth and North and South Lagoons (see Table 2 for site acronyms) for each sampling occasion during the 2012/2013 survey. Grey shading indicates living *F. enigmaticus* were observed at that site at that time. Note: due to high water, reefs were not accessible at all sites, so blank spaces do not necessarily indicate worms were not present. The table is based on a combination of observations from settlement frames, field observations and macroinvertebrate samples.**

Region	Site	Sep-12	Dec-12	Feb-13	Mar-13
MM	MR				
	HC				
	EI				
	PP				
NL	MP				
	LP				
	NM				
	PaPN				
SL	PaPS				
	VdY				

During December 2012, *Ficopomatus enigmaticus* settlement was not observed on any of the 8 tiles deployed at Pelican Point and only one tubeworm was present on one of eight tiles deployed at Long Point. No tiles were collected from Monument Road or Mundoo Channel during December 2012. By February 2013, all tiles deployed at Monument Road and Mundoo Channel were colonised by *F. enigmaticus*, with thick mats of worm tubes covering between 60% and 100% of the surface of tiles at Monument Road, and between 5% and 70% of the tiles surface at Mundoo Channel. Width of tube openings at Monument Road and Mundoo Channel ranged from 0.1mm to 1mm. During February 2012, settlement was observed on only 2/8 tiles at Long Point and 1/8 tiles at Pelican Point. Higher rates of settlement were observed at Mundoo Channel (8/8 tiles) and Monument Road (4/5 tiles). Settlement of tubeworms occurred only at Monument Road and Mundoo Channel in March 2013, and the lower rates of colonisation can be due to the shorter deployment time of the tiles in the field.

These records indicate that *Ficopomatus enigmaticus* is thriving around the Murray Mouth, with live reefs and high rates of larval settlement observed at Monument Road throughout the 2012/2013 sampling period. Observation of living reefs, settlement of larvae and individuals in samples at other Murray Mouth sites (Hunters Creek, Mundoo Channel and Pelican Point) also indicate that populations of this species in this region are healthy. Populations in the North Lagoon do not appear to be thriving to the same degree as those around the Murray Mouth, with few signs of reef growth and low rates of larval settlement observed at Long Point. However, reefs in deeper sections of the channel in the North Lagoon could be doing well, but are methodically out of reach. This survey was designed to determine the current distribution and growth of tube worm reefs, but cannot explain why this species, which has a wide tolerance towards a range of environmental conditions (Dittmann *et al.* 2009)

appears to be currently doing well in the Murray Mouth region. Recent molecular analysis of *F. enigmaticus* has revealed the co-occurrence of genetically different species in other estuaries around southern Australia (C. Styan and E. Kupriyanova, personal communication), which could explain varied responses to environmental conditions.

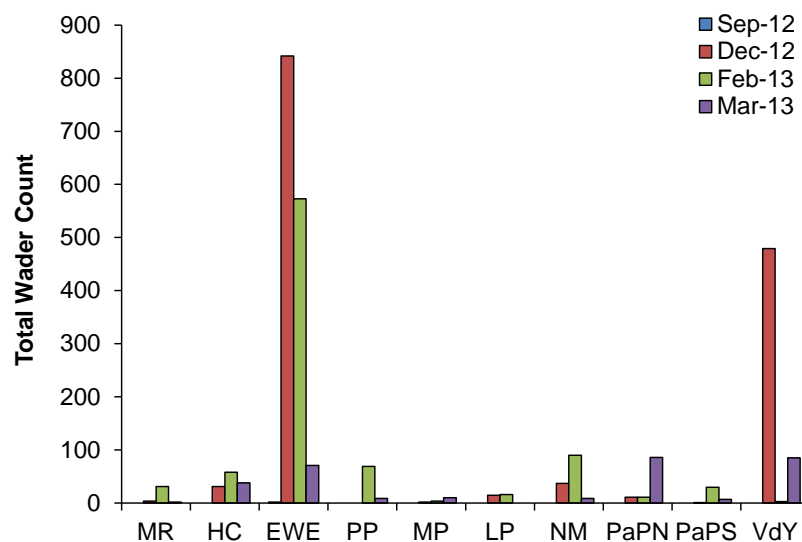
#### 4.5 Shorebird use of mudflats

**Key Question 5) Is there a relationship between diversity, abundance and habitat use of shorebirds and the availability of their macroinvertebrate food source at study sites?**

Shorebirds (waders) were observed at study sites several days prior to macroinvertebrate sampling, and over 2600 birds were counted. Eleven species were recorded, yet abundances were dominated by red-necked stints (1977 individuals) and sharp-tailed sandpiper (248 individuals). Other shorebirds occurred in lower abundance or only as occasional sightings (Table 9). Wader numbers were low in September at the start of the over-wintering period, but increased over the summer months as migratory species arrived in the area, before declining in March as birds started to migrate north (Table 9). Highest counts of waders were at Ewe Island and also Villa de Yumpa (Figure 21),

**Table 9. Species list and total abundance of each shorebird species observed during one day of sampling in each survey month. Bill indicates bill length or foraging depth, with s=short, m=medium and l=long.**

Common Name	Scientific Name	Bill	Sep-12	Dec-12	Feb-13	Mar-13	Total
Red-necked Stint	<i>Calidris ruficollis</i>	s	0	1197	626	154	1977
Sharp-tailed Sandpiper	<i>Calidris acuminata</i>	s	0	85	160	3	248
Red-capped Plover	<i>Charadrius ruficapillus</i>	s	0	71	31	40	142
Common Greenshank	<i>Tringa nebularia</i>	m	0	11	8	67	86
Red-necked Avocet	<i>Recurvirostra novaehollandiae</i>	s	2	2	34	29	67
Curlew Sandpiper	<i>Calidris ferruginea</i>	m	0	35	14	4	53
Black-winged Stilt	<i>Himantopus himantopus</i>	m	0	0	10	14	24
Red-kneed Dotterel	<i>Erythronyx cinctus</i>	s	0	11	0	6	17
Banded Stilt	<i>Cladorhynchus leucocephalus</i>	m	0	9	0	0	9
Golden Pacific Plover	<i>Pluvialis fulva</i>	s	0	0	2	0	2
Sooty Oystercatcher	<i>Haematopus fuliginosus</i>	l	0	1	0	0	1



**Figure 21. Total number of shorebirds counted at the study sites in the Murray Mouth and Coorong (see Figure 2), based on snapshot observations during one day in each survey month from spring 2012 to autumn 2013.**



indicating that shorebirds are using foraging sites throughout the system. Yet, bird counts taken for this study were snapshots and thus lower than in other monitoring efforts (Wainwright & Christie 2008). Between five and eight different shorebird species were recorded in the four surveys from sites in the Murray Mouth and North Lagoon, but less than four just south of Parnka Point. All species were observed to be foraging along the water's edge or in shallow water, and this behaviour was recorded at multiple sites throughout the Murray Mouth and Coorong.

### **Prey availability and shorebird distribution**

Shorebirds are often found where densities of prey in mudflats are highest, yet their intake rate can be independent from prey densities and subject to harvestability (Goss-Custard *et al.* 2006; Geering *et al.* 2007; Finn *et al.* 2008). Patterns in benthic and shorebird distribution and abundance were explored to detect whether shorebirds were attracted to sites with higher food availability. Macroinvertebrate species that are known prey items of shorebirds generally dominated abundances (Figure 22a). Some prey items, such as Amphipoda and Chironomid larvae, occurred at all sites sampled (Figure 22b, c). Other species, such as *Simplisetia aequisetis*, *Capitella sp.* and *Arthritica helmsi* were more restricted in distribution (Figure 22e, f, h, respectively).

*Simplisetia aequisetis* is a preferred prey item of many waders, especially the Curlew Sandpiper (Kent & Day 1983; Dann 1999). This polychaete occurred in relatively high abundances between Monument Road and Long Point, being more abundant at the two ends of this distribution range (Figure 22e). *S. aequisetis* was most abundant at each site in March 2013 (Figure 22e), after shorebirds began migrating away from the Coorong (Figure 21).

Amphipoda, chironomid larvae and *Capitella sp.* are preferred prey items of short-billed waders (Verkuil *et al.* 1993; Hilton *et al.* 2002). Amphipoda and chironomid larvae were collected from all sampling locations, but were most abundant at Pelican Point in September 2012 and March 2013 (Figure 22b), prior to the arrival of migrating shorebirds and as they departed (Figure 21). *Capitella sp.* were most abundant at Long Point and Mark Point, and became more abundant towards March 2013 (Figure 21). Other potential polychaete prey (e.g. *Nephtys australiensis* and *Boccardiella limnicola*) was most abundant at Monument Road in December 2012 and February 2013, and were also collected in large numbers from Pelican Point and Noonameena in February and March 2013 (Figure 22g). Larger polychaetes occurring in greater sediment depths (e.g. *S. aequisetis*, *A. ehlersi* and *Nephtys australiensis*) are accessible for long-billed shorebirds (Kent & Day 1983), whereas smaller species such as *Capitella sp.* are generally accessible to waders with shorter bill lengths (Zwarts & Wanink 1993).

The small bivalve *Arthritica helmsi* (Figure 22h), a prey item preferred by sandpipers (Kent & Day 1983), was abundant at Mark Point but shorebird numbers were low at this site (Figure 23). Other molluscs (*Spisula trigonella*, *Soletellina alba*, Hydrobiidae and *Salinator fragilis*) were collected between Monument Road and Noonameena, and were most abundant at Hunters Creek in February 2013 (Figure 22i).

In the 2012/2013 survey, sites with highest numbers of either shorebirds and highest abundances of macroinvertebrates did not always align (Figure 23). In overwintering grounds, shorebirds can cause seasonal reduction in densities of their preferred prey (Kalejta 1993; Mercier & McNeill 1994; Zharikov & Skilleter 2003). Foraging pressure was high at some sites, such as Ewe Island (Figure 23 and 24), and could mask any relationship between prey availability and shorebird distribution, and tests showed no significant relationships (DistLM, results not shown). Whether depletion of macroinvertebrate prey by intense predation pressure occurred at the study sites in the Murray Mouth and Coorong remains unknown, as a differentiation of cause or consequence for shorebird and benthic distributions and abundances would require experimental investigations.

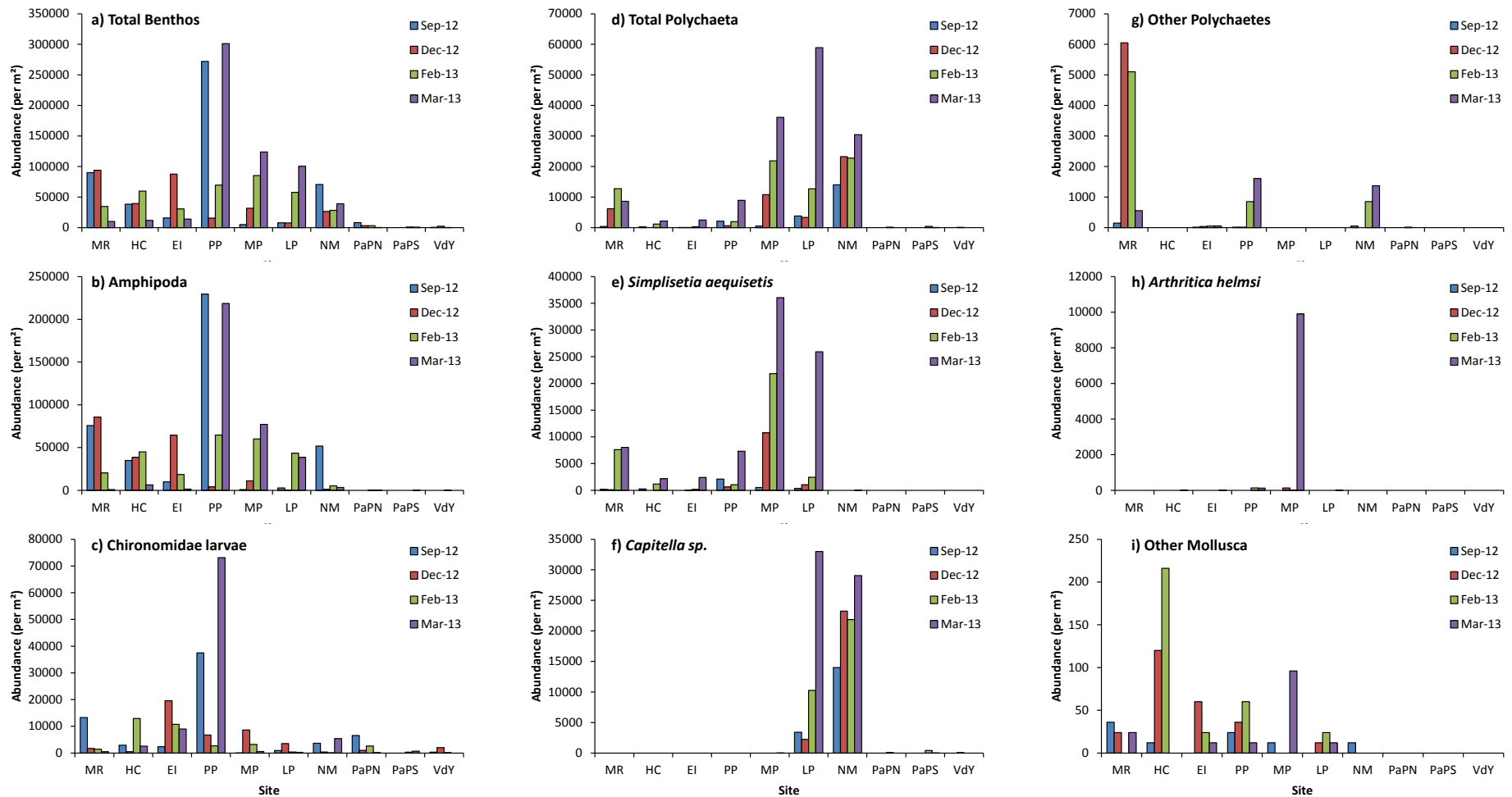
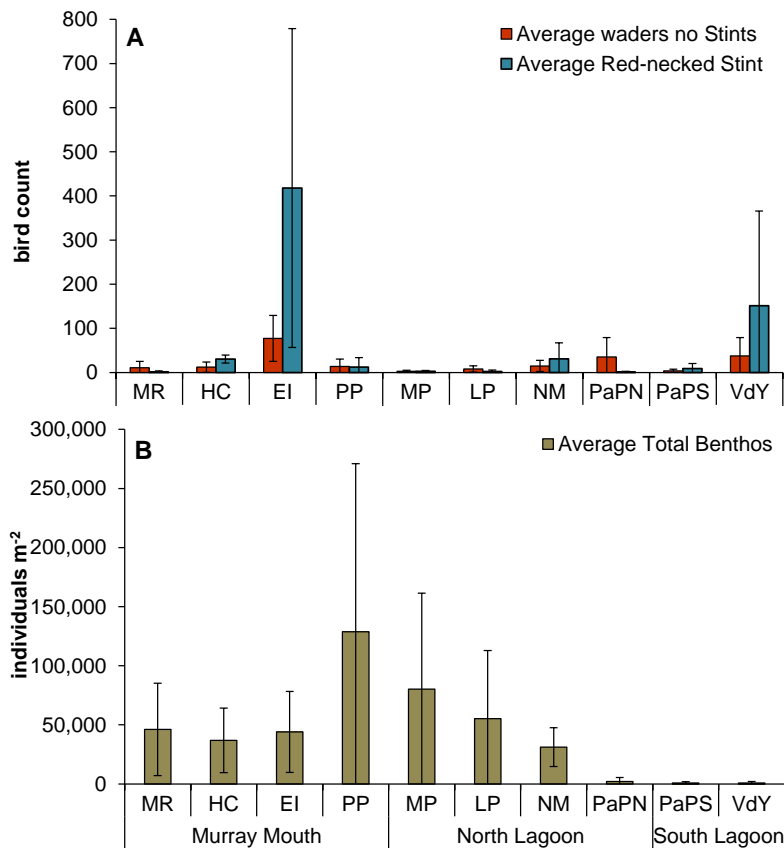


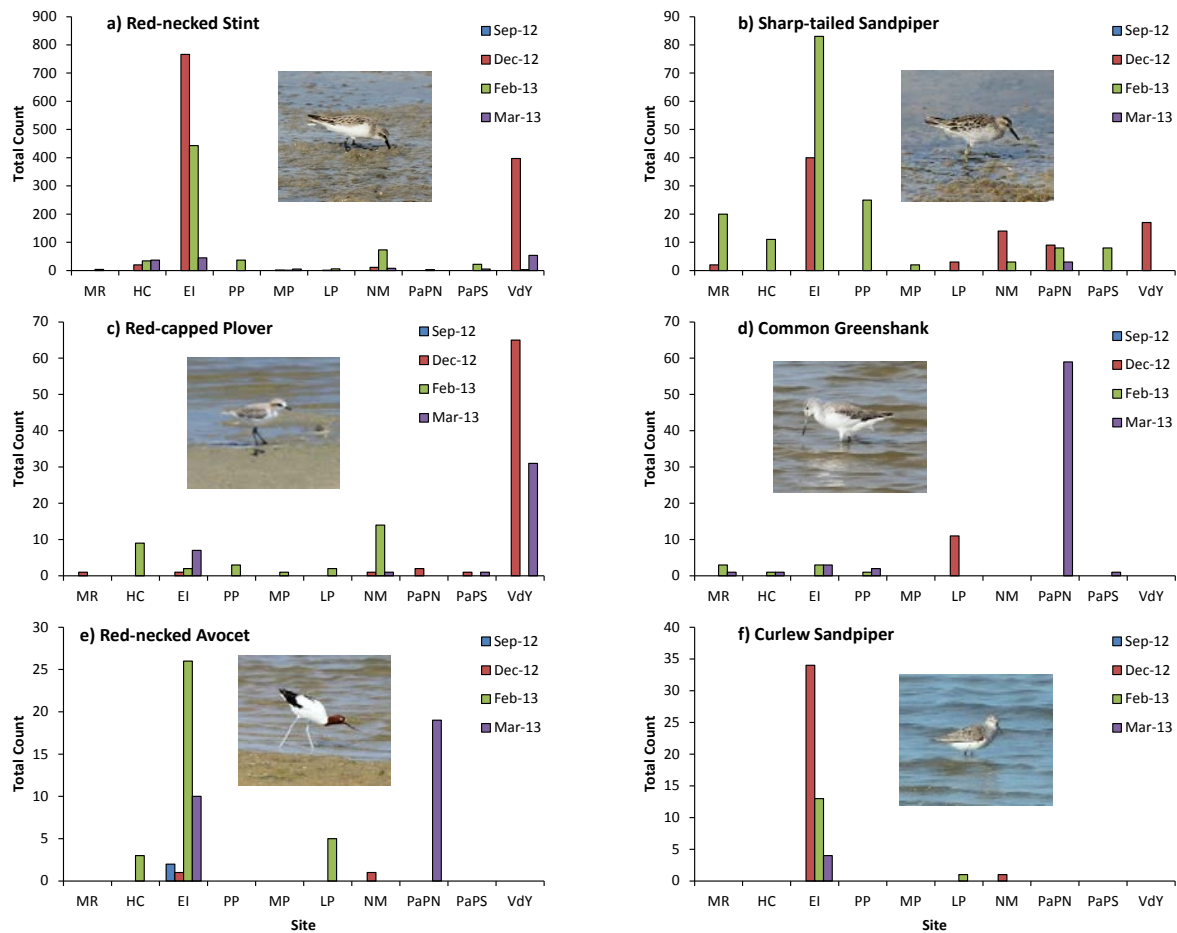
Figure 22. Average abundance during each sampling occasion of a) the total macroinvertebrate community at each site, and that of individual prey items, b) – i).



**Figure 23. Bar-graphs showing averages and standard deviations over three (December, February and March) sampling events in the 2012/13 monitoring for A) shorebirds, with average abundances shown for waders without the most abundant species (Red-necked Stints) and for Red-necked Stints separately; and B) macroinvertebrate abundance. Bird numbers are based on snapshot counts in each survey month. September 2012 was not included as very few shorebirds had arrived.**

Of the six abundant shorebird species (Table 9), the Red-necked Stint, Sharp-tailed Sandpiper, Red-necked Avocet and the Curlew Sandpiper were most abundant at Ewe Island in the December 2012 or February 2013 observations (Figure 24a, b, e, f, respectively). Benthic abundances in sediments at Ewe Island were comparable to adjacent sites in the Murray Mouth in this monitoring period (Figure 22), and food density may not be the only reason for the diverse and abundance shorebird use of this site. Sediment properties can also affect the foraging of shorebirds (Gerritsen & van Heezik 1985), yet the grain size characteristics were similar at Ewe Island to other sites in the Murray Mouth and North Lagoon (Appendix Figure A2).

Two further shorebird species observed in high abundances in the Coorong were the Red-capped Plover and the Common Greenshank, both of which occurred mainly at sites in the southern North Lagoon and South Lagoon. The Red-capped Plover was most abundant at Villa de Yumpa during the December 2012 and March 2013 observations (Figure 24c), while the Common Greenshank was most abundant at Parnka Point (northern sight) in March 2013 (Figure 24d). Few macroinvertebrate species were collected from these locations and in low abundances (Table 4, Figure 22). The Common Greenshank foraging at Parnka Point would have only encountered some amphipods and chironomid larvae. All Red-capped Plover observed at Villa de Yumpa were foraging in wet mud. In February 2013, Red-capped plovers were observed in lower abundances at Hunters Creek and Noonameena (Figure 24c), where they encountered small molluscs and *Capitella sp.* respectively as possible prey items (Figure 22f and I, respectively). Red-capped plover are a short-billed species that prey mostly on small macroinvertebrates that live in surface sediments, such as Amphipoda, *Capitella sp.* and insect larvae, such as Chironomidae.



**Figure 24.** Total counts of the six most abundant shorebird species at each site within the Coorong, based on snapshot observations during one day in each survey month.

### Relationships between bird bill length and macroinvertebrate distributions

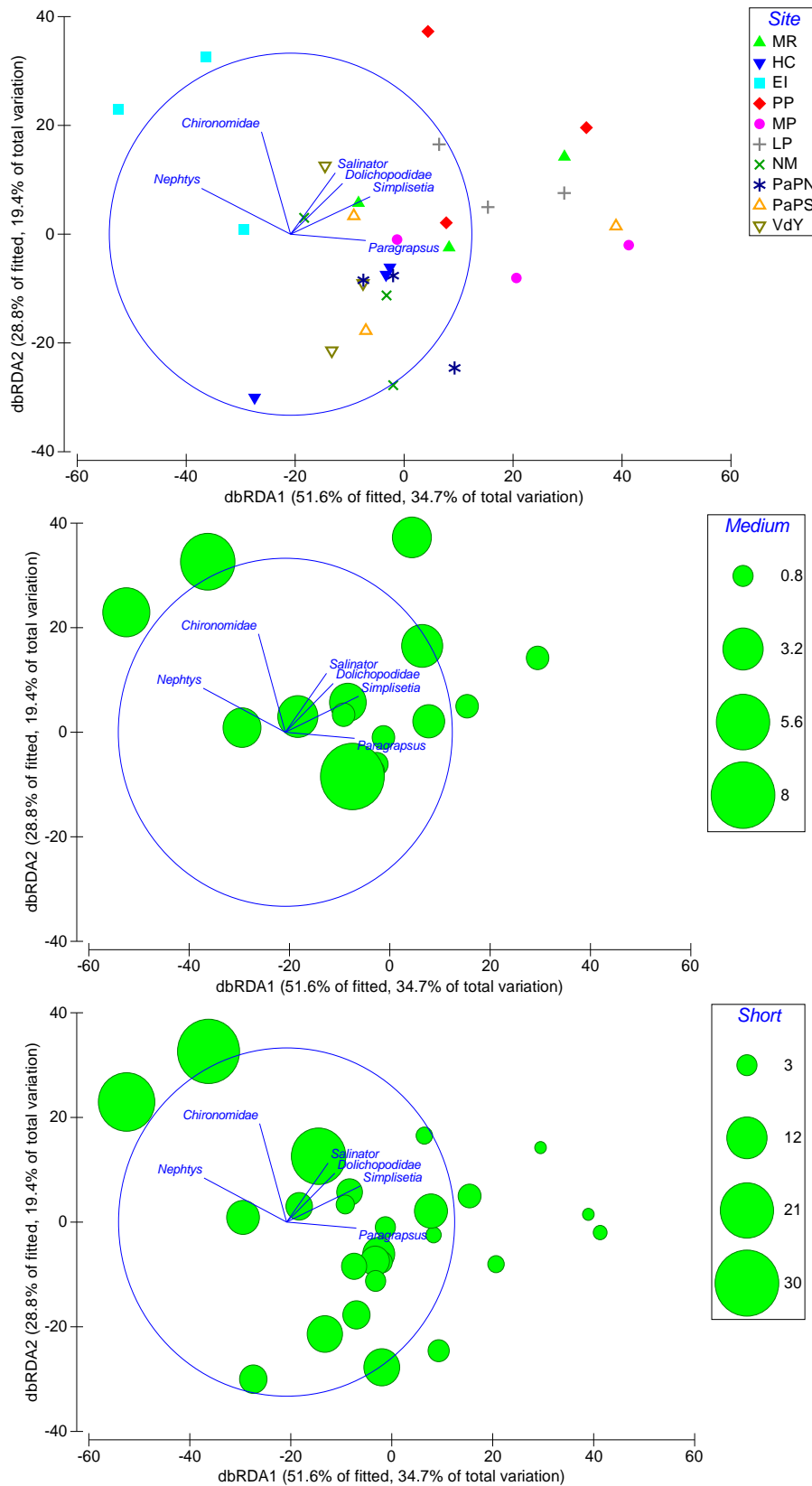
The length of the bill of each bird species will determine what subset of the total macroinvertebrate community is available to that species as a food source (Zwarts & Wanink 1993). Short-billed species, for instance, cannot readily access large polychaetes buried deep in the mudflat, and forage instead on small amphipods, capitellid polychaetes and insect larvae that occur at the surface. Of the eleven species of shorebirds observed in the Murray Mouth and Coorong during the 2012/13 surveys, only the Sooty Oystercatcher is considered 'long-billed' (Table 9). A further four species, the Common Greenshank, Curlew Sandpiper, Black-winged and Banded Stilts, all have medium bill lengths and the remaining six species are short-billed. This includes the Red-necked Avocet, which, although it has a long, curved bill, mainly feeds by sweeping through the water or the surface of sediments (Geering *et al.* 2007).

When the shorebird community was grouped by bill lengths, relationships with macroinvertebrates as predictor variables emerged (Figure 25), yet being only strongest with all 20 macroinvertebrate species considered (DistLM:  $R^2 = 0.67$ ). *Nephtys australiensis*, *Paragrapsus gaimardii* and *Simplisetia aequisetis* contributed significantly to the differentiation of the shorebird distribution. Medium billed shorebirds occurred at sites in the Murray Mouth, especially Pelican Point (Figure 25), whereas short billed shorebirds were more widespread in their occurrence throughout the Coorong and Murray Mouth. Both medium and short billed shorebirds overlapped at Ewe Island, indicating high predation pressure on benthic macroinvertebrates at this site.

### **Relationships between environmental conditions and bird use of habitats**

Most of the shorebirds observed in the Coorong were foraging, with only a few individuals recorded roosting (11 Common Greenshank at Long Point in December 2012; 19 Red-necked Avocet at Parnka Point north in March 2013; and three Sharp-tailed Sandpipers at Long Point in December 2012). The relationship between shorebirds use of habitats and environmental conditions thus mostly reflects presence of foraging birds as recorded during the snapshot observations at the study sites.

The distribution of shorebird species showed little relationship with water chemistry or sediment characteristics at the study sites when tested by DistLM, with only the percentage of silt and clay in sediments ( $R^2 = 0.10$ ;  $p = 0.01$ ) accounting for some of the variation observed in the data. The same results were observed when the two most abundant species were each analysed individually, and there were no relationships in the distribution and abundance of Red-capped Plover (the third most abundant species) and any environmental variables (not shown). Further analysis with more comprehensive shorebird monitoring data may elucidate the relationship between shorebird habitat use and foraging behaviour and macro-invertebrate distributions.



**Figure 25. dbRDA plot of the shorebird assemblage grouped by bill lengths (short, medium and long), based on snapshot counts during three of the sampling occasions (December 2012, February and March 2013), as almost no shorebirds had arrived in September. Benthos data are taken as explanatory variables and the vector overlay (base variables > 0.4) shows macrobenthic taxa contributing to the differentiation of shorebird assemblages. See Table 2 for site name codes. The bubble plot overlays indicate relative abundances of medium and short billed birds at the respective sites.**

## 5 Conclusions

The monitoring of macroinvertebrates over spring/summer 2012/2013 documents further recovery of benthic communities with continued flow, as the communities in the Murray Mouth and North Lagoon became increasingly distinct from those occurring during the drought (Key questions 1 and 3). Yet, recovery at the study sites in the South Lagoon was taking longer. Recruitment of key species was recorded, although at particular sites only for some species, and capture of small-sized polychaetes and bivalves indicated their dispersal capability to recolonise sediments in the study regions. Most of the recruitment occurred in December and February and may be temperature and/or salinity related in response to the flow. Species rarely or not recorded in our previous monitoring were found in this survey period. This included several bivalve species and a sabellid polychaete.

The distribution range of many macro-invertebrate species had extended further south into the North Lagoon, yet some species were recorded at particular sites only (Key question 1). The mudflats in the Murray Mouth were recolonised by benthic organisms, in particular those at Ewe Island and Pelican Point, yet several macroinvertebrate species, such as the bivalves, were only found in sediments of deeper water. General patterns of the response recorded in this monitoring resemble findings after water releases in the 1980s (Geddes & Butler 1984; Geddes 1987).

The recovery of macroinvertebrates concurs with improved environmental conditions, with less hypersaline waters in the Coorong and more estuarine conditions in the Murray Mouth. Salinity, but also sediment characteristics such as the content of organic matter and particular grain size fractions, explained the macroinvertebrate community pattern (Key question 2). Differences between drought and flow conditions were largest for macroinvertebrate communities in the Murray Mouth and North Lagoon, with different species dominating the communities under those conditions (Key question 3). Flow-related salinity emerged as the main driver for these changes.

Tubeworms (*Ficopomatus enigmaticus*) were present throughout the Murray Mouth and North Lagoon, especially at Monument Road and in Mundoo Channel, where they occurred before. Settlement of tubeworms was recorded, thus establishing that the population is reproducing (Key question 4).

The shorebird community recorded was diverse, with two species (Red-necked stints and sharp-tailed sandpiper) dominating shorebird numbers. Shorebirds with medium bill length were most common at Ewe Island and Pelican Point, where larger polychaete prey was available (Key question 5). Short-billed shorebirds occurred throughout, and were common on the mudflats at Ewe Island as well. Assessment of spatial and/or temporal relationships between shorebird and macroinvertebrate communities to fully answer key question 5 requires a larger data set of bird counts than our snapshots observations, and needs to consider both attraction of shorebirds to sites with high harvestable prey availability as well as the effects of foraging, which can cause reduced macroinvertebrate abundances.

As the ecological benefits of restored estuarine character of the Lower Lakes, Murray Mouth and Coorong are becoming evident, continued water release and connectivity between the Lower Murray, the estuarine and lagoon habitats as well as the Southern Ocean will consolidate the recovery of this ecological community recently listed as critically endangered<sup>3</sup>. Further monitoring of macroinvertebrates is needed to document the effects of improved environmental conditions and test whether the ecological character of this iconic estuary has been restored.

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<sup>3</sup> <http://www.environment.gov.au/cgi-bin/sprat/public/publicshowcommunity.pl?id=92>

## 6 Acknowledgements

This monitoring would not have been possible without further helping hands in the field and lab that were provided by George Giatas, Michael Drew, Shea Cameron, Sasha Whitmarsh and Sam Davies. Their assistance in collecting, sorting and identifying samples is greatly appreciated. For longer term comparisons, data obtained through The Living Murray Icon Site Monitoring, funded by the Murray-Darling Basin Authority, were used in this report. The monitoring was funded through the Department of Environment and Natural Resources, and the entire CLLMM team are acknowledged for ongoing discussion and support of the project.

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