

# Seed Bank Assessment of Goolwa Channel, Lower Finniss River and Lower Currency Creek



**Jason Nicol and Rod Ward**

**SARDI Publication No. F2010/000303-1  
SARDI Research Report Series No. 489**

SARDI Aquatic Sciences  
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**August 2010**



**Government of South Australia**  
Department of Environment  
and Natural Resources

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This Publication may be cited as:

Nicol, J. and Ward, R (2010). Seed bank assessment of Goolwa Channel, Lower Finniss River and Lower Currency Creek. South Australian Research and Development Institute (Aquatic Sciences), Adelaide. SARDI Publication No. F2010/000303-1. SARDI Research Report Series No. 489. 39pp.

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Printed in Adelaide: August 2010

SARDI Publication No. F2010/000303-1

SARDI Research Report Series No. 489

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Date: 23 August 2010

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Circulation: Public Domain

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## **Acknowledgments**

The authors thank, Alec Rolston, Susan Gehrig and Luciana Bucater for comments on early drafts of this report, Arron Strawbridge for technical and field support and Pauline Craven for access to the Finnis River through Sterling Downs. This project was funded by the Department of Environment and Natural Resources through the Murray Futures (Lower Lakes Recovery) Program.

## Executive Summary

The aims of this study were to assess the size and floristic composition of the seed bank of Goolwa Channel, the lower Finnis River and lower Currency Creek (using the seedling emergence technique) and investigate the impact of salinity on germination from the seed bank. Sediment samples were collected from +0.6, +0.3 and 0 m AHD from eight sites within the study area (except the Wally's Wharf site, which had steep sides and sediment was only collected from +0.3 and 0 m AHD) that were inundated by the Clayton regulator. The seed bank was subjected to nine salinity regimes: fresh ( $<1,000 \mu\text{S}\cdot\text{cm}^{-1}$ ), 5,000, 10,000, 15,000 and 20,000  $\mu\text{S}\cdot\text{cm}^{-1}$  for the duration of the experiment and 5,000, 10,000, 15,000 and 20,000  $\mu\text{S}\cdot\text{cm}^{-1}$  for six weeks then flushed with freshwater for the remainder of the study. The study ran for 26 weeks.

Seed density ranged from 253 seeds  $\text{m}^{-2}$  in Clayton Bay at 0 m AHD to 14,812 seeds  $\text{m}^{-2}$  in the lower Finnis River at Sterling Downs at +0.6 m with no clear spatial patterns in seed density across the site. A total of 55 taxa (including 26 exotics) emerged from the seed bank and similar to seed density, there were no clear spatial patterns in floristic composition across the site.

Surface water salinity had a significant impact on germination from the seed bank. Based on the floristic composition and number of germinants, two groups of salinity regimes were identified:

- Group 1: Fresh, Continuous 5,000  $\mu\text{S}\cdot\text{cm}^{-1}$ , Flushed 5,000  $\mu\text{S}\cdot\text{cm}^{-1}$ , Flushed 10,000  $\mu\text{S}\cdot\text{cm}^{-1}$ , Flushed 15,000  $\mu\text{S}\cdot\text{cm}^{-1}$  and Flushed 20,000  $\mu\text{S}\cdot\text{cm}^{-1}$
- Group 2: Continuous 10,000  $\mu\text{S}\cdot\text{cm}^{-1}$ , Continuous 15,000  $\mu\text{S}\cdot\text{cm}^{-1}$  and Continuous 20,000  $\mu\text{S}\cdot\text{cm}^{-1}$ .

There was no significant difference in the number or floristic composition of germinants within each group. However, group 1 had a significantly higher number of germinants and the floristic composition that germinated from the seed bank was significantly different than group 2. The differences in floristic composition were driven by significantly higher numbers of *Typha domingensis*, *Sonchus oleraceus*, *Schoenoplectus validus*, *Schoenus* sp., *Lilaeopsis polyantha* and *Aster subulatus* germinating from the seed bank in samples subjected to the salinity regimes in group 1.

The results show that there is a relatively large and species rich seed bank in Goolwa Channel, the lower Finnis River and lower Currency Creek. This has probably contributed to emergent, amphibious and submergent species recruitment in the study area since water levels have been restored by the construction of the Clayton regulator and pumping. For example, the submergent species *Potamogeton pectinatus*, *Myriophyllum salsgineum* and *Vallisneria australis* were present in the seed bank and have colonised areas inundated by the regulator.

Salinities 5,000  $\mu\text{S}\cdot\text{cm}^{-1}$  and below had no impact on germination from the seed bank and six week pulses of higher salinity water followed by flushing also had no impact on germination from the seed bank. Salinities 10,000  $\mu\text{S}\cdot\text{cm}^{-1}$  and higher induced dormancy in some species but six weeks exposure was not sufficient to affect seed viability. Elevated salinity in the study site may have prevented some species present in the seed bank from germinating and recruiting but dormant viable seed from these species may be present in the seed bank.

## 1. Introduction

The seed bank is defined as the reserves of viable seeds (and spores) in and on the soil surface and associated litter (van der Valk and Davis 1976; Thompson and Grime 1979; Roberts 1981; Thompson 1987) and is part of the flora of the system although it is not readily evident (Major and Pyott 1966). The primary role of the seed bank is to ensure the continuation of species after disturbance or natural mortality has killed the extant vegetation; however, population maintenance (Simpson *et al.* 1989) and a reserve of genetic diversity (Templeton and Levin 1979) are also functions of the seed bank. The role of the seed bank in vegetation dynamics varies from system to system. In wetland systems seeds are the only desiccation resistant life cycle stage for the majority of species present; hence, the seed bank plays an important role in the persistence of wetland plant species during unfavourable periods such as drought or elevated salinities (van der Valk and Davis 1978; Keddy and Reznicek 1982; Smith and Kadlec 1983; Leck and Simpson 1987; Thompson 1987; Welling *et al.* 1988; Baker 1989; Leck 1989; Wienhold and van der Valk 1989; Britton and Brock 1994; Wetzal *et al.* 2001; Richter and Stromberg 2005). Therefore, the seed bank contributes to the resilience of a system by providing a local (no reliance on dispersal into the area) source of propagules for recolonisation after disturbance has killed, reduced or significantly changed the extant plant community.

Extended drought and abstraction for domestic, agricultural and industrial purposes in the Murray Darling Basin has resulted in reduced inflows into South Australia since 2007, which in turn has meant flows over Lock and Weir number 1 were insufficient to maintain pool level downstream of the aforementioned structure. Subsequently water levels in Lakes Alexandrina and Albert have fallen to unprecedented lows (<-0.75 m AHD), disconnecting all of the fringing wetland habitats and exposing extensive areas of acid sulfate soils, especially in the reaches of the Finnis River and Currency Creek that were previously connected to Lake Alexandrina (Merry *et al.* 2003; Fitzpatrick *et al.* 2009a; Fitzpatrick *et al.* 2009b). Disconnection and subsequent desiccation of the fringing habitats that historically (pre 2007) contained diverse submergent, emergent and amphibious plant communities (Renfrey *et al.* 1989; Holt *et al.* 2005; Nicol *et al.* 2006) has resulted in a shift to communities dominated by terrestrial species or bare soil (*sensu* Nicol *et al.* 2003; Nicol 2004; Marsland and Nicol 2009).

To mitigate acid sulfate soils in Goolwa Channel and lower reaches of the Finnis River and Currency Creek two regulators were constructed in August 2009 (Figure 1). A high level regulator (sill height 1.8 m AHD) between the mainland and Hindmarsh Island near the township of Clayton and a low level regulator (sill height 1.2 m AHD with a 24 m long spillway

at a height of 0 m AHD) at the mouth of Currency Creek (Figure 1). The Clayton regulator impounds water from the Finnis River and Currency Creek, preventing it from flowing into Lake Alexandrina resulting in higher water levels between the regulator and Goolwa Barrage (Figure 1). The Currency Creek regulator impounds water from Currency Creek when water levels in Goolwa Channel are below 0 m AHD, which was designed to prevent acidic water (and allow it to be treated) from flowing into Goolwa Channel during acidification events (Figure 1). When water levels in Goolwa Channel are above 0 m AHD the spillway is inundated and Goolwa Channel and Currency Creek are partially connected. Furthermore, water was pumped from Lake Alexandrina to raise water levels in Goolwa Channel to +0.7 m AHD in spring 2009, which has inundated the previously desiccated fringing habitats.

Evidence from studies in other wetland systems suggests that hydrological restoration will facilitate establishment of a plant community dominated by amphibious, emergent and submergent species (e.g. Johansson and Nilsson 2002; Mauchamp *et al.* 2002; Nilsson and Johansson 2002; Nicol 2004; Timmermann *et al.* 2006; Hoffmann and Baattrup-Pedersen 2007; Malson and Rydin 2007). However, a source of viable propagules (either the soil seed bank or dispersal into the area) is required for wetland plant species to establish.

The construction of the Clayton Regulator has restored water levels in the Goolwa Channel, lower Finnis River and lower Currency Creek to pre 2007 levels; however, hydrological modelling predicted that surface water electrical conductivity (salinity) throughout much of the study area will exceed 10,000  $\mu\text{S}\cdot\text{cm}^{-1}$  for the first three years. Brock *et al.* (2005) demonstrated that the number of germinants and a diversity of species germinating from seed banks of wetlands in the eastern Murray Darling Basin was significantly lower at salinities above 1,000  $\text{mgL}^{-1}$  TDS ( $\sim 1,500 \mu\text{S}\cdot\text{cm}^{-1}$ ). However, Nielsen *et al.* (2007) showed that short pulses of high salinity followed by flushing with freshwater had no impact on germination from the seed bank. Therefore, the elevated salinity in Goolwa Channel, the lower Finnis River and lower Currency Creek may result in species that are present in the seed bank not recruiting but remaining viable but dormant.

There was no information regarding the seed bank of Goolwa Channel, lower Currency Creek and lower Finnis River and the impact of salinity on germination from the seed bank; therefore, the Department for Environment and Heritage (now the Department of Environment and Natural Resources) commissioned SARDI Aquatic Sciences to undertake an assessment of the seed bank in the aforementioned area. The aims of the study were:

- to determine the size and floristic composition of the Goolwa Channel, lower Finnis River and lower Currency Creek seed bank,

- to examine the impact of different salinities on germination from the seed bank, and
- to investigate the impact of a short pulse of higher salinity water on germination from the seed bank.

## 2. Methods

### 2.1. Field Site

Sediment was collected from eight sites within Goolwa Channel, the lower Finnis River and lower Currency Creek (Figure 1, Appendix 1), in areas that were inundated by the Clayton regulator. Where possible, soil was collected from existing The Living Murray (TLM) (Gehrig *et al.* 2010) or Goolwa Channel (Gehrig and Nicol 2010) vegetation monitoring sites.

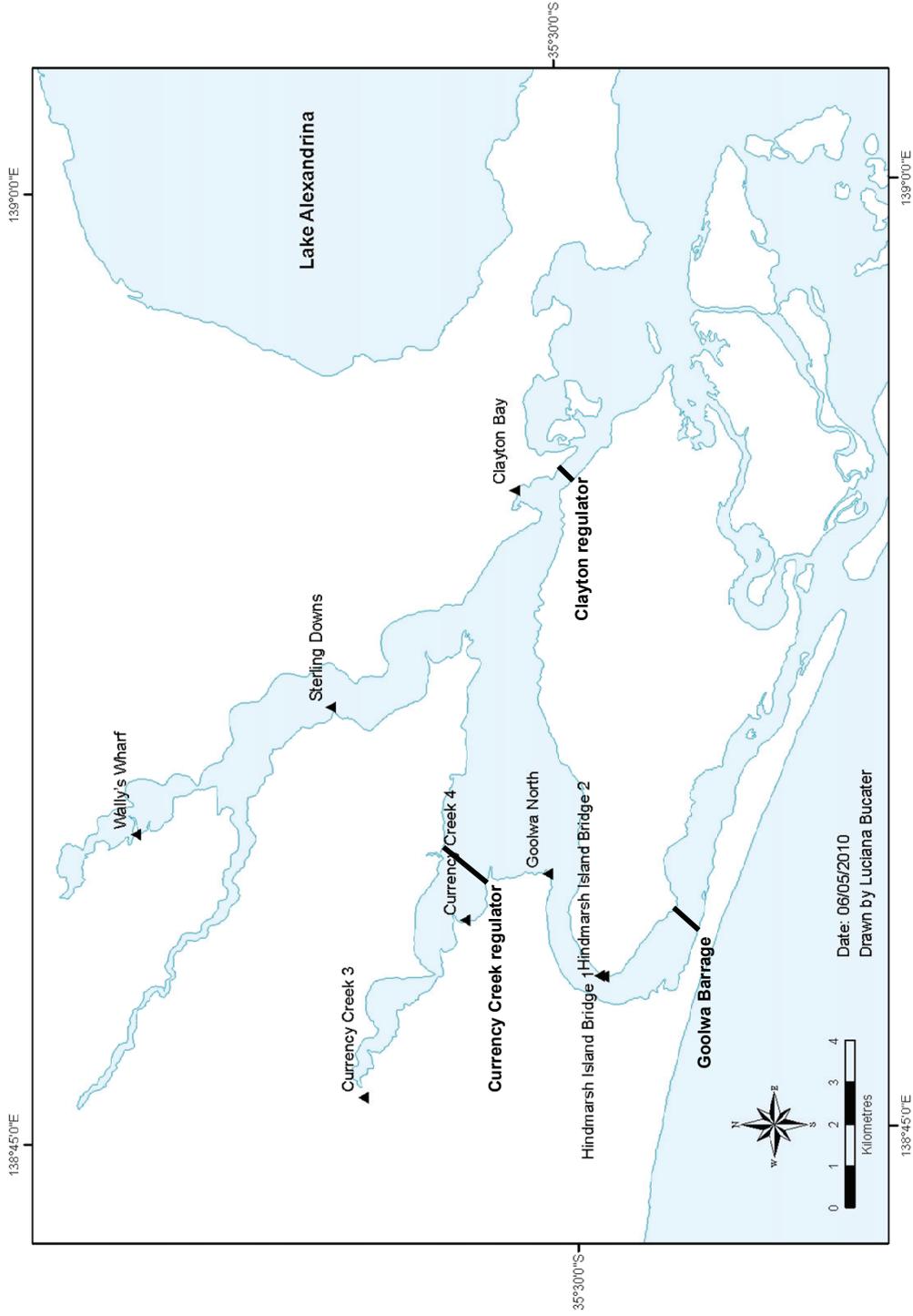


Figure 1: Goolwa Channel, Lower Finniss River and Lower Currency Creek showing sediment sampling locations and regulators.

## 2.2. Sediment Sampling and Pre-treatment

At each location approximately 8 kg of the top 5 cm of sediment (for each elevation) was collected at +0.6, +0.3 and 0 m AHD elevations using a hand trowel (except Wally's Wharf, which had a steeply sloping bank and samples were only able to be taken at +0.3 and 0 m AHD). The sediment was stored in paper bags and transported to South Australian Aquatic Sciences Centre where it was dried at 40° C to a constant weight. Sediment from each site and elevation combination was broken up; gross organic matter (roots and rhizomes) removed and thoroughly mixed to form a homogenous sample for each site and elevation combination. Three subsamples of a known volume were taken from each sample and weighed to calculate bulk density to standardise germination counts to seeds m<sup>-2</sup> (*sensu* Nicol *et al.* 2003).

In addition, sediment pH and electrical conductivity for each site and elevation combination was measured using the 1:5 soil: water extract technique (Rayment and Higginson 1992) (Appendix 2).

## 2.3. Experimental Design and Protocol

To examine the seed bank of each site and elevation and to investigate the impact of different salinities and salinity regimes the seedling emergence technique was used (e.g. Leck and Graveline 1979; Haag 1983; Roberts 1986; Thompson 1987; Poiani and Johnson 1988). A known mass of sediment was spread evenly over a base of 15 cm of sterilised 80:20 sandy loam contained in 225 mm (height) x 150 mm (diameter) nursery bags. Prior to addition of the seed bank, a slow release fertiliser (Osmocote N:P:K=17.4:4.4:8.8 plus micronutrients, Scott's Sierra Horticultural Products, Marysville, OH) was added to the sandy loam to give a loading rate of 100 kg N m<sup>-2</sup> year<sup>-1</sup> to ensure there was no nutrient limitation for plant growth (*sensu* Nicol *et al.* 2003).

Pots were placed in outside tanks (three pots from each site and elevation combination were used in each conductivity and water level combination ( $n=3$ ). Samples were subjected to nine electrical conductivity (salinity) and two water level treatments: <1,000 (fresh), 5,000, 10,000, 15,000 and 20,000  $\mu\text{S}\cdot\text{cm}^{-1}$  for the duration of the experiment and 5,000, 10,000, 15,000 and 20,000  $\mu\text{S}\cdot\text{cm}^{-1}$  for six weeks then flushed and subjected to <1,000  $\mu\text{S}\cdot\text{cm}^{-1}$  for the remainder of the study. The desired conductivity for each treatment was achieved by mixing the appropriate volumes of sea and freshwater in separate tanks then adding it to the experimental tanks. Water level was kept static for the duration of the experiment and pots were placed at +5 cm (the soil

surface 5 cm above the water level) and -20 cm (soil surface 20 cm below the water level). Using two water levels ensured the likely detection of species that require complete submergence or exposure (*sensu* Nicol *et al.* 2007). In addition to the seed bank samples, three control pots containing only sandy loam were placed in each treatment to determine if there was any contamination of the soil or external seed inputs. Water level and conductivity were measured daily and either filled or drained to maintain the correct water level and sea or freshwater added to adjust conductivity. The study ran for 26 weeks between May 7<sup>th</sup> and November 13<sup>th</sup> 2009.

Seedlings present in each pot were counted, identified and removed every two weeks for 20 weeks. Seedlings that could not be identified were transplanted and grown to a stage at which they could be identified.

The number of germinants was converted to seeds m<sup>-2</sup> by calculating the total mass of sediment for 1 m<sup>2</sup> of sediment to a depth of 5 cm in the field using the following equation (Nicol *et al.* 2007):

$$\text{total number of seeds (m}^{-2}\text{)} = \frac{\text{number of germinants (m}^{-2}\text{)} \times \text{mass of sediment (g)}}{\text{sample mass (g)}}$$

#### 2.4. Plant Identification and Nomenclature

Plants were identified using keys in Cunningham *et al.* (1981), Jessop and Tolken (1986), Prescott (1988), Sainty and Jacobs (1981; 2003), Dashorst and Jessop (1998), Romanowski (1998) and Jessop *et al.* (2006). Nomenclature follows Barker *et al.* (2005).

#### 2.5. Data Analysis

Data from the water level treatments for each site and elevation combination were pooled, which enabled comparisons of the entire seed bank (species that require submergence or exposed sediment for germination) between sites and elevations.

The floristic composition and density of the seed bank (fresh salinity treatment only) at each site and elevation was compared by two factor PERMANOVA (Anderson 2001; Anderson and Ter Braak 2003) and NMS ordination (McCune *et al.* 2002), using the package PRIMER version 6.1.12 (Clarke and Gorley 2006).

The impact of salinity on germination from the seed bank (floristic composition and number of germinants) across the study site (data from the different elevations were pooled for each site; hence each site was a replicate) was compared using PERMANOVA (Anderson 2001; Anderson

and Ter Braak 2003), Indicator Species Analysis (Dufrene and Legendre 1997), NMS ordination and group average clustering (McCune *et al.* 2002) using the packages PCOrd version 5.12 (McCune and Mefford 2006) and PRIMER version 6.1.12 (Clarke and Gorley 2006).

Bray-Curtis (1957) similarities were used for all multivariate analyses where species or floristic composition were compared and Euclidean distances were used for PERMANOVA analyses comparing seed density and numbers of germinants.  $\alpha$  for all statistical analyses was 0.05 and was corrected for multiple comparisons using the Bonferroni correction (Quinn and Keogh 2002). Replicates were pooled for NMS ordination and cluster analyses for clarity but all other statistical tests were performed on unpooled data.

### 3. Results

#### 3.1. Seed Density

Seed density across the study site was variable and ranged from 253 ( $\pm$  252) seeds  $m^{-2}$  in Clayton Bay at 0 m AHD to 14,812 ( $\pm$  1,792) seeds  $m^{-2}$  in the lower Finniss River at Sterling Downs at +0.6 m AHD (Figure 2). There were no clear spatial patterns of seed density between elevations or sites (Figure 2), which was confirmed by the significant interaction between site and elevation detected by PERMANOVA (Table 1).

**Table 1:** PERMANOVA *Pseudo-F* statistics comparing seed density between sites and elevations.

<b>Factor</b>	<b>df</b>	<b>Pseudo-F</b>	<b>P</b>
Elevation	2, 46	6.09	0.008
Site	7, 46	17.01	0.001
Elevation x Site	13, 46	2.97	0.004

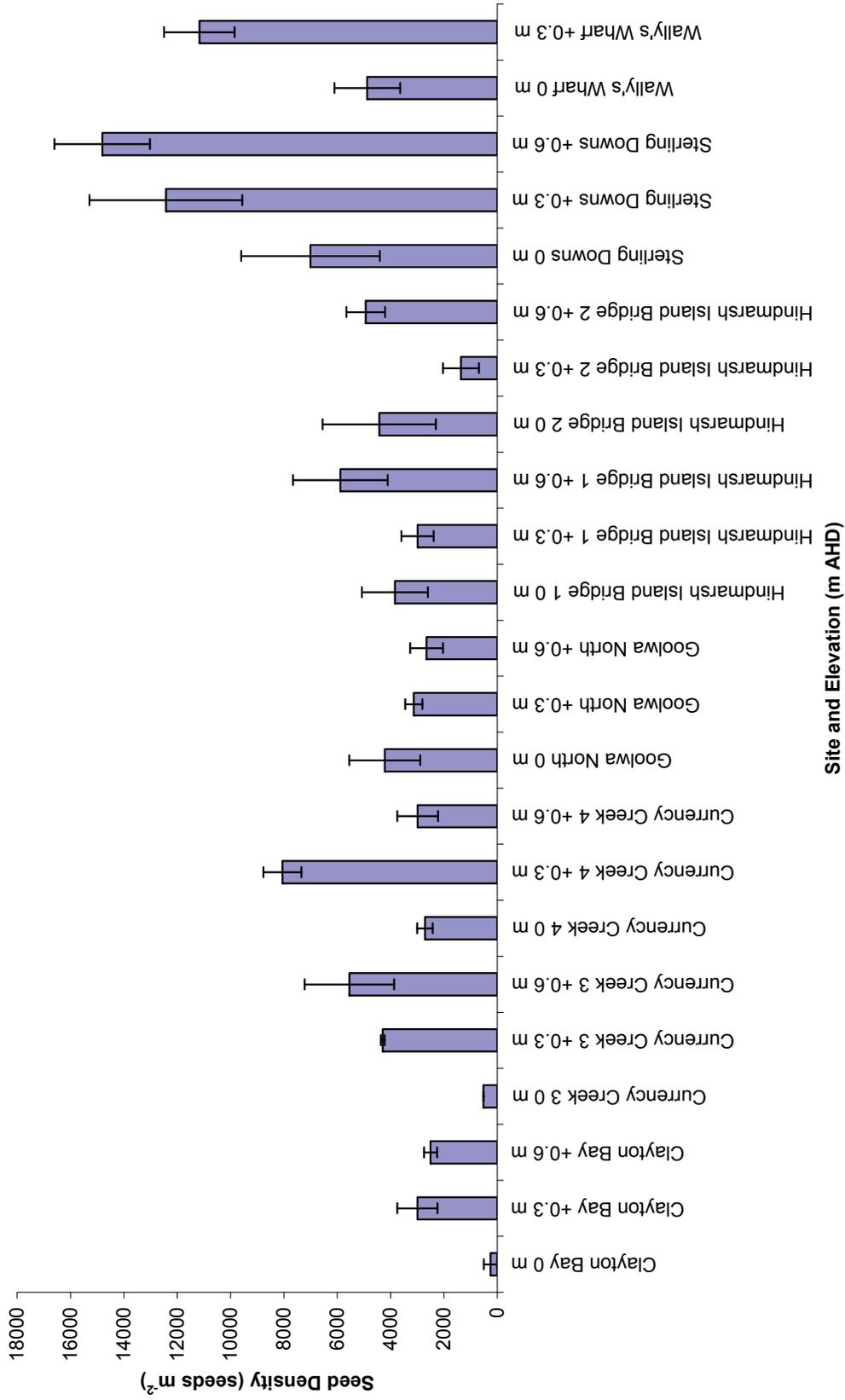


Figure 2: Seed density at each site and elevation (error bars = ± 1 SE).

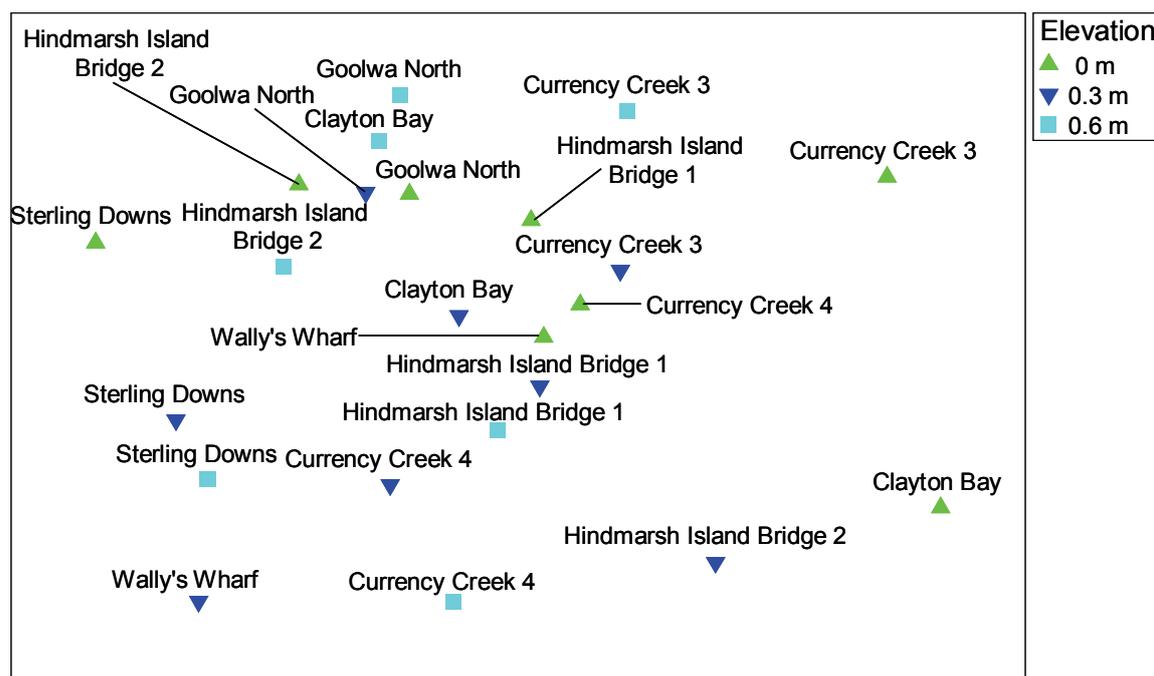
### 3.2. Floristic Composition

A total of 55 taxa including 26 exotics emerged from the Goolwa Channel, lower Finniss River and lower Currency Creek seed bank from all sites and elevations (Appendix 3). The distribution and abundance of species was patchy throughout Goolwa Channel, the lower Finniss River and lower Currency Creek. Across the study area the ten most abundant taxa were *Plantago coronopus* ssp. *coronopus* (1,067 seeds m<sup>-2</sup>), *Schoenus* sp. (534 seeds m<sup>-2</sup>), *Lolium* sp. (343 seeds m<sup>-2</sup>), *Lachnagrostis filiformis* (304 seeds m<sup>-2</sup>), *Sonchus oleraceus* (276 seeds m<sup>-2</sup>), *Crassula helmsii* (195 seeds m<sup>-2</sup>), *Cotula coronopifolia* (187 seeds m<sup>-2</sup>), *Aster subulatus* (184 seeds m<sup>-2</sup>), *Lilaeopsis polyantha* (183 seeds m<sup>-2</sup>) and *Triglochin striatum* (179 seeds m<sup>-2</sup>). Whilst there were high abundances of exotic taxa present in the seed bank (Appendix 3); native submergent, amphibious and emergent taxa were also present (e.g. *Bolboschoenus caldwellii*, *Chara* spp., *Cyperus gymnocaulos*, *Isolepis platycarpa*, *Isolepis producta*, *Juncus kraussii*, *Lycopus australis*, *Myriophyllum caput-medusae*, *Myriophyllum salsugineum*, *Potamogeton crispus*, *Potamogeton pectinatus*, *Ruppia megacarpa*, *Ruppia polycarpa*, *Ruppia tuberosa*, *Schoenoplectus validus*, *Typha domingensis* and *Vallisneria australis* (Appendix 3).

Similar to seed density, there were no clear spatial patterns in floristic composition between sites or elevations (Figure 3) despite the significant interaction between site and elevation detected by PERMANOVA (Table 2).

**Table 2:** PERMANOVA *Pseudo-F* statistics comparing seed bank floristic composition between sites and elevations.

Factor	df	<i>Pseudo-F</i>	<i>P</i>
Elevation	2, 46	2.9932	0.001
Site	7, 46	3.393	0.001
Elevation x Site	13, 46	2.1254	0.001



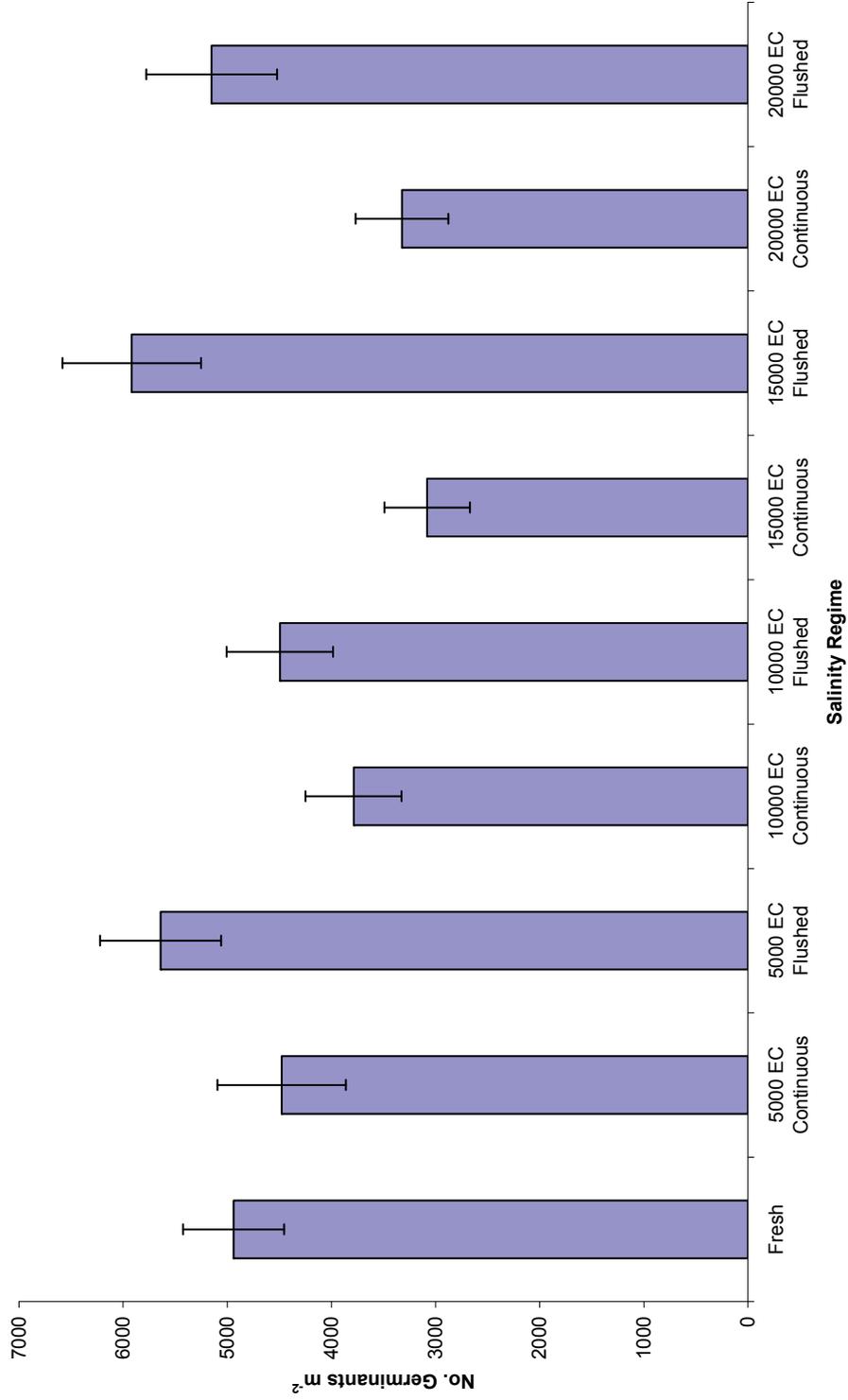
**Figure 3:** NMS ordination of the floristic composition of the seed bank at each site and elevation combination in Goolwa Channel, the lower Finnis River and lower Currency Creek (stress=0.18).

### 3.3. Impact of Salinity on Germination from the Seed Bank

Despite no clear spatial patterns in seed density and seed bank floristic composition in Goolwa Channel, the lower Finnis River and lower Currency Creek there were clear effects of salinity regime on germination from the seed bank. PERMANOVA detected a significant difference in the number of germinants between salinity regimes ( $P_{suedo-F_{8,612}}=3.31$ ,  $P=0.001$ ) (Figure 4). Corrected multiple comparisons showed that there were two groups of treatments:

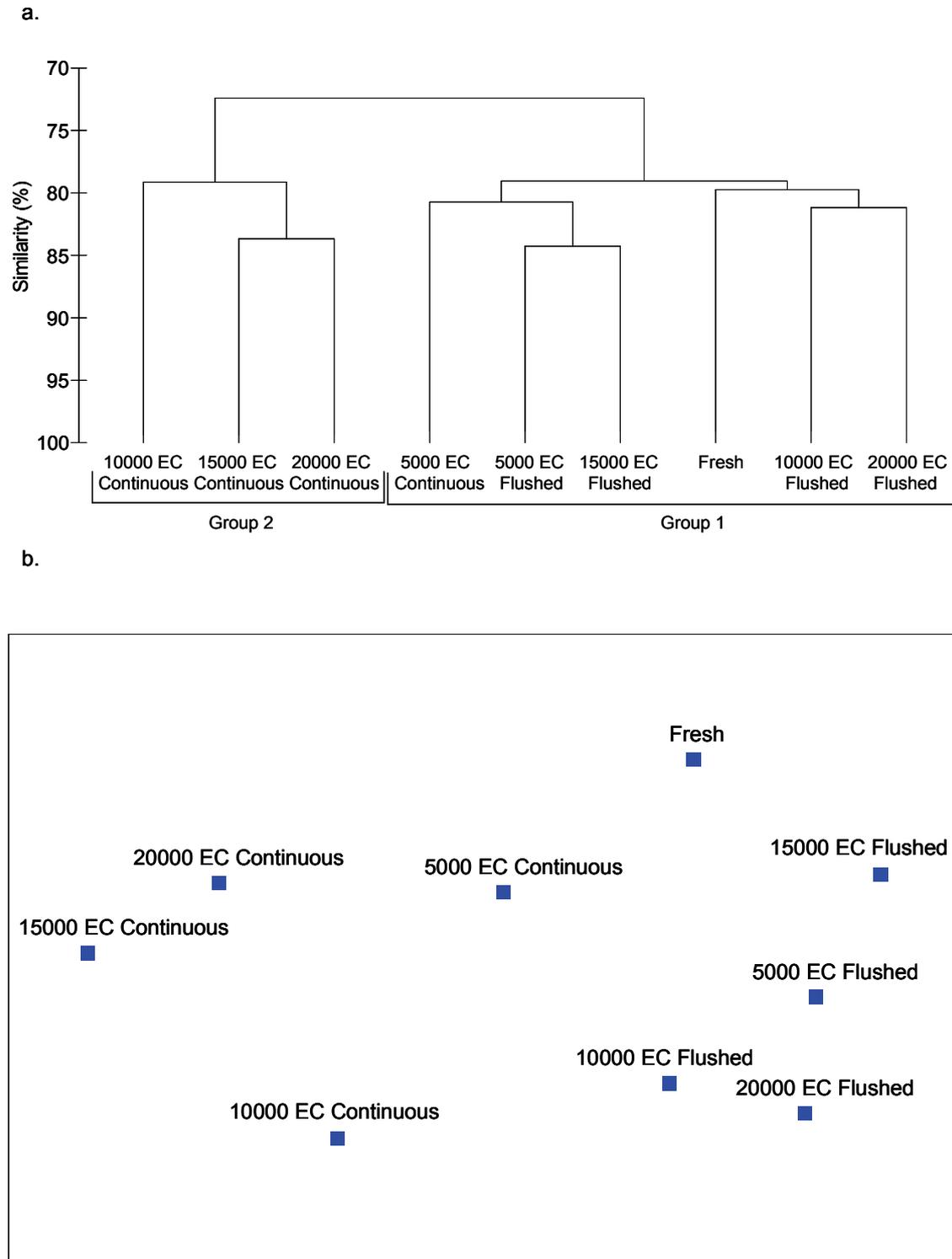
- Group 1: fresh, continuous  $5,000 \mu\text{S}\cdot\text{cm}^{-1}$ , flushed  $5,000 \mu\text{S}\cdot\text{cm}^{-1}$ , flushed  $10,000 \mu\text{S}\cdot\text{cm}^{-1}$ , flushed  $15,000 \mu\text{S}\cdot\text{cm}^{-1}$  and flushed  $20,000 \mu\text{S}\cdot\text{cm}^{-1}$ .
- Group 2: continuous  $10,000 \mu\text{S}\cdot\text{cm}^{-1}$ , continuous  $15,000 \mu\text{S}\cdot\text{cm}^{-1}$  and continuous  $20,000 \mu\text{S}\cdot\text{cm}^{-1}$ .

There were no significant differences in the number of germinants within each group but group 1 had a significantly larger number of germinants than group 2 (Figure 4).



**Figure 4:** Mean number of germinants throughout Goolwa Channel, the lower Finniss River and lower Currency Creek under each of the experimental salinity regimes (error bars =  $\pm 1$  SE).

The floristic composition that germinated from the seed bank under the experimental salinity regimes throughout Goolwa Channel, the lower Finniss River and lower Currency Creek showed the same pattern as the number of germinants (Figure 5). There was a significant difference in the germinable seed bank between salinity treatments (PERMANOVA:  $P_{\text{pseudo-F}_{8,612}}=1.71$ ,  $P=0.001$ ) and corrected multiple comparisons showed there were two groups that corresponded to the groups identified by cluster analysis at a similarity of 75% (Figure 5a). The germinable seed bank was not significantly different within each cluster (group) but there were significantly larger number of germinants (detected by indicator species analysis comparing the two groups) of *Typha domingensis*, *Sonchus oleraceus*, *Schoenoplectus validus*, *Schoenus* sp., *Lilaeopsis polyantha* and *Aster subulatus* in group 1.



**Figure 5:** a. Group average cluster and b. NMS ordination (stress=0.07) of the floristic composition of the germinable seed bank across the Goolwa Channel, the lower Finnis River and lower Currency Creek under the different experimental salinity regimes.

## 4. Discussion and Management Implications

The results show that there is a relatively large and species rich sediment seed bank in Goolwa Channel between the Clayton Regulator and Goolwa Barrage between 0 and +0.6 m AHD. The same submergent species present in the Goolwa Channel seed bank were also recorded in the Dunn's Lagoon and Shadow's Lagoon seed banks (Nicol and Ward 2010), two wetlands adjacent to Goolwa Channel. Only the submergent seed bank was investigated in Dunn's and Shadow's Lagoons; therefore, comparisons of the total seed bank cannot be made. Seed density and species richness is also comparable to natural/semi-natural wetland systems throughout Australia (e.g. Magela Creek Floodplain, NT 3,800-7,100 seeds m<sup>-2</sup> with 33 taxa (Finlayson *et al.* 1990); Menindee Lakes, NSW 500-80,000 seeds m<sup>-2</sup> with 58 taxa (Nicol 2004); Bool Lagoon 20,000-90,000 seeds m<sup>-2</sup> with 31 taxa (Nicol *et al.* 2003). Similar to wetland systems worldwide the seed density and floristic composition is highly spatially variable (e.g. Roberts 1981; Thompson 1987; Baker 1989; Benoit *et al.* 1989; Leck 1989; Thompson 1992; Brock *et al.* 1994; Casanova and Brock 1996; Brock and Rogers 1998; de Winton *et al.* 2000; Stone 2001; Goodson *et al.* 2002; Nicol 2004; Bernhardt *et al.* 2008). There was also no evidence of vertical zonation of species within the seed bank, which has been observed in permanent wetlands (Parker and Leck 1985; Leck and Simpson 1994; Stone 2001).

The size and species richness of the Goolwa Channel, lower Finniss River and lower Currency Creek seed bank has probably been underestimated in this study because the seedling emergence technique was used to assess the seed bank. The seedling emergence technique tends to underestimate the size and species richness of the seed bank compared with seed separation techniques (Benoit *et al.* 1989; Simpson *et al.* 1989; Gross 1990; Brown 1992) because it relies on breaking of dormancy. Seeds may have multiple chilling requirements (Thompson and Grime 1979; Araki and Washitani 2000), lack the proper cues such as light or appropriate redox potential (Froend and McComb 1994; Kalin and Smith 2007), require processes such as the breakdown of fruits by bacterial action (Haag 1979), require passage through the digestive tract of vertebrates (Haag 1979; Anderson *et al.* 2009; Brochet *et al.* 2009) or require multiple wetting and drying cycles (Brock and Rogers 1998) to break dormancy. In addition, viable seeds may be buried deeper than the sampling depth (van der Valk and Davis 1979), varying percentages of viable seeds present from taxa that have evolved persistent seed banks will not germinate in a single season (Thompson and Grime 1979; Thompson 1987; Thompson 1992) and germination from the seed bank has been reported to continue for over three years (Roberts 1981).

Nevertheless, the seedling emergence technique has advantages over seed separation techniques as it only detects viable seeds, is less labour intensive, plants are easier to identify (and can be

grown to flowering stage to make a positive identification), does not discriminate against small seeded species (which are often missed using seed separation techniques) (Roberts 1981; Benoit *et al.* 1989; Simpson *et al.* 1989; Gross 1990; Brown 1992) and the impact of different treatments on emergence from the seed bank can be investigated (*sensu* Brock and Rogers 1998; Nicol *et al.* 2003). Therefore, the seedling emergence technique was the most appropriate method to assess the seed bank for this study.

The presence of a large, species rich seed bank can provide a mechanism for recovery of aquatic plant communities after disturbances such as low water levels (Leck 1989; McKnight 1992; Britton and Brock 1994; Brock and Britton 1995; Vivian-Smith and Handel 1996; Brown and Bedford 1997; Brock and Rogers 1998; Lundholm and Simser 1999; Wetzel *et al.* 2001; Combroux *et al.* 2002; Capers 2003; Leck 2003; Middleton 2003; Weyembergh *et al.* 2004; Adams *et al.* 2005; McKinstry and Anderson 2005; Richter and Stromberg 2005; Robertson and James 2007; Eldridge and Lunt 2010). There is evidence to suggest that the seed bank of Goolwa Channel, the lower Finnis River and lower Currency Creek has provided a mechanism for recolonisation of submergent species. Gehrig *et al.* (2010) and Gehrig and Nicol (2010) recorded *Potamogeton pectinatus*, *Myriophyllum salsugineum*, *Ceratophyllum demersum* and *Vallisneria australis* in areas inundated by the Clayton Regulator, all (except *Ceratophyllum demersum*) were present in the seed bank. In addition, the submergent species *Potamogeton crispus*, *Chara* spp., *Ruppia megacarpa*, *Ruppia polycarpa* and *Ruppia tuberosa* were also present in the seed bank and there is potential for recolonisation of these species in Goolwa Channel, lower Currency Creek and the lower Finnis River.

Whilst the sediment seed bank is important for recolonisation after disturbance it is not the only mechanism for submergent species recolonisation. Many submergent species reproduce primarily via asexual propagules (e.g. Brock 1982; Brock 1983; Brock 1991; Grace 1993; Warwick and Brock 2003; Riis and Sand-Jensen 2006), which can colonise areas by dispersal from refuges. For example, Nicol *et al.* (2007) reported that submergent species were not present in the seed bank of Thegoa Lagoon; however, when the wetland was refilled dense beds of *Vallisneria spiralis*, *Potamogeton tricarlinatus* and *Potamogeton crispus* were observed which had colonised the wetland from upstream areas probably by asexual propagules. Many aquatic species seeds' are also dispersed by water (hydrochory) (e.g. Spence *et al.* 1979; Nilsson *et al.* 1991; Ikeda and Itoh 2001; Lopez 2001; Boedeltje *et al.* 2004; Markwith and Leigh 2008), which may be an important mechanism of recolonisation in the study area because there are several refuge areas in the upper Currency Creek and Finnis River catchments that could act as seed sources (Nicol and Marsland 2010). Aquatic plants are also dispersed by animals (endo or exozoochory) sometimes over very long distances (e.g. Willson 1992; Vivian-Smith and Stiles 1994; Cain *et al.* 2000; Owens *et al.*

2001; Holt Mueller and van der Valk 2002; Figuerola and Green 2004; Figuerola *et al.* 2005; Pollux *et al.* 2005; Green *et al.* 2008; Nathan *et al.* 2008; Casanova and Nicol 2009; de Winton *et al.* 2009) and by the wind (Huiskes *et al.* 1995; Neff and Baldwin 2005; Leyer 2006; Merritt and Wohl 2006; Soons 2006). However, reliance on dispersal into an area for recolonisation after disturbances is a much higher risk strategy than the formation of an in-situ seed bank that can persist through periods that are unfavourable for the survival of adult or juvenile plants.

In addition to submergent plants, the seed bank of the Goolwa Channel, lower Finniss River and lower Currency Creek contained native amphibious and emergent species such as *Bolboschoenus caldwellii*, *Crassula helmsii*, *Cyperus gymnocaulos*, *Isolepis platycarpa*, *Isolepis producta*, *Juncus kraussii*, *Lachnagrostis filiformis*, *Lilaeopsis polyantha*, *Lycopus australis*, *Persicaria lapathifolium*, *Schoenoplectus validus*, *Triglochin striatum* and *Typha domingensis*. The seed bank also provides a mechanism for persistence of these species through drought and recolonisation when conditions become favourable.

The high proportion (47% of taxa) and large numbers of exotic species present in the Goolwa Channel, lower Finniss River and lower Currency Creek seed bank (Appendix 3) is probably the result of adjacent land use. All of the exotic species present (except the amphibious species *Aster subulatus*, *Atriplex prostrata* and *Cotula coronopifolia*) are terrestrial agricultural weeds (Cunningham *et al.* 1981; Jessop and Tolken 1986; Jessop *et al.* 2006) that have probably dispersed into the seed bank from the surrounding agricultural land. In the field, these species are generally restricted to the upper elevations (0.8-0.6 m AHD) and only colonise lower elevations when water levels are low (Marsland and Nicol 2009; Gehrig *et al.* 2010). The exotic amphibious species present in the seed bank are also presently restricted to the upper elevations in Goolwa Channel, lower Currency Creek and the lower Finniss River (Gehrig *et al.* 2010; Gehrig and Nicol 2010). *Aster subulatus*, *Atriplex prostrata* and *Cotula coronopifolia* rapidly colonise areas after water levels are drawn down, their seeds remain viable but dormant whilst submerged (Nicol *et al.* 2003) and as juvenile and adult plants they are restricted to shallow water because they are intolerant of complete submergence for long periods (Sainty and Jacobs 1981; Sainty and Jacobs 2003).

One group of species that was generally absent from the seed bank was the samphire and saltmarsh species (except *Suaeda australis* and *Juncus kraussii*). Two of the dominant samphire shrubs in the Lower Lakes region, *Halosarcia pergranulata* and *Sarcocornia quinqueflora* (Stewart *et al.* 2009), were not present in the seed bank. Little is known about the seed bank dynamics of the aforementioned species, there is evidence from Bool Lagoon that *Sarcocornia quinqueflora* forms a soil seed bank (Nicol *et al.* 2003) but there is no information regarding the seed bank dynamics of *Halosarcia pergranulata*. Why these species were absent from the soil seed bank despite being

common in the extant vegetation of the Lower Lakes (Gehrig *et al.* 2010) and the potential consequences for recolonisation of these species, are unknown. This may be of concern for *Sarcocornia quinqueflora* because it is an important food source of the EPBC listed orange bellied parrot (Phillips and Muller 2006; Mondon *et al.* 2009). Other species that were common in Goolwa Channel, the lower Finniss River and lower Currency Creek that were not detected in the seed bank were *Phragmites australis*, *Paspalum distichum*, *Pennisetum clandestinum*, *Calystegia sepium*, *Azolla filiculoides*, *Isolepis nodosa*, *Juncus usitatus*, *Senecio pterophorus*, *Polygonum aviculare*, *Bromus hordeaceus*, *Bromus diandrus*, *Salix babylonica*, *Mimulus repens* and *Triglochin procerum*. *Salix babylonica* does not reproduce via seed in South Australia (only female plants present and hybridisation is currently prevented due to temporal asynchronisation in flowering times with other taxa) (Cremer *et al.* 1995; Cremer 1995; Cremer 2000; Gehrig 2010), *Pennisetum clandestinum* is a common turf grass and often male sterile clones are grown but can become naturalised (Pessaraki and Pessaraki 2007), *Triglochin procerum* seeds are short-lived in the soil seed bank (Nicol and Ganf 2000) and *Phragmites australis* primarily reproduces asexually (Sainty and Jacobs 1981; Romanowski 1998; Mauchamp *et al.* 2001; Sainty and Jacobs 2003). *Paspalum distichum*, *Azolla filiculoides*, *Juncus usitatus*, *Isolepis nodosa*, *Polygonum aviculare*, *Bromus hordeaceus*, *Bromus diandrus* and *Mimulus repens* (Britton and Brock 1994; Nicol *et al.* 2003; Nicol 2004; Capon 2005; Capon and Brock 2006; Capon 2007; Nicol *et al.* 2007) have been reported to form soil seed banks and it is unknown why they were not detected in this study.

Results from this study show that the seed bank is a mechanism for recovery from disturbance and has probably contributed to recolonisation of submergent, emergent and amphibious species in Goolwa Channel, lower Currency Creek and lower Finniss River after construction of the Clayton Regulator (Gehrig *et al.* 2010; Gehrig and Nicol 2010) but results also show that there is a significant impact of surface water salinity on germination from the seed bank. Constant salinity 10,000  $\mu\text{S}\cdot\text{cm}^{-1}$  and above significantly reduced the number of germinants and species germinating from the seed bank but short pulses (six weeks followed by fresh) of elevated salinity had no significant impact whatsoever. Brock *et al.* (2005) reported a similar decrease in species richness of aquatic plants germinating from the soil seed bank at salinities above 1,000  $\text{mgL}^{-1}$ . Therefore, the species present in the seed bank of Goolwa Channel, lower Currency Creek and lower Finniss River appear to have a higher salinity tolerance for germination than other systems in the eastern Murray-Darling Basin (Nielsen *et al.* 2003; Brock *et al.* 2005; Nielsen *et al.* 2007). However, the elevated soil salinity at some sites (Appendix 2) may have resulted in salt sensitive species that may have been historically present in the seed bank losing viability. In addition, the time taken for *Myriophyllum salsugineum* to germinate from the seed bank (*Myriophyllum salsugineum* germinated under all salinity regimes) increased as salinity increased

(pers. obs.). Sim *et al.* (2006) reported similar results, with the time taken for germination of salt tolerant submergent species in Western Australian salt lakes increased with increasing salinity and concluded that in systems with higher salinities the hydroperiod needs to be longer to enable submergent species to complete their life cycle and replenish the seed bank. *Myriophyllum salsugineum* has been identified as important habitat for threatened small bodied native fish (Bice *et al.* 2008) and with the current elevated salinities in Goolwa Channel between 15,000 and 19,000  $\mu\text{S}\cdot\text{cm}^{-1}$ , water levels will need to remain above 0 m AHD longer to ensure this species can complete its life cycle and replenish the seed bank compared with areas where the surface water salinity is below 10,000  $\mu\text{S}\cdot\text{cm}^{-1}$ .

Short pulses of elevated salinity had no impact on germination from the seed bank (*sensu* Nielsen *et al.* 2007). The species that were significant indicators of group 1 (*Typha domingensis*, *Sonchus oleraceus*, *Schoenoplectus validus*, *Schoenus* sp., *Lilaeopsis polyantha* and *Aster subulatus*) did not germinate in the flushed treatments until after they were flushed with freshwater (pers. obs.) This suggests that salinity induces dormancy but seeds remain viable and can colonise saline areas after the surface water salinity falls to around 5,000  $\mu\text{S}\cdot\text{cm}^{-1}$ ; however, there is no information regarding the longevity of seeds under different salinities.

The submergent species present in Goolwa Channel, the lower Finniss River and lower Currency Creek probably require water levels to remain at or above 0 m AHD to persist. Emergent species are dominant above +0.2 m AHD (Gehrig and Nicol 2010; Gehrig *et al.* 2010); therefore, submergent taxa probably would not be able to colonise these even if permanently inundated. The optimum water regime for Goolwa Channel (and the remainder of the Lower Lakes) to promote aquatic and amphibious plant diversity is probably seasonally fluctuating water levels with high water levels (maximum +0.8 m AHD) in spring and low water levels in autumn (minimum +0.2 m AHD). This will provide permanent areas for submergent vegetation and wetting and drying of the fringes will provide an opportunity for amphibious taxa (e.g. *Persicaria* spp.) that require exposure for seed germination to recruit (*sensu* Nielsen and Chick 1997). Water levels falling below 0 m AHD may result in large beds of submergent species being lost due to desiccation and potential depletion of the seed bank if plants have not set seed.

*Typha domingensis*, *Aster subulatus*, *Schoenoplectus validus* and *Sonchus oleraceus* were common (*Typha domingensis* was the second most abundant species) in Goolwa channel, the lower Finniss River and lower Currency Creek despite surface water salinities exceeding 10,000  $\mu\text{S}\cdot\text{cm}^{-1}$  (Gehrig *et al.* 2010; Gehrig and Nicol 2010). However, all of the *Typha domingensis* and *Schoenoplectus validus* plants had clearly sprouted from rhizomes; *Sonchus oleraceus* was restricted to 0.8 and 0.6 m AHD and only adult plants of *Aster subulatus* were present (Gehrig *et al.* 2010; Gehrig and Nicol 2010).

These results suggest that adult plants and plants sprouting from rhizomes of *Typha domingensis* and *Schoenoplectus validus* have higher salinity tolerances than juveniles because seeds will remain dormant under current salinities in the study site. Similarly, *Aster subulatus* juveniles are probably less salt tolerant than adults because seeds remained dormant but adult plants were present in the field (Gehrig *et al.* 2010). The high elevations (where *Sonchus oleraceus* was restricted to in the field) generally had lower soil salinities than the low elevations (Appendix 2), probably due to flushing by rainfall.

The increase in water levels and reduction in surface water salinity has generally had positive impacts on the plant community (Gehrig *et al.* 2010; Gehrig and Nicol 2010). Nevertheless, salinities were not reduced sufficiently for *Myriophyllum caput-medusae* (a common species in the Lower lakes prior to 2007 (Holt *et al.* 2005; Nicol *et al.* 2006) or *Potamogeton crispus* to recruit. In addition, the elevated salinity prevented the emergent species *Typha domingensis* and *Schoenoplectus validus* from recruiting from seed (although the seed may still be viable). Finally, the construction of a regulator to impound water from the Finnis River and Currency Creek disconnects Goolwa Channel with the remainder of the Lakes. Whilst this is of little consequence for plants, it has a negative impact for animals such as fish that may need to move between Goolwa Channel and the Lower Lakes.

#### 4.1. Key Knowledge Gaps

- Seed longevity of key species under different conditions
- Salinity tolerances and exposure times of the seeds of key species (acute versus chronic exposure)
- Impact of acid on seed viability
- Impact of heavy metals on seed viability
- Seed bank dynamics of samphire species
- Other potential mechanisms of plant colonisation in the Lower Lakes under different conditions
- Germination requirements of key species
- Impact of acid sulfate soil remediation measures on germination from the seed bank
- Impact of sub-lethal salinities on germination, growth and reproductive output of key species
- Hydroperiod required to complete life cycle under different salinities for key species
- Salinity tolerance of different life stages of key species

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## 6. Appendices

**Appendix 1:** GPS coordinates for soil sampling sites (UTM format, WGS84).

Site	Easting	Northing
Clayton Bay	311301	6070626
Currency Creek 3	296772	6074222
Currency Creek 4	301013	6071800
Goolwa North	302130	6069846
Sterling Downs	306108	6074992
Hindmarsh Island Bridge 1	299670	6068521
Hindmarsh Island Bridge 2	299695	6068616
Wally's Wharf	303066	6079631

**Appendix 2:** Soil pH and electrical conductivity values for each site and elevation combination.

Site	Elevation (m AHD)	pH	EC ( $\mu\text{S.cm}^{-1}$ )
Hindmarsh Island Bridge 1	0.6	8.34	4,683
Hindmarsh Island Bridge 1	0.3	7.70	13,650
Hindmarsh Island Bridge 1	0	6.07	18,467
Hindmarsh Island Bridge 2	0.6	5.50	11,017
Hindmarsh Island Bridge 2	0.3	3.06	14,433
Hindmarsh Island Bridge 2	0	8.78	11,633
Currency Creek 3	0.6	5.18	600
Currency Creek 3	0.3	4.39	583
Currency Creek 3	0	3.37	10,283
Currency Creek 4	0.6	4.04	950
Currency Creek 4	0.3	6.58	1,900
Currency Creek 4	0	8.01	13,600
Sterling Downs	0.6	6.95	4,617
Sterling Downs	0.3	5.15	18,783
Sterling Downs	0	3.69	16,800
Clayton Bay	0.6	4.02	500
Clayton Bay	0.3	4.57	1,933
Clayton Bay	0	3.65	9,883
Goolwa North	0.6	8.48	367
Goolwa North	0.3	7.86	300
Goolwa North	0	8.59	950
Wally's Wharf	0.3	3.92	18,500
Wally's Wharf	0	3.88	26,683

**Appendix 3:** Species list of the Goolwa Channel, Lower Finnis River and Lower Currency Creek seed bank from all sites, elevations and water level and salinity treatments (\*denotes exotic species).

<i>Anagallis arvensis</i> *	<i>Myriophyllum caput-medusae</i>
<i>Aster subulatus</i> *	<i>Myriophyllum salsugineum</i>
<i>Atriplex prostrata</i> *	<i>Oenothera stricta</i> *
<i>Bolboschoenus caldwellii</i>	<i>Pennisetum clandestinum</i> *
<i>Bromus diandrus</i> *	<i>Persicaria lapathifolia</i>
<i>Bromus mollis</i> *	<i>Plantago coronopus</i> *
<i>Bromus unioloides</i> *	<i>Poa annua</i> *
<i>Callitriche stagnalis</i> *	<i>Polygonum aviculare</i> *
<i>Centaurea calcitrapa</i> *	<i>Portulaca oleracea</i>
<i>Chara</i> spp.	<i>Potamogeton crispus</i>
<i>Chenopodium album</i> *	<i>Potamogeton pectinatus</i>
<i>Conyza bonariensis</i> *	<i>Ranunculus</i> sp.
<i>Cotula coronopifolia</i> ssp. <i>coronopifolia</i> *	<i>Romulea rosea</i> *
<i>Crassula helmsii</i>	<i>Ruppia megacarpa</i>
<i>Cyperus gymnocaulos</i>	<i>Ruppia polycarpa</i>
<i>Epilobium pallidiflorum</i>	<i>Ruppia tuberosa</i>
<i>Eragrostis curvula</i> *	<i>Schoenoplectus validus</i>
<i>Hypochoeris glabra</i> *	<i>Schoenus</i> sp.
<i>Hypochoeris radicata</i> *	<i>Senecio pterophorus</i> *
<i>Isolepis platycarpa</i>	<i>Sonchus oleraceus</i> *
<i>Isolepis producta</i>	<i>Spergularia marina</i> *
<i>Juncus kraussii</i>	<i>Suaeda australis</i>
<i>Lachnagrostis filiformis</i>	<i>Trifolium</i> spp.*
<i>Lilaeopsis polyantha</i>	<i>Triglochin striatum</i>
<i>Lobelia alata</i>	<i>Typha domingensis</i>
<i>Lolium</i> sp.*	<i>Urtica urens</i> *
<i>Lycopus australis</i>	<i>Vallisneria australis</i>
<i>Medicago</i> spp.*	