Marine Ecosystems

Potential for *Zostera* seagrass recovery and rehabilitation to enhance blue carbon in South Australia. II.

Jason E. Tanner

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July 2022

Report prepared for the Green Adelaide Board Blue Carbon Futures Fund

Government of South Australia
Department of Primary Industries and Regions

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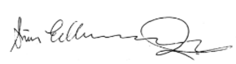
Signed:	
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TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

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EXECUTIVE SUMMARY

The Adelaide coastal waters have experienced a substantial and well documented decline in seagrass cover over the last 50+ years, primarily due to losses of large, long-lived genera (*Amphibolis* and *Posidonia*), which are typically slow to recover. There are also several estuaries along the Adelaide coast which form important habitat for a third group of seagrass: the genus *Zostera*, which is also present in the coastal region, although not as abundantly as the former genera. Little is known about how the cover of this genus has changed over time, or what role it plays in carbon storage, although seagrass meadows of other genera hold some of the largest carbon stores of any habitat. We previously documented a substantial increase in cover of *Zostera* in part of the Port River between 2010 and 2020 (from 1.3-7.9 ha in 2010 to 14-14.3 ha in 2020), using historical aerial photography and newly acquired drone imagery. To assist in determining the potential for rehabilitation to enhance the recovery of *Zostera* in the region, the timing of flowering and fruiting of both *Z. muelleri* and *Z. tasmanica* was also investigated, although these appeared to be rare for both species over both the 2019/20 and 2020/21 putative reproductive seasons. Here, another year of observations are added to the study of reproductive seasonality, although as with previous years, flowering appeared to be patchy, and fruiting almost non-existent. As it does not appear that the use of seeds is a reliable way to restore *Zostera* in the Adelaide region, a small transplantation trial was undertaken. This proved to be highly successful, with both cores and shoots attached to wire grids establishing successfully and greatly increasing in area over 5 months.

Keywords: Blue carbon, Mapping, Restoration, Seagrass, *Zostera.*

1. INTRODUCTION

Seagrasses form important coastal habitats throughout much of the world, providing habitat for a wide range of fish and invertebrates, being sources of high primary production, buffering coastlines against erosion, and providing important stores of blue carbon (e.g. Duarte 2000, Lavery et al. 2019). There have been extensive losses of seagrass meadows worldwide, with 29% of this habitat having been destroyed due to coastal development and other anthropogenic disturbances, and with this decline accelerating (Waycott et al. 2009). South Australia has not been immune to this loss of seagrass, with extensive areas experiencing decline in both Spencer Gulf (Irving 2014) and Gulf St Vincent (Bryars et al. 2008).

Since 1949, there has been a total loss of some 6,200 ha of seagrass from Gulf St Vincent along the Adelaide metropolitan coast, which has been documented through *in situ* sampling and the analysis of aerial photography (Neverauskas 1987a, Shepherd et al. 1989, Hart 1997, Cameron 1999, Cameron 2008, Hart 2013). Much of this loss has occurred in shallow waters, up to ~ 7 m depth, with seagrasses receding seaward, rather than the pattern frequently documented elsewhere of losses due to eutrophication commencing in deep water and proceeding shoreward (Westphalen et al. 2005). Some loss has also occurred within the seagrass meadows, associated particularly with sewage sludge discharges in the 1970s and 80s (Neverauskas 1987b, Shepherd et al. 1989, Bryars and Neverauskas 2004), and more recently, meadow fragmentation is occurring in the shallower remaining seagrasses in more wave exposed areas (Seddon 2002, Fotheringham 2008). The primary causes of loss are generally considered to be the overgrowth of seagrass by epiphytic algae that thrived as a result of anthropogenic nutrient inputs, and to a lesser extent, turbidity associated with stormwater runoff (Fox et al. 2007).

To date, studies of seagrass loss and recovery around Adelaide have focused on the gulf waters, and have not examined local estuaries such as the Port River/Barker Inlet system, West Lakes, The Patawalonga and the Onkaparinga River estuary. All of these enclosed or semi-enclosed bodies of water are potential seagrass habitats, and are highly exposed to human disturbance, either through direct development (e.g. ports), nutrient inputs from stormwater/wastewater, or altered flow regimes. Historically, their importance for seagrasses has been largely ignored, potentially because much of the seagrass was lost well before its importance was understood, and before good quality aerial photography became available, meaning that there is little historical record of what was present. We have incidentally observed that the seagrasses in at least some of these estuaries have expanded considerably over the last 10-15 years, potentially as a result of improved water quality, but as seagrass distribution and cover have not been quantified in any of these estuaries, there are no data currently available to assess the potential extent of this putative recovery.

These enclosed waters are dominated by seagrass in the genus *Zostera*, with *Z. muelleri* occurring intertidally, and the somewhat larger *Z. tasmanica* occurring in the shallow subtidal. There are also Zostera in the shallow waters along the open coast, primarily *Z. tasmanica*. Previously this species was widespread through the deeper waters of Gulf St Vincent, but has been lost over much of this area, probably due to increased turbidity (Tanner 2005).

One increasingly important ecosystem service provided by seagrasses is that they store large amounts of blue carbon (McLeod et al. 2011, Lavery et al. 2013). Continued decline of seagrass habitats therefore has the potential to release large amounts of carbon into the atmosphere. Conversely, recovery and restoration of seagrass habitats can lead to increased carbon storage (Greiner et al. 2016, Serrano et al. 2019). Carbon stored in seagrass habitats also tends to be stored for much longer time periods than that stored in many terrestrial habitats. There has been limited work to understand how well seagrasses in South Australia fit national and global generalisations about the amount of blue carbon that they store. Although early work suggests that they may be low per unit area compared to other regions, the extensive seagrass distribution indicates significant national contribution (Lavery et al. 2019). However, there have only been two samples taken in *Zostera* habitats, which are typical of the shallow estuarine environments in and around Adelaide. Consequently, it is not currently possible to determine the role that any increases in *Zostera* cover will play in blue carbon storage.

The Department for Environment and Water recently released a Blue Carbon Strategy for South Australia, which aims to promote the uptake of projects that lead to increased storage of blue carbon in the state. For this strategy to be effective, it is important that we understand how much carbon is stored in local ecosystems and how recovered/restored habitats compare to habitats that have existed over the longer term. Effective evidence-based strategies for enhancing the extent of these habitats are also required to provide additional options to increase blue carbon storage.

In 2020/21, seagrasses (*Zostera* spp) in the Port River were mapped using drone imagery and historically available aerial photography, with a substantial natural increase in cover in part of the Port River documented between 2010 and 2020 (from 1.3 – 7.9 ha in 2010 to 14 – 14.3 ha in 2020) (Tanner et al. 2021). Sediment carbon stocks in this area ranged from 2.1 to 115.7 tonnes per hectare (T C_{org} ha^{-1}). The average stocks in new (22.9 T C_{org} ha^{-1}) and persistent (52.8 T C_{org} ha^{-1}) *Zostera* patches were lower than the national average for Australian seagrasses (112 T C_{org} ha^{-1}), but higher than the combined seagrass carbon stocks from a previous study on South Australian seagrass blue carbon (16.6 - 31.0 T C_{org} ha^{-1}). To assist in determining the potential for rehabilitation to enhance the recovery of Zostera in the region, the timing of flowering and fruiting of both *Z. muelleri* and *Z. tasmanica* was investigated, although these appeared to be rare for both species over both the 2019/20 and 2020/21 putative reproductive seasons.

Here, we present a continuation and expansion of some of the work in Tanner et al. (2021), to further refine the knowledge and techniques required for any potential future restoration of *Zostera* in the Adelaide region. Reproductive seasonality in both *Z. muelleri* and *Z. tasmanica* is studied further, to assess when mature seeds might be available for seed-based restoration. In addition, small-scale transplantation trials were undertaken to determine the best methods to use.

Figure 1: Map of study area showing major locations referred to in the text, and the transplant site (green dot).

2. METHODS

2.1. Study area

The study's primary area of interest was the intertidal zone of the eastern bank of the Port River between the quarantine station jetty to the north and the southern end of Torrens Island (Figure 1). Tanner et al. (2021) documented *Zostera* recovery spreading south from the quarantine station to slightly north of AGL. Patchy intertidal seagrass at the southern end of this distribution was chosen as the target area for the transplantation trials. To ensure that these trials focussed as much as possible on assessing the transplantation technique, transplants were only moved 1-5 m from the donor site, into an adjacent bare patch (Figure 2). While it is acknowledged that micro-habitat differences may have been responsible for those patches being bare, this was done to minimise the potential for differences in broadscale habitat features to have an effect.

Studies on reproduction focussed on the Onkaparinga, where *Zostera* is easily accessible by foot. Additional observations were made on Torrens Island and at West Beach.

Figure 2: Transplantation site on Torrens Island, showing the recipient site in the foreground and the donor site in the background.

2.2. Transplantation of *Zostera*

Two methods of transplantation were selected (Figure 3), based on reports of success for other *Zostera* species in the literature, with two variations of each. Cores of two different sizes (70 and 88 mm internal diameter) made from PVC pipe were pushed 200 mm deep into the sediment around the edge of an intertidal *Z. muelleri* patch (the donor site), capped and then extracted. As each core was removed, it was immediately transferred to a hole in the chosen bare patch (the recipient site) that was made using the same diameter corer, and extruded from the core tube. In return, the sediment removed from the recipient site was extruded back into the core hole at the donor site. Five replicate cores of each size were transplanted.

The second method involved tying sprigs of *Z. muelleri* onto galvanised wire frames and stapling these to the sediment. Panels were 600 mm x 400 mm, with 100 mm mesh size and 2.5 mm wire. Sprigs were removed from additional cores at the donor site and tied to the wire frames using either plastic cable ties or jute string (Figure 4). A total of 15 sprigs were tied to each frame. Again, five replicate frames of each type were used.

All transplants were established on 22nd September 2021, and qualitatively surveyed on 18th November 2021 and 2nd March 2022. At the same time as the transplants were surveyed, the donor patch was examined to determine if there were obvious signs of holes where the cores had been removed.

Figure 3: Layout of transplants, showing cable tied grids on the left, jute tied grids on the right, and cores in between.

Figure 4: Close-up of (top) jute tied and (bottom) cable-tied grids, showing individual sprigs tied to the corners of the wire frame.

2.3. *Zostera* reproduction

The most successful seagrass restoration program globally relies on harvesting seeds from *Zostera marina* (Tanner et al. 2010, Orth and McGlathery 2012). As an initial step in developing techniques for the restoration of local *Zostera* species, we have surveyed several local populations to assess the timing of seed production. For ease of access, surveys were primarily undertaken in the Onkaparinga estuary, with additional observations at Torrens Island when the transplant experiment was established and monitored. Both intertidal *Z. muelleri*, and subtidal *Z. tasmanica*, were surveyed. A total of 13 surveys were conducted between 15 July 2021 and 18 May 2022 in the Onkaparinga, as well as three surveys on Torrens Island. At both sites, visual surveys were undertaken on foot at low tide to search for the presence of the distinctive spathes, within which the flowers and seeds are contained. Each survey covered at least 500 metres of shoreline, and lasted for at least 30 minutes, unless reproductive spathes were found earlier. In addition, when present, beachcast shoots of *Z. tasmanica* were opportunistically assessed for the presence of reproductive shots during winter surveys for *Amphibolis* seedlings and summer surveys for *Posidonia* fruits at West Beach.

3. RESULTS AND DISCUSSION

3.1. Transplantation of *Zostera*

Both transplantation methods were successful in the short term. All cores and grids were present after two months, although they showed little growth, and some individual sprigs on the grids had perished (Figure 5). After five months, it was no longer possible to distinguish individual cores, and large parts of each grid were obscured, due to extensive growth of seagrass (**Figure 6**). This growth was focused around the transplants, and did not appear to be a result of a general growth of seagrasses into bare patches. At each survey, it was not possible to distinguish where the cores had been extracted from in the donor meadow.

All four techniques trialed were successful, and could potentially be used in future restoration efforts with this species. There was no apparent difference between the two different core sizes, and no apparent difference between the two methods of tying to grids. The grid method was more time consuming to implement than the cores and introduced foreign materials into the environment. Grid transplants, however, cover a larger area than cores. Given the rapid growth of seagrasses in all techniques, the larger area covered per transplant is probably not of great importance.

Figure 5: Transplants after 2 months. Cable-tied grids are at the top of each image, jute-tied at the bottom, and cores in between.

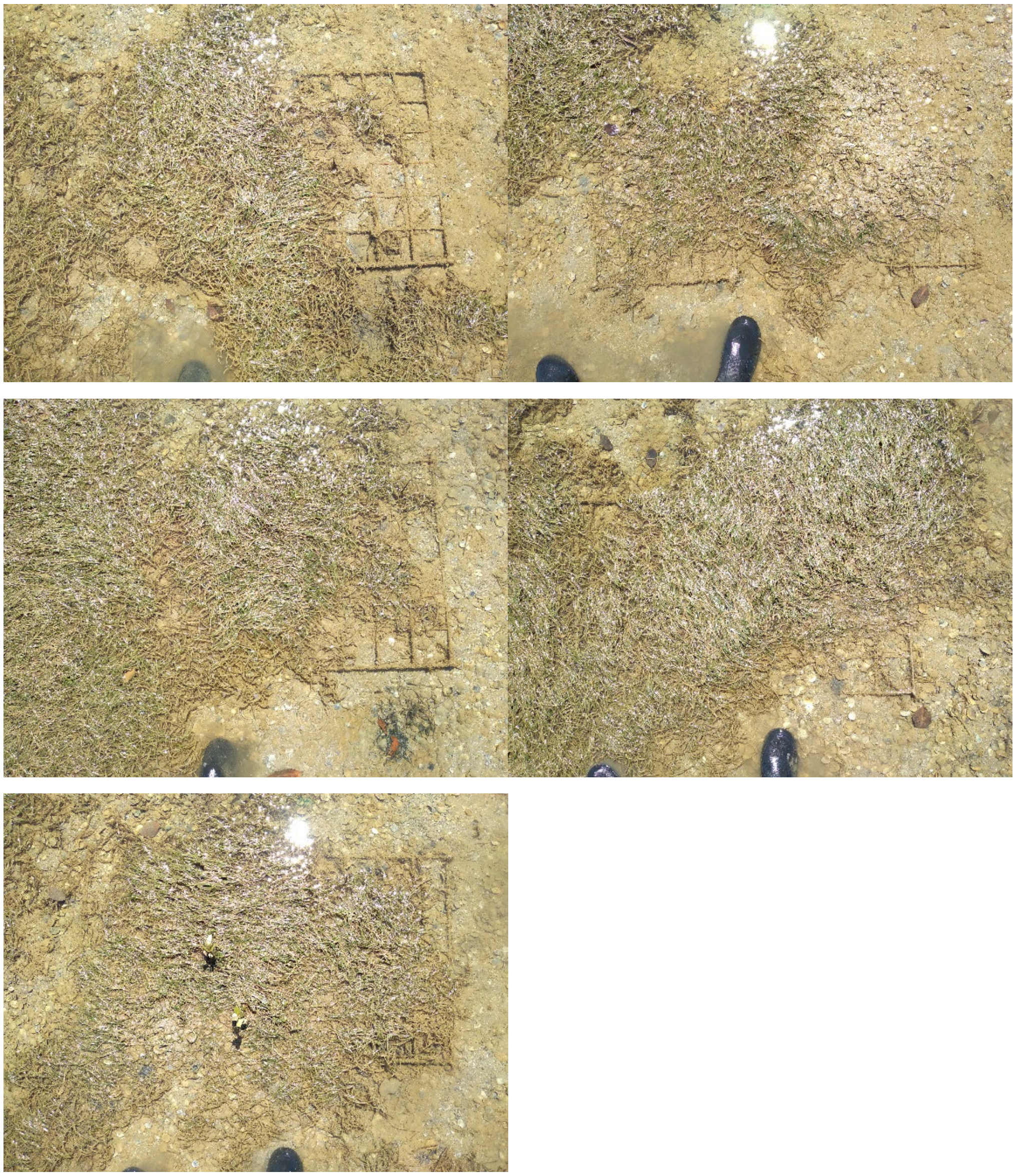

Figure 6: Transplants after 5 months. Cable-tied grids are at the right of each image, jute-tied at the left, and cores in between.

3.2. *Zostera* reproduction

Both *Z. muelleri* and *Z. tasmanica* were found on each survey on Torrens Island and the Onkaparinga (Figure **7**, Table 1). Flowering of *Z. tasmanica* was identified in the Onkaparinga on seven surveys between 7/10/21 and 21/1/22. Reproduction appeared to be absent on surveys in July-September 2021 and February-May 2022. Flower presence in *Z. muelleri* was restricted to the period October-December 2021. Although not quantified, reproductive spathes were noticeably more abundant than in previous years. Despite the observation of flowers, no fruiting was identified in either species at this location. Flowering was observed in *Z. tasmanica* on Torrens Island in September 2021 only, with no fruiting, and no signs of reproduction in

Z. muelleri. Spathes containing flowers and immature fruit were found washed up at West Beach in September 2021, but not at other times of the year.

Reproductive spathes containing flowers were distinct and obvious when present (Figure 8). In all cases when found, spathes were rare, and although not quantified, only appeared to occur at a density of 1-2 per square meter, and in isolated patches. As with Tanner et al. (2021), the current observations do not indicate widespread reproduction, even at the site where spathes were found. Interestingly, previous work in southern Australia has found that species in the *Z. tasmanica* complex flower from August to November, with seeds being present October to December (Parry et al. 2005, Waycott et al. 2014), although there are records of seeds as late as February (Robertson 1984). Thus it was originally expected that surveys early in each year would not detect any reproductive activity in this species. Tanner et al. (2021) speculated that spathes present early in the year were out of season and unlikely to be pollinated. However, the same patterns in flowering have now been documented for another year, with observations in what was previously considered the reproductive season showing no signs of reproduction. Thus. it is possible that shallow subtidal forms of this species have a different reproductive season, or even that it is actually a different species. Interestingly, in both 2020 (Tanner et al. 2021) and 2021, spathes with immature seeds were observed washed up at West Beach in September. These observations support the previously published reports of seeds being present in October to December (Parry et al. 2005, Waycott et al. 2014), and may indicate that the population in open waters off Adelaide is reproductively separate from that in estuarine areas along the same coast.

Conversely, *Z. muelleri* typically flowers from October through to January, with seeds present December to March (Parry et al. 2005, Waycott et al. 2014), although flowering has been documented as early as August (Robertson 1984). Unlike in previous years (Tanner et al. 2021), flowering in this species was observed in the Onkaparinga in late 2021, although again no fruiting was observed. The presence of reproductive activity in this species was much more sparse than for *Z. tasmanica*, and it appears that locally it may be even less reliant on sexual reproduction.

Figure 7: *Zostera* reproductive survey locations.

Table 1: Presence of reproductive spathes in *Zostera* around the Adelaide region. Red cells with 'x' indicate the species was observed but no spathes were found, green cells with '✓' indicate spathes were found, empty cells indicate either no survey at that site, or that the tide was too high to observe *Z. tasmanica*. Data for 17/12/19 to 20/4/21 are from Tanner et al. (2021).

	West Beach	Torrens Island		West Lakes	Onkaparinga	
	Z. tasmanica	*Z. muelleri*	*Z. tasmanica*	*Z. muelleri*	*Z. muelleri*	*Z. tasmanica*
17/12/19		x	x			
5/2/20					x	x
13/2/20		x	x			
25/2/20				x		
5/3/20					x	✓
11/3/20				x		
17/3/20					x	✓
19/3/20		x				
24/3/20					x	✓
7/4/20		x	x			
9/4/20					x	
14/4/20					x	✓
22/4/20					x	✓
4/5/20					x	✓
13/5/20					x	x
16/5/20					x	x
2/9/20					x	x
7/9/20	✓					
21/9/20	✓					
25/9/20	✓					
1/10/20						✓
15/10/20					✓	✓
29/10/20					✓	✓
16/11/20					✓	✓
4/12/20					✓	✓
16/12/20				x		
12/1/21						✓
28/1/21						✓
4/2/21				x		
15/2/21						✓
4/3/21				x	✓	✓
18/3/21						✓
7/4/21						✓
20/4/21					x	x
15/7/21					x	x
26/8/21					x	x
7/9/21				✓		
22/9/21		x	✓			
23/9/21					x	x
7/10/21					✓	✓
8/11/21					✓	✓
18/11/21		x	x			
22/11/21					✓	✓
6/12/21					✓	✓
21/12/21					✓	✓
4/1/21					x	✓
21/1/21					x	✓
17/2/21					x	x
2/3/21		x	x			
17/3/21					x	x
18/5/21					x	x

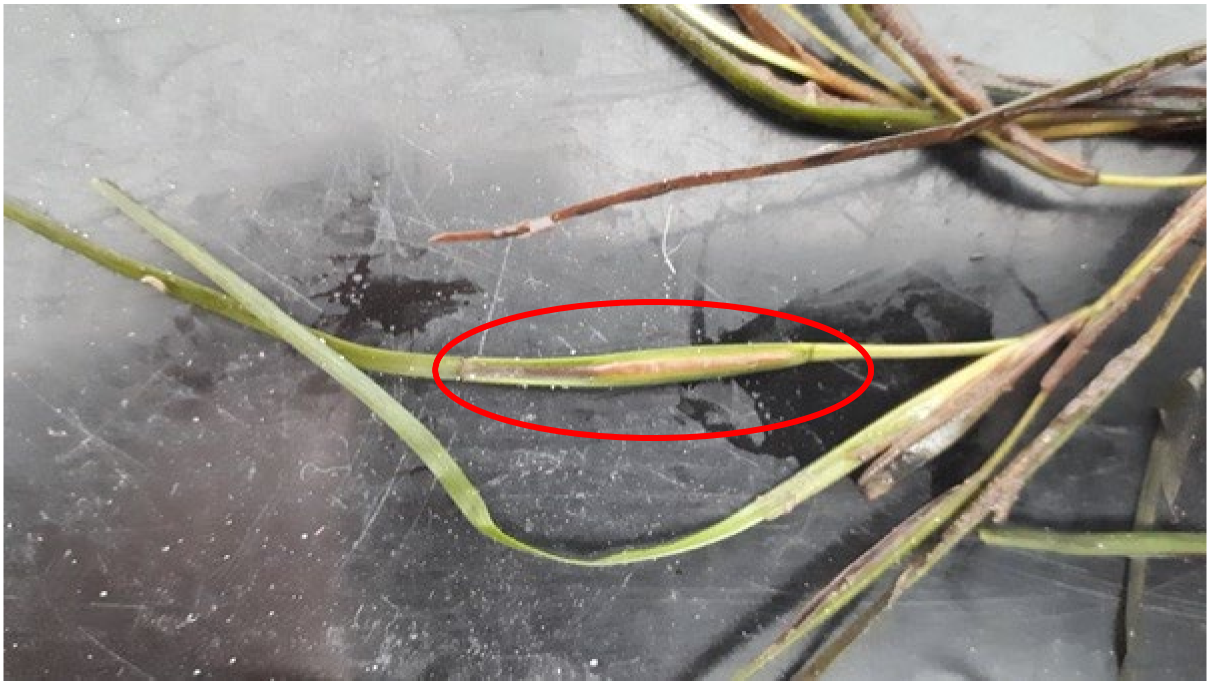

Figure 8: *Zostera tasmanica* shoot showing reproductive spathe (circled) containing flowers.

4. CONCLUSIONS

Although at very small scale, the transplantation experiment was highly successful, with all cores and grids leading to substantial establishment of *Zostera muelleri*. It thus appears that any of the methods employed would be suitable for active transplantation if this is required to re-establish *Z. muelleri* in other areas of the Port River where it is not re-establishing naturally. The quickest method was the use of cores, although these cover a smaller area per transplant than the grids. The grids do have the disadvantage of adding foreign materials to the environment, although they are expected to rust away relatively quickly. The cost of materials for each grid is ~$10, whereas cores are $1-2 each and can be reused.

It was deliberately decided to only move the transplants a few meters from the donor site, so as to examine as much as possible the transplantation technique only, with as little confounding as possible with site suitability. In doing this, it was possible to ensure that the tidal height of the transplants was within a few centimeters of the donor site, and that the broad environmental conditions, such as exposure to wave activity, were the same. This decision also minimized handling effects, as cores were in place at the recipient site within 1-2 min of extraction, and grids within 10 min. If the option of using transplants for restoration is to be pursued further, then the next steps would include examining the effects of handling, site selection, and changes in tidal level between the donor and recipient sites. Although not documented, a small transplantation trial was undertaken several years ago by a community group without success. This earlier trial involved moving cores from Torrens Island to the area around Snowdens Beach, which is on the other side of the Port River and 1-2 km upstream. It is thought that these cores were exposed to greater wave activity from vessel wakes, and that they may have been planted at a different tidal level to where they were obtained from. Either of these factors may have compromised their survival.

The use of seeds does not appear to be a viable option for restoring *Zostera* species seagrasses in the Adelaide region. Repeated surveys at multiple sites over several years and all seasons have failed to find any evidence of large-scale seed production. The most consistent signs of seed production have been in beachcast material of *Z. tasmanica* at West Beach. The source of this material is unknown, but there have been anecdotal observations of this species in shallow waters (3-10+m depth) along the Adelaide metropolitan coast. It is possible that this is actually a different species than the *Z. tasmanica* found in estuaries along the same coast, given that it appears to have different reproductive characteristics. Alternatively, the estuarine populations may be living in stressful environments, and thus not have the resources to reproduce, even though they visually appear to be healthy. It is also possible that this species has irregular ‘mast’ years with high levels of reproduction, and little to no reproduction in other years. Either way, without the regular presence of mature seeds, it would be difficult to undertake seed-based restoration.

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