

# Persistence study:

# Propagules and juvenile mangroves at the St Kilda boardwalk

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Prepared for:



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The sole purpose of this report and the associated services performed by Delta Environmental Consulting is to prepare a brief report containing information on a monitoring program conducted at the St Kilda mangrove boardwalk in accordance with the scope of services set out in the contract between Delta Environmental Consulting ('Delta') and the Green Adelaide Board ('the Client'). That scope of services was defined by the requests of the Client, by the time and budgetary constraints imposed by the Client, and by the availability of access to the site.

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# Persistence study: propagules and juvenile mangroves at the St Kilda boardwalk

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The investigators examined the rate of recovery after a recent hypersalinity event (starting in December 2019) that killed *Avicennia marina* mangroves in South Australia's Barker Inlet. Initially the aims of the study included determining the distance mangrove propagules would disperse through a standing forest of dead wood and the persistence of juveniles in the face of ongoing, although smaller, hypersaline leakage. An unforeseen informal program to augment natural recovery meant the initial question could not be answered, and the study refocussed on the persistence of juveniles and observations of adult trees in the adjacent areas of stressed and healthy trees.

Simple meter square quadrats were used to assess eleven locations at approximately monthly intervals from December 2021 to November 2022. Both quantative and qualitative observations suggest that recolonisation of mangroves has been very slow in the study area and is not typical of colonization rates in other sites in the Barker Inlet and Port River estuary. Slow recolonization of mangroves could have several causative factors. Possible causes include ongoing slow leakage of hypersaline brines liberated from the salt pond system into the groundwater as the ponds are rewet with annual winter rainfall, or residual hypersalinity that has not been ameliorated in low lying areas with fine sediments by daily tidal inundation (Leyden *et al* 2022, Dittmann *et al* 2022). Changes to the geomorphology of the site that make it hostile to the establishment of juvenile mangroves, or a lack of propagule material, are other possibilities.

Observations in the 'dead' zone included the decomposition of the standing dead trees, substantial and ongoing sediment loss from around the dead mangroves, especially along drainage lines, and dislodgement of re-established benthic mat communities. In the adjacent 'live but stressed' portions of the forest observations included reproductive failure and subsequent leaf chlorosis, leaf dieback in winter and increased leaf herbivory by opportunistic species.

The slow rate of recovery and evidence of ongoing stress in live trees within the transition zone led the authors to consider that reducing hypersalinity inputs should be the initial focus of recovery efforts. Possibilities for assisted recolonisation, along with identification of further avenues for investigation, were also explored.

Keywords: Avicennia marina, hypersalinity, recovery, recolonisation

#### 1. Introduction

Mangrove forests in South Australia contain one species, *Avicennia marina*. The habitat created by this species provides several important ecosystem services.

Well documented at the State level are the fisheries values of the mangrove habitat, specifically for blue crab, whose juveniles are found in mangrove creeks in Gulf St Vincent (Dixon and Hooper 2009). There are also correlations recorded in SA between the extent of juvenile habitat with the productivity of the prawn fishery, particularly with respect to mangrove habitat on the West Coast (Dixon and Roberts 2006).

The presence of mangrove forests and their benefits have been studied in the light of sea level rise and the potential for increased storm surges resulting from climate change. In Florida, Zheng *et al* (2012) measured decreased area of inundation and reduction in surge water velocity in areas protected by mangroves. Blankespoor *et al* (2017) quantified the protection services of mangroves for 42 developing countries in the current climate, finding that with the current mangrove coverage 3.5 million people and GDP worth \$400 million are at risk. With ongoing losses of mangroves continuing at the current rate, along with a future sea level rise of 1 meter and a 10% increase in storm intensity, the people and GDP at risk would increase by 104% and 233% respectively. This sea level rise estimation is congruent with that which could eventuate by 2100 according to the IPCC sixth assessment (Fox-Kemper *et al* 2021).

Respiration and transpiration in mangroves contribute to air quality. A commonly recognized but rarely measured benefit of mangroves is the production of oxygen. Calculation of oxygen production from mangroves in India (Pan *et al* 2019) has led to calls for active reforestation, while a study in Indonesia (Junaedi *et al* 2020) found that oxygen production from mature stands of mangroves was twice that of the local terrestrial forest. Water vapor from transpiration will, by its nature, affect local humidity, buffering temperature variations and facilitating the settling of fine particles from the air. While mangroves grow in tidal areas, which are themselves evaporative features, Liang *et al* (2019) found that compared to the wet soil surface, which had evaporation rates varying strongly by season, the evapotranspiration flux from mangrove canopies in China and Florida was more stable, and exponentially of larger magnitude, rising with leaf area index.

As globally reported, carbon sequestration is high in mangroves, even in arid climates. Schile *et al* 2017) in their discussion of the limits to sequestration in arid blue carbon ecosystems found that mature mangroves total carbon stocks (soil, above ground and belowground biomass), as tC/ha, exceeded that of intertidal microbial mats, coastal sabkha, planted mangrove stands, saltmarsh and seagrass beds, in descending order.

Mangroves in Gulf St Vincent occur in climates that range from semi-arid with >400 mm rain pa, to arid with <250 mm rain pa (Commonwealth Bureau of Meteorology 1971, 1972). It is possible to speculate that the mangroves occurring within the less arid Barker Inlet, the subject of this study, could contain larger pools of carbon than those further north in the Gulf, similarly to mangroves in Moreton Bay that show differences in carbon pool magnitude across geomorphological variations (Lovelock et al 2014). In the Moreton Bay study, *Avicennia marina* mangroves backed by *Juncus* sedgelands had notably greater carbon pools. While the authors of that study merely note that *Juncus* has a rhizomatous growth form, it is also worth

noting that *Juncus* sedgelands frequently occur in areas with regular flows of freshwater and generous sediment supply. The Barker Inlet mangrove stands are the best watered in Gulf St Vincent, both from direct rainfall and runoff from rivers, creeks, and stormwater drains sourced across metropolitan Adelaide and the Hills Face Zone. These watercourses frequently debouch large volumes of sediment into the intertidal area (Bone *et al* 2005), and this could reasonably be considered to contribute to increased terrestrialisation of carbon in the mangrove zone of Barker Inlet.

Despite the benefits of intact mangrove ecosystems, around the world mangroves face threatening processes and the habitat is considered by the IUCN to be in global decline (Polidoro *et al* 2010), although *Avicennia marina*, as a species, was considered at the time to be of least concern (Duke *et al* 2010). Despite global programs to conserve mangroves, the loss appears to be continuing. Recent mapping by NASA (Stevens 2020) attributed observed losses to human-driven or natural causes. The mapping revealed mangrove loss to natural causes (defined as erosion, storms, desiccation, or drowning) were extreme in along the Amazonian South American coast, with decreasing but still notable rates of naturally caused deaths in India, Australia, parts of eastern Africa and the United States.

Studies of hypersalinity impacts on *Avicennia marina* in South Africa (Naidoo *et al* 2011) revealed that trees in more saline sites had lower concentrations of chlorophyll in their leaves (29-45% reduction) leading to significantly lower CO<sub>2</sub> exchange rates, In Florida a study of mangroves in an area with seasonal salinity increases found that hypersalinity reduced mangrove forest evapotranspiration by 26% (Barr *et al* 2014). Devaney *et al* (2021), using growth chambers, observed that *Avicennia germinans* exposed to cold conditions (<10°C) responded more poorly in higher salinities than in lower salinities, and worryingly severely stressed plants did not recover even when provided with conditions of low salinity and high humidity.

In tropical Australia, Taimasa events driven by the El Nino-SOI sea-level swings have recently caused extensive areas of death in the Gulf of Carpentaria (Duke *et al* 2022, Abhik *et al* 2021). These deaths appear to be the combined result of desiccation and resultant high salinity soil conditions.

Human-driven impacts reported by Stevens (2020) were greatest in South-east Asia, followed by Madagascar, western tropical Africa, and Central America. Many of these losses resulted from harvesting mangroves for wood or removing them to make way for fish farming and other developments, but the trees are also harvested for fuel wood, fodder, and tannin.

In Gulf St Vincent, South Australia, previous mass deaths of mangroves have resulted from human-driven impacts. Approximately 250 hectares of mangroves adjacent the Bolivar Sewage works outfall between St Kilda and Port Gawler were lost after offshore seagrass beds were impacted by eutrophication and died, changing sedimentation patterns and hydrology in the mangrove forest (Overton 1994, Commonwealth of Australia Senate Environment, Communications, Information Technology and the Arts References Committee 2000). In the southern part of Barker Inlet, at North Arm, approximately 8 hectares of mangroves died when construction of causeways for electricity transmission lines disrupted tidal movements (Commonwealth of Australia Senate Environment, Communications, Information Technology and the Arts References Committee 2000).

## 2. Study area

In the first half of 2020, *Avicennia marina* mangroves edging parts of the Barker Inlet and Port River estuary, near St Kilda in South Australia, were observed to die. Stressed and dead trees were reported by Council workers at the St Kilda boardwalk by July 2020. In early August reports of deaths of trees in the St Kilda township were emailed from residents (L Virgo 20 Aug 2020, K Collins 14 Aug 2020) to the City of Salisbury.



Figure 1 - Map of study area with transect line (boardwalk) in yellow

The death of the mangroves as well as of parks and garden trees within the township occurred subsequent to an adjacent leaseholder disposing of salt, via hypersaline brine, into a series of elevated 'salt ponds' or dams that had been lying moribund for some years. Pumping of the waste brine began in December 2019 (GeoScience Australia 2022) and continued until the leaseholder was directed to stop pumping in November 2020 (DEM 2020). While this paper does not attempt to establish a causative link, research published by Dittmann *et al* (2022) found that dead trees at this site occurred in areas where soil porewater was hypersaline.

By spring 2021 there was little visible recruitment of mangroves or saltmarsh in the affected area. Seeps of hypersaline brine onto the marsh surface were still evident and were observed precipitating salt onto the soil surface. In contrast, salinity of surface waters measured in the creeks and pools was euhaline, most probably due to regular daily tidal movement. The initial aim of this study was to determine how far into the "dead zone" new mangrove propagules from the adjacent forest would penetrate, and to determine their persistence over time. The information obtained would inform planning to augment, or not, the recovery of the mangrove forest.

#### 3. Method

A transect, formed by the mangrove boardwalk, was established with 1m quadrats laid out at 50m intervals. We chose a distance-based spacing to ensure no possibility of researcher bias in selection of quadrat locations. The downside of this selection method is that it was not possible to ensure there were similar numbers of sites in each 'health' class of the mangrove habitat being assessed. Additionally, the selected layout resulted in only 11 locations, providing low statistical power for the study, given the wide variations in site conditions across the study area. In retrospect, more robust quantitative results could have been produced by either using triplicate sites at each 50m location or by reducing the intervals to 15m. Constraints on time and labour availability precluded that. Despite these limitations the study has provided some useful qualitative observations, particularly when combined with findings from other researchers.

The removable quadrat used was a 1m square made from PVC electrical conduit subdivided with a 10 x 10 grid made of builders' twine. Each quadrat location was 50m seaward of the one before. The boardwalk was painted with a mark on its outside edge at the designated intervals. These marks were unobtrusive, so that there would be less likelihood of the quadrat locations drawing unwanted attention. The removable quadrat was placed at each location and a reference photograph taken so that on each return visit the quadrat could be placed exactly again.

The quadrat names include chainage (e.g., Q350 is 350m along the transect). The sampling method notes contain coding that specifies, for each quadrat, its spatial relationship to the mark on the boardwalk (before, after or centred, on left or right of boardwalk). The choices for which side of the boardwalk and relationship to the mark were based on random selection, where possible, or the choice was made judgmentally in areas where trees and other obstacles prevented quadrat placement.

After placement it was determined whether each quadrat was in the "dead". transition" or "healthy" zone of the mangrove forest. The initial quadrat, placed at the start of the boardwalk, was classed as "embankment" as trees in this zone were not typical. Some, with roots in the natural mangrove sediment, were dead. Some others had roots in the built embankment and those trees were alive, accessing the freshwater lens present in the earthen structure. The "dead zone" was taken to mean the area of totally dead mangroves, mapped as such by DEW (2021) and by ARA and EcoProTem (2021). "Transition zone" in this study means the additional area of mangroves impacted by this event but not killed outright, identified through a reduced chlorophyll presence on 2020 hyperspectral aerial imagery flown by ARA and corrected to remove any pre-existing areas of reduced chlorophyll identified in 2018 hyperspectral aerial imagery (also flown by ARA), and analysed by EcoProTem. "Healthy zone" refers to areas where ARA's hyperspectral imagery shows "normal" concentrations of chlorophyll. Trees in these areas may have other health problems and environmental challenges, however their chlorophyll concentrations are similar to mangroves that occur in the Gawler River delta, River Light delta, and on Torrens Island.

During the process of developing an understanding of the total area of mangroves impacted in this event, areas of poor health/low chlorophyll were observed in the hyperspectral imagery of "eastern shore" mangroves between Middle Beach and Wingfield, in some earlier years. Speculatively, these transient areas of poor health may reflect the multitude of stressors Barker Inlet mangroves face on an ongoing

basis. Potential stressors include reduced catchment flows and sediment supply at Salt Creek and Gawler River (due to impoundments across these waterways), numerous vestigial tidal creeks (creeks cut off from catchment inflow by the embankments of saltfields and other land reclamations), ongoing variable levels of hypersalinity impacts from saltfield operations since the 1930s, sewage outfall impacts between Chapman Creek and the historic outlet of Thompsons Creek, and pollutants from the urban catchments, rubbish tips and industry.

Barker Inlet mangrove habitats may not be as resilient as pristine mangrove forests. In unflushed vestigial creeks the development of deposits of monosulfidic black ooze mean the mangroves lining those creeks may be at risk from events that disturb those deposits. Mangroves lining vestigial creeks may not receive sufficient catchment sediment keep pace with local sea level rise. Trees growing along the seaward edge of the forest may be impacted when sediments released by the eutrophication death of offshore seagrasses is relocated by storms into the forest, smothering the breathing roots of the trees.

The classified quadrats along the transect were revisited approximately monthly, from December 2021 to the first week of November 2022 (eleven visits). At each visit the quadrat was placed and photographed sequentially at each location. One quadrat was not photographed on one occasion (Q200, 28 August 2022), although the field journal was filled in with the details of propagules and juveniles.

The records collected included number of individuals (the total of juveniles and propagules within the quadrat), separate counts for juveniles and propagules, leaf counts for juveniles and the average height of juveniles. We defined juveniles as having set a root and unfurled at least 1 true leaf from their stem, held above the cotyledons. Propagules were all the earlier stages - they were not yet established. "Average height" refers to the average height of the juveniles, and propagules were ignored for this measure. Leaf counts were undertaken for all juveniles and excluded the cotyledons. Opportunistic notes were made about the occurrence of juveniles outside the quadrats and the presence of invertebrates, seagrass wrack and other transitory features.

All data was recorded in a field notebook and transferred, on return to the laboratory, to an online data repository, the BioCollect platform of the Atlas of Living Australia (ALA 2022). The project was made publicly accessible so that the monthly data could be viewed by other researchers and interested members of the public.

### 3.1 A confounding issue

It became apparent early in 2022 that sometime earlier an informal propagule relocation program had started at the boardwalk. After talking to the people undertaking the program, it seems that approximately 5000 propagules were picked and/or retrieved from those parts of the boardwalk (and adjacent mud) where trees were alive and producing propagules. The program continued from sometime in December 2021 to late March 2022. Both picking and retrieving of propagules only occurred within arm's length of the Boardwalk. The picking and retrieving occurred in the "healthy", "transition", and 'embankment' zones. Handfuls of the propagules were then thrown from the boardwalk into the "dead" zone.

Our quadrat locations were marked in early December 2021. Researchers from Flinders University and the University of Adelaide had selected their sites earlier in the year. Research locations were selected using differing rationales, as detailed by the different researchers.

As our location markers were on the boardwalk, and the use of the marks to locate the quadrats was coded in our methodology, they were not easy to see when walking along the Boardwalk. It was necessary to take reference photos on to site to relocate our quadrats exactly each time. Some of the other researchers' quadrats along the boardwalk were marked with corner pins, ribbon tags and flagged posts. All research locations were selected before the 'propagule relocation' program started. It is possible that some of the more easily visible 'marked' quadrats in the dead zone may have had handfuls of propagules relocated to them. There may have been relocations to our less easily visible quadrats too, but they may have only received random, not determined, relocation.

As a result of the relocation program there was no way of knowing whether propagules recorded in an area were naturally occurring (and reflect penetration range) or relocated. There was also no way of knowing whether the propagule counts in the stressed and healthy zones reflected the actual propagule load for the season – it is likely that most quadrats in these two zones would have had freshly fallen propagules removed from the mud, and nearby trees may have been picked, reducing fall into those areas.

The overlap of the study area and the relocation program meant the initial question about the penetration distance of mangrove propagules into the 'dead' zone could not be answered. Survival rates of propagules that established (regardless of their source) could be measured, and so the monitoring of the quadrats continued.

#### 3.2 Description of individual quadrats

**Q0** was located at the intersection of the wooden boardwalk and the St Kilda-Dry Creek Embankment. At the start of this study there were a mixture of live and dead trees in a narrow band along the embankment. It is likely that trees with their roots inside the freshwater lens that forms from rainfall within raised near-coastal features like cheniers and embankments (Stuyfzand 2016, Röper *et al* 2013) were able to survive the hypersalinity event, while those slightly "downslope" had their roots in soil impacted by the hypersalinity event. There were juveniles in this quadrat pre-dating the impacting events, and these presumably survived because they had their roots in the freshwater lens. On our initial survey we recorded 13 iuveniles with 188 leaves between them.

**Q50** was within the dead zone. All mangroves in this area were dead. At the start of the study there were no live juveniles in this quadrat.

**Q100** was located within the dead zone and once again, all mangroves in this area were dead. At the start of the study there were no live juveniles in this quadrat.

**Q150** was located inside the dead zone and close to the margin between the dead zone and transition zone. The edge of the live trees began about 15m away from the boardwalk, just visible through the dead trees in Figure 1.

Q200 was within the transition zone. The adult trees adjacent the quadrats from here to the end of the board walk were alive. Within the transition zone leaf reflectance on hyperspectral imagery appeared to show lower chlorophyll concentrations (Airborne Research Australia and EcoProTem 2021). Many trees were displaying other signs of stress, which were noted in the field, and which are detailed here in the discussion. There were initially 35 juveniles in this quadrat that had survived through the recent impacting events.

**Q250** was the second quadrat located within the transition zone.

Q300 was the first site located in the healthy zone, defined as the zone where calculated chlorophyll reflectance from the trees matched other stands of mangroves in the Barker

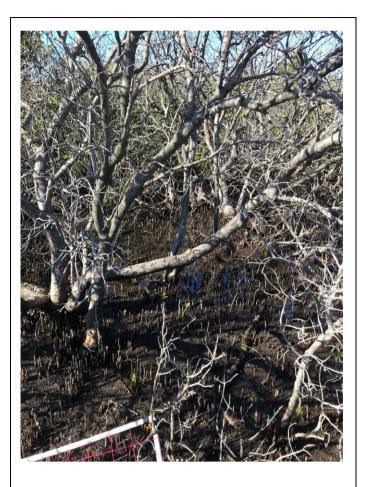


Figure 2 - Q150, with the margin of the transition zone just visible through the dead mangroves.

Inlet that were distant from the site of the hypersalinity impact. The healthy sites were all close to the seaward side of the forest. Adjacent to the quadrat, the presence of a bird hide suggests the site had been a low chenier in the early 1990's when the boardwalk was built. The hides were located where they had views across open saltmarsh or pools. Vegetative remnants near the hide confirm that, at that time, this area supported shrubby samphire (*Tecticornia arbuscula*) and bearded glasswort (*Salicornia quinqueflora*). The site had no existing juveniles at the start of the study.

**Q350** was within the healthy zone and demonstrates that the better flushed portions of the forest face other challenges than salinity. Water movement through the forest, from daily tides and storms, can be rapid. This has implications for propagule establishment. Accumulations of seagrass cover the pneumatophores of adult trees and create unstable temporary mounds. This location, and the other healthy zone quadrats (**Q400**, **Q450** and **Q487**), had no existing juveniles at the start of the study.

**Q487** was the final quadrat. The distance between **Q450** and **Q487** was only 37m as the boardwalk comes to an end here.

#### 4. Results

Quantitative data from the eleven monitoring events are presented in Table 1. Graphs of propagules, juveniles and leaf counts are appended.

The pre-existing juveniles on the embankment at **Q0** increased by 1 plant and then decreased by one plant over the period. Very few propagules were seen in this quadrat until Spring of 2022. There is a note in the field book to the effect that the winter fall of propagules were small thinnings, dropped out of season. Leaf counts in the juveniles increased over the period (from 188 to 349 leaves) while average height increased from 0.3m to 0.41m before some loss of upper leaves and loss of 1 plant saw average height decrease to 0.35m.

**Q50** and **Q100** in the dead zone did not gain any propagules or juveniles over the monitoring period.

An occasional propagule (assumed to be one of the relocated ones) established close to these quadrats. In the last few monitoring events it was noted that some of the newly established juveniles had started to lose their upper leaves. Adult trees in this zone continued to degrade – the outer parts of pneumatophores rotted away, and the lateral roots, normally covered in sediment became exposed. Detritivores including marine pill bugs and snails were observed grazing, in winter.

**Q150** in the dead zone gained occasional propagules in the first summer, and 2 juvenile plants established inside the quadrat, with more establishing outside the quadrat. The 2 juveniles gained leaves and height until the 29<sup>th</sup> of September, when they had 25 leaves between them and an average height of 0.19m. By early November the plants had lost 1 leaf, other leaves showed signs of tip burn, and the average height calculation was omitted. Several of the juveniles that had established outside the quadrat were dead.

The first quadrat in the transition zone, **Q200**, started with 35 juveniles that had 95 leaves between them and an initial average height of 0.08m. Over the period of monitoring there was a peak of propagules washing into the quadrat in the initial summer months and again in November 2022. Leaf counts peaked in winter at 179 leaves, before varying through late winter and spring, ending the period with 136 leaves. The average height of the juveniles increased over the period, from 0.08m to 0.15m.

The transition zone quadrat **Q250** saw an increase in juveniles (10-14), an increase then decrease in leaf counts (44, 74, 56) and an increase then decrease in average height (0.18m, 0.19m, 0.15m). The leaf counts and average height variations did not match directly – at these small heights and leaf numbers the loss or gain of a few large leaves can produce a noticeable difference in average height, and conversely the loss of gain of small leaves can make a smaller difference.

The **Q300** quadrat marked the start of the healthy zone. None of the occasional propagules that washed into this site established.

**Q350** gained 4 juveniles over the monitoring period but 2 of these died. Leaf counts rose to 13 at the peak in April and decreased to 5 by the end of the monitoring period. The average height peaked at 0.12m and decreased slightly to 0.11m by November 2022.

In **Q400** and **Q450** a peak of 16 and 2 juveniles, respectively, established, but all were lost over the monitoring period, while in **Q487** no juveniles established.

Propagules											
Date	Q0	Q50	Q100	Q150	Q200	Q250	Q300	Q350	Q400	Q450	Q487
12/12/2021	0	0	0	1	12	13	4	2	9	11	15
13/01/2022	1	0	0	2	12	6	2	3	25	13	13
12/02/2022	0	0	0	0	16	6	0	6	16	13	0
26/03/2022	0	0	0	0	5	0	0	5	7	9	13
8/04/2022	0	0	0	1	6	0	0	0	4	26	7
28/05/2022	0	0	0	0	2	2	0	0	1	3	0
25/06/2022	0	0	0	0	1	3	0	1	0	1	0
27/07/2022	1	0	0	0	3	2	0	1	0	4	2
28/08/2022	7	0	0	2	3	4	0	4	3	3	0
29/09/2022	2	0	0	1	2	1	3	8	3	0	1
11/11/2022	17	9	0	2	21	23	0	35	7	8	13
Juveniles											
Date	Q0	Q50	Q100	Q150	Q200	Q250	Q300	Q350	Q400	Q450	Q487
12/12/2021	13	0	0	0	35	10	0	0	0	0	0
13/01/2022	13	0	0	2	39	10	0	0	1	0	0
12/02/2022	13	0	0	2	46	15	0	2	8	0	0
26/03/2022	13	0	0	2	46	16	0	4	16	1	0
8/04/2022	14	0	0	2	47	15	0	4	15	2	0
28/05/2022	14	0	0	2	48	14	0	3	7	1	0
25/06/2022	13	0	0	2	49	16	0	3	5	0	0
27/07/2022	13	0	0	2	51	15	0	2	1	0	0
28/08/2022	13	0	0	2	44	15	0	2	0	0	0
29/09/2022	13	0	0	2	36	15	0	2	0	0	0
	13	0	0	2	44	14	0	2	0	0	0
11/11/2022	15	U	U		44	14	U	Z	U	U	U
Leaf counts Date	Q0	Q50	Q100	Q150	Q200	Q250	Q300	Q350	Q400	Q450	Q487
12/12/2021	188	0	0	1	95	44	0	0	0	0	0
13/01/2022	224	0	0	4	126	51	0	0	2	0	0
12/02/2022	250	0	0	8	158	70	0	5	70	0	0
	261	0	0	12		71	0	12	50	4	0
26/03/2022		0	0		151		0		49	4	0
8/04/2022	292	0	0	12	156	69	0	13 12		2	0
28/05/2022	308			16	159	69			22		
25/06/2022	318	0	0	16	167	74	0	11	16	0	0
27/07/2022	301	0	0	16	179	70	0	7	4	0	0
28/08/2022	304	0	0	22	141	70	0	6	0	0	0
29/09/2022	301	0	0	25	117	67	0	5	0	0	0
11/11/2022	349	0	0	24	136	56	0	5	0	0	0
Average height	Q0	Q50	Q100	Q150	Q200	Q250	Q300	Q350	Q400	Q450	Q487
Date	-	<b>Q50</b>	-	Q150 0	-	-	Q300 0	Q350 0	Q400 0	Q450 0	-
12/12/2021	0.3		0		0.08	0.18					0
13/01/2022	0.35	0	0	0.05	0.1	0.18	0	0	0.05	0	0
12/02/2022	0.35	0	0	0.08	0.1	0.2	0	0.05	0.05	0	0
26/03/2022	0.4	0	0	0.15	0.14	0.19	0	0.08	0.11	0.1	0
8/04/2022	0.4	0	0	0.18	0.16	0.14	0	0.08	0.13	0.15	0
28/05/2022	0.4	0	0	0.2	0.16	0.16	0	0.12	0.11	0.15	0
25/06/2022	0.41	0	0	0.2	0.15	0.16	0	0.12	0.1	0	0
27/07/2022	0.4	0	0	0.2	0.15	0.15	0	0.11	0.1	0	0
28/08/2022	0.35	0	0	0.18	0.15	0.17	0	0.1	0	0	0
29/09/2022	0.35	0	0	0.19	0.15	0.15	0	0.11	0	0	0

Table 1 – Raw data

#### 4.1 Ad hoc observations

During the monitoring program ad-hoc observations outside the monitoring quadrats were recorded.

The establishment of juveniles from the approximately 5000 propagules informally relocated into the dead zone occurred in the early months of the monitoring program. While the numbers of propagules were large, the areal magnitude of the dead zone resulted in new juveniles being sparse at best, spaced meters apart. From the 28th of August 2022 observations of juveniles close to the boardwalk revealed that some were showing tip burn of their upper leaves. The 11th of November observations recorded that some of the new juveniles in the dead zone had died.

Two trees at the border of the dead and transition zone were observed closely, initially to gain an indication of whether the dead zone was spreading. These trees displayed reproductive failures in early 2021 and early 2022. The trees seemed to excrete large amounts of salt through the lower leaves on the twigs that were flowering. These leaves eventually became pale (chlorotic) and developed small black necrotic spots on the undersides.

In Tree 1, flowering in early 2021 was followed by a complete reproductive failure in autumn 2021. The tree maintained its canopy of pale green leaves with "dusty" spots underneath. Flowering in 2022 produced some propagules in winter, but leaves yellowed significantly after that, some fell, and the remaining leaf undersides had many small black spots. The propagules continued to be much smaller than propagules on trees in the healthy zones, as of November 2022 (Figure 3, left).



Figure 3 - Tree 1 (left) and Tree 2 (right)

Tree 2 flowered in 2021, and most flowers did not set propagules. The few tiny propagules that were produced were aborted early in winter 2021. The tree's leaves became yellowed with black spots on the underside. Many yellowed leaves fell as it grew new dark green leaves in early summer. In 2022 the tree flowered very lightly and set no fruit. The new, fully grown leaves remained dark green, however when observed on 27<sup>th</sup> July 2022 it was noted that the youngest leaves at the tips of the branches were dying (Figure 3, right).

Over winter and into spring of 2022 several trees further into the transition zone, closer to the healthy zone displayed leaf damage. In some cases, the trees' leaves turned brown, from the tip back towards the petiole (Figure 4, left). There was also a swathe of trees with noticeably heavy herbivory of leaves. Some caterpillar damage of mangrove leaves is common, but in this case the grazing impact occurred on most leaves on many of the branches. The damage was seen easily from the boardwalk (Figure 4, right). Active herbivory appeared to cease by mid-December (new leaves were ungrazed), and a search for larval shelters revealed several empty shelters that had been made by joining leaves together, with empty remnants of silk cocoons and frass inside.



Figure 4 - Transition zone trees displaying leaf dieback (left) and herbivory (right)

Sediment loss (erosion) around the tree skeletons in the dead zone has become more visible over the period of monitoring. Incremental loss of sediment in an area without any monitoring posts or sediment elevation tables often passes unnoticed. In the case of the dead mangroves at St Kilda, the gradual erosion of sediments previously held around the trees became noticeable when that loss was sufficient to expose the lateral roots of the trees, in late 2021. Lateral roots are usually buried, with only the vertical breathing roots (pneumatophores) protruding above the

surface. At the edges of drainage lines, the sediment loss occurred both above and below the lateral roots, undermining the table of roots holding the trees upright.

Within open pools in the mangroves and saltmarshes loose sediment is often partitioned from the overlying water by benthic mat communities. Such communities may form complex layers comprising filamentous and colonial cyanobacteria, and layers of overlying filamentous green algae and periphytic diatoms. Besides stabilizing the sediments, the mat communities support complex food chains including grazing and predatory protozoa and invertebrates. The autotrophic components of the mats ensure that the shallow waters of these pools are saturated with oxygen in the daylight hours.

During the monitoring on 11<sup>th</sup> of November it was noted that the contiguous benthic mat in the main pool in the dead zone had been 'rolled up' and large areas of fine sediment were exposed. Touching these areas of sediment even lightly resulted in the sediments resuspending into the water column above (Figure 5).



Figure 5 - Benthic mat communities displaced in open pool area

Decomposition of the pneumatophores of the dead trees continued throughout the period of observation. Initially the lenticel-rich epidermis and cap of the pneumatophores became loose and the fleshy cortex and pericycle containing the spongy gas-filled aerenchyma started to degrade. Once this material was shed, the central stele comprising xylem and pith remained standing, until winter and spring of 2022, when these last remnants of the pneumatophores started to break off at the mud surface.

#### 5. Discussion

#### 5.1 What individual constraints exist in specific quadrats?

The embankment area seemed to produce very few propagules into quadrat **Q0**, despite live mangroves occurring very close by. This may support the supposition that propagules from mangroves on the landward side of a mangrove forest do not disperse far from their parent trees, possibly as water velocity tends to be less so far from the open water of the Barker Inlet.

It was originally estimated that propagules would be unlikely to reach the dead zone sites in any numbers. The informal relocation program makes it impossible to determine whether this was so. The presence of juveniles at **Q150** in the dead zone near the edge of the transition zone, both inside the quadrat and adjacent, suggests that at least some propagules may have penetrated the area naturally. While scant juveniles were observed near the **Q50** and **Q100** quadrats in the dead zone, no juveniles established inside the quadrats during the monitoring period.

The quadrats from **Q200** (start of the transition zone) to the end of the boardwalk are likely to have lost fertile propagules to the harvesting for the relocation program, rather than gaining them. This would be especially so in those quadrats where the boardwalk is low lying and the mud surface easy to reach. From where the trees with green leaves start, to the end of the boardwalk, ripe propagules in the tree canopy were also harvested. It may be supposed that propagules from trees in the central part of the forest would be unlikely to disperse far from their parent trees, due to calm water and wind conditions as well as the density of adult stems and pneumatophores. Some propagules in these areas managed to establish as juveniles in the monitored quadrats, between periods of propagule collection. It is therefore unknown whether the numbers of juveniles that did establish at these sites is typical or not.

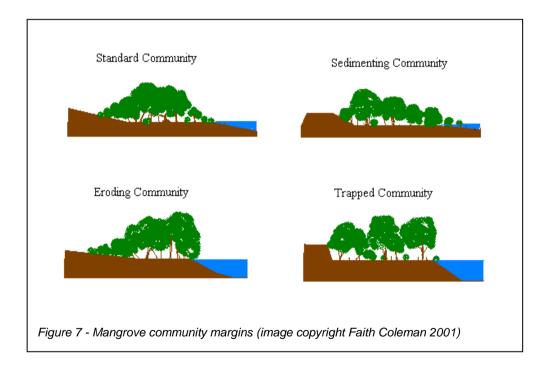
Between **Q200** and **Q250** the carpet of juveniles is similar in appearance to the central parts of most mangrove forest near Adelaide. Most of these juveniles are several years old, pre-dating the hypersalinity event. *Avicennia* trees produce relatively few, large viviparous propagules, each of which has a high probability of establishment. Typically, the initial fast growth (anywhere up to 0.2m in their first year) of juvenile mangrove plants slows and the juveniles await the development of a gap in the canopy (Burns and Ogden 1985). Many very old trees are present in this part of the forest. When one of them is damaged, large numbers of juveniles will grow rapidly to fill the open space in the canopy.

Low numbers of new, and older, juveniles in the healthy zone (Q300, Q350, Q400, Q450 and Q487) could be the results of effects other than harvesting of propagules. Hydrogeomorphic features of the forest closer to the open waters of Barker Inlet may also impact reproductive success. For example, Q300 (on the remains of an old chenier) has an elevation slightly higher than its surroundings. This site may experience rapid water movement as tidal inundation on this high land drains away towards the nearby lower land. This seems a reasonable supposition, because there was rarely any build-up of leaf litter or seagrass wrack observed here, and the substrate was often visible, revealing the relatively coarse shelly soil of the remnant chenier.

Burial of juveniles by seagrass wrack deposited in storms seems the reason the juveniles in **Q350** and **Q400** decreased in numbers over winter.



Figure 6 – Typical mid-forest carpet of juveniles



The quadrats closest to the sea edge (Q450 and Q487) are very exposed to wave action. In a mangrove forest where accretion of sediments is occurring, young mangroves grow seaward of the adult trees, colonizing the mud banks fringing the

forest. This phenomenon is easily recognized in aerial imagery, with the seaward margin of the mangroves invaginated where small trees' canopies do not touch, and even smaller 'islands' where juvenile trees stand separated from the forest. In contrast the margin of a forest being eroded by sea level rise tends to be smoother, comprised of the joined canopies of mature trees (Figure 7).

In much of Barker Inlet, the seaward forest margin comprises mainly old trees, which suggests an eroding community, most likely the result of relative sea level rise which has been calculated at nearly 15cm at nearby Torrens Island (Coleman et al 2017). The exposed areas on the edge of a mangrove forest that is experiencing landward progradation and seaward erosion are the sites that tend to be the donor sites for widely dispersed propagules. Their furry coating allows the propagules to float significant distances and the open water allows the action of currents and winds to aid dispersal. Propagules in open waters may travel quite long distances over some time. MacMillan (1971) found that the furry coating did not drop off the propagules until they were in still conditions, so it is possible that many of the propagules that fell from the trees nearest the sea may have been removed from the area with the outgoing tides. It is also likely that those that remained did not sprout if water movement and seagrass deposits prevented them from settling.

In late March and early April quadrat **Q450** gained 2 juveniles (one each month). It would seem unlikely that these late starters were the result of the cessation of propagule harvesting – this site would take a long stretch from the Boardwalk to harvest. It was a challenge to retrieve the quadrat each month. It may reflect some settlement in the dense seagrass deposits that had not previously provided a stable base for juveniles, combined with calmer weather in autumn. The juveniles did not remain, however, being dislodged by rough weather before the 25<sup>th</sup> June 2022.

In summary, two of the three dead zone quadrats gained no propagules or juveniles. The third dead zone quadrat gained two juveniles, but these were beginning to lose leaves by the end of the monitoring period.

Other quadrats that did not gain permanent new juveniles were the three seaward-most healthy zone quadrats, the Q300 healthy zone quadrat and the embankment quadrat. The lack of establishment on the three seaward-most quadrats was most likely a function of the eroding nature of the seaward edge and the excess of seagrass wrack impacting those propagules that established. The Q300 quadrat appears to also suffer from water movement speed that prevented juvenile establishment. The embankment quadrat did not appear to receive many propagules in the 2021-22 summer and the one juvenile that established was lost in early winter. There appeared to be an adequate supply of propagules in the most recent monitoring, and so this quadrat may gain juveniles next year.

Both transition zone quadrats had existing juveniles at the beginning of the monitoring period, and both gained more juveniles over the same period. In the healthy zone, Q350 retained two of the juveniles that established during the monitoring period, the rest being lost to burial or removal in the seagrass wrack.

These results suggest that in the dead zone, three years after the actions that resulted in the initial hypersalinity event, there is little evidence of significant reestablishment of mangroves.

Is this slow rate of recolonization typical for mangroves in the Mediterranean conditions of South Australia? A comparison could be made with other local sites

where mangroves have established. Mutton Cove was a controlled tidal inundation site on Lefevre Peninsula, managed to maintain a saltmarsh habitat with mangroves edging a creek line, in a subsided landform. In May 2016 the levee surrounding the site breached and daily partial inundation of the site began. In August 2017 the site was examined to see how many mangrove juveniles may have established (Coleman and Coleman 2017). At that point, only one propagule drop had occurred since the levee breach. In the area where the breach had resulted in twice daily tidal flooding, many juvenile mangroves were present. Depending on the location, across the site juvenile density varied from 25-250,000 plants per hectare. Lowest concentrations of juvenile growth occurred in elevated areas not receiving frequent inundation, or in creek lines, or near the breach where excessive water velocity was causing scouring. The areas of dense juvenile growth were generally sheltered locations on the lower marsh plain with regular tidal inundation, and where existing mangroves or structures like embankments slowed water velocity. The authors estimated that within three years, as more breaches formed, the existing breach widened and inundation affected more of the site, juveniles would colonize across the entire marsh plain. They estimated that Avicennia marina would dominate the site within a decade.

The latter forecast appears to have been conservative. Satellite imagery viewed online (Airbus 2022), showed that for at least half the 34 hectares of marsh plain that would flood twice daily once the site becomes completely tidally unrestricted, mangrove tree canopies were touching, or separated by less than the average width of the canopies. In the remainder of the slightly more elevated marsh plain tree canopies were separated by 1-10 canopy widths. Across the site, six years after the first embankment breach, the juvenile mangroves have canopy widths from 1-2m diameter, where the canopies are sufficiently separated to measure. Where trees form a contiguous cover, the canopy diameters may be larger. Avicennia marina is already the dominant feature of the Mutton Cove marsh plain.

Mutton Cove is not the only location within Barker Inlet and eastern Gulf St Vincent where mangroves have colonised quickly. Mangrove incursion landward in response to sea level rise has been recorded in Barker Inlet at rates of up to 18m per annum (Fotheringham 1994). Landward progradation has also occurred very rapidly at Port Gawler, where mangroves have established on land that previously supported saltmarsh, reducing the saltmarsh habitat values for migratory bird species (Purnell *et al* 2012). Purnell identified juvenile mangroves establishing at Port Gawler on a wide strip of land along the landward edge of the forest. In the last 18 years (since 2004) mangroves have enlarged their cover until now the landward edge of the closed canopy is 100m east of its 2004 location, and further east for another 100m there is an open canopy of younger trees. Recently Lees *et al* (2020) estimated that 75% of the tidal flat at Port Gawler is now colonised by mangroves.

These comparisons suggest that the slow rate of recolonization at St Kilda is not typical of mangrove recolonization rates in the nearby environment. Some consideration of why that may be so follows, along with a discussion around possible causation of the other observations made over the year, including poor adult health, sediment erosion, and disturbance of benthic mat communities.

#### 5.2 General discussion of observations

Slow recolonization of mangroves into the site may have several causative factors. Ongoing pollution with hypersaline brines, either newly liberated from the salt pond system into the groundwater as the ponds are rewet with annual winter rainfall, or residual hypersalinity that has not been ameliorated by daily tidal inundation of fine sediments in lower lying areas, is one possibility. Changes to the geomorphology of the site that make it hostile to the establishment of juvenile mangroves could also be implicated, as could a lack of propagule material.

A recent conceptual model of the St Kilda mangrove and saltmarsh hypersaline brine contamination (Leyden et al 2022) states that due to hydraulic connectivity with the salt pond surface water, the groundwater underneath the salt ponds also became extremely hypersaline. This groundwater moved towards the intertidal zone under the increased hydraulic gradient of the brine-filled salt ponds. Leyden's report further discusses the potential drainage of the salinized sediments via creeks and tidal exchange and points out that while coarse grained elevated sediments and the surficial waters in the creeks have reduced in salinity over time, areas of lower elevation, fine grained sediments appear to remain hypersaline. Dittmann et al (2022) conducted soil coring in 2021, to ascertain soil salinity at depth in healthy, stressed, and dead portions of the forest. That study found that soil salinity in the dead zone remained hypersaline in 2021 and the hypersalinity increased with depth. Soil salinity conditions in the stressed zone were intermediate compared to the healthy and dead zones but were enriched with sulfate and chloride at the shallower depths. Sampling of porewater across the saltmarsh habitat as well as the mangrove habitat (Jones et al 2022, Wood 2022) found more than 80% of porewater samples had salinity greater than seawater (with a maximum of 278 ppt). Deeper soils returned a higher salinity than surface soils, indicating hypersaline groundwater.

Geomorphological changes that are possible include changed hydrological conditions, sediment loss (erosion), sediment instability and decomposition of the standing biomass. None of these have yet been measured at the study site. These factors have been shown to impact the recolonization rates of mangroves in the Gulf of Carpentaria (Duke 2019) and Sydney Olympic Park (Paul 2019).

Clarke's 1993 study of *Avicennia marina* propagule dispersal found that most propagules strand within 500m of their release, with fewer stranding at 1km and very few dispersing 10km. There are many live mangroves within 500m of the study area, however Clarke's study did not look at the penetration of mangrove propagules through a standing dead wood forest, rather it looked at strandings onto nearby open beaches adjacent to mangrove forests. At the St Kilda study site, the propagule penetration aspect of the study was confounded, providing no further illumination.

The ad-hoc observations recorded on the site over the monitoring period may be qualitatively considered in the light of the aspects outlined above, to determine whether the observations are consistent with the constraints to mangrove establishment and growth identified by other authors.

#### 5.2.1 Decomposition of dead trees

During the period of observation, the standing dead mangroves have started to decompose. *Avicennia marina* wood is unusual in that it contains successive cambia – the secondary phloem tissue in the alternating rings of xylem and phloem

(concentric internal phloem) within the trunk continues to transport and store water (Robert *et al* 2011, 2014). This feature allows the trees to store water in periods of physiological drought, but also means that the trees can withstand damage to their outer bark, a benefit in an environment where storm-driven flotsam could otherwise ringbark the trees.

The internal phloem makes *Avicennia marina* timber porous and the wood rots relatively quickly in damp or waterlogged conditions, through the differential swelling of the concentric rings of successive cambia. Studies in New Zealand, from a similar temperate climate zone to Barker Inlet, found that broken-off pneumatophore material lost about 50% of its weight in about 316 days and wood lost about 50% of its weight in 460-613 days. Leaf litter was removed from the system much quicker, with a  $t_{(50)}$  of only 63-88 days (Gladstone-Gallagher *et al* 2014).

The process of decomposition observed at the study site is compatible with the New Zealand findings. In the first year after the hypersalinity event, drifts of leaves fell from the dying trees, and were utilized by leaf rolling spiders that lived in the *Tecticornia arbuscula* shrubs along the embankment. By the second and third summers after the event (2021-2 and 2022-3), the leaf deposits were gone, and the large populations of spiders along the embankment were no longer observed.

By the summer of 2021-2 almost all the pneumatophores in the dead zone had lost the softer materials surrounding their central steles. Broken off steles (the central cores of the pneumatophores) were widely evident by spring of 2022 (Figure 8).

In the winter and spring of 2022 several of the dead mangrove trees started to fall, and the standing trees had lost bark and, in several cases, some outer layers of wood.

#### 5.2.2 Sediment loss

While the accumulation of sediment in mangrove systems is widely recognized as a function of the trees' and pneumatophores' presence reducing the speed of tidal flows and allowing fine sediments to settle, the process does not appear to be merely a passive one. Furukawa and Wolanski (1996) found that tidal frequency works as a pump that preferentially transports fine cohesive sediment from coastal waters into the mangroves. Kazemi *et al* (2021) used high resolution particle velocimetry with structures similar to pneumatophores in a flume, to develop an understanding of particle transport through patches of mangrove. They found both the spacing and diameter of mangrove root structures were important, and that a patch "porosity" of 47% marked the change between accretion (<47%) and erosion (>47%).

It is also possible to speculate that something more than the physical impediments provided by trunks and pneumatophores is at play, based on the work of Reef and Lovelock (2015). They found mangroves need to maintain water potentials lower than those of the saline water of the soil, to prevent desiccation. They found that *Avicennia marina* maintained hydrostatic potentials between -3 MPa at night and -5 MPa at midday when transpiration was greatest. Similar findings were recorded by Scholander (1968) in their study of mangroves' ability to desalinate seawater. Such potentials would assist in stabilising sediments around the roots of the trees.

Mangrove sediments are frequently the destination for contaminated sediments in waterways near human populations. Srikanth *et al* (2016) report widespread

findings of heavy metals accumulated in mangrove sediments, particularly lead. Harbison (1986) and by Dittmann *et al* (2022) reported similar findings within the study area and Barker Inlet more widely.

Very little research has been undertaken to establish exactly why sediment may be lost from a dead forest, although it is commonly reported that dead mangrove forests lose sediment (Tregenza 2022, Slezak 2016). If the accretion were purely passive, one may expect the sediment to remain in place. After all, tidal pumping and flocculation are not processes that would change because the trees were dead. Observations in the study areas revealed that while the tree skeletons remain, their presence alone has not been sufficient to prevent loss of sediment. It may be that the loss of the spongy material around the steles of the pneumatophores resulted in increased spacing between the remnant steles, leading to erosion. Or it may be that the loss of the strong negative hydrostatic potentials driven by transpiration of the living trees could play a part, as could any changed chemistry of the soil particles resulting from the exposure to hypersaline brine.

The loss of sediment from around the dead mangroves has been sufficient to reveal the lateral roots of the trees (Figure 8). This is more obvious along drainage lines where the velocity of the water has cut into the banks and removed sediment from under some trees. It is not known how much sediment has been lost, nor its ultimate destination. Options include the sediments being redeposited in the open water pools in the mangroves, or being flushed along the creek lines towards Barker Inlet, in a similar manner to the surficial expressions of the hypersaline brine (Leyden *et al* 2022). The presence of offshore seagrass beds at an elevation of approximately mean sea level (Leyden *et al* 2022) is, however, a barrier to



Figure 8 - Left: 1 = broken-off pneumatophores leaving circular scars, 2 = exposed lateral roots, 3 = juvenile established between pneumatophore steles, small unnumbered yellow circles are propagules that have shed their pericarps.

Right: propagule attempting to establish in unstable sediment

extensive removal of sediments as only the upper part of the tidal prism would be available to carry the material.

Should the sediment settle into the pool areas, the benthic mat community may partition the sediments (and any included heavy metals) from the water column. Recent observation of the dislodgment of a large benthic mat highlight how fragile such partitioning may be, compared to the sedimentation processes provided by live mangroves.

In mid-November 2022 the benthic mat in the large pool between Q0 and Q50 appeared to have lost its purchase on the benthos, in the shallows close to the dead trees where it was growing amongst the pneumatophores. Shallow water that is unshaded can become hot and this may explain the phenomenon – it is possible the mat edges died in the hot weather preceding the cool windy change that occurred in early November. Once the mat was not well entangled with the pneumatophores, the strong wind could dislodge the remaining mat, rolling it up across the pond and exposing the labile sediment below, which resuspended with very slight disturbance.

#### 5.2.3 Propagule settlement

Settlement and establishment of *Avicennia germinans* propagules in the central Texas coast were studied by McMillan (1971) who discovered that salinity was not a limiting factor to establishment, at least between fresh water and twice seawater salinity. McMillan stated that *Avicennia marina* propagules from Darwin exhibited similar salinity tolerances. Saenger's 1982 research found that salinity's main impact related to pericarp shedding. The fuzzy pericarp is buoyant and is important for dispersal of the propagules. Brackish water led to the fastest shedding of the pericarp. Fresh or hypersaline water led to longer maintenance of the pericarp, increasing dispersal distances.

The main driver of emergence and establishment appeared to be cessation of water turbulence followed by shallow water depths (McMillan 1971). Lodgement is important – the propagules were observed not to extend their roots and leaf shoots while they were still rocking.

Saenger (1982) reported that high temperatures caused rapid pericarp shedding and therefore limited propagule dispersal and McMillan (1971) found that high temperature was lethal to stranded propagules that had not yet established roots and leaves, but once these were established the juveniles could tolerate high temperatures for several days. Temperature during the period between propagule stranding and establishment is likely to be important for South Australian mangroves, with propagule drop occurring in the hottest time of the year (December to March).

The site visit on 17<sup>th</sup> December 2022 revealed 'propagule drop' had started. Many propagules were present floating in the water, and some were attempting to establish. Decomposition of the mangrove skeletons edging the pool areas (stripping of pneumatophores and steles breaking off at surface level) were visible. This feature may make juvenile establishment challenging, this season.

The underlying sediment in the open pools appeared to have very poor integrity, resuspending at the lightest touch. It is not likely that the loose sediment in the open pool areas would support the growth of mangrove juveniles.

#### 5.2.4 Juvenile growth

Research on salinity tolerances in juvenile *Avicennia* has shown that juveniles under three months old may withstand short periods of hypersalinity better than adult trees (McMillan 1974). In the longer term, in hypersaline environments, it appears that while hypersalinity does not necessarily prevent initial establishment, it does inhibit rooting, root growth, and photosynthesis in cotyledons and the early leaves, possibly due to a difficulty at high salinities in the uptake of magnesium (Barhoumi *et al* 2021, McMillan 1974).

The very large cotyledons on juvenile *Avicennia marina* can provide sustenance to the plant for between 45 days and four months (Nguyen *et al* 2015). This enables the plant to develop leaves and start to photosynthesize even when conditions around the plant root system may be hostile. Observations of tip burn and sudden death of some of the juveniles in the dead zone in early November 2022 could be explained by the plants reaching the end of the reserves provided by the cotyledons while the soil conditions were still inhospitable.

Early observations of the dying mangroves at the study site in 2020 led to an initial suspicion of hypersalinity as the cause, rather than waterlogging. This evaluation was based on the death of all the juvenile mangroves in the dead zone, along with the adult deaths. Juvenile *Avicennia marina* in their first year do not possess pneumatophores and the plants may be completely submerged for long periods. Research in Botany Bay (Ashford and Allaway 1995) using 10-month-old juveniles found a continuum of interconnected gas space inside the juvenile plants (including the roots) that has direct connection with the atmosphere via the stomata and spongy mesophyll of the leaves and via lenticels that occur on all internodal stems and on the hypocotyl (stem below the cotyledon scar). The stem lenticels and leaf stomata provide continuity with the atmosphere when tides are low at any time of the day or night, and the leaves may also photosynthesize when under water in daylight hours. The large, interconnected gas space acts as a reservoir allowing gas to be transported quickly to the root zone at any time.

This leads to the estimation that it is unlikely to be waterlogging currently causing tip burn and death of the newly established juveniles. The tolerance of juveniles to high temperatures has been mentioned already, and the deaths did not occur in remarkably hot weather. Hypersalinity or possibly sunburn resulting from the lack of adult canopy, could be a potential cause.

#### 5.2.5 Stress in mature trees

Mature trees in the transition, or stressed zone, exhibited features such as reproductive failure, pale leaves (Figure 3), black spots under leaves, tip burn/dieback in leaves and evidence of heavy herbivory (Figure 4). Some of these may be directly related to hypersalinity in the subsoil, others may be less direct signals of stress.

Studies of *Avicennia marina* growing in naturally hypersaline conditions in South Africa found that trees are significantly stressed in hypersaline sites and show changes to the leaf ultrastructure (Naidoo *et al* 2011). Chlorophyll concentrations in the leaves of trees in hypersaline conditions were much lower than those of trees in fresher conditions. A similar reduction of chlorophyll concentration was observed in the St Kilda mangroves by Dittmann *et al* (2022).

Despite root exclusion of salts as well as leaf excretion of salts, Naidoo *et al* (2011) found the leaves from trees in hypersaline sites in South Africa retained much

higher quantities of sodium ions while displaying lower concentrations of potassium, calcium, and magnesium. Appropriate ionic balances within leaf ultrastructures are essential for, and intimately involved with, all physiological processes conducted within the plants.

Small numbers of leaves from the healthy and stressed zones of the current study area were analysed for several ions in a study reported by Dittmann *et al* (2022). Statistically significant differences recorded by those authors included leaves from stressed areas having higher concentrations of arsenic, iron, sulfur, and zinc than leaves from trees in the healthy zone. The stressed trees had lower concentrations of calcium, copper, manganese, molybdenum, sodium, and phosphorus. The direction of the differences match some, but not all, of the findings of Naidoo *et al* (2011). That may be related to the age of the leaf samples being tested, geology of the sites or to the ionic balance and salinity of the hypersaline brines being observed. In both cases, however, a significant difference was noted between healthy and stressed trees in each study area.

One specific leaf structural change noted by observers at St Kilda related to tiny black spots on the underside of mangrove leaves in the transition (stressed) zone. Dittmann *et al* (2022) identified these tiny spots as a type of necrosis that forms in tissues around the salt excreting glands. As *Avicennia marina* excretes salt through these salt glands from the water they take in daily, it would not be an unreasonable speculation to think that the necrotized leaves may explain McMillan's 1974 observation that *Avicennia* adults are salt excreters and secrete salt in increasing amounts as the salinity increases, up to a tolerance level. It may be that the physical death of these cells marks that threshold.

Devaney *et al* (2021) recently published work on several mangrove species, including *Avicennia marina*, that suggests hypersalinity impacts may also reduce mangrove trees' capacity to cope with low temperatures. This reduction in resilience remained even many months after lower salinity and higher humidity was provided, suggesting recovery from such damage may not be easy. The research did not suggest a causative mechanism. At St Kilda the loss of adult tree leaves in the transition zone in late winter and early spring (Figure 4, left) could possibly reflect some similar type of loss of resilience to extreme weather.

The lack of successful reproduction and presence of chlorotic leaves in the two observed trees in the transition zone over both 2021 and 2022, along with the symptoms of leaf dieback in other trees in the transition zone would be consistent with research on the physiological costs of hyperexcretion of salt.

In late winter and early spring, a swathe of mangroves where the transition zone is closer to the healthy zone were observed to be heavily predated by insects. Stressed plants may mobilise nutrients sought by herbivores, which then attack the plants, further stressing them. In extreme situations the positive feedback between physiological stress and insect attack can cause the plant to succumb (McQuillan, email dated 26 November 2022).

Recently, McQuillan collected an as yet unnamed phycitine moth of the genus *Ptyomaxia* at Port Gawler. This was an unusual finding, as known members of the genus are tropical (SE Asia). The caterpillars are nocturnal, and shelter in the daytime in shelters made by pulling two or three leaves close together with silk. The damage from feeding appears as discoloured patches on the leaves, sometimes with clustered "windows" of missing leaf tissue. This description matched the damage that had been reported at the study site and so a site visit was made in

December 2022, to see whether any caterpillars could be caught. Many old shelters were found, but all were vacated, and the leaf damage was present only on older leaves. The newest growth had not been grazed (Figure 4, right).

#### 5.2.6 Conclusion

Both quantative and qualitative observations suggest that recolonisation of mangroves has been very slow in the study area and is not typical of colonization rates in other sites in the Barker Inlet and Port River estuary. There are also signs of continuing or derivative impacts on the plant life and sediment physical characteristics of the area.

A reason for the current study was to help land managers decide on what actions could be taken to facilitate recovery of the forest. The slowness of natural recovery, low success rates of juveniles (even with the informally augmented propagules), the ongoing indicators of stress in adult trees in the transition zone and the lack of resilience in the benthic mat communities, leads one to caution that removal of the source of the ongoing hypersalinity may be needed, prior to active revegetation. This conclusion is not dissimilar to that reached by Leyden *et al* (2022) who recommended immediate efforts be concentrated on reducing the residual hypersalinity by lowering the hydraulic head difference between the salt ponds and the intertidal forests, and in the longer term assessing the removal of the hypersaline pollution pathway.

It is probable that propagule augmentation, either manually distributed or delivered via drone, may have a beneficial effect on areas of dead mangrove south of Burrows Creek. Those areas were exposed to hypersalinity for a much shorter period, the forest is located further from the source of the pollutant, and the affected areas are a very narrow band of trees. However, based on the speed of mangrove colonisation in other parts of Barker Inlet, if subsoil conditions there have already ameliorated, it is likely the forest could recover relatively quickly.

Barker Inlet contains several locations that host healthy populations of mangroves. Areas including Mutton Cove, Garden Island, Torrens Island, Dry Creek and North Arm could be useful sources of propagules.

Much remains unknown about the impacts of the hypersalinity event in the intertidal habitats at St Kilda. Further work could be undertaken to determine the ultimate receptor for resuspended sediment and any mobilized contaminants. Other aspects that have not been investigated include the impacts on faunal biodiversity, in particular invertebrate and fish communities. Further investigation of the species of moth grazing the mangroves may be warranted, to establish whether the species is native, or a recent introduction from the tropics.

#### 6. References

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# 7. Appendix - Graphs

■12/12/2021 ■13/01/2022 ■12/02/2022 ■26/03/2022 ■8/04/2022 ■28/05/2022 ■25/06/2022 ■27/07/2022 ■28/08/2022 ■29/09/2022 ■11/11/2022



