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Review of past seagrass restoration projects and guidelines for restoration in Cockburn Sound

Theme: Benthic Habitats and Communities
WAMSI Westport Marine Science Program



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ABOUT THE MARINE SCIENCE PROGRAM

The WAMSI Westport Marine Science Program (WWMSP) is a \$13.5 million body of marine research funded by the WA Government. The aims of the WWMSP are to increase knowledge of Cockburn Sound in areas that will inform the environmental impact assessment of the proposed Westport development and help to manage this important and heavily used marine area into the future. Westport is the State Government's program to move container trade from Fremantle to Kwinana, and includes a new container port and associated freight, road and rail, and logistics. The WWMSP comprises more than 30 research projects in the biological, physical and social sciences that are focused on the Cockburn Sound area. They are being delivered by more than 100 scientists from the WAMSI partnership and other organisations.

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DATA

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1 Stage 1: Review of past seagrass restoration projects and guidelines for restoration in Cockburn Sound

Authors

Gary A Kendrick, The University of Western Australia

Rachel Austin, The University of Western Australia

Giulia Ferretto, The University of Western Australia

Mike van Keulen, Murdoch University

Jennifer Verduin, Murdoch University

Project

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Executive Summary

We review past and continuing seagrass restoration projects (1990s to 2020s) in Cockburn Sound and Owen Anchorage with the aim to collate and assess their major outcomes to assist in planning for Westport Project 2.3: Seagrass Restoration. The Westport program would like to 1) with some confidence be able to rehabilitate seagrasses after local and regional impacts of dredging and port development are concluded, and 2) address concerns that seagrass restoration can be shown to be able to restore seagrasses in Cockburn at appropriate scales of 10s to 100s of hectares and the benefits of a seagrass restoration and rehabilitation program outweighs the costs.

There have been over 110 restoration efforts, including experiments and programs, in Cockburn Sound and Owen Anchorage since the 1990s. The review summarises the aims, methods, results and outcomes from the major restoration efforts. Methods of seagrass restoration have been diverse and included using shoots (sprigs, plugs, cores, sods, hessian bags), seedlings and seeds. Over time methods have moved to address cost and scale. Two studies in the 2000s, in Albany and Cockburn Sound, restored 1 to 3 hectares using shoots, respectively. In the 2010s, seed based restoration research became a major focus as shoot (sprig) based methods still were unable to address broader losses at 10s to 1000s hectares and seeds were an alternative cost effective way to address these broader scales. To scale up to 10s to 1000s of hectares, a community-based citizen science restoration program OZFISH 'Seeds for Snapper' has been developed and has demonstrated that with focus and effort this approach is cost effective. We also revisited 31 individual restoration sites and assessed their success.

The major outcomes of the review were: present environmental conditions and processes are extremely important in determining success; success of restoration is both site and time specific and determine the most suitable type of restoration units to be used (shoot, seedling or seed); scaling up requires the environment to be suitable for natural seagrass colonisation or will need modification, and; researchers focus on small scale experiments has not prepared us for scaling up. We also found the language of restoration and rehabilitation did not appear appropriate given that most restoration approaches are augmenting (enhancing) natural recovery processes rather than rehabilitating or restoring a completely modified environment.

Guidelines for present and future work include:

1. Address the suitability of the environment to be restored though: 1) development of restoration suitability modelling and mapping for Cockburn Sound that incorporates our understanding of the biological and physical processes that impact successful restoration, and 2) explore large scale modification to the environment to be restored, like sand capping and top dressing with dredge products, linking artificial reefs to seagrass restoration sites, and removal of bioturbators and grazers.
2. Careful selection of restoration units to be at an appropriate spatial scale and targeted for the environmental conditions of areas to be restored. This will include mechanisation of present units, methods and processes.
3. Understand the bottlenecks in the seagrass life history and enhance survival of restoration units through modifying the environment. Co-planting, seeding and multispecies approaches are to be explored.
4. Increase social capital for restoration through community-based restoration programs and modifying unrealistic expectations about what is a successful restoration program. Community engagement in restoration success builds a large community of practice that influences the political and economic imperatives to restore seagrasses.

In conclusion, our guidelines focus on addressing major limitations to scaling and success in seagrass restoration. Present methods of seed, seedling and shoot-based restoration have been successful in enhancing natural seagrass recovery but need to be now scaled from 1,000m² - hectares to 10s - 1,000s hectares. From a cost-benefit viewpoint, involving community in restoration programs creates a large and scale appropriate community of practice. Community engagement plays a fundamental role in the scaling up of seagrass restoration in Cockburn Sound and Owen Anchorage and should be fostered and grown.

2 Introduction

Cockburn Sound, Owen Anchorage, Parmelia Bank and Success Bank in the southern metropolitan coastal waters near Perth, Western Australia have been the focus for seagrass restoration in Western Australia since the 1980s. Most of the 110+ restoration programs have been small scale covering m² to 1,000 m² with a single exception of 3 hectares of seagrass that was transplanted in the late 2000s (Oceanica 2013) (Figure 1). This review has focused on work since the 1990s to present day (2022) where we have described the goals of the programs, summarised their methods, results and implications, and then we produce a list of outcomes for each study, that can be interrogated when moving from experiment to actual scaled on the ground seagrass restoration.

Cockburn Sound was declared a future industrial zone and major port for Western Australia in the 1950s. Rapid industrial development and uncontrolled effluent release from industry and from the southern metropolitan sewage outfall at Woodmans Point, resulted in a loss of 80% of seagrasses on shallow banks that ring the sound by 1972 (Kendrick et al. 2002). Subsequent public outcry resulted in the state government funding a multidisciplinary scientific study, the Cockburn Sound Study. The Cockburn Sound Study was released in 1978 and recommended stronger management of effluent entering the Sound to control nitrogen and pollutants through tighter environmental regulation of industrial activities.

The loss of seagrasses in Cockburn Sound and Owen Anchorage from the 1950s through to the 1990s set up a political incentive for maintaining seagrasses in these shallow coastal marine ecosystems, to remediate environmental impacts and to rehabilitate seagrass meadows. Large investments and opportunities for seagrass restoration have occurred, including the Cockburn Cement funded programs that scaled up restoration across Success and Parmelia Banks in the 1990s, and the Seagrass Research and Rehabilitation Program (2004-2013) (Oceanica 2013) that addressed the science and technology behind seagrass restoration and rehabilitation. The Oceanica 2013 project resulted in a best practice seagrass manual and a comprehensive report on activities from 2004 to 2012. It is a unique document as it addressed how to restore in an Australian context, rather than the focus on Northern Hemisphere species, especially *Zostera marina*, that accounted for 80% of published literature in seagrass restoration at that time.

After the restoration work summarized in Oceanica 2013, researchers at the University of Western Australia have focused on seed-based restoration and have addressed recruitment bottlenecks in the life history of seagrasses. This has led to the Ozfish led 'Seeds for Snapper' community seed-based seagrass restoration program now active for 4 years with a goal to restore *Posidonia australis* meadows to Cockburn Sound and Owen Anchorage. BMT has also been active in hectare-scale, shoot-based restoration in southern Cockburn Sound and in deeper waters on dredged areas of Success and Parmelia Banks, further developing cost effective commercial approaches to restore seagrasses.

This review is timely, as it aims to collate and assess outcomes from past and existing research and development on seagrass restoration into Cockburn Sound and Owen Anchorage to address:

1. Whether with some confidence we can recommend a restoration package to rehabilitate existing and planned losses of seagrasses in Cockburn Sound and Owen Anchorage
2. Whether that restoration package is at scale (10s-100s hectares) and cost - benefit compatible.



Figure 1: Map of sites where seagrass restoration experiments and trials have been conducted in the greater Cockburn Sound area over the past 40+ years. Such restoration involves a wide range of species, methodologies and results.

3 Globally Recognised Limitations to Seagrass Restoration

The major limitations to existing seagrass restoration activities globally are:

Seagrass restoration rarely has been at the scale of seagrass loss. Exceptions include:

- a. Successful seed-based restoration of eelgrass (*Zostera marina*) to the coastal bays of Virginia, USA (Orth et al. 2020) where eelgrass was restored from seeds over a period of 20 years and now covers 1,000s hectares.
- b. Three ha of *Posidonia sinuosa* and *P. australis* was restored onto Southern Flats, Cockburn Sound from 2006-2012 (Oceanica 2013).

Suitability of the habitat for restoration is rarely addressed.

- a. Light plays a major role in the loss of seagrasses associated with dredging and port activities (Wu et al. 2017) and can impact the success of restoration programs but is rarely documented.
- b. Sediment health, or condition, and scaled approaches to sediment modification need further intensive research. Recent experiments to replace anoxic muddy sediments from years of eutrophication with coarse sands from dredging programs have resulted in diversification and increased abundances of benthic invertebrates (Oncken et al. 2022). Whether sediment modification could be used for seagrass restoration has yet to be tested at scale.
- c. Hydrodynamics at the scale of the seagrass shoot or seedling drives morbidity and mortality in restoration programs but is rarely determined before restoration (Statton et al. 2017a).
- d. It is important to factor in natural recruitment in restoration programs, and this requires an understanding of connectivity within the system (Ruiz-Montoya et al. 2015, Kendrick et al. 2016, Sinclair et al. 2018).

Monitoring of restoration has been generally limited to months to a few years.

- a. Comparisons with Australian and US restoration projects indicated a median of 6 months to 2 years (Statton et al. 2012, van Katwijk et al. 2016) suggesting little investment in quantifying the ecological value of the restoration activity.
- b. This has been attributed to the poor social perceptions of success that drive social license, and cost to restore (Abelson et al. 2020).

Successes in seagrass restoration have not been front and centre in community perceptions.

- a. Recent reviews of seagrass restoration in Australia have included social perceptions and citizen science as well as addressing cost effectiveness (Tan et al. 2020, Sinclair et al. 2021).

4 A History of Restoration Research and Development

4.1 Pre – 1990s

Until the mid-1990s, many seagrass transplantation techniques used successfully elsewhere in the world were attempted in Western Australia, but with limited success (Gordon, 1996). The difficulties and knowledge gaps surrounding seagrass transplantation in Australia were reviewed by Kirkman (1992) and he emphasised slow rhizome growth in our major meadow-forming species as a particular bottleneck towards large scale restoration projects. For example, Kirkman (1998) documented numerous unsuccessful transplantation experiments, using a variety of seagrass species, and a variety

of transplanting methods. Paling et al. (2000) noted that, of some 7,500 planting units (PUs) transplanted, most had washed away due to high levels of water motion. Many of these studies were not formally published and exist only in the grey literature; a substantive review was conducted by Paling and Gordon (1995) but is itself difficult to access. An overview of the early Australian literature on seagrass restoration was provided by Paling & van Keulen (2002).

The outcomes of these studies were:

- Seagrasses were hard to restore using seedlings and shoot transplants
- The environmental drivers that disrupted successful seagrass seedling and adult transplanting were: 1. physical exposure of sites to shear stress that removed units, and; 2. Disturbance by marine animals (crabs, rays).

4.2 1990s

4.2.1 Early Sprig and Plug Transplanting on Success Bank

Experiments showed that firmly anchoring planting units (PUs) improved survival, and that sediment stability was an important factor in transplant success (Hancock, 1992; Nelson, 1992). In 1994 a study was conducted to examine the influence of PU size and sediment stability on transplant survival (Walker, 1994). In areas of higher water movement, plugs were found to perform better than sprigs. It was hypothesised that the larger the PU the better the survival, through improved sediment stability as a result of increased biomass, and reduced disturbance to the roots and rhizomes. The influence of sediment stabilisation was also examined, including the fluidisation of sediment that can occur during the transplantation process. Improved survival of PUs was also achieved by transplanting them into existing meadows of minor species, such as *Heterozostera nigricaulis* (Walker, 1994; van Keulen et al., 2003) or *Halophila ovalis* (Parker, 2020) (Figure 2). These initial studies, examining the role of transplant size and sediment stabilisation on survival, led to the conclusion that larger transplants survive better, and that sediment stabilisation is an important factor in transplant survival (van Keulen et al. 2003) (Figure 3).



Figure 2: *Posidonia coriacea* transplanted into three substrates (1) *P. coriacea* meadow, (2) *Halophila ovalis* meadow and (3) bare sand (from van Keulen et al. 2003)

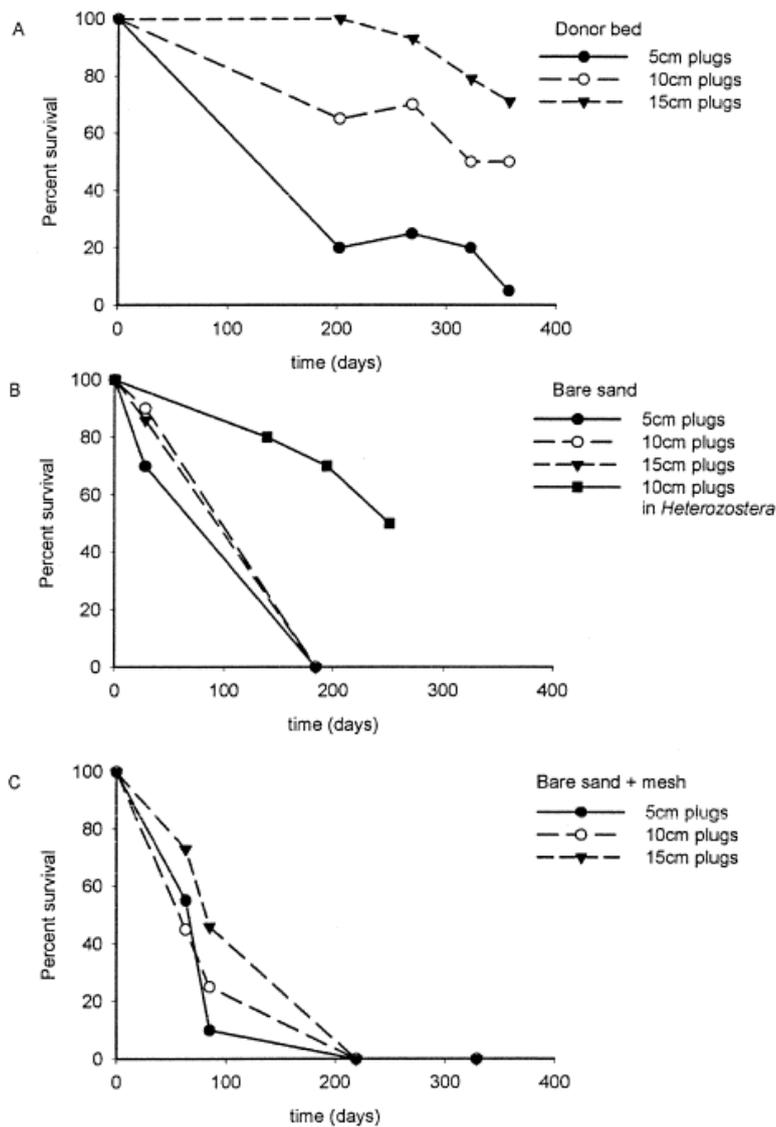


Figure 3: Percent Survival of *Posidonia sinuosa* plugs of diameter 5, 10 and 15 cm at treatments of (A) donor bed, (B) transplanted to bare sand and, (C) transplanted to bare sand with mesh. Forty replicates were used in the 5 and 10 cm plugs and 28 replicates of the 15 cm plugs. 10 cm plugs were used in the *Heterozostera nigricaulis* meadow (from van Keulen et al. 2003). Periodic burial of transplants is shown by declines and increases in the number of surviving transplants over time.

The outcomes of these studies were:

- Larger plugs (10-15cm diameter) had greater survival rates
- Plugs transplanted into donor meadow had greatest survival rates
- Transplanting plugs into existing meadows of minor species also improve survival rates

4.2.2 *Ecosub I and II and the Planting of Sods*

This preliminary work gave rise to the concept of mechanical transplantation of large blocks (sods) of seagrass (Paling et al. 1998). ‘ECOSUB1’ was the prototype developed as part of the Environmental Management Plan for Cockburn Cement Ltd., which mines shell sand on Success Bank. Improvements in the technology led to ECOSUB2, which consisted of two machines operating simultaneously, one

harvesting seagrass and the other planting it (Figure 4 top). A sod shuttle was used to transfer sods from one machine to the other which greatly increased the efficiency of the operations (Figure 4 bottom). The theoretical maximum output of ECOSUB2 was 75 sods, or 40m², per day (Paling et al. 2001a b). Survival of transplanted sods was generally high, although this varied depending on planting season (Figure 5).

Experimental work to supplement the mechanical transplantation program included studies on transplanting seagrass to different depths (Paling et al. 2000), and an examination of wave energy effects on survival (Paling et al. 2003). The depth studies suggested that it was possible to transplant seagrasses to greater depths than donor sites, although wave energy was still instrumental in lowering survival. To examine wave energy effects, mechanically transplanted sods were deployed in an area of high wave energy, with spacing being used to modify the hydrodynamic regime. The results indicated no significant differences in survival or shoot density as a result of the spacing treatment, with poor survival once winter storms impacted the area (Paling et al. 2003). Monitoring of fluctuations in sediment height showed the presence of large sand waves passing through the region, and this is clearly a factor that needs to be considered when transplanting seagrass to areas of high wave energy.

Transplanting seagrasses in the approach to winter has not been successful, largely because of the erosive effect of winter storm events (Paling et al. 2003). The sediment surrounding PUs can remain fluid for long periods of time after transplantation (in the order of many weeks), making the PUs highly susceptible to exposure through erosion, and survival is reduced as a consequence (van Keulen & Paling 2002). Sediment dynamics therefore should be a major consideration in any seagrass rehabilitation program, particularly in high energy areas.

The studies suggest that larger transplants (sods) are required when transplanting to areas of high wave energy, although smaller transplants (plugs and sprigs) may be appropriate in more moderate conditions (Paling et al., 2007). Major factors to consider in enhancing rehabilitation success are the appropriateness of the technique in regard to species, seasonality of weather and the range of sediment level fluctuation.

The outcomes of these studies were:

- Survival of transplanted seagrass is reduced during winter months as sediment remains fluid for many weeks following transplantation
- Sediment dynamics was an important consideration, particularly in high energy regions
- Larger transplants (sods) are required for areas of high wave energy and plugs and sprigs are appropriate in more moderate conditions
- When transplanting you need to consider the appropriateness of the technique in regard to species, seasonality of weather and range of sediment level fluctuation



Figure 4: ECOSUB 2 was an example of mechanising and industrialising seagrass restoration and rehabilitation. Top image shows the harvester (front) and planting unit (back) on a truck trailer. Bottom left shows the planting unit and a line of transplanted sods. Bottom right shows the shuttle unit and sods being placed into the planting unit.

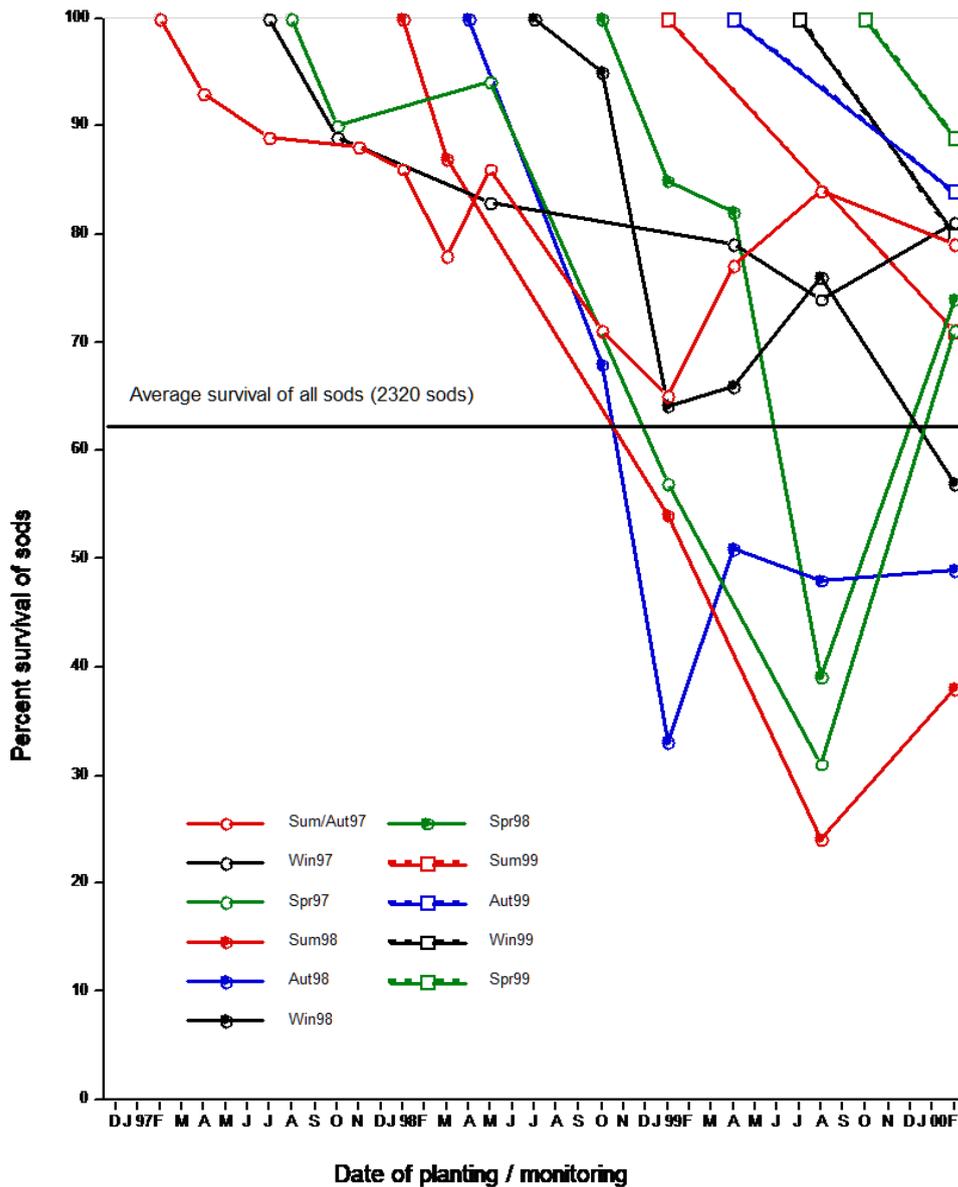


Figure 5: Mean percent survival of all sods transplanted mechanically. For clarity, planting years have different symbols (1997 open circles, 1998 closed circles, 1999 open squares), and planting seasons have different colours (summer red, autumn blue, winter black, spring green) (Paling et al. 2001a).

4.3 2000s

4.3.1 Southern Flats Large Scale Transplant Trial

Southern Flats, Cockburn Sound is where the large-scale seagrass rehabilitation area is located (Figure 6) (Oceanica 2013). This area provided the greatest opportunity for success in relation to planting conditions (i.e., depth, hydrodynamics, large unvegetated sandy areas, water quality) and field logistics. There have been continual advancements in techniques associated with large-scale transplanting exercises. These include sprig removal, handling and tying techniques, alignment of plant growth axes into the dominant direction of swell, sprig spacing, protection from the sun and retaining

harvested sprigs underwater. The greater biomass of *P. australis* made it more robust for handling and anchoring than *P. sinuosa*, resulting in it being the target species for these transplantation exercises.

Donor sprig material was harvested from an area partially dredged on Parmelia Bank at a depth of 5–6 m (Oceanica 2013). Sprigs were harvested from the donor material after it was brought to the surface. Each sprig was then tied to a purpose designed wire peg (30 cm in length) using two or three biodegradable cable-ties. Sprigs were kept under water as much as possible and wire pegs were collated into groups of five before being secured with string to enable accurate quantification, transport, handling and planting at the recipient site.

Initially, configurations of two hectares consisting of 20,000 sprigs placed 1 m apart were completed in the 2005/06 season at the rehabilitation site (Oceanica 2013). However, due to sprig survival issues and the desire to maintain the two hectare planting configuration, a total of 36,000 sprigs were added into the 2 hectare area. In 2006 it was decided that doubling the density of transplanted sprigs within the target area would increase both efficiency and survival. This resulted in the density changing from 1 sprig m^{-2} (i.e. 1 m spacing) to 4 sprigs m^{-2} (i.e. 0.5 m spacing). In areas where survival was high, this consisted of planting between existing sprigs (i.e. planting took place to configure a meadow with 0.5 m spaced sprigs). In areas with low survival, planting took place at 0.5 m since there were no existing sprigs to plant in-between.

Large-scale seagrass rehabilitation exercises at Southern Flats occurred over four separate summers between 2004 and 2008, yielding a total area of 3.1 hectare. As it is difficult and impracticable to regularly assess the entire number of sprigs planted, randomly selected 10 m x 10 m plots were monitored. Survival counts of these plots were undertaken to determine the number of sprigs in each of the plots. These plots were not infilled at any stage in order to accurately produce long-term survival data, however they may underestimate true (total) sprig survival as some of the material used was not subject to improved techniques.

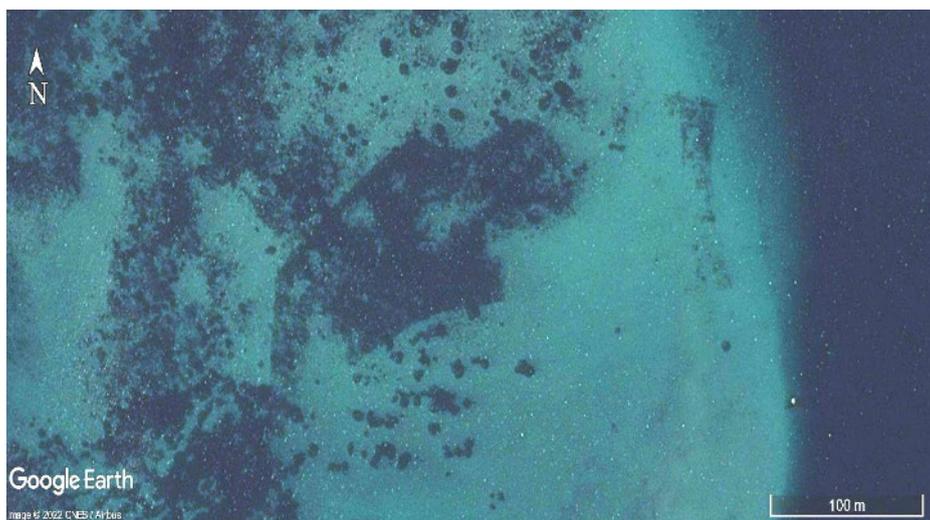


Figure 6: Aerial imagery of the Southern Flats transplant site from 28/2/2013 showing the significant growth after 6.5 years. The straight lines of the three 1 hectare plots can be seen in contrast to the round naturally recruited seagrass patches.

In addition to the monitoring of plots, a monitoring exercise was carried out by divers, using three joined 1 m quadrats in order to monitor survival of transplanted sprigs. The western hectare, which

was most recently planted in the summer of 2007/08 was monitored for growth using 10 m x 10 m plots. In addition, a count of the two previously planted hectares (middle and eastern) was undertaken in May/June 2009 and a holistic count of the third hectare finalised in April 2010.

In April 2010, monitoring results of selected 10 x 10 m plots of the westernmost hectare showed survival rates of 100%, 25–29 months after planting (Figure 6.7). This indicates that most sprigs in this hectare have continued to grow and merge over the 2009/10 period. During the monitoring of the representative plots in July 2010, many sprigs were flowering for the first time. The density of shoots in these plots was 100 ± 10.4 shoots m^{-2} .

Overall survival of the eastern hectare is 23.3%. In comparison, the middle and western hectares (planted between 2005 and 2008) were growing well, with overall survival rates of 86% and 87%, respectively. Shoot density of sprigs spaced 0.5 to 1 m apart were 103.2 ± 11.2 to 109.6 ± 2.98 shoots m^{-2} , respectively during July 2010. These results approximate naturally occurring shoot densities of 100.8 ± 7.08 shoots m^{-2} in *P. australis* during winter.

Improved transplanting techniques have enhanced the survival of transplanted sprigs. This was particularly prevalent in the middle and western hectares. Visual observations, validated by aerial photography from April 2009 and 2010 (e.g. Figure 6), found a change in survival rates for eastern and middle hectares in 2009 and for all three hectares in 2010. The middle hectare had notably expanded and adjoining sprigs accompanied by a survival rate of >90%. The western hectare also showed high overall survival, with survival increasing to >85% after three years of growth. The eastern hectare had lower sprig survival (23%) compared to that of the middle and western hectares. Some variability in survival and growth was evident across the hectares and within years. Lowest survival in the eastern hectare was found in the eastern part of the hectare. Sprig growth in this area was affected by algal growth. Differing biological and abiotic conditions appear to factor into sprig survival rates. Although grain size and chemical analyses of sediment sample investigations were not conclusive, results demonstrate that the recipient site is as important as healthy donor sprig material in maximising sprig survival and growth.

The outcomes of this study were:

- The western hectare had survival rates of 100% 25-29 months after planting. Since has reduced slightly to 86% and 87%, respectively, after three years of growth
- Eastern hectare had survival of 23.3%. Seagrass growth may be affected by algal growth, as occurred in eastern hectare
- Many sprigs were flowering during July 2010 monitoring
- Shoot densities similar to that of naturally occurring shoot densities of *P. australis* during winter
- Recipient site is important and the health of donor sprig material maximises survival and growth

4.3.2 Donor Meadow Recovery Experiment

It may not always be possible or appropriate to source transplantation material from seagrass meadows that are to be dredged. Prior to removal of materials from natural meadows it is recommended that investigations are undertaken to ensure donor meadow recovery is viable. This is particularly important for large-scale extraction projects where mechanical extraction (e.g. plugs) of natural meadows may result in considerable short-term damage to the donor meadow. To understand this issue, two years of data have been collected from previous donor meadow recovery trials, with a final round of monitoring undertaken in December 2008 (Verduin et al. 2011).

Previous donor meadow recovery trials have been set up in Owen Anchorage in 2004 and expanded in 2005. In December 2006, an alternative donor meadow recovery trial was set up in an area south of the mass transplant hectares in Southern Flats. Prototype III of the pre-mechanical SeaPot corer (8.3 cm diameter) was used to remove cores of *Posidonia australis* and *P. sinuosa*. Three density configurations of plug removal were examined: “Line 5” (five plugs were removed in a 1.25 m row), “Block 5” (five plugs removed in a square metre) and “Block 9” (nine plugs were removed in a square metre) (Figure 7). Metal rings (8.3 cm Ø) were placed where the cores were removed to monitor shoot recovery into the newly bare area, and were also placed into the adjacent undisturbed meadow to act as controls for shoot density. The recovery of shoots over 24 months into hole configurations of Line 5, Block 5 and Block 9 were compared for *P. australis* and *P. sinuosa* using a two-way ANOVA (species x configuration) and a post-hoc pair-wise comparison of the means was performed using either Tukey’s HSD test or the Student’s t-test.

After 24 months of monitoring both species showed an increase in shoot density in the first 13 months. *P. sinuosa* showed significantly better recovery than *P. australis* in the latter 11 months. There was no difference between Line 5, Block 5, and Block 9 configurations for either species. *P. sinuosa* shoot recovery into the rings for each configuration was an average of 2.2 shoots over 24 months. The controls showed a change of ~2.3 shoots over the same period. *P. australis* shoot recovery into the rings for each configuration was an average of 0.8 shoots in 24 months with the controls showing a change of ~1.5 shoots over the same period.

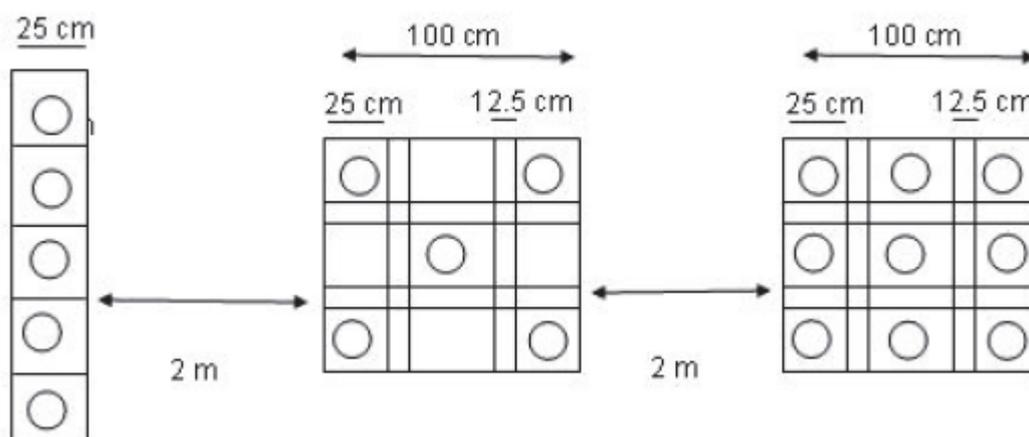


Figure 7: Experimental layout of plug removal to test donor meadow recovery for three treatments: (from left to right) “Line 5”, “Block 5” and “Block 9” (Verduin et al. 2011).

The outcomes of these studies were:

- No significant differences became apparent between the three densities of core removal after two years of observation.
- This suggests that using different coring density configurations does not appear to impact meadow recovery.
- Predicted recovery time for *P. australis* and *P. sinuosa* based on 24 months of monitoring is estimated at 3 years and 2.5 years, respectively.

4.3.3 *Depth-Dependent Seagrass Rehabilitation*

Cost-effective techniques, such as the one developed for the Southern Flats rehabilitation exercise, needed to be tested across the depth range of naturally occurring meadows. To investigate this, a transplantation experiment was conducted south of Woodman Point. This consisted of transplants to five different depths (2-9 m), to test if depth is a limiting factor in newly transplanted sprig survival and growth (Oceanica 2013) (Figure 8).

In May 2009 individual *Posidonia australis* sprigs were collected from an area designated to be dredged on Parmelia Bank, Cockburn Sound. Sprigs were sorted on board the vessel on the basis of length of rhizome (minimum 15 cm) and condition of shoots, and then attached to purpose-built iron staples (Oceanica 2013). Three replicate 5 m x 5 m plots were set up at each depth (2, 4, 6, 8 and 9 m) situated along a gradient approximately 200 m long. A spacing of 0.5 m between individual sprigs was used within these plots. Monitoring counts were undertaken after five months, 12 months, 24 months and again in June 2012 three years after initial planting. Individual shoots were counted for each of the 5 m x 5 m plots and divided by the original number of shoots initially planted to obtain percentage survival. Shoot densities were obtained using fifteen 25 cm x 25 cm quadrats per replicate plot at each depth.

Shoot density at all depths increased over the first year to 18 months. However, during the winter period in June 2011, aside from the 2 m depth, all shoot densities decreased in the 6 months following the last monitoring period. In June 2012 further reduction in shoot densities, including the 2 m site occurred. Shoot density at 2 m depth increased steadily from the initial 11 shoots m⁻² to 370.7 ± 33.4 shoots m⁻² over the first two years, 2013 monitoring saw a slight decline from 370.7 ± 33.4 (or 91.2 % survival) to 284.8 ± 21.8 (or 71.2 % survival, similar to the survival rate one year after initial planting).

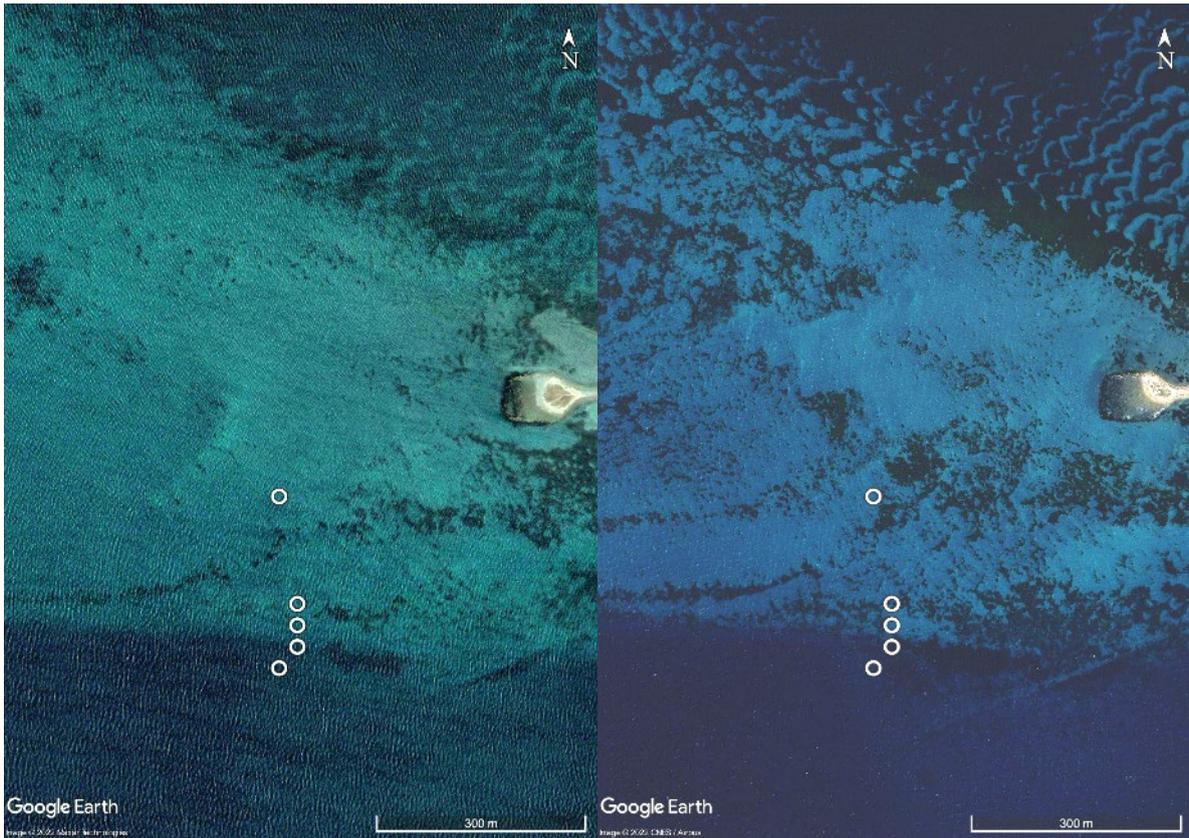


Figure 8: Aerial imagery of the Woodman Point area from 24/12/2006 (left, before) and 06/2022 (right, after) where the depth dependent seagrass trials were conducted. Since the time of the trial the transplants have continued to grow in the shallower sites, but a large amount of natural recruitment and growth has also occurred in the area.

Survival, expressed as the percentage of the total of planting units (PUs) that were still present, remained high for 2 m depth (71 %), however at all other depths survival was < 40% after three years. There is a significant difference in shoot density between all depths ($p < 0.001$) (Oceanica 2013). During monitoring in June 2012, visibility was limited to a maximum of 1.5 m. A large plume extended along the entire length of the experimental site set-up. The sprigs at 6 m appeared healthy but at 8 m and 9 m plants appeared stressed, and whereas in June 2011 they often had only one shoot or leaf per sprig, in June 2012 many plants had perished. *Heterozostera* was often found between the sprigs at the 9 m site. Sprigs were merging with adjacent sprigs at 2 m, 4 m, and 6 m, but not at 8 m and 9 m. In June 2012 sprigs were looking healthy at 2 m, 4 m and 6 m depth. The most western replicate 5 m x 5 m plot at 8 m depth, which had a significantly lower survival rate of 15 % in the previous year has now perished bringing the average survival down from 30.1 % in 2011 to a total of 15.2 % in June 2012 at 8m.

Marked differences between depths after 36 months of growth showed a tendency for shoot numbers to decrease with depth, with the exception of 8 m and 9 m. Growth rates appeared to be greatest at 2 m, 4 m and 6 m, with up to 24 shoots per sprig. This suggests that depth is a limiting factor on the survival and growth of seagrass sprigs. As light is a key factor limiting natural depth of many seagrasses (Cambridge and Hocking, 1997). Considering donor sprigs were collected from 5-6 m depth and both donor and recipient sites experienced similar wave energy, it was suggested that replanted sprigs planted at similar recipient depths may experience less stress and hence have greater survival. However, this trend was not maintained across subsequent monitoring periods with only sprigs

planted at 2 m consistently increasing in size and shoot density.

The transplanted areas were close to natural densities reported for *P. australis* at depth (9 m, 190 shoots m⁻²: Cambridge and Hocking 1997). Survival at depths excluding 2 m was less than 40%. The data suggests that rehabilitation sites on Southern Flats should remain around 2-4 m, which appear to be optimum for new growth. The ability of *P. australis* to recover to near natural densities at deeper sites proves promising with regard to potential effects of sea level changes and the ability of seagrass to adapt and inhabit to deeper areas in the future.

The outcomes of this study were:

- At all depths shoot densities increased until 18 months
- After 18 months, all depths except 2 m decreased in shoot density
- After 36 months, 2 m showed decreased shoot density
- Survival remained high (>70%) at 2 m but was low (<40%) for all other depths after 3 years
- Significant difference in shoot density across all depths (p<0.001)
- Depth and light are likely limiting factors on sprig growth

4.3.4 Amphibolis Seedling Recruitment using Spiral Pegs in Shoalwater Bay

Seagrass rehabilitation often focuses on the use of apical sprigs or plugs from healthy donor meadows. Alternative, non-destructive methods may be necessary for successful rehabilitation such as of seedlings. *Amphibolis* spp. have seen rehabilitation efforts using seedlings trialled in Australia. The success of these methods vary, with the most efficient method in allowing seedlings to anchor appearing to be hessian bags (to mimic the fibrous *Posionia* rhizome material which they are often seen attached to), though survival rates still appear to be quite low (Wear et al., 2010).

Another possible method is using a specially designed spiral wire peg into which a seedling is inserted then pegged into areas that need enhanced recruitment (Figure 9). In June 2010 this was trialled in Shoalwater Bay using *Amphibolis antarctica* seedlings (n = 85) collected from nearby beaches. The site that was selected was protected in the lee of an existing *Amphibolis antarctica* meadow. After one week all 85 seedlings were still present. After two years 29.4% of seedlings were still present and had become well established.

The outcome of this study was:

- Using coiled wire for *Amphibolis* seedling recruitment found a survival rate of 29.4% after two years.



Figure 9: Coiled wire design for *Amphibolis* seedlings for secure attachment of seedlings to sediment (Shoalwater Bay, June 2010) (Wear et al. 2010).

4.3.5 *Posidonia sinuosa* and *P. australis* transplants at Garden Island

Donor and transplant recipient sites were identified on the eastern edge of Garden Island in October 2003 and two transplant sites were set up in December 2003. One site was located within the old urchin scar in Buchanan Bay and the other within a 'blow out' in Luscombe Bay. Two sites were established in Luscombe Bay, within 50 m of each other, as the original site was too small for all treatments. Experimental transplanting of *Posidonia* plugs and sprigs was carried out in January 2004. A total of 192 plugs (96 of each of *Posidonia australis* and *P. sinuosa*) and 432 sprigs (216 of each of *Posidonia australis* and *P. sinuosa*) were collected from one of the donor sites on Parmelia Bank (S 32.13494°, E115.70646°) and transplanted to the two recipient sites. For each species, and at each site, the following were transplanted:

Plugs:

- 8 plugs, to be monitored regularly for survival, growth rate and shoot density.
- 40 plugs to be removed at predetermined intervals and measured for changes in root and rhizome growth.

Sprigs:

- 18 sprigs, to be monitored regularly for survival and growth rate.
- 90 sprigs, to be removed at predetermined intervals and measured for changes in root and rhizome growth.

The sprigs were harvested from the middle of the meadow (non-apical growth) and from the edge of the meadow as single-shoot and 4-shoot sprigs. Sprigs and plugs were secured with plastic-coated wire bent into a U shape. Initial samples were taken of plugs and sprigs to monitor morphometric changes such as root length, development and weight. Monitored attributes included:

- Survival of transplants (including those yet to be removed),
- Shoot counts in both sampled and remaining plant units (early spring, late autumn, to include main growth periods),
- Extension rate (based on the datum peg placed initially to anchor the units),
- Morphometric data (root and shoot development) on sampled plant units, and
- Sediment stability.

Monitoring was conducted regularly for non-destructive sampling. Destructive sampling occurred at one month, 6 months, one year, two years and three years. The sampling protocol is described in detail in Oceanica (2013). The sites were visited one, six and 12 months after establishment and all transplants were monitored. Six- and twelve-monthly sampling data were collected. Survival varied from 0 to 92% depending upon transplant type. After the first month, 75-92% of *P. australis* transplants had survived, and 25-67% of *P. sinuosa* transplants had survived. At Buchanan Bay, urchins had grazed many of the transplants, and while *Posidonia australis* was relatively unaffected, showing only partial grazing on the leaf edges, *Posidonia sinuosa* transplants were badly grazed. This apparent difference in urchin grazing preference could explain the difference in the survival rates, indicating that grazers can have a significant impact on restoration success which needs to be investigated prior to ensure success. Four shoot sprigs had a better survival than one-shoot or non-apical sprigs and this is probably due to more leaf material being present. Plugs of both species did not fare well in relation to sprigs at this site. Survival after six and twelve months for *P. australis* was 0 to 83% survival and 0 to 66.6% survival respectively. *P. sinuosa* sprig survival after six months was 16.6 to 50% and after twelve months 0 to 33%.

Plug survival in both species at Luscombe Bay (50 to 81%) was far better than at Buchanan Bay (12.5 to 50%). However, sprig survival in both species at this site (28 to 67%), where no urchins were apparent, was comparable to the grazed *P. sinuosa* at Buchanan Bay for the first month (25 to 67%, Table 1). A probable reason for the poor sprig survival at Luscombe Bay, in the absence of grazing, was the development and migration of sand ripples through the site. These ripples, which were approximately 10 to 15 cm high, were not present when the transplants were planted. A sediment height variation of this magnitude would be sufficient to dislodge sprigs held down by the wire pegs. It was originally considered that this site was in a calm area and it appears that ripples developed in response to east or north-easterly winds.

Table 1: Percentage survival of plug and sprig transplants at Buchanan Bay and Luscombe Bay after 1, 6 and 12 months (Oceanica 2013)

Species	Buchanan Bay (Urchin scar)			Luscombe Bay		
	1 month	6 months	12 months	1 month	6 months	12 months
Posidonia australis						
1 shoot sprigs	81	67	17	31	0	0
4 shoot sprigs	92	83	67	67	33	17
Non-apical sprigs	75	33	17	28	17	17
plugs	50	12	12	81	62	50
Posidonia sinuosa						
1 shoot sprigs	36	33	33	28	17	17
4 shoot sprigs	67	50	0	61	50	33
Non-apical sprigs	25	17	17	47	33	33
plugs	31	25	12	77	62	62

Results from the morphometric analysis illustrated that there were differences in growth patterns between the two species at the different sites. For example, *P. sinuosa* exhibited greater rhizome biomass, number of roots and leaves at the Buchanan site compared to Luscombe Bay. On the other hand, *P. australis*, demonstrated higher rhizome growth at Luscombe Bay in contrast to Buchanan Bay, with *P. sinuosa* producing the most rhizomes overall.

The outcomes of these studies were:

- Survival was variable, and for different reasons at the two sites studied. Biological (grazing) and physical environment (hydrodynamics) factors affected the two sites differently.
- The Buchanan Bay site was affected by urchin grazing, which appeared to affect *P. sinuosa* more than *P. australis*. Plug transplants were grazed more severely than sprig transplants.
- The Luscombe Bay site was affected by sediment movement related to seasonal wind-wave action, with *P. australis* more severely affected than *P. sinuosa*. Sprigs (both single- and four-shoot transplants) were more severely affected than plug transplants.
- Growth of the two species varied at the two sites, with *P. sinuosa* growing better at Buchanan Bay than Luscombe Bay, and *P. australis* performing better at Luscombe Bay than Buchanan Bay.
- Multiple-shoot sprig transplants performed better overall than single-shoot sprigs for both species.

4.3.6 Seagrass Function and Growth – Macronutrients (N and P) and chelated iron additions, Albany

One of the discussion points during the development of the Seagrass Research and Rehabilitation Plan was about the role of macronutrients (N and P) in growth and development of seagrass transplants. In March, 2004 field experiments were set up in Albany to address the effects of sediment enrichment with macro-nutrients and chelated iron on seagrass growth in two different coastal embayments,

Princess Royal Harbour and Oyster Harbour. Princess Royal Harbour suffered from seagrass loss in the 1980s associated with port expansion, dredging and nutrient pollution from industries but is a marine embayment without large riverine inputs. Oyster Harbour also suffered seagrass die-off in the 1980s but the driver for that loss was poor catchment management of the Kalgan and King Rivers and over fertilisation with macro-nutrients that then washed from catchment to the estuary.

A detailed outline of methods is described in the 2013 Seagrass Rehabilitation Report (Oceanica 2013). We ran two levels of experiments: a longer-term two-year experiment focused on N, P and chelated Fe additions to *P. australis* transplants in Princess Royal and Oyster Harbour and; 2 one-year long experiments comparing transplant and root growth in *P. sinuosa* and *P. australis*. The results from the two-year experiment indicated that nitrogen limitation was occurring in Princess Royal Harbour (Figure 10) whereas in Oyster Harbour, phosphorus was limiting plant growth (Figure 11) (Cambridge & Kendrick 2009). The addition of chelated Iron (Fe EDTA) produced equivocal results in both Harbours and may be related more to the interactions between the macro-nutrients and the chelator EDTA than with Fe.

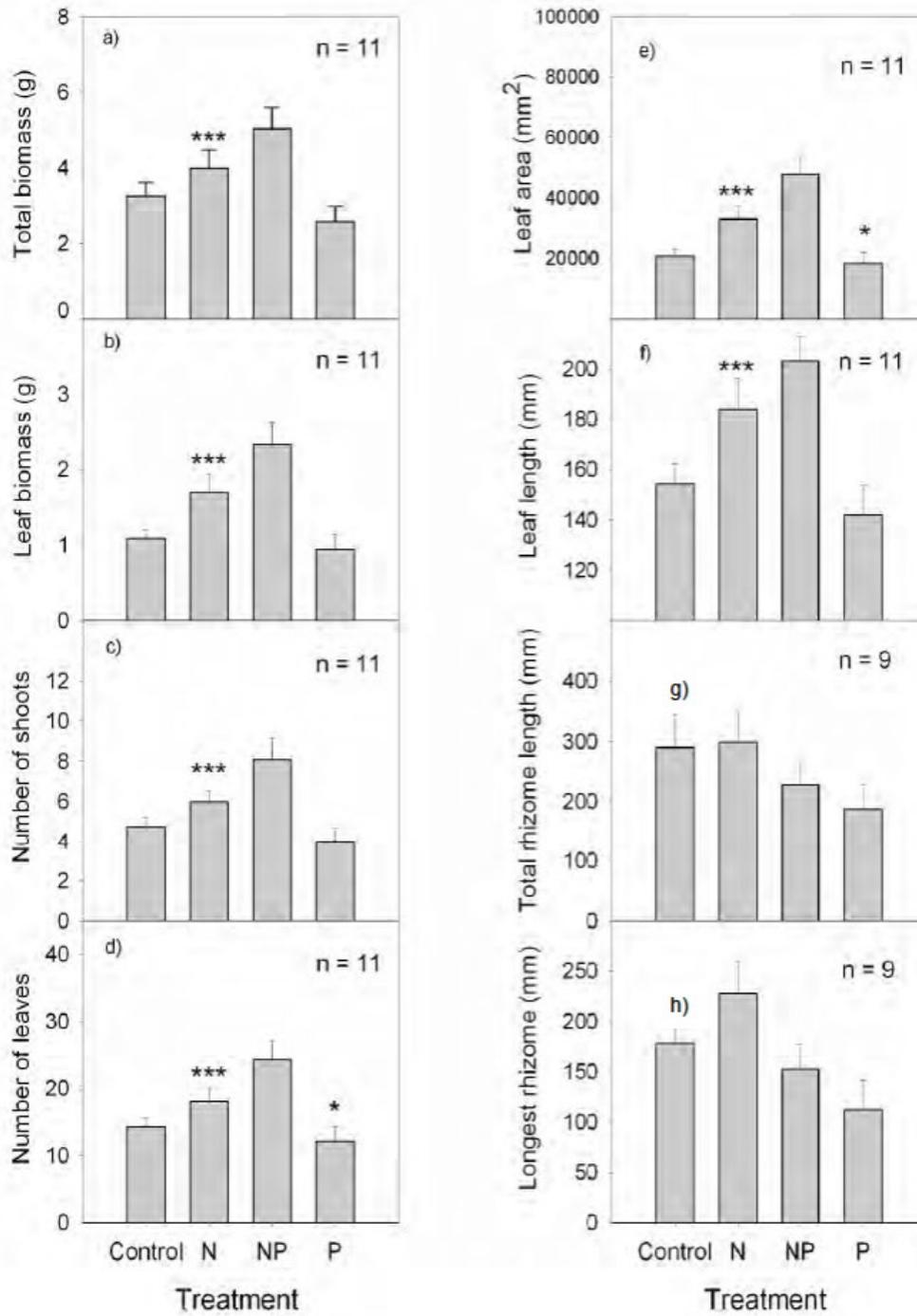
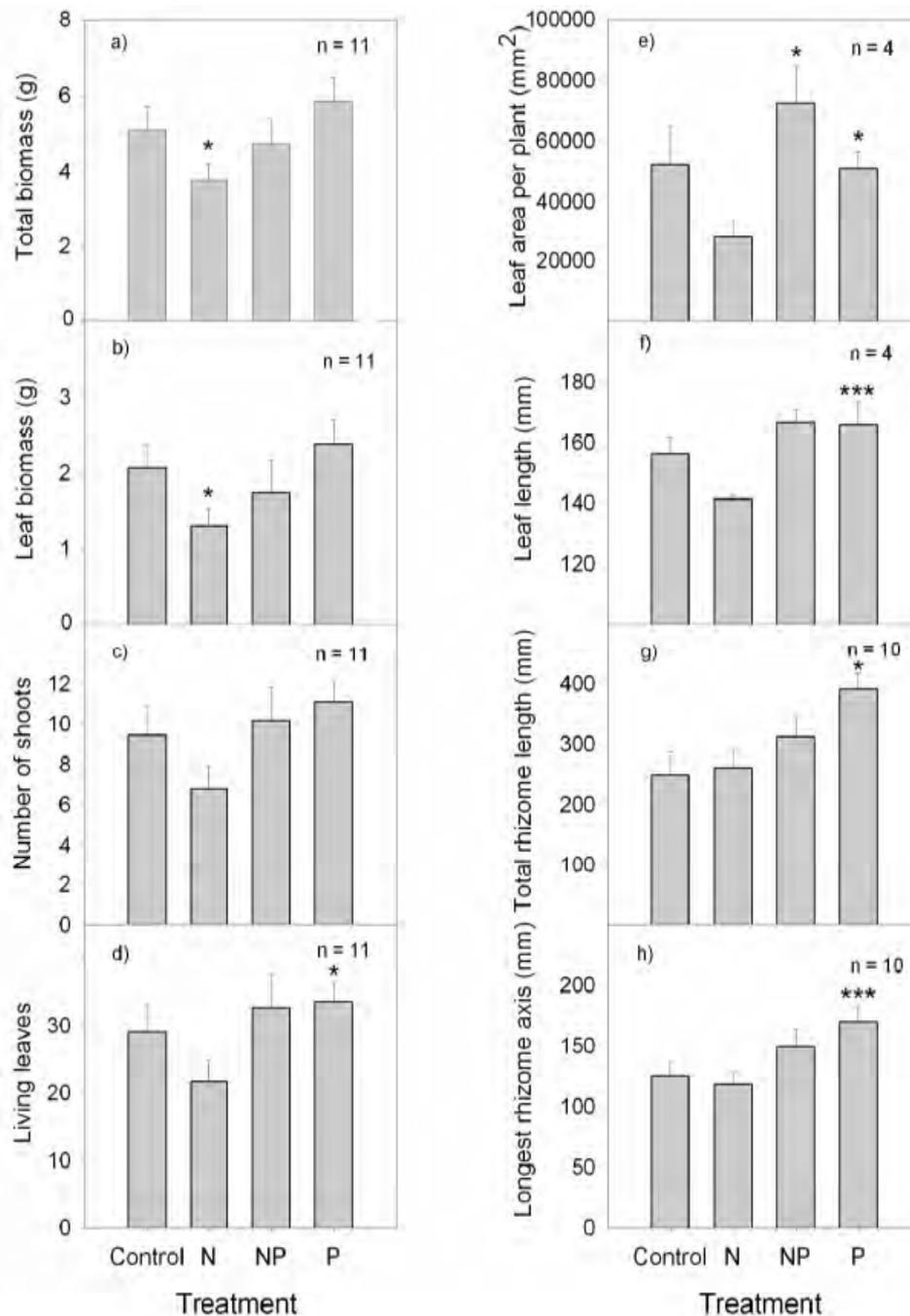


Figure 10: Princess Royal Harbour: Responses in growth and morphology of *Posidonia australis* transplants to sediment nutrient addition (means \pm SE) (from Cambridge and Kendrick 2009)



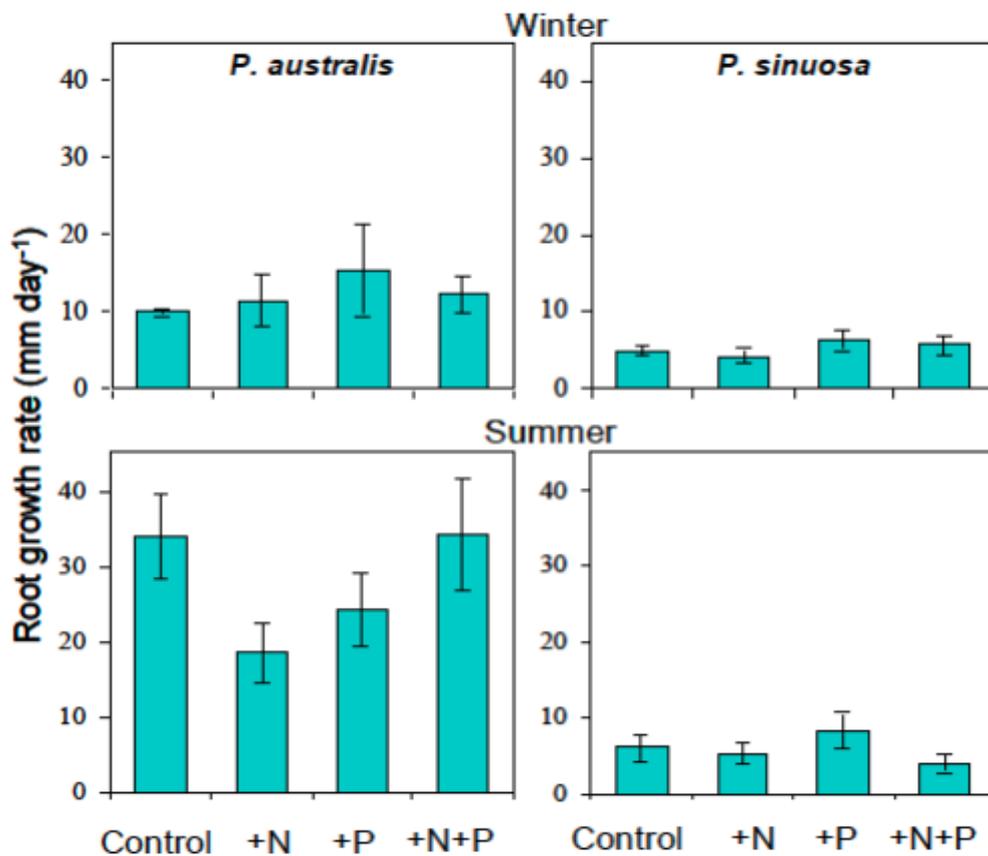
Notes:

1. Total biomass (g dw) per transplant
2. Leaf biomass (g dw) per transplant
3. Number of shoots per transplant
4. Number of living leaves per transplant at harvest
5. Leaf area (mm²) per transplant
6. Leaf length (mm) per transplant
7. Total rhizome length (mm) per transplant
8. Longest rhizome axis (mm) per transplant
9. Statistically significant differences indicated * ≤0.05, ** ≤0.01, *** ≤0.001

Source: UWA

Figure 11: Oyster Harbour: Responses in growth and morphology of *Posidonia australis* transplants to sediment nutrient addition (means ± SE) (from Cambridge & Kendrick 2009).

A large database was also made of sediment nutrients that supported the experimental outcomes. The 1-year comparison of growth and nutrient addition to transplants of *P. sinuosa* and *P. australis* indicated no significant difference in rhizome, shoot, leaf or root growth among transplants with the addition of nutrients or iron, compared to control treatments (Hovey et al. 2012). In the second experimental planting, roots grew over the year, *P. australis* roots were twice as long as *P. sinuosa*, and *P. australis* had more complexity with substantial secondary and tertiary roots (Figure 12) (Hovey et al. 2011). In these experiments, root growth was not nutrient-limited. Based on the trends observed in the study, *P. australis* transplants were better planted in the spring whereas there were no significant differences between spring and autumn plantings in *P. sinuosa*. Root growth was observed to be reduced under high ambient nutrient levels.



Source: UWA

Figure 12: Top, Response of growth of roots of *P. sinuosa* and *P. australis* to nutrient additions (mean \pm SE, $n = 4$) and Bottom, differences in total root length and production of secondary and tertiary root between the two species (from Hovey et al. 2011, 2012)

The outcomes of this study were for *Posidonia australis* and *P. sinuosa*:

- The addition of nutrients to sediments was of little or no benefit to the growth of transplants and sometimes was detrimental.
- The results indicated that knowledge of background sediment nutrient levels is required as adding nutrients to sediment with high ambient nutrient levels may reduce growth. An understanding of nutrient limitation in the ecosystem was paramount to determining if there was any value to fertilising the sediments. Princess Royal Harbour showed N-limitation whereas Oyster Harbour was P-limited.
- Root growth was not nutrient-limited and adding nutrients may inhibit growth.
- The time of year when planting occurs also appears to be an important factor for *P. australis* but not for *P. sinuosa*.

4.3.7 Seagrass Rehabilitation using Sprigs in Albany

Seagrass rehabilitation in Albany was co-ordinated and undertaken by Bastyan & Associates (2007). Here we report on the sprig transplant trials in Oyster Harbour and Princess Royal Harbour, and the large, 1 hectare rehabilitation of Oyster Harbour. Other studies included impacts of harvesting on donor beds and seed culturing and are presented only in the outcomes.

Sprig transplant trials were established in 2003/04 at four sites: two in Oyster Harbour (OH_S and OH_D), and; two in Princess Royal Harbour (PRH-S and PRH_D). All sites were previously vegetated by dense seagrass meadows, and the sediments still contained dead rhizome. Survival over 4.2 years to June 2008 was high, ranging from 17 to 100% (Table 2). At the deeper site in Oyster Harbour and shallower site in Princess Royal Harbour severe grazing occurred on *P. australis* with just 19% and 56% survival by 2008 (4.2 years later), respectively. Whereas *P. sinuosa* was more impacted by grazers in the shallow site in Princess Royal Harbour (PRH_S) with 39 % survival after grazing but the grazer was not identified. The deeper site at Princess Royal Harbour also showed high levels of bioturbation.

Shoot production in the more successful sprig trials at PRH_D and OH_S varied for both *P. australis* and *P. sinuosa*, with the larger the sprig when planted the greater the number of shoots produced by age 6.2 years. *P. sinuosa* had the highest shoot production at both sites (Table 3).

The rehabilitation exercise focused on Oyster Harbour after preliminary results from the sprig transplant trial indicated that transplants for both species had highest survival and shoot production in this location. The target was to plant 0.5 hectare in Oyster Harbour in 2004-2005 and a further 0.5 hectare in 2005/06. Approximately 7000 sprigs were planted at locations in bare areas ranging in mean depth from 1.2–2.3 m. *P. australis* was predominantly used, with a smaller amount of *P. sinuosa* to provide heterogeneity. A planting pattern of 1.5 m between plants in staggered rows 1 m apart was used.

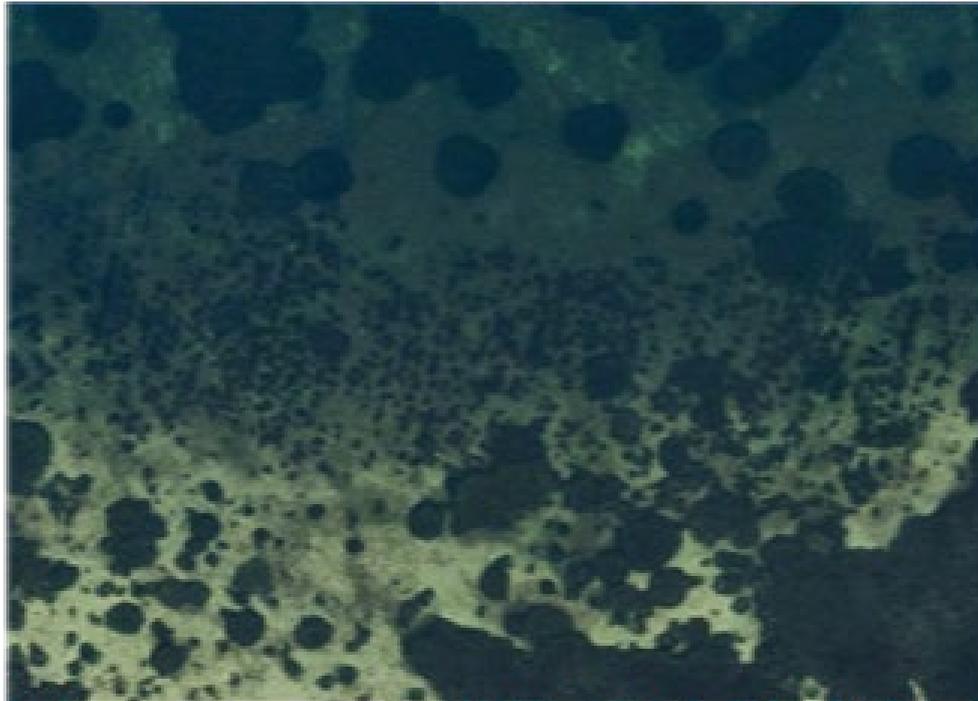
Table 2: Percent survival over 4.2 years since December 2003 for sites in Oyster Harbour (OH) and Princess Royal Harbour (PRH) (From Oceanica 2013)

Site	<i>Posidonia australis</i>			<i>Posidonia sinuosa</i>		
	June 2006	June 2007	June 2008	June 2006	June 2007	June 2008
OH_S	97	94	89	83	81	81
OH_D	89	17	19	84	61	64
OH non-apical	100	100	100	100	100	100
OH new plots	81	64	53	81	78	64
PRH_S	67	56	56	66	44	39
PRH_D	92	89	86	81	81	81

Table 3: Mean shoot production over 6.2 years for sites planted December 2003 in Oyster Harbour (OH_S) and Princess Royal Harbour (PRH_D) (From Oceanica 2013)

Species	Sprig shoot #	Shoot#	Shoot#
		OH_S	PRH_D
<i>Posidonia australis</i>	2	186.5	33.6
	4	215.5	32.2
<i>Posidonia sinuosa</i>	2	224.5	48.0
	4	342.5	53.2

High survival and growth rates were reported with 90% survival and the prediction that transplants would start to coalesce after 5 years. This proved to be accurate (Figure 13; 2011) and in 2022 there is now a continuous meadow over both transplant locations. In Albany's OH, seagrass rehabilitation has demonstrated that highly successful rehabilitation is feasible. The results of the rehabilitation efforts are visible from the air and in 2022 have coalesced into a continuous meadow.



Source: Bastyan & Associates

Figure 13: Aerial photograph of the rehabilitation site in Oyster Harbour, March 2011

The outcomes from this project are:

- One hectare seagrass rehabilitation in Oyster Harbour has been successful
- Transplanting sprigs of *P. australis* and *P. sinuosa* at 1 m spacings was successful with >90% survival and transplants coalesced within 5 years,
- The transplant trials showed that the larger the sprig when planted, the greater the number of shoots produced by ~5 years of age.
- At both Oyster Harbour and Princess Royal Harbour, *P. sinuosa* had the highest shoot production of the two species.
- Trials investigating the impacts of donor removal have suggested recovery in less than 2.5 years for cores and less than 2 years for leading edge removal.
- Preliminary transplanting trials were also undertaken using both wild and laboratory-cultivated seedlings. There was high survival of seedlings raised from seed in culture, and high survival of transplanted seedlings (wild and cultured) after 6-12 months.

4.3.8 Cockburn Cement Mooring Scars

Cockburn Cement changed their dredging process from a in situ dredge and barges to a single bottom-opening dredge in the 1990s and decided to use the Seagrass Research and Rehabilitation Plan (Oceanica 2013) to experiment and rehabilitate the scars in the seagrasses north of Woodmans Point caused by barge moorings (Figure 14). In 2008, shoots of the seagrasses *Posidonia australis* and *P. sinuosa* were transplanted into scars 2 and 3 to test their capacity to enhance natural seagrass shoot growth, seedling colonisation and meadow infilling. In 2012, the UWA team also investigated the use of sandbags and artificial seagrass to enhance survival of sprigs and shoots directly planted into unvegetated sediment. Sandbags and artificial substrata have been used extensively to modify

terrestrial environments to enhance restoration. For example, sandbags are exclusively used in South Australia to act as a substrate for *Amphibolis* and *Posidonia* restoration (Statton et al. 2018). Artificial seagrass has also been used as a hydrodynamic buffer and to protect against high levels of grazing (Statton et al. 2018).

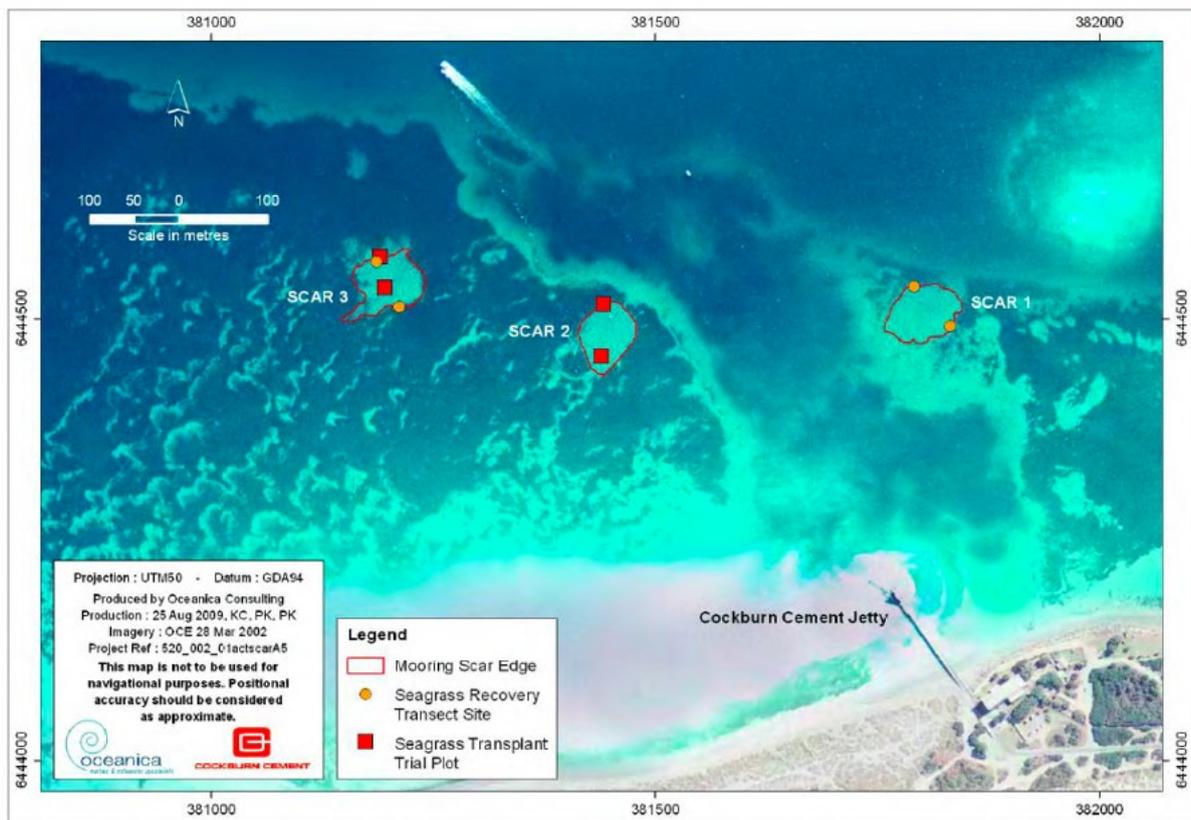
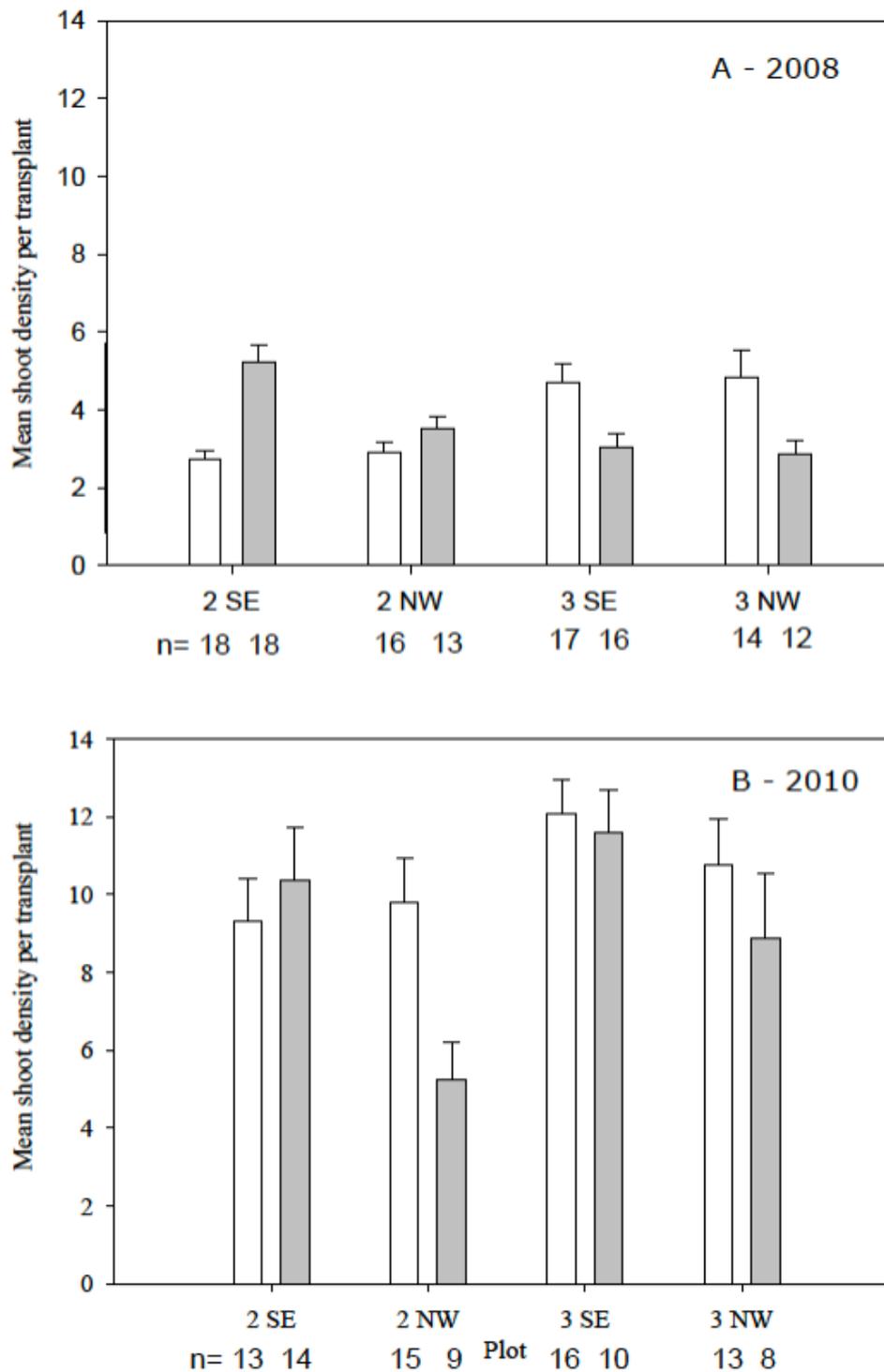


Figure 14: Location of experimental plots for seagrass transplants in barge mooring (From Oceanica 2013)

The results of the 2008 transplant trial demonstrated survival of seagrass transplants in the barge mooring scars two years after planting. Survival of seagrass transplants planted in March 2008 were 44-89% two years later (March 2010). Eighty six percent of seagrass transplanted into the scars survived the first winter, and 68% survived after two years.

Growth of transplants was evident with transplants more than doubling their shoot density between November 2008 and March 2010. Shoot density in transplants increased from 2–5 shoots per transplant (November 2008) to 6–12 shoots by March 2010 (Figure 15). Shoot densities were similar to natural meadows nearby. In March 2010, mean shoot density per transplant was similar across all transplant plots for *P. australis* but *P. sinuosa* transplants had significantly less shoots in the exposed NW plot in scar 2 (Figure 15).



Source: UWA

Figure 15: Mean (\pm SE) shoot density for *Posidonia australis* (clear) and *P. sinuosa* (shaded) transplants in each plot in (A) November 2008 and (B) March 2010 (From Oceanica 2013)

Sandbags and artificial substrata have been used extensively to modify terrestrial environments to enhance restoration. We tested their applicability to seagrass restoration by transplanting shoots into sandbags with and without artificial seagrass then comparing them to seagrass transplants, with and without artificial seagrass, planted into unvegetated sand (Figure 16).



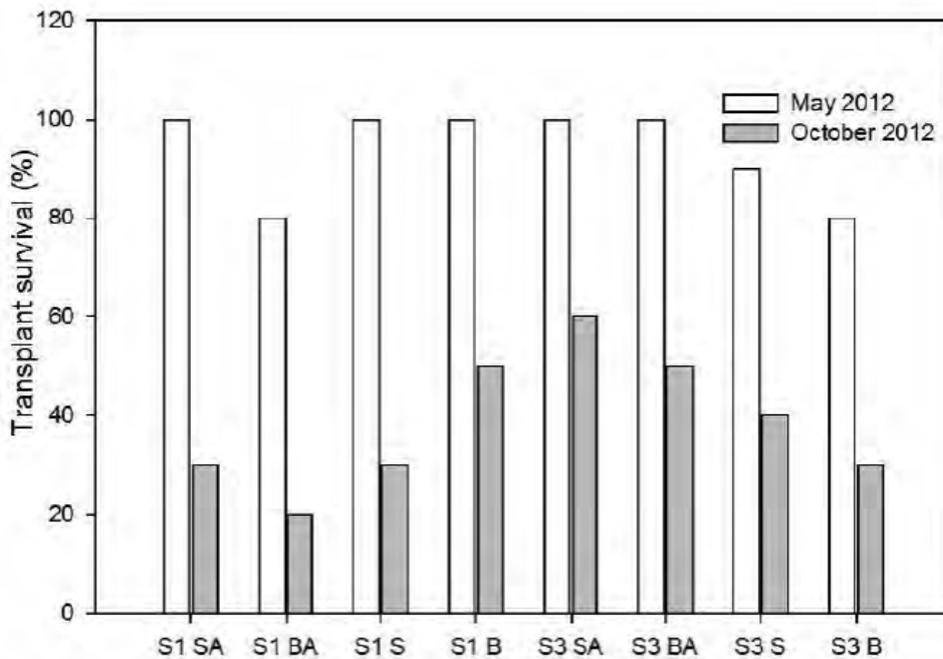
Source: UWA

Figure 16: *Posidonia* transplants planted into a sandbag and artificial seagrass leaves (left) and planted into bare sand with artificial seagrass attached to a plastic grid (right) (From Oceanica 2013)

Sandbags are often used in rehabilitation to help stabilise sediments and promote the establishment of transplants, but the results obtained during barge mooring scar trials were equivocal. Survival of transplants planted into sandbags in February 2012 were recorded in early May and October 2012. In May, survival was $\geq 80\%$, and not consistent among the treatments (Figure 17). By October, between 20 and 60% survival was observed with higher transplant survival in scar 3 (30-60%) than scar 1 (20-50%). Artificial seagrass leaves appeared to have a positive effect on transplant survival in scar 3 with both sandbag and bare sand treatments demonstrating 60% and 50% survival, respectively. In contrast, scar 1 transplants planted in the bare sediment with no artificial seagrass leaves had the highest survival (50%) (Figure 17). Results appear to have been affected by other factors influencing the survival of transplants in sandbags; for example, removal of transplants due to crabs using sandbags as habitat. Note that there were differences in sediment accretion and sand wave formation between the high sediment accretion over bags at scar 1 versus less bag accretion at scar 3, suggesting small scale hydrodynamics play a major role in the efficacy of using sandbags underwater.

The outcomes of this study were for *Posidonia australis* and *P. sinuosa*:

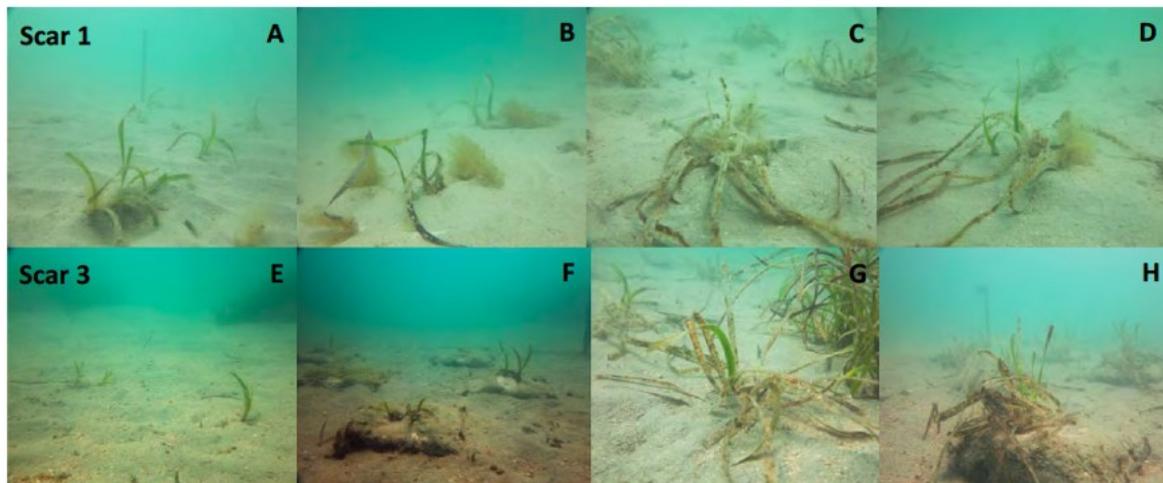
- Survival of transplants was high within a year of planting but within two years was 30-50% of transplant densities.
- Surviving transplants doubled or trebled their shoot densities within 2 years, thus were effectively spreading into unvegetated sediments.
- Results using hessian bags and/or artificial seagrass to protect shoots and sprigs were equivocal and site specific, suggesting strong local drivers to survival and growth. They showed promise but the effort was not justified in the present outcomes.



Notes:

1. sandbags with artificial seagrass (SA)
2. bare sand with artificial seagrass (BA)
3. sandbags only (S)
4. bare sand only (B)

Source: UWA



Note:

1. Sediment height around sandbags with scar 1 showing significant sediment accretion (up to 15 cm) compared to scar 3 that showed less sediment movement. Sand ripples were also more prominent in scar 1 (A) compared to scar 3 (E) suggesting greater sediment movement.

Source: UWA

Figure 17: Survival (%) of *Posidonia australis* transplants 3 months (May 2012; clear bars) and 8 months (October 2012: shaded bars) after planting in the four treatments in scars 1 and 3 (Top graphs), and; Images of *Posidonia* transplants in scar 1 (top) and 3 (bottom) during October sampling: bare sand with no artificial seagrass (A and E), sandbag with no artificial seagrass (B and F), bare sand with artificial seagrass (C and G) and sandbag with artificial seagrass (D and H) (Bottom graphs) (From Oceanica 2013).

4.3.9 Seedling Nurseries

The growth of seeds in land-based aquaculture nurseries to prepare seedlings before they are transplanted *in situ* was investigated in a collaborative study between the University of Western Australia and the Kings Park Botanic Gardens and Parks Authority between 2005 and 2010. The research investigated the growth requirements of seagrasses at the seedling stage and assessed their value as propagules for restoration. A detailed research plan included planting seeds directly and growing seeds into >6 mo seedlings in aquaculture tanks on land and then transplanting them. The flow chart below outlines the series of field and lab experiments and whether they were successful or not (Figure 18).

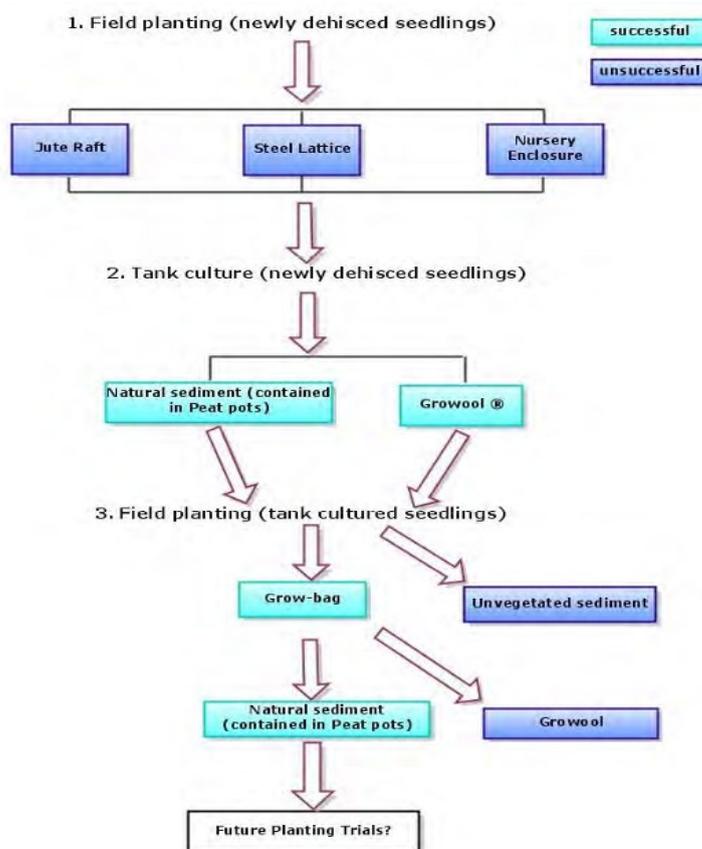


Figure 18: Flowchart of trials conducted on *Posidonia australis* seedlings in the development of an effective strategy to enhance establishment success in Cockburn Sound, WA (from Oceanica 2013).

The seedling nursery had 12 tanks (Figure 19 d) and trays with plant pots (Figure 19e, f) were set up at the University of Western Australia Watermans lab and seedlings were grown under continuous flowing seawater. The growth of seedlings was monitored for 6 to 8 months before they were transplanted *in situ* (Figure 19)

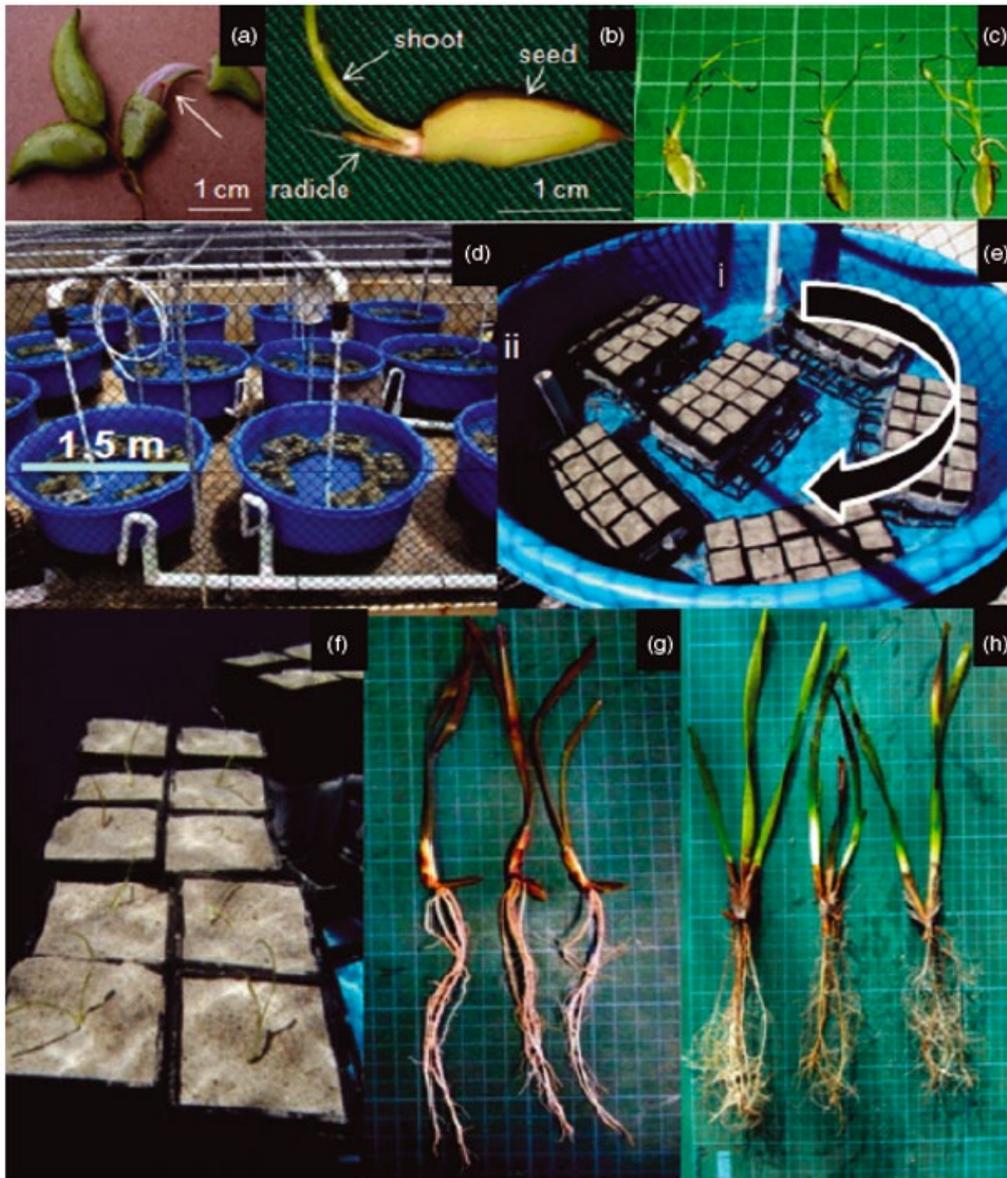


Figure 19: Stages of *P. australis* seedling development and tank culture environment. Initially, seedlings were collected as mature fruit attached to the inflorescence: (a) shows an inflorescence with three mature fruit (up to 20) with a lateral section of one fruit removed to reveal a developing seedling inside (arrow), (b) newly released seedling (1-day-old) with a well-developed and intact shoot, radicle, and seed, (c) 1-week-old seedlings before planting (each small square represents 1 cm²), (d) whole tank system (12 tanks), (e) tank design for culturing seedlings, (i) spray bar directing water in a circular manner (arrow), (ii) internal stand-pipe to remove surface particles (marine and aeolian) to maintain light penetration, (f) week-old seedlings planted into mesh-lined pots, (g) 7-month-old seedlings with intact root system grown in the land-based aquaculture facility at Watermans Bay facility (each small square represents 1 cm²), and (h) 18-month-old seedlings with multiple shoots and extensive root development, grown in the land-based aquaculture facility at Watermans Bay facility (each small square represents 1 cm²). (From Statton et al. 2013)

Seedlings of both *P. australis* and *P. sinuosa* were able to be grown from viviparous seeds thus breaking the seed to seedling bottleneck in establishment (Statton et al. 2017a). Early establishment relied on both carbohydrate resources in the seed and photosynthesis but after 6 months when starch stored in the seed was exhausted, seedlings required more carbon from the environment. This was observed in the growth of seedlings in tank culture where augmenting sediment carbon with dried seagrass leaves from beach wrack when planting resulted in larger seedlings in the 7th month of culture (Statton et al.

2013) (Figure 20). There was also little difference in the effect of sediment type with equal increases in biomass with carbonate and silicate sediments.

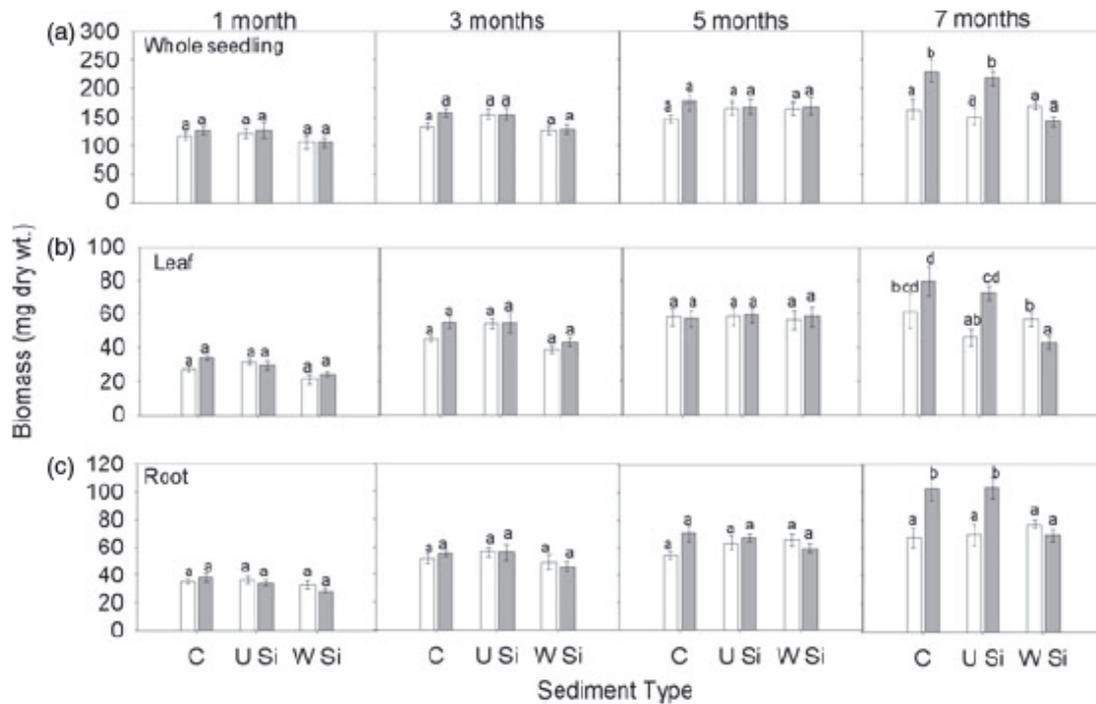


Figure 20: Whole seedling (a), leaf (b) and root (c) biomass (mg dry wt.) of *P. australis* seedlings grown in three sediment types: unsorted calcareous (C), unsorted silica (U Si) and well-sorted silica (W Si), without (white columns) and with (grey columns) organic matter. Biomass was measured at 1, 3, 5, and 7 months after planting (Interaction $S \times O \times T$; $p < 0.05$). Different letters indicate significant differences between each sediment type, without and with organic matter, at a particular month. Columns represent means \pm 1SE (from Statton et al. 2013).

Nutrient additions to sediment had the opposite effect with increased mortality and lower growth rates, suggesting that increases in nutrients like that observed in Cockburn Sound have negative effects both on healthy adult meadows (Kendrick et al. 2002) and the process of recruitment from seeds (Statton et al. 2014). Growout trials found that hessian grow bags had greater survival and growth than grow-wool and planting into unvegetated sediment (Oceanica 2013). Mortality in seedlings was higher than that observed from transplanting shoots. They were preferentially predated on by grazers and were more easily removed by currents and waves

The outcomes of this study were:

- *P. australis* seedlings represent a potentially useful source of propagules for seagrass restoration programs.
- Growing seedlings in tanks prior to transfer to the field reduces seedling mortality and improves establishment.
- Transferring tank grown seedlings into sand-filled biodegradable hessian bags (Grow-bags) is crucial for seedling success as they provide a stable sedimentary environment for seedlings to be established.

- Sediment composition (sediment texture and organic matter content) had a greater influence on seedling establishment (i.e. initial anchorage success) than adding inorganic nutrients (N and P).
- The optimal light condition for growing seedlings in tank culture was 70% of the surface irradiance. Seedlings had greater biomass and larger root systems than seedlings grown in higher and lower light regimes.
- Growing seedlings in low water movement regimes is more desirable for production of larger seedlings with greater root development.
- Seedlings appear to be heavily reliant on seed stores in the early stages of growth. However, growth can be uncoupled from seed stores when grown under optimal conditions. Seed starch content may be used as a surrogate for assessing seedling health, where healthier seedlings retain the greatest amount of starch.
- Site selection plays an important role in determining suitable sites for seed-based restoration methods. Successful seedling establishment may be linked to sediment characteristics and water depth. It also appears that water motion could significantly influence seedling establishment (growth and anchorage potential). Preferential grazing by predators requires careful consideration during the site selection process.

4.4 2010 - 2020s

4.4.1 Seed-based Restoration Research

In the early 2010s, conversations started around using seeds directly, rather than the nursery approaches used in the Seagrass Research and Rehabilitation Plan (Oceanica 2013), with RJ Orth, Virginia Institute of Marine Science. Dr Orth was successfully restoring the eelgrass *Zostera marina* in Virginia coastal bays using seeds. He had come to the conclusion that nurseries were ineffectual as large losses in the seagrass life history occurred at seed and early recruiting seedlings stages (Kendrick et al. 2016). Like most plants, seagrasses have a type III survivorship curve. It is a numbers game where millions of seeds were required to be dispersed for 100s to 1000s of seedlings to persist. *Posidonia australis* was chosen as the initial test seagrass for seed-based restoration as it was under threat in parts of its geographical distribution and it had large showy inflorescences and produced a large number of fruit in the Cockburn Sound, Owen Anchorage and Rottneest Island regions. Research into seed-based restoration focused on the demographic bottlenecks in the life history of *P. australis* (Statton et al. 2017a). A multidisciplinary research program was developed and focused seeding experiments in Cockburn Sound and Owen Anchorage and on natural recruiting seagrass populations at Rottneest Island.

The multidisciplinary program addressed:

- Restoration and population genetics of *P. australis* populations and how this may affect seed and shoot sourcing for restoration (2010-2017) (Sinclair et al. 2009, 2014)
- Demographic approaches to seed-based restoration (Ruiz-Montoya et al. 2012, 2015, Statton et al. 2017a, Sinclair et al. 2018).
- Seed-based restoration experiments. Three locations were started in 2014 and another added in 2015. (2014-2018).

- Use of seed-based methodologies with other seagrass species. The focus has been on using seeds of the colonising *Halophila ovalis* which differs from *P. australis* as it has a hard dormant seed and can be stored for a few months (Statton et al. 2017b, 2018, Waite et al. 2021)
- Scaling up seed-based restoration (Tan et al. 2020, Sinclair et al. 2021).

The seed-based restoration focus has been productive, is relatively cheap to maintain and can be repeated for many years. It is appropriate as a restoration method where seeds and fruit are produced in large numbers and can be scaled to hectares or can be used to add to existing restoration using other methods. The major results of the research programs will now be summarised.

4.4.2 Population Genetics of *Posidonia australis*

Major genetic issues with seagrass restoration that are rarely addressed are: that restoration does not move populations from their genetic provenance; that genetic diversity in restored areas is maintained or even enhanced, and; that the propagules used in restoration, whether they be shoots, seeds, plugs, sods etc., are representative of how seagrasses naturally colonise the restoration site.

Cockburn Sound and Owen Anchorage fit within a broader provenance for *Posidonia australis* (Sinclair et al. 2014) they show similarities in population genetics between populations from Lal Bank to the north, Rottnest Island to the west and Shoalwater Islands to the south (Figures 21, 22). This led to substantial effort to characterise seed movement (Ruiz-Montoya et al. 2012, 2015) and to address whether seeds from a broad geographical area recruit into natural meadows in Cockburn Sound and Owen Anchorage (Sinclair et al. 2018).

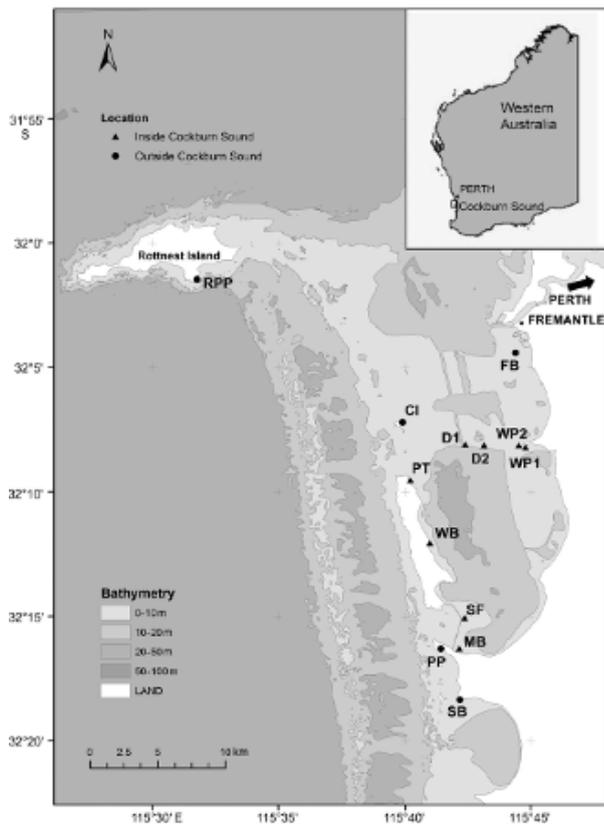


Figure 21: Population genetic sampling of 13 *Posidonia australis* meadows across the Cockburn Sound area.

Onshore, a weak linear north to south isolation by distance in genetic similarity is shown in Cockburn Sound which indicates a barrier to dispersal and connectivity in this geographically isolated system has occurred (Figure 22), with southern locations more isolated from northern locations (from Sinclair et al. 2018).

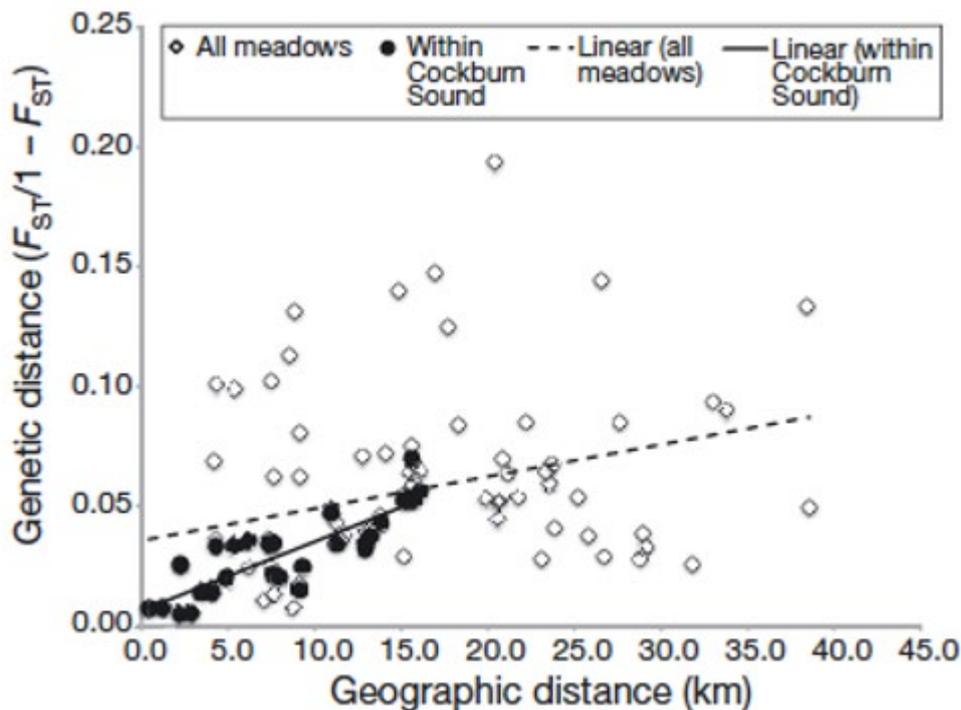


Figure 22: Relationship between genetic distance and geographical distance over all pairs of sampled *Posidonia australis* meadows and among the 8 meadows sampled within Cockburn Sound (from Sinclair et al. 2014).

The recent 3 hectare shoot-based restoration program on Success Bank was also tested for genetic diversity and found to be diverse, and that it reflected the clonal diversity of the source meadows from Parmelia Bank (Sinclair et al. 2013). It was recommended that population genetics needs to be assessed for both source meadows and the restoration site prior to any translocation of sprigs, to reduce genetic contamination and to enhance genetic diversity in transplants.

The outcomes of these studies were:

- An understanding of the population genetics of natural populations in the area to be restored dramatically reduced risk of introducing plant genotypes that will not survive in the restored area (genetic provenance)
- Seeds are a valuable source of genetic diversity and connectivity that naturally recruit in small numbers into seagrass meadows within Cockburn Sound and Owen Anchorage. Seeds can move >84 km and recruitment has been determined from genetic studies to be at much as 34 km in this region. From a population genetic basis, seeds are viable as a restoration technique across Cockburn Sound and Owen Anchorage.
- Shoot-based restoration in Cockburn Sound should focus on sourcing transplants from populations that have high clonal richness, to maintain or increase genetic diversity in restored areas.

4.4.3 *Demographic Bottlenecks in the Life History of P. australis*

Seagrasses are highly clonal but also produce an abundance of seed in some locations. The interplay between vegetative and sexual reproduction determines the maintenance of natural meadows and overall seagrass resilience in the region (Figure 23). A demographic approach to assess bottlenecks in the life history of *P. australis* led to our understanding of the effect of the environment on the production of seeds, dispersal of fruit, settlement of seeds, early seedling mortality, mortality of 1 year old seedlings and subsequent survival to adulthood.

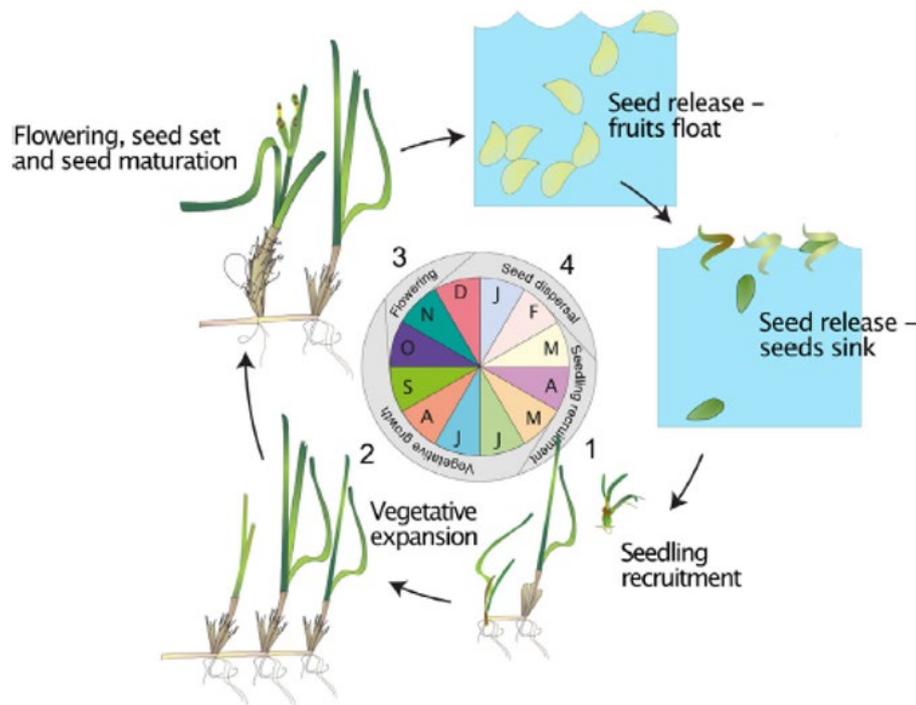


Figure 23: Demographic Approach for understanding the bottlenecks in recruitment from seeds (Sinclair et al. 2021)

These studies included determination of seedling survival across gradients in hydrodynamics and grazing pressure in Cockburn Sound and Owen Anchorage (Statton et al. 2017a). This was combined with an understanding hydrodynamics of dispersal of *P. australis* fruit, to address source-sink relationships among seagrass meadows and to assess the impact of harvesting fruit and seed from natural populations (2012-2014) (Ruiz-Montoya et al. 2012, 2015, Sinclair et al. 2018). And associated experiments of seed predation and bioturbation (Orth et al. 2002, 2007, Johnson et al. 2018). A detailed study of flowering seed set and seedling recruitment in *P. australis* also occurred at Rottneest Island (Kendrick in review) to assess interannual variation in flowering, seed set and fruit production, seed settlement and seedling recruitment (2013-2018).

The outcomes of these studies were:

- In sheltered waters (e.g. Cockburn Sound) herbivory by crustaceans and bioturbation affect the transition from seed settlement to early seedling recruitment. Their effect can result in 100% loss in this transition from seed to early recruit.
- In swell-influenced environments (e.g. Owen Anchorage, Carnac Island) the frequency and duration of winter storms result in high mortalities between early (3 month) to 1 year old recruiting seedlings and can result in complete loss.
- Seed settlement densities highly influence these transitions. Increasing seed settlement via seed-based restoration strongly influences survival from seeds to 1 year old recruiting seedlings.
- Flowering and seed production is high at Rottneest Island, Cockburn Sound and Owen Anchorage. There is geographic variation among locations within each of these locations.

- Many of the seeds produced are transported out of their bed of origin and numbers from Rottnest Island suggest that only 10 % stay within shallow seagrass habitats. Thus, collecting seeds for restoration has little or no impact on source meadows.

4.4.4 Seed-based Restoration across Cockburn Sound and Owen Anchorage

From the demographic analysis, we found that seedling recruitment in *P. australis* populations was affected by environmental conditions and frequency of disturbances within the first month after seeding and some 4–6 months later when established seedlings were physically removed by winter storms (Statton et al. 2017a). The differences observed across the gradients of disturbance by grazers, bioturbators and physical disturbance from swells generated by winter storms in Cockburn Sound and Owen Anchorage suggested that the constant pressures of disturbance across relatively local scales limit seagrass population growth and restoration outcomes. We also modelled the influence of seeding density on survival probabilities especially around the seed to early seedling and established seedling stages and found that maintaining high seeding densities resulted in higher seedling recruitment.

Armed with this information we decided to test the Cockburn Sound - Owen Anchorage north-south gradient in disturbance and to re-seed over two annual flowering and seeding events. Seed-based restoration was scaled to 3 replicated 5 x 5 m plots across 3 locations in 2014: one on Kwinana Shelf, Cockburn Sound, and; two in Owen Anchorage. A further location was added in Owen Anchorage near Coogee Beach in 2015 (Figure 24). In each region a seeding density of 5,000 seeds m⁻² was maintained for 2014 and 2015.

These locations showed a gradient of increased seedling survival from south to north. A seeding rate of 5,000 seeds across 25 m⁻² (200 seeds m⁻²) was applied in November 2014 and November 2015 resulting in 0.5 plants m⁻² at Cockburn Sound East, 1 plant m⁻² at Owen Anchorage Scars, 5 plants m⁻² at Owen Anchorage North, and 15 plants m⁻² at Coogee Beach (Figure 25).

The outcomes of these studies were:

- Fruit collection was scaled up such that 10,000s to 100,000s of seeds were collected. This involved design of rapid fruit collection methods, both boat and diver-based.
- Seedling recruitment seems to be context dependent and greater in Owen Anchorage than Cockburn Sound.
- Seedling survival was significantly greater in seeded plots than natural controls across all restoration sites, justifying the approach to seed rather than other approaches, like seedling nurseries, etc.

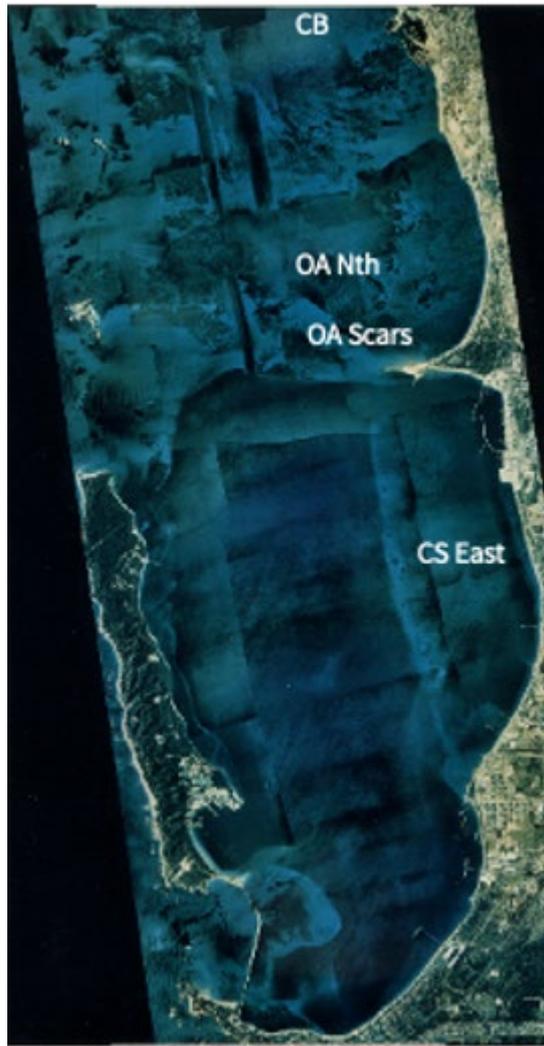


Figure 24: Locations used for seed-based restoration experiments. CS East is on the outer edge of Kwinana Shelf in 7 m depth; OA Scars is Dredging Barge Scar 3 near the Cockburn Cement Wash Plant in 4.5 m depth; OA Nth is due west of the Woodmans Point Ammunitions Jetty in 6.5 m depth; CB is north west of Coogee Marina in 6.8 m depth.

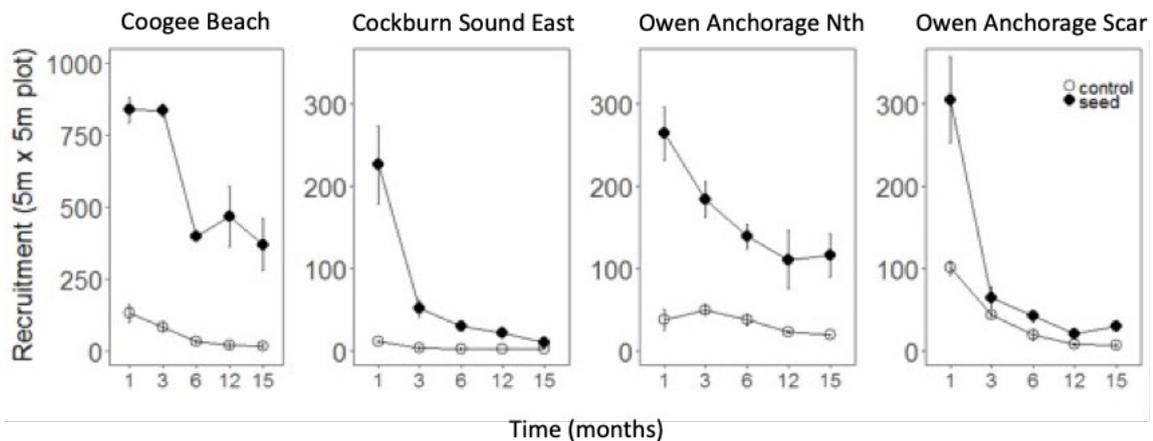


Figure 25: Recruitment in experimental and control (natural recruitment) plots ($n = 3$ experimental and 3 control) between seeding in December 2015 and counts of recruits made in February 2017 (Kendrick pers comms)

4.4.5 Use of seed-based methodologies for other species

One of the outcomes of the demographic study (Statton et al. 2017a) was the suggestion that in wave exposed locations mixtures of seeds for different species that overlap in their environmental niches may enhance survival of both species. Similarly, van Keulen et al. (2003) observed that sprigs transplanted into meadows of the faster growing, colonising genera *Halophila* and *Heterozostera* were more likely to survive than if planted into unvegetated sands. These observations led to experiments in seed-based restoration using the colonising *Halophila ovalis* which differs from *P. australis* in that it has a hard dormant seed and can be stored for a few months.

Halophila ovalis is annual to pseudo-perennial where many beds will persist for optimal growing months of spring and summer and die back over winter. Annual recolonisation relies mainly on recruitment from vegetative fragments in the sediment or the sediment seed bank. Despite this, there is little understanding of seed dormancy releasing mechanisms and germination cues. We addressed that knowledge gap by experimenting with temperature and light pre-treatments with the aim to determine optimal conditions for high rates of seed germination (Statton et al. 2017b, Waite et al. 2021). Trials were maintained in both the laboratory and we ran field trials with seeds where dormancy had been broken.

Dramatic increases in seed germination (on-demand germination) in *H. ovalis* was observed in the field after seeds deployed were initially exposed to gradual increase in temperature from 15 to 25°C and to red spectrum light in the laboratory (Figure 26).

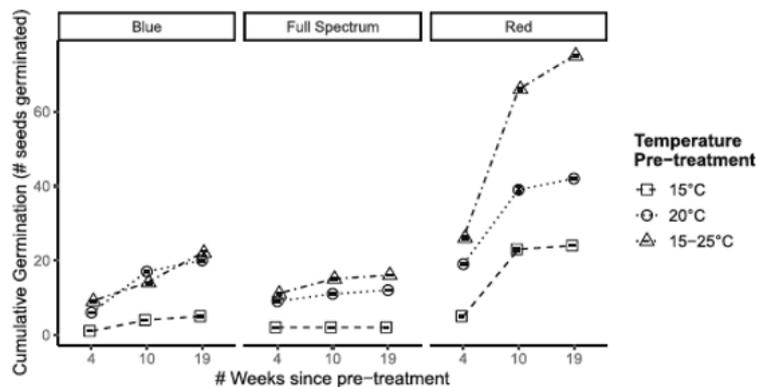


Figure 26: Cumulative germination counts of both field sites for each of the nine pre-treatments over the three collecting periods with error bars showing standard error. Collection 1 took place 4 weeks after pre-treatment (August), collection 2 followed 10 weeks after pre-treatment (October), and collection 3 took place 19 weeks after pre-treatment (December). The plots are faceted by light quality. Each light quality facet contains three lines for each temperature treatment, differentiated by symbol (from Waite et al. 2021).

The outcomes of these studies were:

- For *Halophila ovalis*, increasing temperatures in a stepped fashion from winter (15°C) to summer (20-25°C) slowly (Statton et al. 2017b) and changing spectral irradiance from blue dominated to red-yellow dominated (Waite et al. 2021) resulted in >35% germination of dormant seeds.
- Colder temperatures (<15°C) and either no light or blue light were effective in arresting germination and is the preferred environment for seed storage in *H. ovalis*.

4.5 2020s

4.5.1 *Seeds for Snapper - a UWA, OZFISH, and RecFishWest community restoration program*

Our experience with seed-based approaches in Cockburn Sound and Owen Anchorage is that they offer the opportunity to scale-up seagrass restoration to address the loss of seagrasses in Australia at the ecosystem scale. Seed-based approaches also address the cost of more traditional seagrass restoration approaches, seen to be excessive by some (Bayraktarov et al. 2016), through developing integrated community-based restoration approaches (Tan et al. 2020, Sinclair et al. 2021).

The OZFISH 'Seeds for Snapper' program (Figure 27) started in 2018 and developed from discussion at the Marine Restoration Workshop (McLeod et al., 2018b) held by the Marine Biodiversity National Environmental Science Program at the Department of Agriculture Water and Environment offices in Canberra in 2018. These discussions were between Craig Copeland (CEO - OZFISH), Gary Kendrick and John Statton (Researchers - UWA). The first two years built up the community group, coordinated by Andrew Matthew and diver coordination by Tania Douthwaite, and developed a community-based methodology through John Statton. By 2020 the scale of operations had increased to 100s of participants and required a revisit of the technology being employed and the aquaculture facility holding fruit and seeds.

The 'Seeds for Snapper' project uses volunteers to scale up seagrass restoration using *Posidonia australis* fruit and seeds to restore seagrass to sand patches. There are now well-developed steps that community volunteers drive (Figure 27). Firstly, we determine seed production in donor meadows using 5 x 2 m belt transects. We then collect fruit with free divers and SCUBA divers by hand and net which is then transferred into aquaculture tanks that are circulated and aerated with pumped seawater. When the fruit splits open, the seeds sink and are collected from the bottom of the tanks and placed in holding aquaria until counted (volumetrically) and bagged for delivery to the restoration sites via the UWA team or by volunteer recreational fishers and boaters. Present activities are highly targeted to occur every day over a 6-week period between late-October to mid-December each year.

The 2021 season has proven that we can scale up restoration using seeds and a dedicated community group. Between 18th November and 15th December 2021, the OZFISH 'Seeds for Snapper' Community group collected 1.18 million fruit (Figure 28). We obtained 375,000 viable seeds that were dispersed to 6 restoration sites (Figures 28 & 29). A total of 42 dive sessions were conducted with over 300 individual collection dives done by the community. Over 1,000 hours were volunteered during this time. Our group grew during the season and now the 'Seeds for Snapper' Facebook Group page has 645 active members.

We did have some breakdowns during the activity. Collection of seeds far outpaced our ability to process the fruit and seed, combined with equipment failure and extremely hot daytime temperatures, this resulted in the loss of many fruit. To prevent this from reoccurring, we have now bought purpose-built tanks for faster processing and larger pumps to increase circulation in tanks. We have also considered having two facilities to reduce the risk of another catastrophic breakdown of pumps.

The first two years (2018 and 2019) of the project were the proof of concept and trial stage. Using a

small group of university volunteers, ~200 000 seeds were dispersed each year across four sites totalling over ~1 000m² using a seeding density of ~200 seeds m⁻². The monitoring of these sites showed seedling densities of ~20-40 seedlings m⁻², compared to 0-2 seedlings m⁻² in the control sites. If you multiply the total seeded area by seedling density, the “Seeds for Snapper” sites resulted with between 18,900 and 34,200 seedlings were established compared to natural recruitment from seeds which would have produced 0 to 1,800 seedlings in an equivalent area. These levels clearly demonstrate that this project and its methodology is effective at increasing seed recruitment.



Figure 27: The ‘Seeds for Snapper’ fruit collection, seed release, seed delivery and seedling development process. From top left to right: *Posidonia australis* fruit, collection in nets, volume of fruit in collection net, transfer to buckets, counting fruit to determine daily amounts collected. From bottom left to right: fruit in aquarium tanks, released seeds (handful), tossing seeds from boat, seeds settling onto sea floor, a 1-2 day old seedling on seafloor. Photos: Rachel Austin, Tania Douthwaite, Andrew Matthews, Marta Sanchez Alarcon, Lara Oppermann, Sharmini Jayasinghe

The next two years (2020 and 2021) of the project focused on scaling up via increasing the number of volunteers. The volunteers are primarily used for the collection of fruit and the preparation of seed prior to dispersal. Volunteers include certified SCUBA and free divers from the universities and from the public. In 2020 a small group of community and university volunteers allowed us to disperse ~350,000 seeds across three sites with an area of substrate of ~1,100 m² using a seeding density of ~200 seeds m⁻² (Figure 28). In 2021 the volunteer group grew substantially and we collected ~1.18 million fruits and dispersed ~375 000 seeds at ~85 seeds m⁻² into six new sites with an area of substrate of ~2,100 m² (Figure 29). The monitoring of these six new sites showed that after ~10 weeks seedling densities were 5-45 seedlings m⁻², with control sites having 0-2 seedlings m⁻². If you multiply the total seeded area by seeding density the “Seeds for Snapper” sites between 10,545 and 94,905 seedlings were established compared to natural recruitment from seeds of 0 to 4,218 seedlings. These levels clearly demonstrate that involving the community allows seagrass restoration to be scaled up with relative ease but high effectiveness with high recruitment success. Note that most plants and

seagrasses have a type III survivorship curve where there is an exponential decline between the population density of seeds, early recruits, 1 year old recruits and multi-aged individuals.

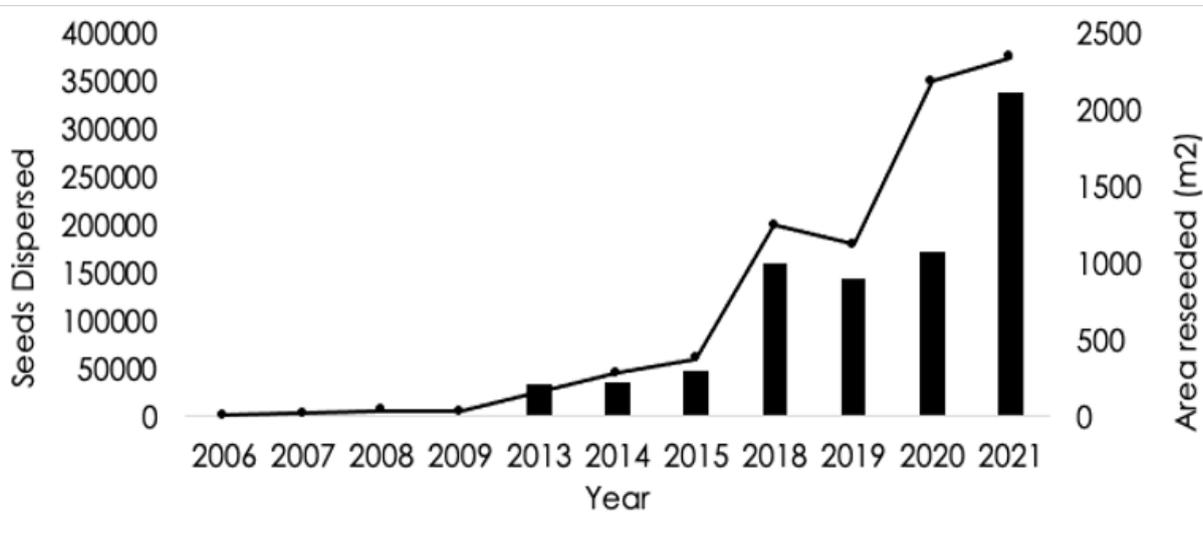


Figure 28: Progress with scaling up seeding through the OZFISH ‘Seeds for Snapper’ community restoration program. UWA trialed diver-based seeding of small trial plots between 2013-2015 for proof of concept, then in 2019 UWA-Ozfish volunteer divers became involved and larger plots with boat seeding were set up, in 2020 we trialed the Ozfish volunteer involvement, scaled it up for 2021 which success. Line with points = seeds dispersed, Columns = area reseeded



Figure 29: Location of Seeds for Snapper collection sites (filled circles), 2021 restoration sites (open circles) and community and previous years site (open squares).

The outcomes of seed based restoration and the OZFISH ‘Seeds for Snapper’ restoration program were:

- The OZFISH ‘Seeds for Snapper’ community-based restoration program is growing and with further refinement and will hopefully reach their goal of 1,000,000 seeds sowed in 2022 and 1 hectare restored.
- Kwinana Shelf is promising as are sites in deeper waters of Owen Anchorage. Our goal is to increase sites in Cockburn Sound (presently we have 2: S4S sites 4 and 5), maintain Owen Anchorage sites (S4S sites 1,2 and 3) and reduce effort in areas that have shallow reef pavement and where sand waves are observed (restoration site 2B18).
- The largest bottlenecks to recruitment remain at the seed to 3 month old seedling and the 3 month old seedling to 1 year old seedling stages as shown in Section 4.4.4. above.
- In the seed to early seedling stage, disturbance of seeds before roots are established occurs in shallow waters (<3 metres) affected by the seas created by summer sea breezes, and from 0.5 metres to 10 metres in sheltered waters, where grazers and bioturbators are common.
- In the early seedling to year old seedling stage, the greatest threat to seedling survival is the frequency direction and duration of winter storm swells.
- All water activities are run through the OZFISH membership and their OH&S policy. All OZFISH volunteers are insured for all activities and they also need to be registered with OZFISH as qualified divers.

5 2022 Revisiting Past Restoration Programs

The team revisited a number of restoration and rehabilitation sites across Cockburn Sound and Owen Anchorage in early 2022. We focused on seed, transplant and sod- based restoration methods and a timeline from 0.5 to 26 years (Table 4). Cockburn Sound is dominated by temperate, slow growing seagrasses, primarily *Amphibolis* and *Posidonia* species, that can take decades to grow into sizable meadows. However, most projects only last 1-3 years, which makes it difficult to determine if they have been successful at the ecosystem scale. To further complicate this, there are also several different ways you can measure success, and the most ecologically relevant method changes with time. For *Posidonia* spp. and *Amphibolis* spp. the relevant measure of success in the first few years is survival, shoot growth and rhizome expansion. After 5+ years increase in seagrass cover is a better indicator of restoration success and the more plant specific variables, like transplant survival and shoot density, become difficult to accurately determine and represent a significant investment of time underwater (see Oceanica 2013 for examples). A proportion of the old restoration sites we revisited also were either indiscernible from natural recolonization and/or there was no evidence of previous restoration activities at that site.

We revisited sites of previous restoration activities to determine their success (or failure) after ecologically relevant time periods have passed (Tables 4, A1). The restoration sites we revisited were selected from a larger pool based on our ability to obtain accurate information on the restoration activity and to obtain accurate GPS locations. Please note that the restoration sites we visited were originally sand patches that showed low to no natural recovery of seagrasses since seagrass were lost across Cockburn Sound in the 1960s and 1970s.

There were 23 examples of small (10-100 m²) to medium (1,000 m² - 1 hectare) scale, partial or full success, 2 complete failures and 6 where no evidence of the actual restoration was found although seagrass cover had increased (Table 4). Natural recruitment in areas of restoration were high suggesting the environmental pressures have been removed and there has been high connectivity across the system for seagrass recruitment and propagation. A selection of the seed, sprig and sod-based restoration programs reported in Table 4 (and Appendix Table A1) have been investigated in detail.

Table 4. Restoration Locations visited in 2021-2022 to determine the present status of past restoration activities. More details found in Appendix table A1

Restoration Location	Site Code	Depth	Species	Method	Time of Restoration	Age Today	Current Site Status
Cockburn Sound	S4S - 4	8	<i>Posidonia australis</i>	Seeds	December 2021	2 months	~33 seedlings m ⁻²
Cockburn Sound	S4S - 5	7	<i>Posidonia australis</i>	Seeds	December 2021	2 months	~23 seedlings m ⁻²
Cockburn Sound	CSE14	8	<i>Posidonia australis</i>	Seeds	December 2014	7.5 years	Three seeded plots varied in cover from small patches to 15% coverage of the 25m ² seeded area. 12-15 individuals 25 m ⁻² , Shoot densities of 129 to 249 shoots 25m ⁻²
Cockburn Sound	CSH3B	5	<i>Posidonia sinuosa</i>	Sprigs, Plugs	February 2000	22.3 years	No evidence found, solid seagrass meadow present
Cockburn Sound	CSH3C	3	<i>Posidonia sinuosa</i>	Sprigs, Plugs	February 2000	22.3 years	No evidence found, solid seagrass meadow present
Cockburn Sound	CSH6B	5	<i>Posidonia sinuosa</i>	Sprigs, Plugs	February 2001	22.3 years	No evidence found, solid seagrass meadow present
Cockburn Sound	CSH6C	3	<i>Posidonia sinuosa</i>	Sprigs, Plugs	February 2002	22.3 years	No evidence found, patchy seagrass meadow present
Cockburn Sound	CSH7B	5	<i>Posidonia sinuosa</i>	Sprigs, Plugs	February 2000	22.3 years	No evidence found, sandy
Cockburn Sound	CSH7C	3	<i>Posidonia sinuosa</i>	Sprigs, Plugs	February 2000	22.3 years	No evidence found, sandy with some small patches
Cockburn Sound, Southern Flats	R1/2	2	<i>Posidonia australis</i> , <i>Posidonia sinuosa</i>	Sprigs, Plugs	September 2006 onwards	15.8 years	Large meadow present, significant infilling and expansion, shoot densities similar to surrounding meadows

Woodman Point	2m	2	<i>Posidonia australis</i>	Sprigs	May 2009	13 years	Sprigs have formed circular patches
Woodman Point	4m	4	<i>Posidonia australis</i>	Sprigs	May 2009	13 years	Sprigs have merged, plots have formed square meadows
Woodman Point	9m	9	<i>Posidonia australis</i>	Sprigs	May 2009	13 years	No evidence found - covered in <i>Posidonia sinuosa</i>
Woodman Point	WP14	4.5	<i>Posidonia australis</i>	Seeds	December 2014	7.5 years	Recovery has been so great that we could not discern our seedling from rhizome spreading into our seeded plots. Cover ranged from 25% to 45% in plots. Controls were highly variable as rhizomes from nearby meadows had grown into the plots, few seedlings were found, cover ranged from 5% to 70%.
Woodman Point	scar 1	5	<i>Posidonia australis</i> , <i>Posidonia sinuosa</i>	Seeds, Sprigs	March 2008	14 years	Not revisited. After 4 years there were few surviving transplants and they were not healthy.
Woodman Point	scar 2	4.5	<i>Posidonia australis</i> , <i>Posidonia sinuosa</i>	Seeds, Sprigs	March 2008	14 years	Recovery observed and the area. Approximately 70% filled with seagrass. We found star pickets at the site but no evidence of bags or artificial leaves.
Woodman Point	scar 3	4.5	<i>Posidonia australis</i> , <i>Posidonia sinuosa</i>	Seeds, Sprigs	March 2008	14 years	Recovery observed with 45% of the area covered in seagrasses. Stakes were still present. photos only were taken
Owen Anchorage	S4S - 1	7	<i>Posidonia australis</i>	Seeds	December 2021	2 months	~5 seedlings m ⁻²
Owen Anchorage	S4S - 2	6	<i>Posidonia australis</i>	Seeds	December 2021	2 months	~17 seedlings m ⁻²

Owen Anchorage	S4S - 3	8	<i>Posidonia australis</i>	Seeds	December 2021	2 months	~45 seedlings m ⁻²
Owen Anchorage	S4S-community	2	<i>Posidonia australis</i>	Seeds	December 2021	2 months	~5 seedlings total
Owen Anchorage	BA19	7	<i>Posidonia australis</i>	Seeds	December 2019,2020	1.5-2.5 years	~7 seedlings m ⁻²
Owen Anchorage	BA18	7	<i>Posidonia australis</i>	Seeds	December 2018	3.5 years	Survival was 42 seedlings 225m ⁻² with shoot density of 1333 shoots 225m ⁻² . There was some drift of seeds in a southern direction resulting in a line of seedlings just outside the southern boundary. No seedlings were observed in two smaller control plots.
Owen Anchorage	2BA18	9	<i>Posidonia australis</i>	Seeds	December 2018,2020,2021	0.2-3.5 years	Survival was 1.64 seedlings m ⁻² for 2020 seeding and 0.64 seedlings m ⁻² for 2019 seeding. There were older >3 shoot recruits and patches in the study area with 0.6 individuals m ⁻² . Sand waves (<15 cm) observed throughout the site.
Owen Anchorage	BA15	7	<i>Posidonia australis</i>	Seeds	December 2015	6.5 years	Three seeded plots varied in cover from large patches to 75% coverage of the 25 m ² seeded area. 20-40 individuals 25 m ⁻² , Shoot densities of 277 to 1022 shoots 25m ⁻²
Owen Anchorage	OA14	6.5	<i>Posidonia australis</i>	Seeds	December 2014	7.5 years	Three seeded plots varied in cover from large patches to 80% coverage of the 25m ² seeded area. 29-113 individuals 25 m ⁻² , Shoot densities of 540 to 2065 shoots 25m ⁻² . Note there was a lot of infilling of sand patches by <i>Posidonia sinuosa</i> .
Success Bank	ecosub 1	5	<i>Posidonia sinuosa</i> , <i>Posidonia coriacea</i> , <i>Amphibolis griffithii</i>	Sods	1996-2001	26 years	Square edged patches, width increased 2-3x, mixed meadow still, good shoot densities

Success Bank	ecosub 2	6	As above	Sods	1996-2001	26 years	Square edged patches, width increase in some, mixed meadow still, good shoot densities
Oyster Harbour, Albany	OH_97	2	<i>Posidonia australis</i>	Sprigs	December 1997	25 years	~239 shoots m ⁻² , not significantly different from natural meadow
Oyster Harbour, Albany	OH_03 /04	2	<i>Posidonia australis</i>	Sprigs	December 2003	19 years	~197 shoots m ⁻² , is significantly different from natural meadow
Princess Royal Harbour, Albany	PRH_D	4	<i>Posidonia australis</i>	Sprigs	December 2003	19 years	~99 shoots m ⁻² , is significantly different from natural meadow

5.1 Seed Based Restoration

The potential for seed and seedling-based restoration was investigated during the Seagrass Research and Rehabilitation Plan (Sinclair et al. 2009, Statton et al. 2013) by University of Western Australian researchers and has continued through Australian Research Council Industry Linkage grants partnering with Cockburn Cement/Adelaide Brighton Pty Ltd (LP100200429, LP130100155, LP130100918, LP160101011). Also, the Seeds for Snapper seed-based community restoration program grew from these initial experimental studies and has been running for 4 years. From these studies we are able to assess the success of seed-based restoration across a timeline from ~2 months to 6.5-7.5 years after seeding. We will be presenting data for central Owen Anchorage off Coogee Marina at a site called BA15 (Northern Owen 2015), where the highest rates of seedling survival and growth were recorded and from Cockburn Sound on the edge of the Kwinana Shelf at a site called CSE 14 (Cockburn Sound East 2014) where, despite annually seeding in 2014 and 2015, recorded the lowest seedling survival and growth. Mortality in BA 15 was driven by winter storms that penetrated 6.5 m depth and created 10 cm standing waves across unvegetated sands. Mortality at CSE 14 was due to predation and bioturbation from invertebrates (sand dollars and blue manna crabs) and from rays that create pits throughout the restoration area. Note CSE 14 was also the deepest site at 7.8 m and growth and shoot production was much reduced compared with the Owen Anchorage sites.

The area of seabed that was seeded in 2021 by Seeds for Snapper was 2,100 m² and they dispersed ~375 000 seeds at ~85 seeds m⁻². When we assessed the Seeds for Snapper sites in February 2022 (~2 months old), the range in natural recruitment from seeds was 0-2 seedlings m⁻², compared to 5-46 seedlings m⁻² in our seeded plots (Figure 30). The variability in seedling densities among seeded locations demonstrates the strong site-specific nature of recruitment from seeds. 2BA18 is the most northern site and is heavily impacted by swell from storms and 10-25 cm sand waves are common. Despite seeding occurring at this site in 2019 and 2021 at >100 seeds m⁻² recruitment from seeds is among the lowest we have recorded. Similarly, S4S 1 which is 1.5 kilometres offshore of the Ammunitions Jetty on Coogee Beach (Figure 29), has low seeding recruitment but in this case the sediment was covering shallow reef pavement restricting root penetration. Nearby in southern Owen Anchorage, at Seeds for Snapper sites S4S 2 and S4S 3 recruitment from seeds was exceptionally high and densities of recruiting seedlings were recorded at 17 and 46.5 seedlings m⁻², respectively from a single seeding in November 2021 of 80 seeds per m⁻². The two seeded sites in Cockburn Sound S4S 4 and S4S 5 (80 seeds per m⁻²) also had high recruitment from seeds with seedling densities of 34 and 20 seedlings m⁻², respectively. S4S 5 was also heavily bioturbated by sand dollars so we expect high seedling mortalities over the first year at this site (Johnson et al. 2018).

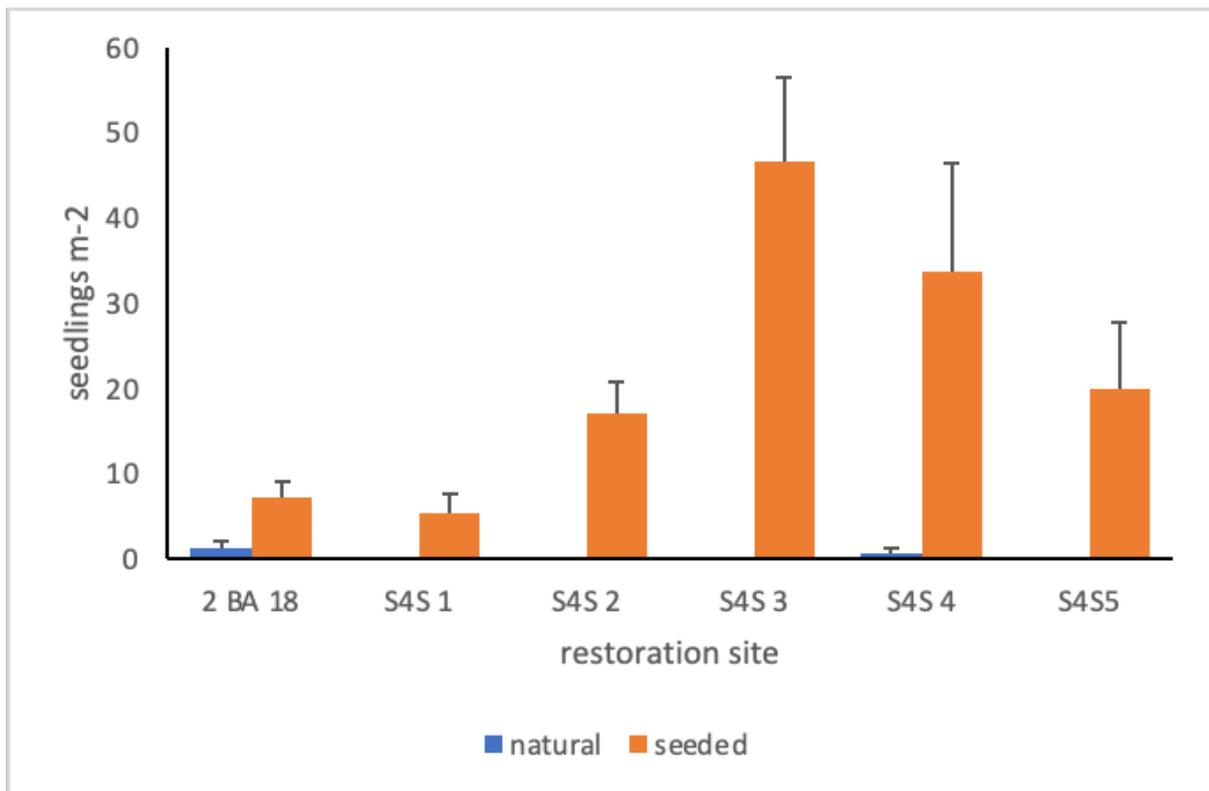


Figure 30: Top: Numbers of seedlings m⁻² recorded at the 6 sites seeded in the S4S 2021 restoration season, and; Bottom: Image taken in 2022 at a restoration site that was seeded ~2 months earlier with *Posidonia australis* seeds as part of the Seeds for Snapper project, surrounding control areas showed no to low natural recruitment from seeds (quadrat is 50x50cm). (data and imagery from Rachel Austin)

The Coogee (BA15) seed-based site was seeded in November 2015 (6.5 years ago) with 5,000 *P. australis* seeds 25 m² for each of 3 experimental plots, we found dense patches of seagrass that were in the process of merging into meadows (Figure 31, bottom) but the natural seed recruitment plots (controls) had few to no patches. This site was monitored for seedling survivorship and shoot numbers were also counted for each seedling between December 2015, February and April 2016, December 2017, October 2018 and February 2022. Survivorship followed a type III survivorship curve with

mortalities highest in the first two years of seedling life in both seeded and naturally recruited experimental plots (Figure 31A, C). In the seeded plots, 20 to 40 individuals survived 6.5 years whereas in the naturally recruited plots, 0 to 2 individuals survived. Interestingly after 6.5 years, shoot densities per plot in seeded plots were at least 5 times and as much as 25 greater than naturally recruiting plots (Figure 31B, D). Also, densities of shoots in seeded plots after 6.5 years were on average as high as initial 1 month survivorship of seedlings.

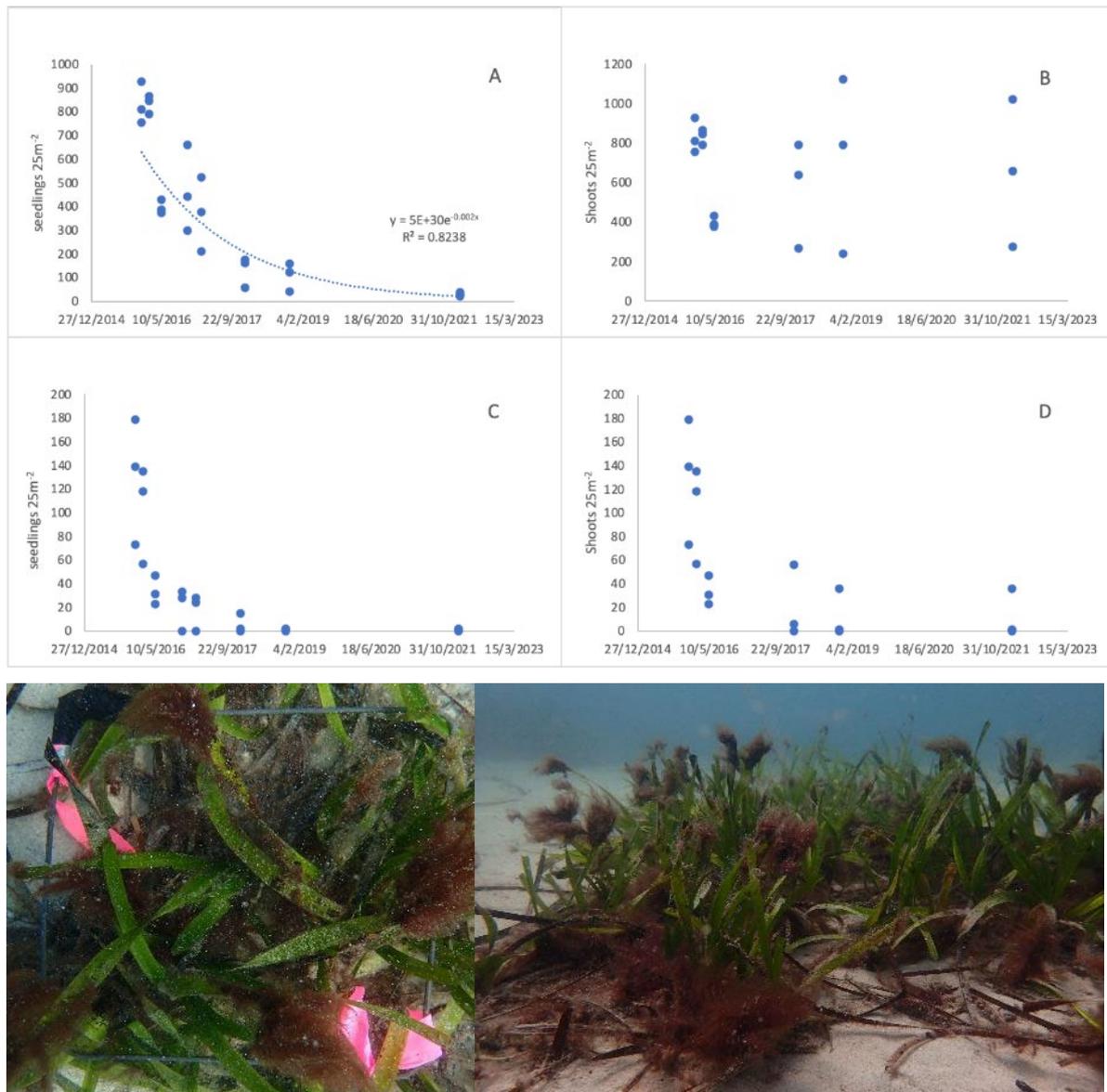


Figure 31: Top: Coogee Beach (BA R15) seedling survival (seedlings 25m⁻², ± SE, n=3) and shoot density (shoots 25m⁻², ± SE, n=3) in seeded (A and B) and natural recruited (C and D) 5 x 5 m experimental plots. Bottom: Images taken in 2022 at Coogee Beach (BA) that were seeded with *Posidonia australis* seeds in November 2015 (7.5 years old) showing that given time this method will result in the formation of meadows with adequate time (quadrat is 25x25cm). 5,000 seeds were delivered to each of the experimental plots (Photos by Rachel Austin).

The Kwinana Shelf, Cockburn Sound (CSE 14) seed-based site was seeded in November 2014 and 2015 (7.5 and 6.5 years ago) with 5,000 *P. australis* seeds 25 m⁻² (total 10,000 seed deployed) for each of the 3 experimental plots (5 m by 5 m). We found a few small patches of seagrasses in seeded plots that

had not grown together and natural seed recruitment plots (controls) had few to no plants. Survivorship followed an extremely steep type III survivorship curve with mortalities highest in the first two years of seedling life in both seeded and naturally recruited experimental plots (Figure 32A, C). In the seeded plots, 9 to 15 individuals survived 7.5 years whereas in the naturally recruited plots, 1 to 2 individuals survived. Interestingly after 7.5 years, shoot densities per plot in seeded plots were 4 X to 15 X greater than naturally recruiting plots (Figure 32B, D). Also, densities of shoots in seeded plots after 7.5 years were on average as high as initial survivorship of seedlings. Survivorship and shoot density at CSE14 on Kwinana Shelf were 1.5 - 3 and 2.2 - 8 X greater but less than that observed at BA15 in Owen Anchorage, respectively.

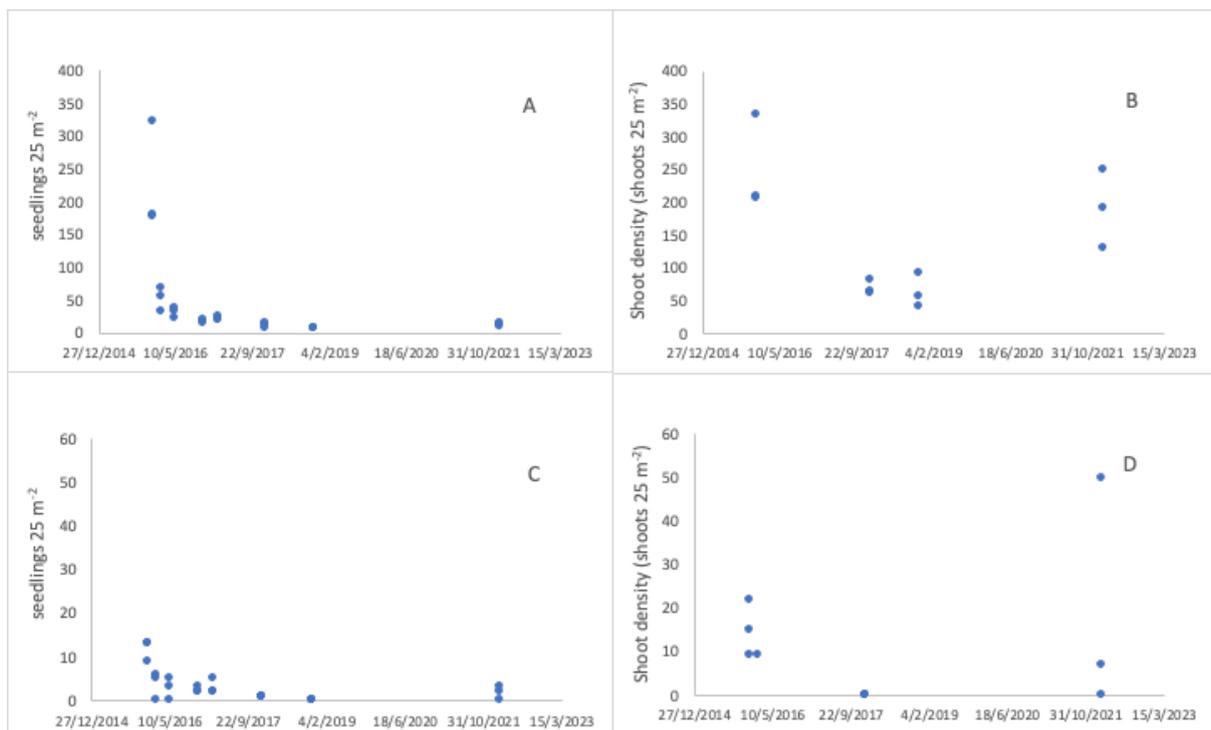


Figure 32: Top: Cockburn Sound East (CSE 14) seedling survival (seedlings 25m⁻², \pm SE, n=3) and shoot density (shoots 25m⁻², \pm SE, n=3) in seeded (A and B) and natural recruited (C and D) 5 x 5 m experimental plots.

The graphs and images in Figures 21 and 32 demonstrate that seed-based restoration programs have the capacity to restore areas of seagrass in Cockburn Sound with sustained effort and allocation of time for growth. Environmental drivers are location specific, and in this comparison the more successful location in Owen Anchorage was impacted by ocean swells from winter storms whereas the more sheltered location in Cockburn Sound was influenced by high levels of bioturbation at both the individual seed and plot scale, with individual seeds redistributed and removed by sand dollar bioturbation and plots destroyed by sand ray feeding on sediment invertebrates. Habitat suitability is paramount to determining long term success of seed-based restoration.

Seagrasses, like most plants, demonstrate a Type III survivorship curve. To effectively restore unvegetated areas and enhance natural seed recruitment, large losses between the time of seeding with seeds and the survival of seedlings to 1 to year old plants occurs. We have recorded mortalities of >90% of original numbers of seeds and those losses need to be acknowledged and expected in seed-

based restoration programs.

5.2 Transplant Based Restoration

There have been many restoration projects that have utilised transplanting throughout the past 30+ years allowing us to thoroughly assess the long-term success of this method. One Murdoch University led project looked into transplant success over a depth gradient from 2-9m at Woodman Point spit in 2009. After 13 years of growth, in line with the initial monitoring results, the transplants in the shallow waters (2m and 4m) showed significant growth, forming dense patches well on their way to long term meadows (Figure 33). But those in deeper waters did not survive nor develop patches (8m and 9 m). It should be noted that the location for this project is highly exposed to all wind and swell conditions which does slow growth and must be considered when interpreting or comparing results.



Figure 33: Images from the 'Woodman Point Depth Transect' transplant trials show good shoot density and the formation of sizable patches (the square shape of the plot is still visible) as a result of the transplants (quadrat is 25x25cm). Photos by Gary Kendrick.

Another Murdoch University led project located on Southern Flats transplanted 3.1 hectares of *Posidonia australis* (at 0.5-1m spacing) between 2006-2008. We compared natural and restored meadows in 2022 (15th February, 2022). Shoot density and % cover was measured at two of the 1 hectare plots (T2 and T3) and at a natural meadow nearby (C1) (Figure 34 A, B). Neither shoot density or % cover in transplant sites were statistically significantly different compared to the natural meadow, with shoot densities (Figure 34A) from 445 to 589 shoots m^{-2} and % cover (Figure 34B) from 75% to 78%. The growth of transplants that were planted between 2006 and 2008 have resulted in the development of extensive meadows over the last 16 years and can be seen clearly from aerial imagery (Figure 34 Bottom). Sexual reproduction has been observed in the restored meadows with flowers recorded in July 2010 after just 4 years of growth. This project in particular demonstrates that large scale (at 1-10 hectare) seagrass restoration in Cockburn Sound is possible and meadows can be restored within a decade.

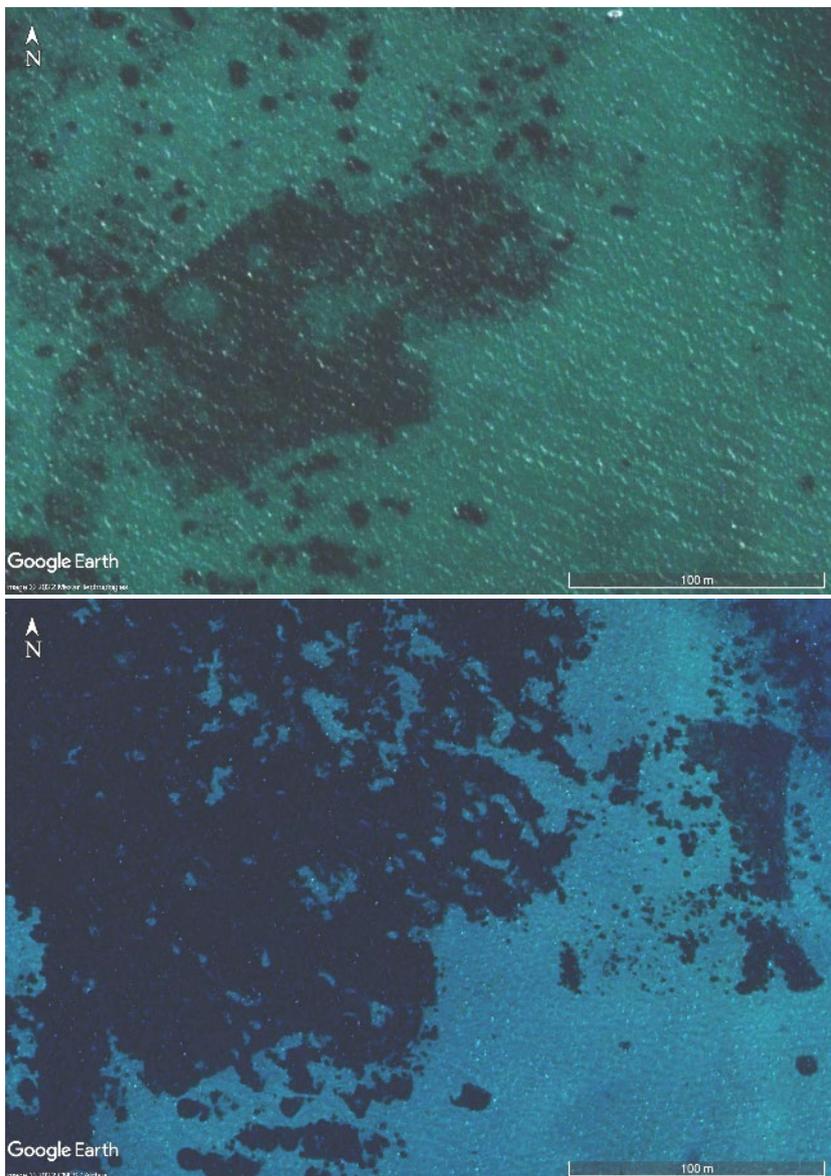
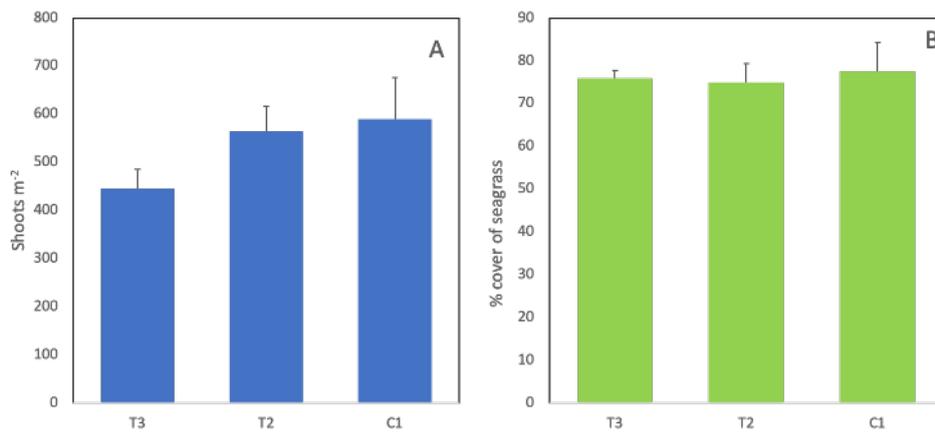


Figure 34: Success of the 3 x 1 hectare rehabilitation sites at Southern Flats. Top: Shoot density (shoots m⁻²) and % cover of seagrass (mean ± SE, n = 6) measured at transplant sites 2 (T2) and 3 (T3) and a control site (C1) in a natural meadow on 15th February, 2022. Bottom: the top aerial image is from March, 2011 and the bottom image is from November 2021 (Images from Google Earth).

5.3 Restoration in Albany

Seagrass restoration at scale has been conducted in Albany with *Posidonia australis* and some of these sites were recently visited by third-year students from UWA (Table 5). In Oyster Harbour large transplanting efforts were conducted in 1997 and 2003/4 led by Geoff Bastyan (Oceanica 2013). Compared to surrounding natural meadows (276±11 shoots m⁻²), shoot densities in the 1997 planted meadows were not significantly different (238±11 shoots m⁻²), but those planted in 2003/4 were (197±13 shoots m⁻²). Transplanting also occurred in Princess Royal Harbour in 2003/04 and shoot densities in that meadow (99±12 shoots m⁻²) were significantly lower compared to surrounding natural meadows (222±16 shoots m⁻²). While the Albany Harbours system is quite different to Cockburn Sound, it further demonstrates that given ecologically relevant lengths of time, seagrass restoration does lead to functioning meadows.

Table 5: 2022 Shoot densities (shoots m⁻²) at Control (naturally recruited) meadows and 1997 and 2004 restoration programs in Oyster Harbour and Princess Royal Harbour (Oceanica 2013).

	Oyster Harbour			Princess Royal Harbour	
	Control	1997	2004	Control	2004
Mean	276.8	238.93	197.33	222.4	99.2
Std Dev	60.62	60.88	70.73	86.96	38.31
Median	280	224	208	224	104

5.4 Sod based Restoration, Success Bank

The Ecosub project (I and II) used mechanical engineering to transplant large cubes (named sods, 0.25 m² and 0.55 m² in seagrass area) of seagrass over a 5 year period between 1996 and 2001 (Paling et al. 2001b). Note the Ecosub was designed to transplant into hydrodynamically active environments where up to 25 cm of sediment burial and erosion have been recorded and related to ocean swell and storm waves and currents (van Keulen et al. 2003). The sods were planted in either rows 0.5m apart, or in grids with varying distances apart to test infilling rates. Today, after 21-26 years, the surviving sods have grown extensively, merging with their neighbour sods, but interestingly they have kept their square shape in some areas (Figure 35).

Recent measurements of 4 replicate 0.25 x 0.25 m quadrats were taken from a number of sods randomly across the original transplant sites (Site 1 n=8: site 2: n=7) (Table 6). It is difficult to ascertain the actual planting size from records so our interpretation is limited to observation of present size of sods and whether sods have coalesced. Some sods have increased their width by up to 300% and there has been some coalescence. They have also maintained the original mixed species composition, with shoot densities reaching up to 1108 shoots m⁻² for mixed *Posidonia* species (*P. coriacea* and *P. sinuosa*) and up to 504 shoots m⁻² for *Amphibolis* species.

Table 6: Shoot density (shoots m⁻²) in the ecosub sods counted in May 2022. Sods were planted in rows of varying length, within an identified row four replicate 25x25cm quadrats were randomly sampled.

Ecosub Site	Sod Rows Assessed	Mean Shoot Density		Range in Shoot Density	
		Posidonia	Amphibolis	Posidonia	Amphibolis
1	8	397 ± 116	230 ± 51	72-1108	16-380
2	7	498 ± 94	127 ± 67	0-752	8-504



Figure 35: Images of the Ecosub sods showing considerable growth and merging into small meadows after 21-26 years. (Rachel Austin)

The outcomes from the 2022 revisit of restoration sites were:

1. Twenty three of the 31 areas that were restored with small (10-100 m²) to medium (1,000 m² - 1 hectare) scale restoration and rehabilitation programs showed partial or full success in restoring seagrass cover with shoot densities that were comparable to natural seagrass habitats.
2. Natural recruitment was also high across the southern and western banks in Cockburn Sound, on Parmelia Bank and in the shallower eastern parts of Owen Anchorage suggesting environmental conditions were favourable for recolonisation.
3. Seed based programs that focus on larger sand habitats had both initial (2 months) and longer term (6.5 - 7.5 years) successes. The seed-based programs follow a type III survivorship curve and from millions of seeds only 10s of patches develop. These form the nucleus for further natural recolonization.
4. Sprig based restoration programs are highly successful if placed in the right environment and the largest successful restoration programs in Cockburn Sound (3 hectares) and Oyster Harbour, Albany (1 hectare) have worked as nucleation sites for high levels of natural recruitment and growth.
5. Sod based industrial planting may have a role in highly disturbed environments like Success Bank, but they are hard to justify as a broad scale restoration method because of cost.

6 Discussion

The outcomes from the historical and present-day restoration programs summarised in this review (Table A2) indicate that:

1. **Environmental conditions and processes are extremely important.** These include levels of light (depth), seasonal and depth-related variation in water motion (orbital: and benthic shear velocity), seabed and sediment movement associated with hydrodynamics, sediment anoxia, organic C and S, and grazers and bioturbators.
2. Our monitoring shows in seed, seedling, sprig and plug restoration programs that **mortality is both site and time specific** and quite unpredictable over local scales. Most studies that monitor for greater than 1 year indicate a >30% survival and a doubling or trebling of shoot density within 2 years for those that survived.
3. **Scaling up requires the environment to be modified** such that natural recruitment matches or exceeds that of restoration. This was noted in Southern Flats, Oyster Harbour and Princess Royal Harbour restoration programs.
4. **Our focus** on small scale experiments **does not prepare us for scaling up** and/or language of restoration and rehabilitation also seems inappropriate given that we are building resilience and augmenting natural recovery processes rather than rehabilitating or restoring a lost habitat.

Also, we worry too much about individual survival from single restoration programs when spatial and temporal patchiness in the environment at the local scale derails us again and again. **We should measure but not expect high survival of transplanted propagules but compare restoration survivorship to local natural population dynamics as our indicator of restoration success.**

Seed and sprig-based methods have been the most successful in the context of survival, growth and increased seagrass cover and is also cost-appropriate. Scaling up will require community volunteers with citizen science approaches and industrialization of the process of seeding and planting.

Modifications to enhance survival of restoration units have also been studied and/or need more detailed study. Some modifications include: seed nurseries to grow out seeds to 6-month old seedlings before transplantation (Statton et al. 2013); hessian bags (Tanner 2015) or tubes (Statton et al. 2020) to both modify hydrodynamics and stabilise sediments, meshes (MacDonnell et al. 2022) and artificial reefs to protect seedlings and sprigs during early development; plugs and cores to enhance survivorship and to stabilise sediments in wave swept environments (van Keulen et al. 2003), and; control of grazers and bioturbators in sheltered environments (Statton et al. 2015, Johnson et al. 2018).

Sprigs are easy and quick to collect and plant, and therefore cost-effective at scales of 1,000 m² to hectares. They also have the highest recorded survival, growth and infilling, and return to ecological function of the methodologies we have reviewed. Yet there are major limitations to industrialising sprig-based restoration including: the damage to donor beds when collecting wild-grown sprigs; the effect of disturbance to the surrounding sediment and to the rhizosphere of the sprig when planting, and; species-specific differences in the morphology of the planting units (e.g. *Posidonia* and *Amphibolis*) and the effect that has on the rate of survivorship and growth when sprigs are recovering from harvesting and planting.

Seeds are also easy to collect and disperse to restoration sites, are seasonal in their availability and most seeds would be lost to either marine environments unsuitable for establishment and growth or

wash up on beaches, so collecting and dispersing them into suitable coastal marine habitats enhances regional recruitment. There is the existing citizen science program in Cockburn Sound and Owen Anchorage called ‘Seeds for Snapper’ and represents a collaboration between Ozfish, BCF, RecfishWest and the University of Western Australia. The focus in Cockburn Sound has been on *Posidonia* seeds and *Amphibolis* seedlings but *Halophila ovalis* has been studied and can be developed further through mixed species seeding and co-planting. Also, there are limitations in seed-based restoration including: higher rates of mortality in the first 2 years of a recruiting seedlings life; dependence on good flowering and seed set to maintain levels of collection and dispersal, and; they are more influenced by hydrodynamics, grazing and bioturbation (Statton et al. 2017a).

7 Conclusions

Both existing sprig and seed-based restoration programs have developed viable methods to use for revegetation of hectares of unvegetated seafloor. Methodologies have developed from small experiments of 1-10s m in size testing species, methodology, donor meadow impacts, and environmental drivers of success and failure (Figure 36). These programs have led to larger scale (0.3 - 3 hectares) demonstrations of restoration at scale and cost. The **3 big drivers of restoration success** are: **the environment** needs to be suitable for seagrass colonisation; **selection of transplant unit**, either sprigs, seedlings or seeds, needs to be made with consideration of biological and physical environmental drivers, and; consideration of **bottlenecks in the seagrass life history**, where predicted effects on survival of transplant units should determine the milestones of the program. We also recommend community-based citizen science and community restoration as the most cost-effective approaches to increasing scale of restoration.

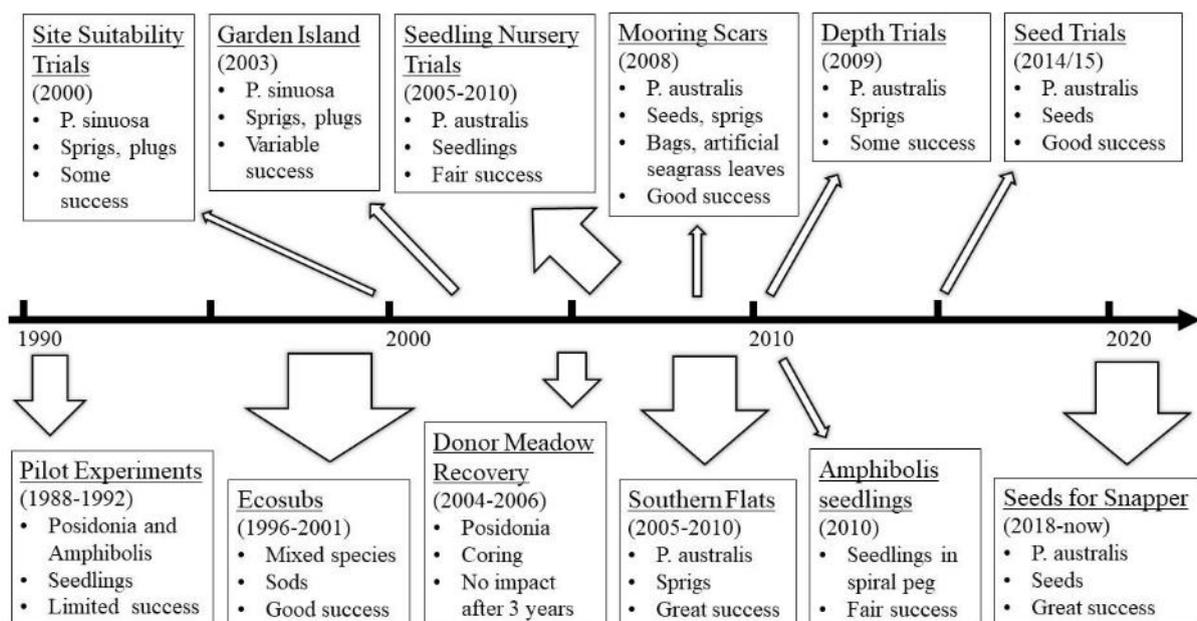


Figure 36: Timeline of major restoration projects conducted in the Cockburn Sound region.

Restoration programs have clearly shown that the shallow coastal environment into which sprigs or seeds are placed needs to be suitable for natural seagrass colonisation. Our revisiting of seagrass restoration programs demonstrated that natural recolonisation had also occurred, sometimes obscuring the restoration results. In reality, a restoration action should be designed to **increase the rate of recovery** of seagrasses into areas lost, thus more accurately we are **ameliorating loss and augmenting recovery of impacted seagrass habitats** and therefore **building resilience in seagrass dominated coastal ecosystems**. With removal of the environmental drivers that caused seagrass loss and system collapse we have shown restoration programs form the nucleus to rapid natural recovery.

The present OZFISH 'Seeds for Snapper' seed-based restoration program is cost effective as it does not invest in early life history survival, does not grow out seedlings in costly nurseries (e.g. Statton et al. 2013) and accepts that the >90% natural losses of seedlings in the first year of life will also occur with seeds and transplanted seedlings in restoration programs (e.g. Statton et al. 2017a). Therefore, seed based restoration is a numbers game where millions of seeds will be required to restore hectares of unvegetated sand. We also built in a process of seeding over multiple years to reduce the impacts of interannual biological and physical drivers of seed mortality. The checks and balances in the restoration method and the cost effectiveness of this approach in Cockburn Sound, an area with high levels of natural flowering and seed production, is advantageous for rapid recolonisation of impacted areas in the Sound.

The sprig-based restoration programs are also successful at scale, are cost effective and can be used to target areas of high intrinsic value or areas where more environmental modification is required. Transplant success using community volunteers has been shown both at Southern Flats, Cockburn Sound (3 hectare transplant area) and Oyster Harbour, Albany (1 hectare area). Sprig survival has been generally as high as 90% and their growth over 15-20 years has resulted in new meadows being formed. Mechanisation of sprig planting and hessian bag preparation and delivery to the bottom would be areas of savings to an already reasonable cost (approx \$35,000 community volunteers: approx \$300,000 commercial).

Monitoring for restoration outcomes regularly ignores the life history imperatives of natural populations. Seagrasses, like most plants, have a type III survivorship curve, an exponentially declining survivorship (Statton et al. 2017a). We need to utilise this curve to our advantage in planning and managing restoration efforts. We should not be concerned with substantial loss of transplants and seeds in the first years after the restoration activity but focus on maintaining expected losses as predicted from the seagrass survivorship curve. After 2 years most sprig transplanting programs described in this review have had >30 % survival of the restoration units, which fits expected survivorship and we recommend any survivorship greater than this level is the benefit the restoration program brings on top of natural recruitment and growth.

8 Guidelines

Our guidelines for developing a broad spatial scale multi-year plan for restoring seagrasses to Kwinana Shelf and Owen Anchorage has been developed from our critical assessment of the outcomes of 10s of small-scale restoration experiments and 4 broader-scale restoration programs in the area. These activities date back to the 1980s and demonstrate the increase in our joint understanding of how to restore seagrasses (e.g. Oceanica 2013). A compilation of the outcomes of previous and existing restoration programs described in our review (Table S2) indicated that restoration success was most influenced by the following major factors.

1. **The suitability of the environment to be restored.** This includes an understanding of the biological and physical processes that may impact successful restoration.

The biological processes that have disrupted restoration efforts include grazers and bioturbators, competition with epiphytic and drifting algae, and status and condition of sediment microbial processes. The physical processes include light availability, wind exposure, and water motion, including orbital velocity and shear velocity at the sediment.

Natural recruitment into restored areas indicates that the environment is suitable for restoration. Scaling up sometimes requires the environment to be modified such that natural recruitment can also occur. Natural recruitment was observed in Southern Flats, Oyster and Princess Royal Harbours, Albany during restoration programs and recently mapped for Adelaide Coastal Beaches (Fernandes et al. 2022).

2. **Selection of transplant units.** Existing monitoring indicates that in seed, seedling, sprig and plug restoration methods, mortality is both site and time specific and unpredictable over local scales. Interestingly, success in sprig transplanting has been greater in sheltered shallow water (<3m) environments whereas seed-based methods have been more successful in deeper (>4.5m) environments below effects of wind and waves. This depth separation of the methods should drive selection of methodology and future efforts.

3. **Understanding the bottlenecks in the seagrass life history.** Here we suggest we use a type III survivorship curve common to terrestrial and marine plants as our benchmark and continue the modelling of Statton et al. (2017a) to determine the most sensitive stages of the life history when predicting suitability of a restored seagrass habitat.

When determining whether sprig restoration has been a success, comparing the restoration outcomes to natural colonisation and spread should set the benchmark, not 100% survival as it is at present. Most sprig restoration studies that monitor for greater than 1 year indicate a >30% survival combined with a doubling or trebling of shoot density within 2 years for those that survived. An adaptive strategy where sprig transplanting is done every spring for a few years to address losses is also recommended.

Similarly, the seed to early recruit (2 month old seedling) and the early recruit to 1 year old seedling are the most sensitive stages in the life history of seagrasses. Increasing the density of seeding (50-100 seeds m⁻²) and the number of seeds we use (millions), restoration can be scaled up to hectares of unvegetated bottom restored.

4. **Expectations of restoration success were unrealistic.** The language of restoration and rehabilitation seems inappropriate given that we are building resilience and augmenting natural recovery processes, not rehabilitating and restoring an altered environment.

Restoration programs have been driven by government decision making and planning that have been specific to individual coastal developments without a broader whole of Cockburn Sound/Owen Anchorage context. The milestones were onerous and resulted in efforts without the adaptive decision making required when restoring large areas. Expectations for 100% success spatially to be met with tight timelines was not representative of natural processes of seagrass recruitment and colonisation that require us to accept and understand the critical bottlenecks in seagrass life histories and the slow growth rates of the temperate seagrasses restored. We need to regroup and revise government guidance around seagrass loss and restoration.

Partly to address the timeline and budgetary issues, the Oceanica 2013, 3 hectare sprig restoration program and the present seed-based restoration program have heavily depended on community volunteers to meet their outcomes and targets. Building community capital and good community outreach through citizen science programs like the OZFISH 'Seeds for Snapper' is imperative to shift community perceptions about seagrass and restoration (Abelson et al. 2020, Sinclair et al. 2021).

Considering these 4 major factors identified from this review of restoration activity in Cockburn Sound and Owen Anchorage our intention is to follow an existing decision-making framework for seagrass restoration (Statton et al. 2021) to plan the activities for WAMSI Westport project 2.3 (Figure 37). There are five key areas to consider in a restoration program: planning for restoration; sourcing plant material; optimising establishment; facilitating growth and survival, and; developing ecosystem function, sustainability and landscape integration. Through these key areas we have identified where additional research is required to achieve successful, long-term restoration.

Guideline 1 (Planning for Restoration)

To address the suitability of the environment and for long term planning and direction of restoration efforts, spatially explicit restoration suitability models need to be developed with a focus on the Kwinana Shelf and Owen Anchorage regions, but also need to incorporate all Cockburn Sound including Southern Flats, Success and Parmelia Banks. The restoration suitability model will allow selection of the most likely environments for restoration based on major environmental drivers like light climate, hydrodynamic forcing, sediment quality and biological impacts of grazing and bioturbation. The modelling will also allow us to set goals for environmental thresholds for successful seagrass restoration (Figure 37: Understanding the Problem).

The overarching goal in the next 2 years is to demonstrate the capacity to scale up restoration such that both historical and contemporary losses of seagrasses in Cockburn Sound and Owen Anchorage can be addressed (Figure 37: How Much to Restore)

Guideline 2 (Sourcing Plant Material)

Initially, plant material will be sourced from wild populations that have similar genetic provenance to the restoration location (Figure 37: Genetics). Significant genetics research has shown the genetic diversity and relatedness among populations of *Posidonia australis* in Cockburn Sound and Owen Anchorage (Sinclair et al. 2014) including the potential drivers and source-sink relationships that have resulted in the genetic connectivity of *P. australis* in Cockburn Sound (Ruiz Montoya et al. 2012; Sinclair et al. 2018). Population genetics of *P. sinuosa* will be determined through WAMSI Westport Project 2.3 and *Amphibolis* is being investigated through an ECU PhD student project.

Posidonia and *Amphibolis* will be the main genera used for restoration and the plant sources will be sprigs, seeds and seedlings given their historical success in the Cockburn Sound region. To scale up to 10s -100s of hectares we need to assess the volume of source materials required (Figure 37: Transplant adult shoots, source seeds and seedlings). The WAMSI Westport restoration program will focus on collecting whole plants for our sprig experiments and collecting seed and seedlings from existing flowering populations in Cockburn Sound and Owen Anchorage. We are also planning to collect ocean drifting and beach cast sprigs and seeds of *Posidonia* and sprigs and seedlings of *Amphibolis*. Beach cast sprigs have already been used successfully in the project 'Operation *Posidonia*' which is a community-based restoration project in NSW (Ferretto et al. 2021). This will require setting up a year-round aquaculture facility and will require discussion with the Westport leadership team and industries. Over time we may need to create ocean nurseries in Cockburn Sound from successful restoration programs but this will not be immediate nor is it initially important.

We will also be assessing natural recovery as a baseline to measure restoration success and to focus restoration where recruitment and recovery is possible. Before we expend too much effort on large scale restoration programs we will be deploying smaller restoration units as indicators of suitability of the environment for restoration (Figure 37: Natural Recovery).

Guideline 3 (Optimising Establishment and Facilitating Growth and Survival)

To optimise establishment and facilitated growth and survival of seagrass units we have developed a series of cross theme experiments in the following areas: plant level hydrodynamics to assess both suitability of restoration habitat and to assess modifications required to enhance seed, seedling and sprig recruitment and growth (Figure 37: Above Ground Environment, Surface and Landforms); experiments on modifying the influence of sediment processes and sediment health on seed, seedling and sprig survival (Figure 37: Below Ground Environment); and to develop methods to control grazing and bioturbation (Figure 37: Biotic Interactions). These will be studied for seeds, seedlings and adult transplants.

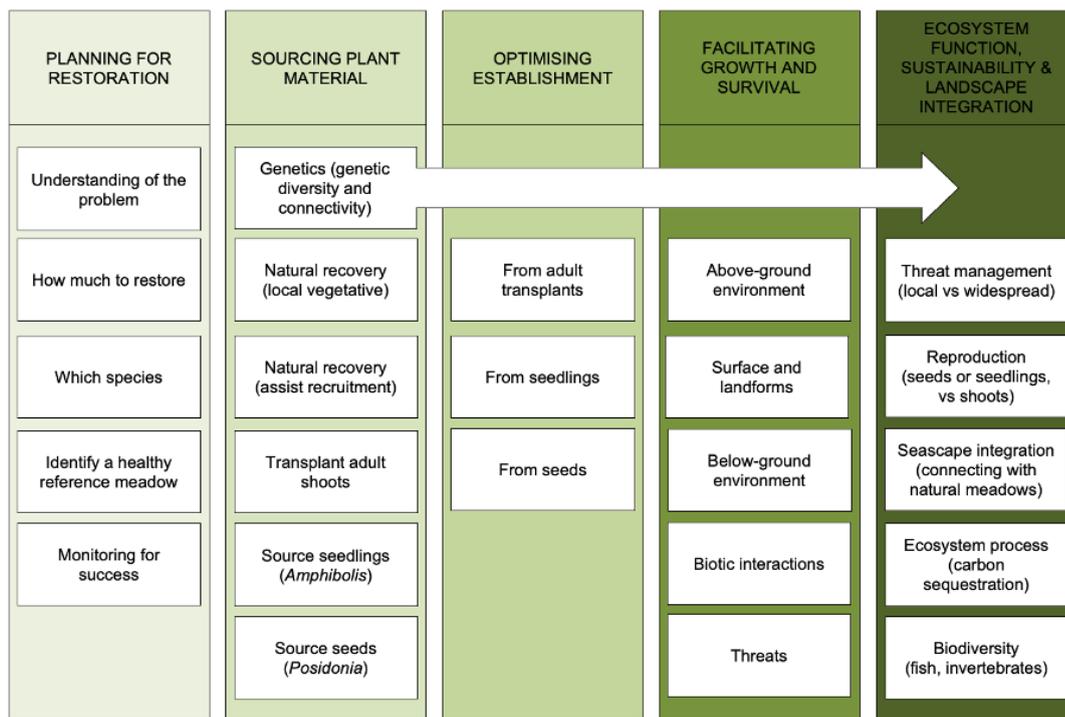
To scale-up our efforts, we propose three foci: 1) to build the existing seed-based OZFISH 'Seeds for Snapper' restoration program, and incorporate some industrialisation in the existing 3 step process; 2) to experiment scaling up sprig-based restoration with multiple species and by optimising establishment using environmental modifications, and; 3) by trialling use of dredge spoil to shallow restoration sites and to cap anoxic sediments with dredging sands. The environmental modifications may include hessian bags, artificial substrata and artificial reefs to enhance seed, seedling and sprig establishment.

Guideline 4 (Ecosystem Function Sustainability and Landscape Integration)

Threat management across Cockburn Sound will be required for return of ecosystem function through restoration and some protection and management of restoration locations is necessary given the diverse and conflicting uses of the shallow coastal waters in the Cockburn Sound region (Figure 37: Threat Management). This is an area beyond the scope of the seagrass restoration project alone but needs to be highlighted as a concern from the WAMSI Westport Program.

We recommend monitoring and working with all projects in the Benthic Habitat Theme to assess seascape integration, ecosystem processes and biodiversity (WAMSI projects 2.1, 2.2, 2.4). Planned activities include: annual monitoring of reproduction across the Cockburn Sound region as a part of the 'Seeds for Snapper' program; seascape integration from past and planned restoration programs through fine scale mapping in collaboration with WAMSI project 2.1, and; deployment of benthic chambers to address ecosystem processes in restored versus natural meadows (carbon and nitrogen release from sediments into the water column) partly funded by CSIRO and the Australian Research Council Linkage Grant Scheme.

The development and maintenance of close relationships with the local community including indigenous groups, fishers, boaters, divers and beach walkers, and other users will determine community uptake of seagrass restoration as both appropriate and viable to maintain ecosystem health and function. We already have strong collaborations with OZFISH and RecFishWest with the 'Seeds for Snapper' program but believe an onshore aquaculture facility is required to drive year-round collection of storm cast sprigs of *Posidonia* and seedlings of *Amphibolis*. Storm cast fragments have better survivorship and growth if kept in seawater aquaculture tanks for 3-4 months before transplanting (Ferretto et al. 2021). Clearly a centralised aquaculture facility offers an opportunity to increase our citizen science activities from the months of October to December to occur throughout the year.



(adapted from Miller et al. 2017 for marine restoration)

Figure 37: Framework as a practical guide to decision making for appropriate restoration activities. Adapted for marine restoration from Miller et al. 2017 by Statton et al. 2021.

In summary, our guidelines focus on the major limitations outlined and summarised in this review. We propose that to address environmental limitations we need to measure the environment to model restoration suitability and to map suitable habitats for restoration. This will aid in site selection and assessment and monitoring (Figure 38). Another approach we will explore is large-scale modifications of the environment. Previous successes led us to propose sprig, seed and seedling-based restoration approaches with modifications and enhancements through sandbags and socks, artificial substrata, artificial reef, sand capping using dredge products, and controlling grazers and bioturbators (Figure 38: Methods and Enhancements). We emphasize that we need to use a more biological focused approach to restoration where the life history and bottlenecks associated with the life history are built into our expectations and predictions of successful restoration (Figure 38: Restoration Genetics and Assessment and Monitoring). Survivorship curves from natural and restored meadows give us insight into the value added by restoration to natural biological processes and will be incorporated in both restoration suitability assessments and in monitoring for success. Finally, community engagement plays a fundamental role in the scaling up of seagrass restoration in Cockburn Sound and Owen Anchorage and should be fostered and grown for cost effective restoration (Figure 38: Community Engagement).

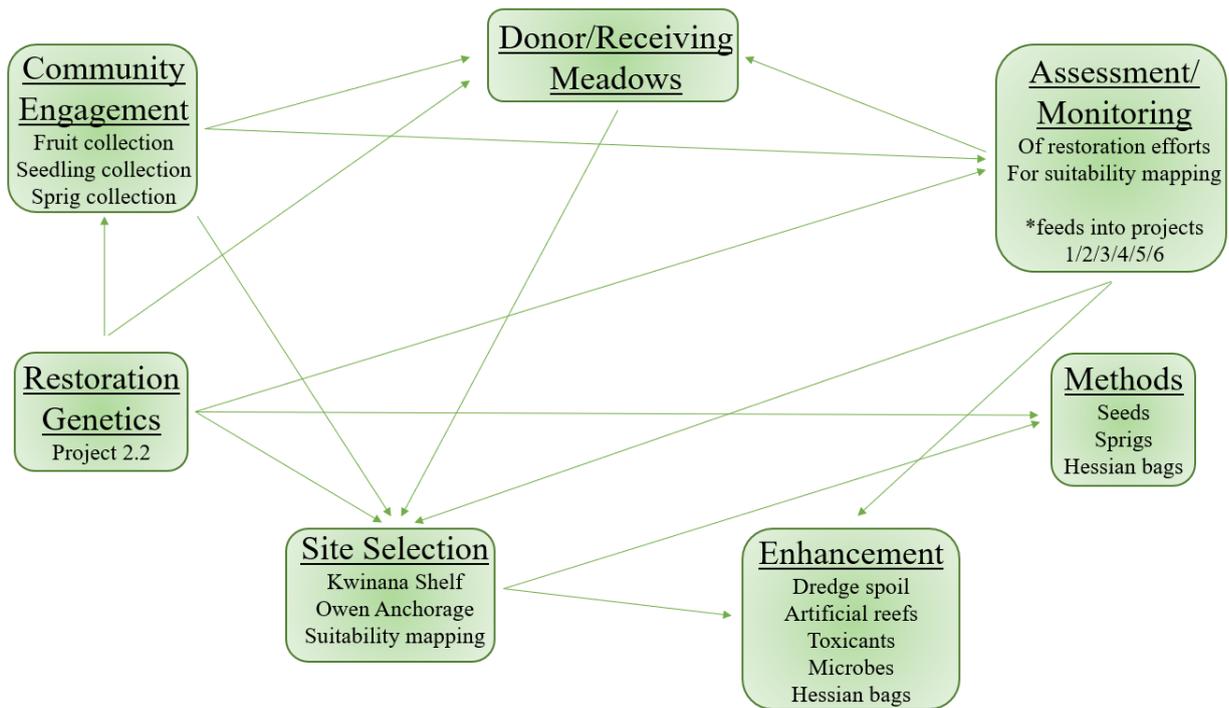


Figure 38: Network diagram summarising major components of a restoration program and their connectivity.

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10 Appendices

- 10.1 Appendix A1: Restoration Locations visited in 2021-2022 with details including Location, site code, latitude and longitude, lead researcher, depth, species restored, method, experimental design, date started, age today (2022), current site status and indication of success (1) or failure (0).

Location	Site Code	Latitude	Longitude	Lead Researcher	Depth	Species	Method	General Experimental Design	Set-up	Monitoring	Age Today	Current Site Status	Success/ Failure
Woodman Point	2m	S 32° 8' 10"	E 115° 44' 1"	Jennifer Verduin	2	P. australis	Sprigs	three 5x5m plots, 0.5m apart, 1 5cm sprigs with peg	May 2009	3 years	13 years	Sprigs have formed circular patches	1
Woodman Point	4m	S 32° 8' 15"	E 115° 44' 2"	Jennifer Verduin	4	P. australis	Sprigs	three 5x5m plots, 0.5m apart, 1 5cm sprigs with peg	May 2009	3 years	13 years	Sprigs have merged Plots have formed square meadows	1
Woodman Point	9m	S 32° 8' 18"	E 115° 44' 1"	Jennifer Verduin	9	P. australis	Sprigs	three 5x5m plots, 0.5m apart, 1 5cm sprigs with peg	May 2009	3 years	13 years	No evidence found Covered in Posidonia sinuosa	0
Southern Flats	1SW/2SE corner	S 32°15.142'	E 115°43.364'	Jennifer Verduin	2	P. australis P. sinuosa	Sprigs Plugs	three x 1 hectare plots in line, planted 0.5m apart, 8.3cm & 10cm diameter plugs	2004-2008	1.5 years	15.8 years	Large meadow present Significant infilling and expansion Shoot densities similar to natural meadows	1
Owen Anchorage	S4S - 1	-32.120625	115.7531556	Gary Kendrick	6	P. australis	Seeds	~85 seeds/m2, centre point with ~15m dispersal radius	December 2021	Ongoing	2 month	~5 seedlings per m2	1
Owen Anchorage	S4S - 2	-32.119025	115.7536889	Gary Kendrick	6	P. australis	Seeds	~85 seeds/m2, centre point with ~15m dispersal radius	December 2021	Ongoing	2 month	~17 seedlings per m2	1
Owen Anchorage	S4S - 3	-32.11358611	115.7487639	Gary Kendrick	6	P. australis	Seeds	~85 seeds/m2, centre point with ~15m dispersal radius	December 2021	Ongoing	2 month	~45 seedlings per m2	1
Cockburn Sound	S4S - 4	-32.174057	115.739325	Gary Kendrick	6	P. australis	Seeds	~85 seeds/m2, centre point with ~15m dispersal radius	December 2021	Ongoing	2 month	~33 seedlings per m2	1
Cockburn Sound	S4S - 5	-32.179095	115.742101	Gary Kendrick	6	P. australis	Seeds	~85 seeds/m2, centre point with ~15m dispersal radius	December 2021	Ongoing	2 month	~23 seedlings per m2	1
Owen Anchorage	S4S - com	-32.132857	115.74101	Gary Kendrick	2	P. australis	Seeds	~85 seeds/m2, centre point with ~15m dispersal radius	December 2021	Ongoing	2 month	~5 seedlings total	1
Owen Anchorage	BA 19	-32.10008799	115.74145	Gary Kendrick	7	P. australis	Seeds	200seeds/m2, 15x15m plot	December 2019,2020	Ongoing	1.5-2.5 years	~7 seedlings per m2	1
Owen Anchorage	BA18 SW corner	-32.10028203	115.74163	Gary Kendrick	7	P. australis	Seeds	200 seeds/m2, 15x15m plot, control plots 5x5m on SW, S & SE sides	December 2018	Ongoing	3.5 years	~42 seedlings 225m-2 ~1333 shoots 225m-2 Evidence of seeds drifting outside plot No seedlings in control plots	1
Owen Anchorage	2BA18 SE corner	-32.096121	115.744448	Gary Kendrick	7	P. australis	Seeds	200 seeds/m2, 20x20m plot, 3 control plots	December 2018,2020,2021	Ongoing	0.2-3.5 years	~1.64 seedlings m-2 in 2020 ~0.64 seedlings m-2 in 2019 ~0.6 individuals m-2 of 3+ yo recruits Sand waves (<15 cm) observed	1
Owen Anchorage	BA R 15	-32.10007299	115.742693	Gary Kendrick	7	P. australis	Seeds	200 seeds/m2, three 5x5m plots	December 2015	6.5	6.5 years	Large patches to 75% coverage 20-40 individuals over 25 m-2 277-1022 shoots 25m-2	1
Owen Anchorage	OA_R14	-32.11842223	115.7463461	Gary Kendrick	6	P. australis	Seeds	200 seeds/m2, three 5x5m plots	December 2014	7.5	7.5 years	Large patches to 80% coverage 29-113 individuals over 25 m-2 540-2065 shoots 25m-2 Infilling by P. sinuosa	1
Cockburn Sound	CS R 14	-32.19086694	115.7463298	Gary Kendrick	6	P. australis	Seeds	200 seeds/m2, three 5x5m plots	December 2014	7.5	7.5 years	Small patches to 15% coverage 12-15 individuals over 25 m-2 129-249 shoots 25m-2	1
Woodman Point	WP_R14	-32.129219	115.740656	Gary Kendrick	4.5	P. australis	Seeds	200 seeds/m2, three 5x5m plots	December 2014	7.5	7.5 years	Significant recovery and infilling 25-45% seagrass cover in plots 5-70% seagrass cover in controls	1
Woodman Point	scar 2	-32.129819	115.743134	Renae Hovey	5	P. australis P. sinuosa	Seeds Sprigs	Transplants into sand and bags with or without artificial leaves	March 2008	4 years	14 years	Recovery observed ~70% filled with seagrass Stakes still present No evidence of bags or artificial leaves	1

10.2 Appendix A2: Collation of outcomes reported from restoration programs discussed in the review. Details include: date; researchers, environmental drivers (biological), environmental drivers (physical), donor meadows, and outcome by each method.

Time	Researchers	Environmental Drivers (Biological)	Environmental Drivers (Physical)	Donor Meadows	Seeds	Seedlings	Sprigs	Plugs Cores Sods	Wire Pegs Hessian Bags Artificial Seagrass
1980s	Kirkman et al.	Disturbance by marine animals (crabs, rays).	Physical exposure of sites to shear stress that removed units			Seagrasses were hard to restore using seedlings	Seagrasses were hard to restore using shoot transplants		
1990s	Paling van Keulen et al.		Sediment dynamics important consideration, particularly in high energy areas					Larger plugs (10-15cm diameter) had greater survival rate	
			Survival of transplanted seagrass is reduced during winter months as sediment remains fluid for many weeks following transplantation					Transplanting plugs into existing meadows of minor species also improve survival rates	
			When transplanting seagrass you need to consider the appropriateness of the technique in regard to species, seasonality of weather and range of sediment level fluctuation					Larger transplants (sods) are required for areas of high wave energy and plugs and sprigs are appropriate in more moderate conditions	
2000s	Verduin van Keulen Paling	Epiphytes and algal growth may impact seagrass growth	variation in survival may be small scale hydrodynamic differences	Predicted recovery time for <i>P. australis</i> and <i>P. sinuosa</i> after 24 months is estimated at 3 years and 2.5 years, respectively. No difference in configuration.			Difference in survival across the 3 1 ha areas: eastern was 23.3%, western was 86-%		
	Verduin van Keulen		Depth therefore light a major limiting factor				Significant differences in shoot density of transplants with depth Survival high at 2m (>70%) but low at all other depths (<40%)		
	Verduin					Amphibolis seedlings			Coiled wire equals survival <30% after 2 years
	van Keulen	Urchin grazing preference for <i>P. sinuosa</i> , not <i>P. australis</i>					<i>P. australis</i> 4 -shoot spigs survived better (67%) than <i>P. sinuosa</i> (0%) sprigs probably as <i>P. sinuosa</i> at Buchanan Bay was grazed more heavily - yet at Luscombe Bay they both did poorly over 12 months. Clearly a site effect.	<i>P. australis</i> plugs (50%) and <i>P. sinuosa</i> plugs (62%) had high survivorship over 12 months at Luscombe Bay but they both did poorly (12%) at Buchanan Bay	
	Statton et al. 2013, 2014	Preferential grazing by predators requires careful consideration during the site selection process.	1. Sediment composition (sediment texture and organic matter content) had a greater influence on seedling establishment (i.e. initial anchorage success) than adding inorganic nutrients (N and P). 2. water depth is important for both light (70% Surface Irradiance) and water motion thresholds (growth and anchorage potential) and seed survival once transferred from tanks to the field..			Growing seedlings in tanks prior to transfer to the field reduces seedling mortality and improves establishment.			Transferring tank grown seedlings into sand-filled biodegradable hessian bags (Grow-bags) is crucial for seedling success as they provide a stable sedimentary environment for seedlings to be established.
	Cambridge Hovey Kendrick 2009, 2011, 2012		Adding nutrients can cause reduction in growth. It is important that limiting nutrient is determined for both <i>P. australis</i> and <i>P. sinuosa</i> .				Time of year (spring) for planting was important for <i>P. australis</i> but not <i>P. sinuosa</i> . High ambient nutrient levels may limit transplant growth		
	John Statton Hovey Cambridge						Survival of transplants was high with a year of planting but within two years was 30-50% of transplant densities. Surviving transplants doubled or trebled their shoot densities within 2 years		Results using hessian bags to protect shoots and sprigs were equivocal and site specific, suggesting strong local drivers to survival and growth. Results using artificial seagrass to protect shoots and sprigs were equivocal and site specific, suggesting strong local drivers to survival and growth.
2010	Snclair et al. 2013, 2014				From a population genetic basis, seeds are viable as a restoration technique across Cockburn Sound and Owen Anchorage.		Shoot-based restoration in Cockburn Sound should focus on sourcing transplants from populations that have high clonal richness, to maintain or increase genetic diversity in restored areas.		

	Statton et al. 2017	In sheltered waters (e.g. Cockburn Sound) herbivory by crustaceans and bioturbation affect the transition from seed settlement to early seedling recruitment. Their effect can result in complete loss of this transition from seed to early recruit.	In swell-influenced environments (e.g. Owen Anchorage, Carnac Island) the frequency and duration of winter storms result in high mortalities between early (3 mo) to 1 yo recruiting seedlings and can result in complete loss.	Many of the seeds produced are transported out of their bed of origin and numbers from Rottneest Island suggest that only 10 % stay within shallow seagrass habitats. Thus, collecting seeds for restoration has little or no impact to source meadows.	Seed settlement densities highly influence these transitions. Increasing seed settlement via seed-based restoration strongly influences survival from seeds to 1 yo recruiting seedlings.				
	Statton Kendrick Orth Dixon				In 2021 the volunteer group grew substantially and we collected ~1.18 million fruits and dispersed ~375 000 seeds at ~85 seeds per m ² into six new sites totalling ~2,100m ² . This involved design of rapid fruit collection methods, both boat and diver-based.	Seedling recruitment seems to be context dependent and greater in Owen Anchorage than Cockburn Sound. Seedling survival was greater in seeded plots than natural controls across all restoration sites, justifying the approach to seed rather than other approaches, like seedling nurseries, etc.			
	Statton Waite Kendrick Dixon				For <i>Halophila ovalis</i> , increasing temperatures in a stepped fashion from winter (15oC) to summer slowly (20-25oC) (Statton et al. 2017b) and changing spectral irradiance from blue dominated to red-yellow dominated (Waite et al. 2021) resulted in >35% germination of dormant seeds. Colder temperatures (<15oC) and either no light or blue light were effective in arresting germination and is the preferred environment for seed storage in <i>H. ovalis</i> .				
2020	UWA OzFish	In the seed to early seedling stage, disturbance of seeds before roots are established occurs from 0.5 m to 10 m in sheltered waters, where grazers and bioturbators are common.	In the early seedling to 12 mo seedling stage, the greatest threat to seedling survival is the frequency direction and duration of winter storm swells. In the seed to early seedling stage, disturbance of seeds before roots are established occurs in shallow waters (<3m) affected by the seas created by summer sea breezes.		The largest bottlenecks to recruitment remain at the seed to 3 mo seedling and the 3 mo seedling to 12 mo seedling stages.				

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