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Appendix A: Ephemeral seagrass meadow assessment to improve understanding of resilience of ephemeral seagrasses on Kwinana Shelf for Environmental Impact Assessment and Management

Theme: Benthic Habitats and Communities
WAMSI Westport Marine Science Program

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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FUNDING SOURCES

The $13.5 million WAMSI Westport Marine Science Program was funded by the Western Australian Government, Department of Transport. WAMSI partners provided significant in-kind funding to the program to increase the value to >$22 million.

DATA

Finalised datasets will be released as open data, and data and/or metadata will be discoverable through Data WA and the Shared Land Information Platform (SLIP).

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FRONT COVER IMAGE

Theme: Benthic habitats and communities
Front cover image: Seagrass (Posidonia australis) in Cockburn Sound. Photo courtesy of Rachel Austin (The University of Western Australia).

Contents

Tables

Figures

The WAMSI Westport Marine Science Program is a $13.5 million body of research that is designed to fill knowledge gaps relating to the Cockburn Sound region. It was developed with the objectives of improving the capacity to avoid, mitigate and offset environmental impacts of the proposed Westport container port development and increase the WA Government's ability to manage other pressures acting on Cockburn Sound into the future. Funding for the program has been provided by Westport (through the Department of Transport) and the science projects are being delivered by the Western Australian Marine Science Institution.

Appendix A: Ephemeral seagrass meadow assessment to improve understanding of resilience of ephemeral seagrasses on Kwinana Shelf for Environmental Impact Assessment and Management

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Project 2.2 Pressure-response relationships, building resilience and future proofing seagrass meadows

Executive Summary

- This study was conducted to generate knowledge to inform the environmental impact and prediction process for seagrass species that form ephemeral meadows in Cockburn Sound and Owen Anchorage.

- Local light thresholds for the opportunistic species, *Syringodium isoetifolium* were generated at summer and winter temperatures, with higher light requirements in summer compared to winter, and compared to larger, more slow-growing persistent species such as *Posidonia sinuosa*. The half-saturating irradiance (I_k) was 115 µmol m^{-2} s^{-1} at 23 °C.

- Based on the dynamics at three sites from December 2023 to April 2024, *Halophila ovalis* forms ephemeral meadows in Cockburn Sound and Owen Anchorage with meadows disappearing between February and April. Flowering was observed at all meadows in December but only one of the three meadows produced a viable seed bank. Therefore, the recovery potential of these meadows is likely to vary between sites and assessment of flowering or seed banks should occur to aid prediction of recovery potential and timescales.

- One colonising species that has been recorded in the past, *Zostera nigricaulis,* was rare within Cockburn Sound and Owen Anchorage, and there was not enough material available to conduct light threshold experiments or meadow dynamics, including recovery potential assessment. It was identified in another component of this WAMSI Westport Marine Project 2.2 Pressure-response relationships, building resilience and future proofing seagrass meadows, that *Z. nigricaulis* is currently growing in conditions in summer that are above its thermal optima and is of high concern with warming oceans due to climate change. The low abundance could be due to impacts from ocean warming that have already manifested.

1 Introduction

Colonising seagrasses, usually *Zostera* (*Heterozostera*) and *Halophila*, are observed on Kwinana Shelf and form ephemeral meadows (Hovey and Fraser, 2018). Due to this dynamic meadow form, the area where the seagrass fluctuates is often considered 'potential seagrass habitat' and based on the seagrass mapping work conducted in the WAMSI Westport Marine Science Program (WWMSP), this habitat is likely to be located adjacent to the Westport development footprint on Kwinana Shelf. Therefore, for the Environmental Impact Assessment (EIA) for the port development, there is a need to be confident of the biological thresholds for these species to dredging pressures and the potential for recovery. This knowledge gap was identified during the WWMSP and this body of work proposed as an appendix to the project *Pressure-response relationships, building resilience and future proofing seagrass meadows* to fill this gap.

There are some data on biological thresholds to dredging pressures for seagrass species that can form ephemeral meadows (*Halophila, Zostera, Syringodium*) in Cockburn Sound and Owen Anchorage, as well as temperature which is a key pressure being considered in the current WWMSP project (Table 1, Said et al., 2024a, Said et al., 2024b). Currently, there is some local data available for *Halophila ovalis* and *Zostera nigricaulis* (previously called *Heterozostera tasmanica*) for optimum temperature and maximum temperature, as well as light requirements for *H. ovalis* based on compensating and half-saturating irradiance (Table 1). Light and burial thresholds have been developed for all three species in other locations, as well as temperature thresholds, except for *Syringodium isoetifolium*. There is some recovery potential data for *H. ovalis* to light stress and for *Z. nigricaulis* to burial stress.

Table 1. Biological threshold information and current gaps for ephemeral seagrasses. Abbreviation legend: compensation irradiance (I_c), half-saturating irradiance (I_k), % surface irradiance (SI) and thermal optima (T_{opt}).

Species	**Locally derived light thresholds**	**Locally derived burial thresholds**	**Locally derived optimum temperature**	**Light thresholds developed in other locations**	**Burial thresholds developed in other locations**	**Temperature thresholds developed in other locations**
Halophila ovalis	I_c & I_k, but not mol m^{-2} d^{-1}	No data	Project 2.2 T_{opt} 32.1 °C	Yes as I_c & I_k & mol m^{-2} d^{-1}	Yes – tolerates 0.25 cm to 4 cm of burial depending on study	Yes , T_{opt} varies from 25-31 °C
Zostera nigricaulis (or *Heterozostera tasmanica*)	No data	No data	Project 2.2 T_{opt} 22.3 °C	Yes as % SI or instantaneous light (umol m^{-2} s^{-1}) but not mol m^{-2} d^{-1}	Yes – Victoria tolerates up to 2cm	Victoria, T_{opt} 30 °C
Syringodium isoetifolium	No data	No data	No data	Yes as I_k	Yes – tolerates up to 4 cm of burial	No data

As the mechanism for recovery for colonising seagrass species involves either clonal growth from remaining plants and/or from the recruitment of seeds stored in the sediment (i.e. seed banks) and the establishment of seedlings, the presence of a seed bank indicates the potential for recovery. Smaller, fast growing seagrass species are less tolerant to pressures, and therefore can be impacted faster and with lower levels of pressure (Kilminster et al., 2015). Thus, understanding the recovery potential of these species is important for impact prediction and management, especially in the context of the Western Australian Environmental Protection Authority's Technical Guidance: Environmental Impact Assessment of Marine Dredging Proposals (EPA, 2021), which provides a framework for Environmental Impact Assessment (EIA) of dredging. To be able to predict recovery potential for ephemeral seagrasses, the EPA Dredging Guidance document recommends that the presence of seed banks should be considered. There have been a few studies that have assessed sexual reproduction and seeds banks in Perth waters, however, data is lacking on the Kwinana Shelf, Cockburn Sound. Where data is available, and flowering was assessed over multiple years, flowering was not observed every year, indicating that sexual reproduction is unlikely to occur every year to generate a seed bank (Campey et al., 2002). However, when flowering and seed production was observed, for *Z. nigricaulis* seed production was recorded from Nov - Jan (Campey et al., 2002) and for *H. ovalis* from Feb – May (Webster et al., 2021). In some cases, seed banks have been detected, but they are of very low density.

The aim of this additional work is in relation to colonising seagrass species that grow in Cockburn Sound and Owen Anchorage, to, 1) fill gaps in the local light requirements based on thresholds for compensating and half-saturating irradiance (I_c and I_k), and (2) investigate meadow dynamics and recovery potential through estimates of flowering, seed bank density and viability of seeds. The outcomes will inform the environmental impact assessment and mitigation of ephemeral seagrasses in relation to dredging pressures.

2 Materials and Methods

2.1 Site selection

The location of colonising species that form ephemeral meadows in Cockburn Sound and Owen Anchorage was identified from the information collected by the WWMSP project: *Benthic Habitat Mapping* during field work. These sites were ground truthed to check if they were suitable for the purposes of this study. For Aim 1 the priority was to target *Z. nigricaulis,* however, at the time of sample collection this species was only found in 20 m of waters to the north of Owen Anchorage. This depth range was outside of both the approved dive depths and the Cockburn Sound and Owen Anchorage dredging footprint, so it was decided to refocus the local light thresholds to *S. isoetifolium* on the Kwinana Shelf, which also had no local threshold data developed. Samples consisting of whole plants for Aim 1 were collected from Kwinana Shelf in 8 m of water (-32.185, 115.750) in April 2024 (Figure 1). For Aim 2 as there were no suitable sites to target *Z. nigricaulis*, three *H. ovalis* meadows (Woodman A; Woodman B and Kwinana Shelf; details in Section 2.3.1) were selected to assess meadow dynamics (Figure 1). These three meadows were sampled over three time periods, stratified to the times when flowering and seed set were most likely, December 2023, February 2024 and April 2024.

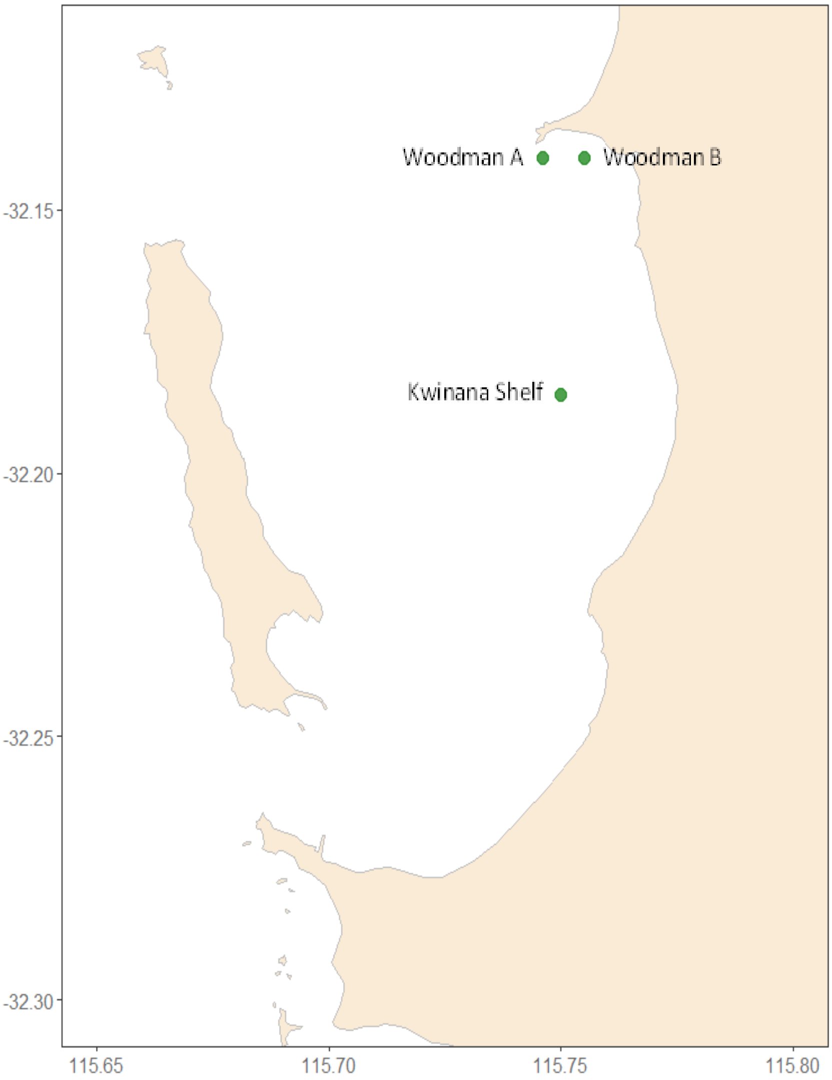

Figure 1. Location of sample collection points and study sites.

2.2 Development of light thresholds for *Syringodium isoetifolium*

2.2.1 Plant collection and acclimation

To assess the photosynthetic-irradiance (P-I) relationship of *S. isoetifolium*, the P-I relationship was determined for two temperatures (16 °C and 23 °C) to mimic Cockburn Sound ambient summer and winter water conditions. Whole plants were collected by gently fanning sediment away to expose the rhizome, extracting whole plants with a minimum of four shoots. Plants were placed in an aerated cooler box filled with seawater for transportation to the laboratory. At time of plant collection ambient water temperature was 20 °C and salinity was 35.1 ppt. In the laboratory, experimental plants were gently cleaned to remove epiphytes and were planted as per field conditions into 50 L aquaria containing a 7 cm layer of washed, quartz river sand. A further 50 L sump tank underneath the aquaria contained a pump to circulate water from the sump to the tank with seagrass for aeration. Water temperature was controlled using aquarium heaters and chiller units. Plants were provided with 180 µmol photons m^{-2} s^{-1} (approximating saturating irradiance; Pollard, 1999) using marine aquarium Light Emitting Diode (LED) modules with a full spectrum light (MarinTech™ Pty Ltd) on a 12/12 light:dark cycle. Light intensity was measured at the base of the canopy using a micro-PAR sensor (In-Situ Marine Optics™). Plants were left to acclimate for two days before increasing or decreasing the temperature by 1 °C per day until the experimental temperatures (16 °C and 23 °C) were reached. Plants were then left for a further 24 hours at the experimental temperature, before taking dark adapted maximum quantum yields on five separate plants to assess their acclimation. Plants were considered healthy and acclimated if they had a dark-adapted yield of 0.73–0.75 (Ralph and Burchett, 1995).

2.2.2 Photosynthetic-irradiance determinations

Seagrass respiration and photosynthesis were measured via the consumption or production of O_2, following the methods in Said et al. (2021). This was repeated for each temperature treatment. Plants were incubated in sealed acrylic chambers (50 cm internal diameter, 20 cm length, 440 ml volume of cylinder and ~110 ml additional water in tubing). Water within the chambers was circulated using small submersible pumps with a flow rate of 7000 ml h-1. Dissolved oxygen concentrations within the chambers were measured using FireSting™ 3 mm robust REDFLASH technology sensors (Pyroscience) inserted through the chamber wall and connected through a 4-channel meter to a computer recording mg of O_2. To maintain a stable temperature (±0.25 °C), chambers were submerged in a 300 L tank containing 150 L of seawater, which was circulated through a chiller-heater unit set to the appropriate experimental temperature. The internal temperature of the chamber was also measured using a submersible temperature sensor connected to the FireSting O_2 meter. Light was provided by full spectrum LED light units (GrowPro 320; MarinTechTM Pty Ltd) suspended above the chambers, providing intensities from 30 to 300 µmol photons m^{-2} s^{-1}. Prior to incubation, the oxygen electrodes were calibrated using the manufacturer's 2-point method (0% and 100% air saturated water). Four replicate plant chambers, each containing four *S. isoetifolium* ramets, and a fifth 'blank' chamber (containing no plant material) were established and placed into the temperature-controlled tank. The chambers were then covered to exclude light, and the inlet of each chamber was connected to its individual pump, to allow the chamber to be flushed whilst the plant material was left to dark adapt for 30 minutes. Once dark adapted, the chamber outlet was connected to the pump to create a sealed system and the dissolved oxygen concentration monitored every second. Monitoring continued in the dark for at 10 minutes after the slope of dissolved oxygen vs time stabilised. Once the dark step was complete, shading was removed and photosynthetic rates were measured for 10 minutes (once the slope had stabilised) at each of 7 light intensities (30, 60, 90, 120, 180, 230, and 300 µmol photons m^{-2} s^{-1}). At the end of the experiment, the plants were removed from the chambers, separated into above-ground (leaves) and below-ground (roots and rhizomes) tissue, dried at 60 °C for 48 hours and weighed to determine biomass dry weight (g).

2.2.3 Photosynthetic-irradiance curve fitting

Standard curve-fitting methods described in Said et al., (2021) were followed. Briefly, for each unique temperature-light intensity combination, oxygen concentration for each replicate incubation was plotted over time, following a 2 min stabilisation period. Rates of oxygen exchange were calculated and normalised to g DW^{-1} hr^{-1} of whole plant material. Oxygen concentrations within the control chamber that contained no seagrass were measured throughout the experiment and experimental units were only used if there was no significant change in control chamber oxygen concentrations. Photosynthetic parameters were extracted from the P-I curves (Figure 2) using the least-squares method. For each incubation, P-I curves were fitted to the data using the hyperbolic tangent model equation of Chalker (1981):

$$P = GP_{max} \text{ X } \tanh\left(\alpha \times I / GP_{max} \right) + R,$$

where P is the rate of photosynthesis (mg O_2 . g DW^{-1} . hr^{-1}), GP_{max} is the maximum gross photosynthesis (mg O_2 g . DW^{-1} . hr^{-1}), α ((mg O_2 . g DW^{-1} . hr^{-1})/(µmol m^{-2} s^{-1})) is the photosynthetic efficiency estimated as the slope for the linear portion (light-limited portion) of the P-I curve, I is irradiance (µmol photons m^{-2} s^{-1}), and R (mg O_2 . g DW^{-1} . hr^{-1}) is the rate of oxygen consumption in the dark.

NP_{max}, the maximum net rate of photosynthesis (mg O_2 g DW^{-1} hr^{-1}) was calculated as:

$NP_{max} = GP_{max} + R$

The saturating irradiance (I_k; µmol photons m^{-2} s^{-1}) was calculated as:

$I_k = GP_{max}/ \alpha$

The compensation irradiance (I_c; µmol photons m^{-2} s^{-1}) was calculated via a mathematical approximation that assumes I_c is much smaller than I_k:

$I_c = (0 – R) / \alpha$

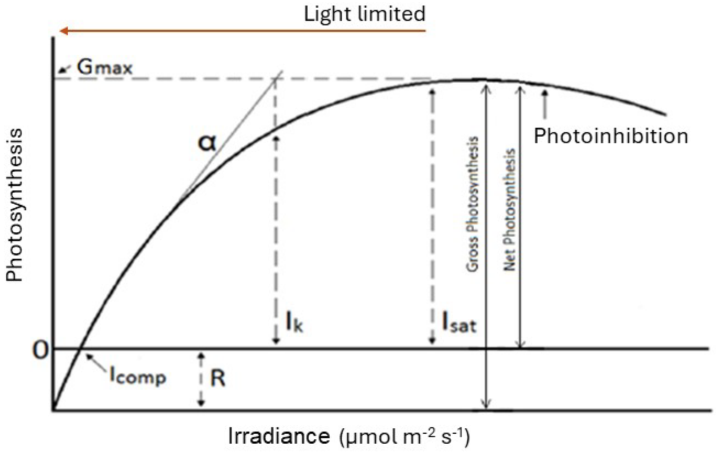

Figure 2. Photosynthesis vs. irradiance (P-I) curve and associated photosynthetic parameters. Figure adapted from Beer et al. (2014). In the absence of light, the rate of O_2 gas exchange is taken as a measure of a plant's respiration rate (R). P-I curves generally show a linear increase in photosynthesis as light intensity increases, with the initial slope (α) a measure of the plant's photosynthetic efficiency. The light compensation point (I_c) refers to the point where photosynthesis and respiration equals zero, while the half saturation point (I_k) indicates the start of light saturation of photosynthesis. GP_{max} and NP_{max} indicate the maximum gross and net primary production rates, respectively.

2.2.4 Statistical analysis

To test for significant effects of temperature on *S. isoetifolium* plants for each P–I parameter, a one-way PERMANOVA were performed, based on Euclidian distances with 9999 permutations using the PRIMER™ software (Anderson et al., 2008). Temperature was treated as a fixed effect. PERMDISP was used to test for homogeneity of variance (p= 0.05). The Sums of Squares was set to Type III (Partial) SS. Where the PERMANOVA main test indicated a significant main effect or interaction ($p < 0.05$), pair-wise comparisons were performed to understand the nature of the effect or interaction. All data had unique permutations less than 100, therefore, a Monte Carlo (MC) simulation was run, and data interpreted in accordance with the MC P-value (Anderson et al., 2008).

2.3 Ephemeral meadow dynamics and recovery potential

2.3.1 Seagrass collection and processing

To assess if biomass, flowering and seed banks for the seagrass species *H. ovalis* varied spatially and temporally within Cockburn Sound, Western Australia, three sites were assessed (Figure 1); Kwinana Shelf (-32.185, 115.750; 8 m water depth), Woodman A (-32.140, 115.746; 12 m water depth) and Woodman B (-32.140, 115.755; 12 m water depth) at 3 time points (Dec-23, Feb-24, Apr-24). The timing of sampling was selected to determine seedbank densities after seed release when the viability of seeds would likely be greatest and to test peak densities of germinable seeds. At each site and time point, ten cores (diameter: 9.5 cm; area: 71 cm^2; max depth: 10 cm where possible) were haphazardly collected within *H. ovalis* dominant meadows. In April 2024, no cores were collected at the Kwinana Shelf site as the above-ground portion of the seagrass meadow was no longer present. Cores were positioned over an area of *H. ovalis* ensuring that all plant material that was rooted inside the circle of the core was inside the core and material rooted outside the core remained outside. The seagrass was placed inside a calico bag along with the sediment sample. As the survey sites consisted of mixed-species meadows, sub-dominant seagrass species may have also been present within the core. Seagrass cores (including sediment and biomass) were refrigerated at 4 °C and processed immediately, except for samples collected in December 2023 which were frozen at -20 °C for processing at a later date. In the laboratory, all plant material was thawed and rinsed in fresh water. The abundance of flowers, fruits, seeds in sediment and or seeds in fruit were recorded. For seed assessment, the sediment sample was sieved in the laboratory into >1 mm and >500 µm fractions and both fractions visually assessed for *H. ovalis* seeds. For the time points when the samples were not frozen, the viability was tested in accordance with Marion and Orth (2010) via the squeeze test. The seagrass material from each core was separated into above ground biomass consisting of leaves, stems and sheaths, and below ground biomass consisting of roots and rhizomes. Each sample was then dried at 60 °C for at least 48 hours, and weighed (g) to four decimal places. The reported biomass only included the target species (*H. ovalis*).

3 Results

3.1 Light thresholds for *S. isoetifolium*

At both summer and winter temperatures, *Syringodium isoetifolium* exhibited the typical relationship between photosynthesis and light, without any photoinhibition (Figure 3). The P–I relationships varied across the two temperatures (16 °C and 23 °C) tested.

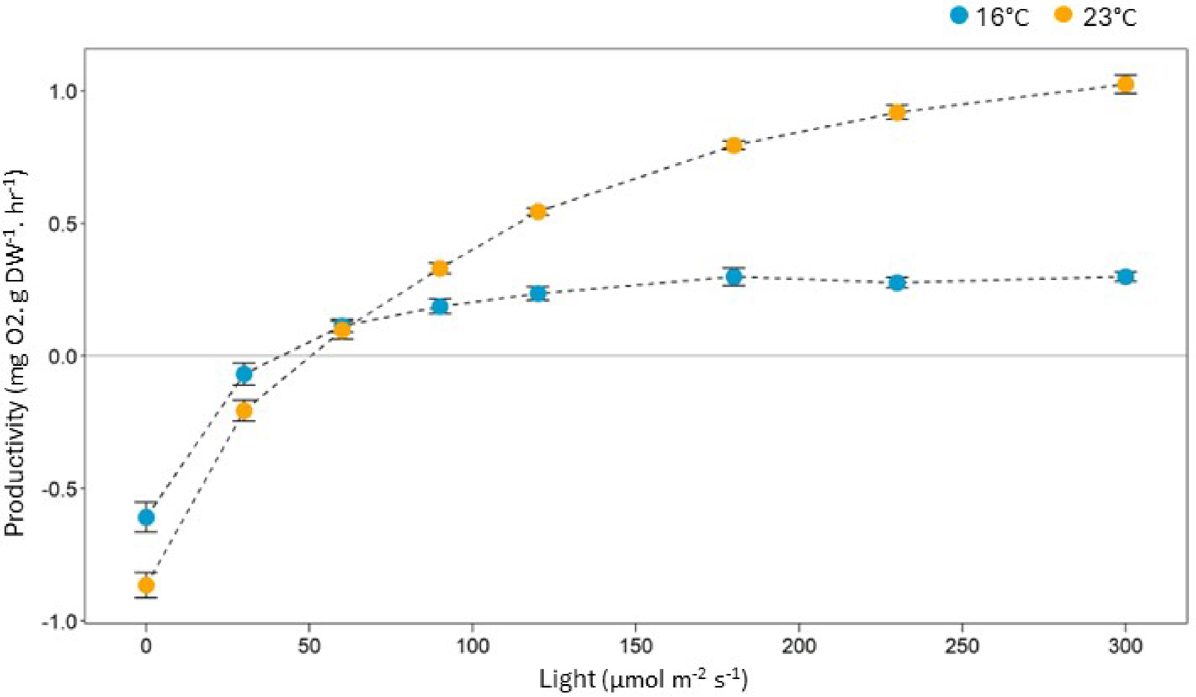

Figure 3. Photosynthetic-irradiance curves at two temperatures (16°C and 23°C) for *Syringodium isoetifolium* in Cockburn Sound. Means and SE, n = 4.

All photosynthetic parameters (except for respiration) significantly varied across the two temperatures tested for *S. isoetifolium* (p= 0.05; Figure 4). The maximum gross photosynthesis rate (GP_{max}) and the maximum net photosynthesis rate (NP_{max}) was lower at 16 °C (0.48 and 0.1 mg O_2 . g DW^{-1} . hr^{-1}, respectively) compared to 23 °C (1.0 and 0.54 mg O_2 . g DW^{-1} . hr^{-1}, respectively). The half saturation irradiance (I_k) and the compensation irradiance (I_c) were also lower at 16 °C, 47 µmol m^{-2} s^{-1} and 33 µmol m^{-2} s^{-1}, respectively, compared to at 23 °C, 115 µmol m^{-2} s^{-1} and 51 µmol m^{-2} s^{-1}, respectively. Conversely, alpha (α) was higher at 16 °C (0.011 mg O_2 g DW^{-1} hr^{-1}/µmol m^{-2} s^{-1}), compared to 23 °C (0.008 mg O_2 . g DW^{-1} . hr^{-1}/µmol m^{-2} s^{-1}). Respiration (R) was not significantly different, but had a trend for being lower at 16 °C (0.33 mg O_2 . g DW^{-1} . hr^{-1}), compared to 23 °C (0.43 mg O_2 . g DW^{-1} . hr^{-1}).

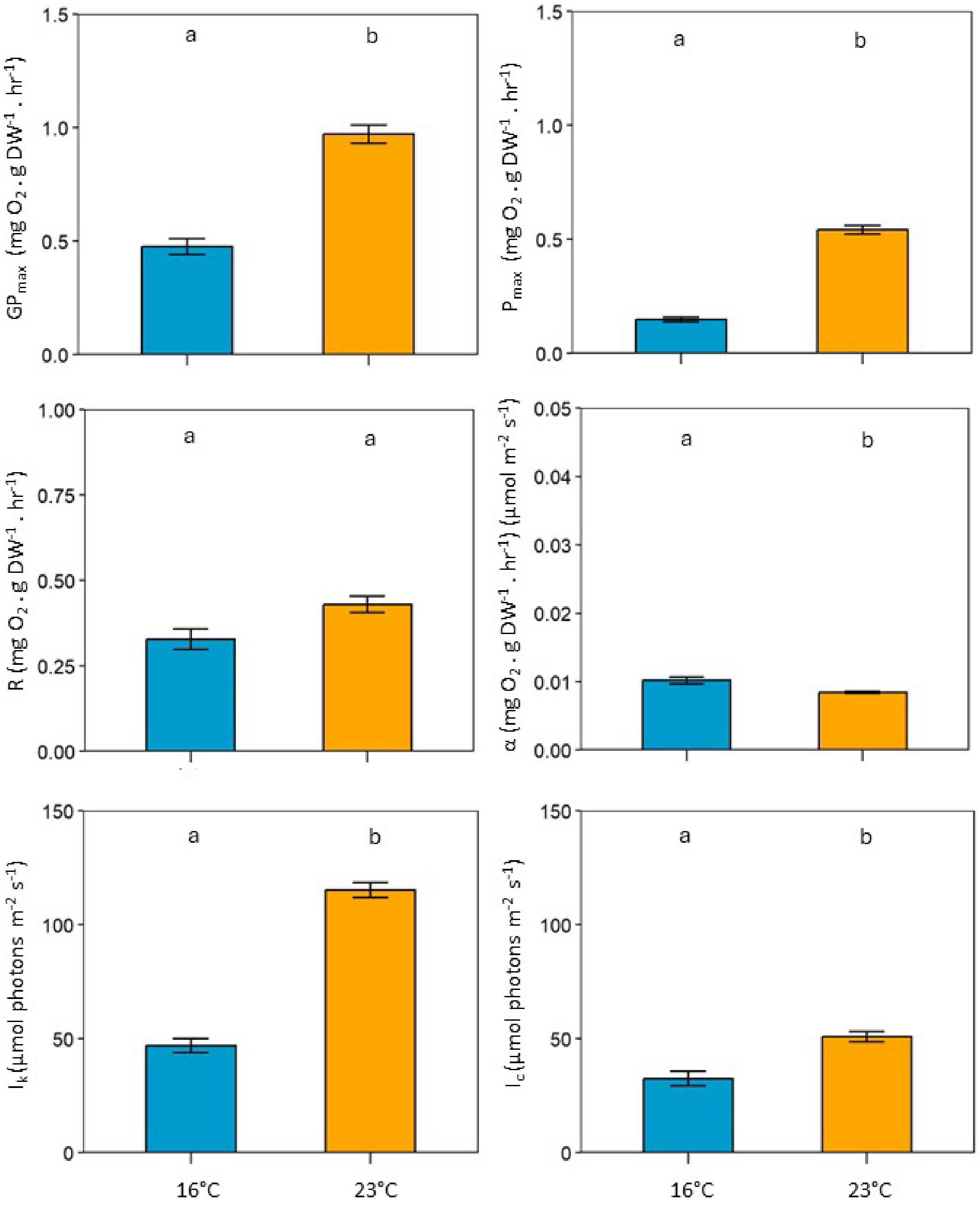

Figure 4. Effect of temperature (16 °C and 23 °C) on photosynthetic parameters (GP_{max}, NP_{max}, R, α, I_k, and I_c) of *Syringodium isoetifolium* in Cockburn Sound. Letters on graphs represent pairwise PERMANOVA tests. Means and SE, n = 4.

3.2 Ephemeral meadow dynamics and recovery potential

Halophila ovalis meadows were present at two (Woodman A&B) of the three sites over all sampling periods. *Halophila ovalis* were absent from the Kwinana site from February to April, indicating the ephemeral nature of these meadows. At the other two sites, total biomass was highest in February and declined in April (Figure 5).

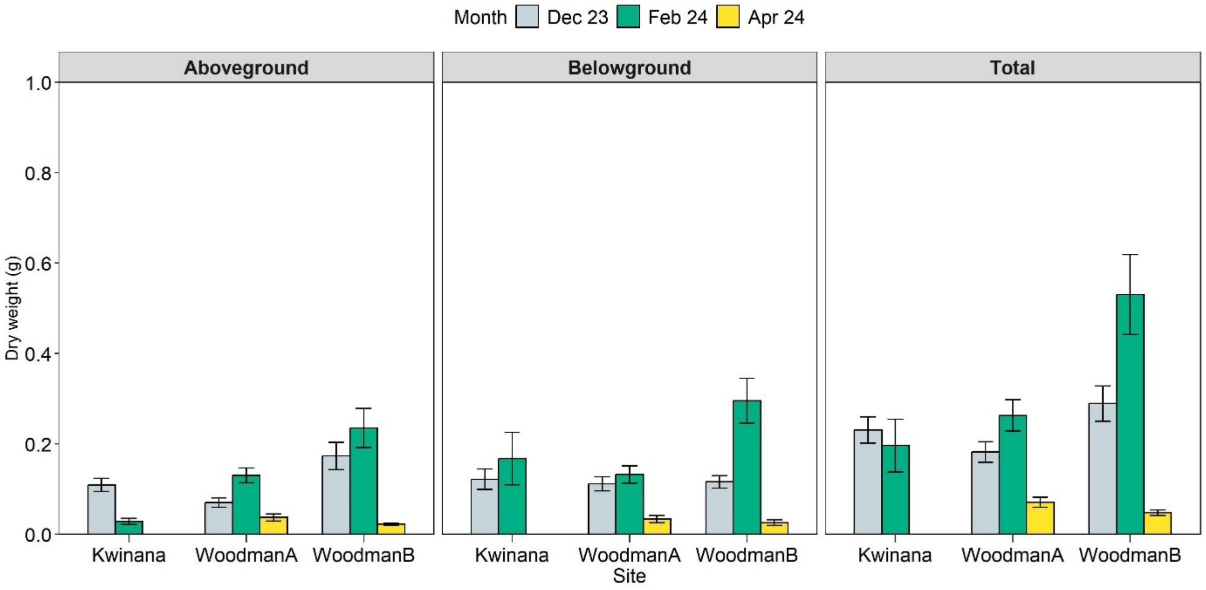

Figure 5. Dynamics of *Halophila ovalis* biomass over time (December 2023 to April 2024) at three sites in Cockburn Sound and Owen Anchorage. Biomass is g DW core^{-1} and based on 10 replicates per site.

Flowering of *H. ovalis* was observed at all sites in December 2023 and only at Woodman B in February 2024 but by April 2024, no flowers or fruits were detected (Table 2). The reproductive biology of *H. ovalis* is such that male and female flowers are on separate plants, and once pollinated the female flowers develop into fruits, therefore, both male and female flowers are required to set seed. Male and female flowers were only observed at Woodman B, and in December there was already an immature fruit developing at the Kwinana site, indicating that previously male flowers were present, and pollination had occurred. Flowering was most extensive at Woodman B with 50% of the cores containing flowers and ranging from 0.2-1.7 flowers on average per core, or up to a maximum of 11 male flowers per core and three female flowers or fruits; *H. ovalis* male flowers were more abundant than female flowers.

A seed bank was only observed at Woodman B in February and April 2024 where the greatest flowering of both male and female flowers was observed (Table 3). Generally, the viability of these seeds was high (90-100%), with the exception of one sample where the seeds were in the fruit and only 9% were viable. The Woodman B site had the greatest density of seeds (as well as being viable), and the greatest number of cores with seeds were observed in February, with declines into April.

Table 2. Density of *Halophila ovalis* flowers at three sites and three times from December 2023 to April 2024. Mean (stdev) from n = 10 cores per time/site.

Time	Male flowers			Female flowers & fruits			% cores that contained flowers		
	Kwinana	Woodman A	Woodman B	Kwinana	Woodman A	Woodman B	Kwinana	Woodman A	Woodman B
Dec 2023	O	0.4(1.0)	1.7(3.5)	0.1(0.3)	0	0.2(0.6)	10%	20%	50%
Feb 2024	0	0	0	0	0	0.3(0.9)	0%	0%	10%
April 2024	NA	0	0	NA	0	0	NA	0%	0%

Table 3. Density of *Halophila ovalis* seeds and seed viability at three sites and three times from December 2023 to April 2024. Mean (stdev) from n = 10 cores per time/site.

Time	Seeds			Viability (%)			% cores that contained viable seeds		
	Kwinana	Woodman A	Woodman B	Kwinana	Woodman A	Woodman B	Kwinana	Woodman A	Woodman B
Dec 2023	0	0	0	NA	NA	NA	0%	0%	0%
Feb 2024	0	0	3.6(6.0)	NA	NA	9-100%	0%	0%	30%
April 2024	NA	0	0.2(0.6)	NA	NA	100%	NA	0%	10%

4 Discussion

This additional scope of work has achieved its aim of providing light threshold data for species of seagrass that form ephemeral meadows that are potentially in the Westport development footprint, and therefore, provides insights into tolerances to dredging pressures and recovery potential of ephemeral meadows in Cockburn Sound. This will help to inform the EIA of the proposed Westport development.

The summer light thresholds estimated for *S. isoetifolium* in this study, I_k of 115 µmol m^{-2} s^{-1} at 23 °C were slightly lower than what is reported in the literature of I_k of 180 µmol m^{-2} s^{-1} from tropical Fiji. This is not surprising, as this study and others have demonstrated higher light requirements with warmer waters, and Fiji is in a tropical setting (Said et al., 2021, Said et al., 2024a). In addition, the winter I_k of 47 µmol m^{-2} s^{-1} at 16 °C supports the lower light requirements in winter. This aligns with the global analysis of environmental windows for dredging, that suggests seagrass meadows are more resilient to dredging pressures during winter when temperatures are lower (Wu et al., 2017). These results also support previous studies where colonising and opportunistic species tend to have higher light requirements than persistent species such as *Posidonia*, I_k of 55 µmol m^{-2} s^{-1} for *P. sinuosa* in Cockburn Sound at 23 °C (Said et al., 2024a; Masini and Manning, 1997). Therefore, where EIA has a need to manage impact to colonising or opportunistic species, such as *H. ovalis* or *S. isoetifolium,* more conservative light thresholds should be considered as these species are less tolerant to light reduction.

Initially, this project planned to focus on the dynamics of two colonising species, *Z. nigricaulis* and *H. ovalis,* but *Z. nigricaulis* was excluded due to its location in deep waters (20 m) north of Owen Anchorage, and outside of the proposed dredging footprint. The absence of *Z. nigricaulis* on the Kwinana Shelf could be due to inter-annual variability that has been observed previously (Campey et al., 2002); however, Said et al., (2024b) predicted *Z. nigricaulis* was at risk with ocean warming, as its thermal optima (22 °C) is already below summer ambient temperatures in Perth (23 °C). Therefore, there is the potential that this species has already become restricted in its distribution due to ocean warming. Further assessment of the distribution over time and comparison with historical records will help to explore this hypothesis.

For the colonising species (*H. ovalis*) where dynamics and recovery potential could be assessed, there was clear evidence to suggest that *H. ovalis* in Cockburn Sound forms ephemeral meadows. For example, above and below ground biomass at one out of the three sites assessed (Kwinana) disappeared from February to April, and meadows at the remaining two declined in biomass over this time. A seed bank was detected at only one of the two sites (Woodman B), indicating that the recovery potential is variable amongst locations, and recovery from seed banks is not guaranteed. Higher abundance of flowering and the presence of male and female plants resulted in the presence of a seed bank. Therefore, if it is of importance to understand the recovery potential of a site with colonising or opportunistic species that have the potential to form ephemeral meadows, either the presence of male and female flowers, and/or the presence of a seed bank should be assessed. Ideally, this should be monitored around January to March to detect flowering or a seed bank. Understanding the link between environmental factors that induce flowering (and female/male ratios on adult plants) in seagrasses is a relatively understudied topic. Some work demonstrates that changes in light quality and temperature induce flowering in some species, although understanding these dynamics in a field setting is difficult, and should be a topic of future research.

From the perspective of managing these ephemeral meadows with dredging pressures, the timing of the pressure is an important consideration and the concept of environmental windows could be incorporated (e.g. Fraser et al., 2017). Pressures could be timed to occur in autumn and winter when the meadows naturally declining and risk from light reduction is of lower concern. In contract, the management of pressures should be considered during the summer, which is a sensitive time when the plants flower and set seed to enable the next seasons growth. Wu et al. (2017) identified that timing of pressures was very important for annual meadows of the seagrass *H. ovalis* in tropical settings. Future environmental impact assessment should assess each proposed dredging footprint on a case-by-case basis, including site assessments at multiple depths and times of year in order to capture the meadow variability within an area of interest.

Overall, this work has led to a greater understanding of the dynamics of opportunistic species on Kwinana Shelf and the development of local light thresholds at summer and winter water temperatures for a new species of seagrass, *S. isoetifolium* that occurs within the Cockburn Sound region. Additional research that could complement these results would be assessing the effects of multiple stressors (e.g. low light and high temperature) in a mesocosm setting (particularly for *Z. nigricaulis*), and field setting (i.e. particularly for *H. ovalis*), as well as incorporate additional species relevant for the area (e.g. locally-derived burial and thermal thresholds for *S. isoetifolium*).

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6 Appendices

Table A1. PERMANOVA main test results for *Syringodium isoetifolium* photosynthetic parameters at 16 °C and 23 °C. Significant P values (P <0.05) are noted in bold.

Source	df	SS	p-value	Unique perms	Monte carlo	df	SS	p-value	Unique perms	Monte carlo
	GP_{max}					**P_{max}**				
Temperature	1	1.6157	0.0289	35	**0.0004**	1	0.99552	0.0312	15	**0.0001**
Residual	6	0.15133				6	0.050189			
Total	7	1.7671				7	1.0457			
	R					**α**				
Temperature	1	0.067609	0.0565	35	0.0622	1	2.0637E-05	0.0569	35	**0.0327**
Residual	6	0.078224				6	1.6082E-05			
Total	7	0.14583				7	3.6719E-05			
	I_k					**I_c**				
Temperature	1	9088.6	0.0275	35	**0.0002**	1	683.94	0.0295	35	**0.0058**
Residual	6	287.65				6	242.28			
Total	7	9376.3				7	926.22			

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