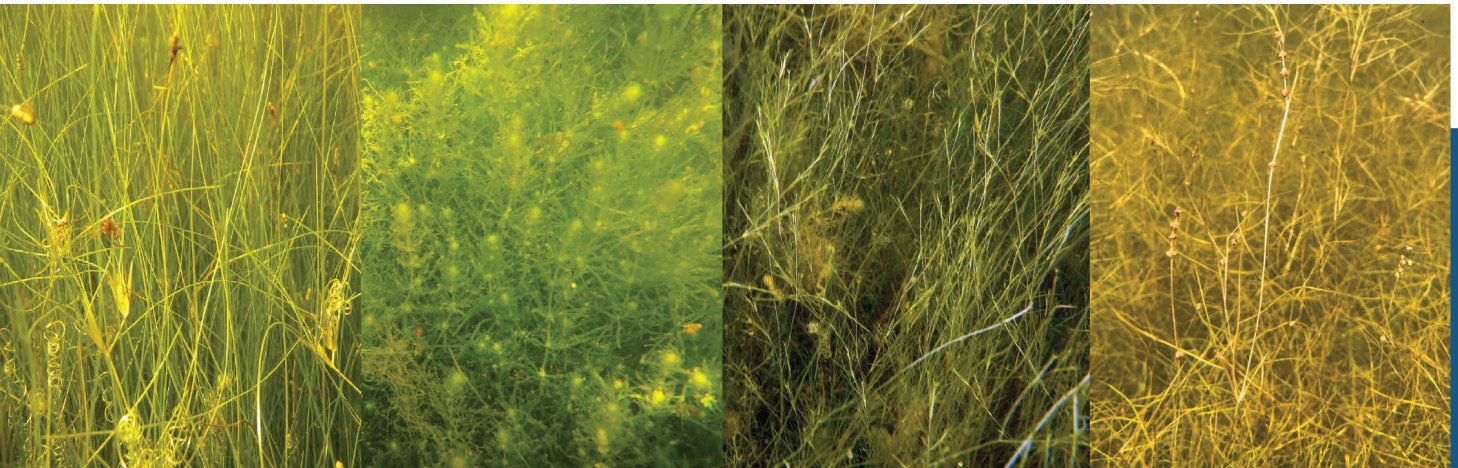# Macrophytes and Macroalgae in the Vasse-Wonnerup Wetland System 2017-2021:

Distribution, seasonal and annual variation, and relationship to water levels and water quality

R.L. Paice & J.M. Chambers
Murdoch University
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Revitalising Geographe Waterways

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Acknowledgement

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Summary

The Vasse-Wonnerup Wetland System is of international significance as a bird habitat but is threatened by poor water quality owing to alteration of hydrology and increased nutrient loads. Submerged aquatic plant communities support the bird populations by providing food resources, habitat and nesting materials. The plants also have critical interactions with water and sediment quality, buffering against degradation.

Aquatic plant communities, including submerged macrophytes (seagrasses), charophytes and macroalgae, were sampled on a seasonal basis from March 2017 to March 2021. Sampling encompassed four ecological regions in the system, defined *a priori*: the upper and lower Vasse Estuary; and the upper and lower Wonnerup Estuary. This work was undertaken as part of the Revitalising Geographe Waterways' Integrated Ecological Monitoring Study (IEM) that aims to better understand the relationships between water regime, macrophytes and the abundance of benthic macroinvertebrates, fish and birds utilising the range of habitats (regions) present in the Vasse-Wonnerup[1].

The objectives of the macrophyte component of the IEM were to improve understanding of spatial distribution and seasonal growth patterns of aquatic plant community assemblages, and identify important changes over the four years of monitoring. This report aims to:

- present and analyse the results of aquatic plant monitoring for the IEM Program over four complete years, from winter 2017 to autumn 2021;
- examine the relationships between plants, water levels and water quality, with consideration of managing seawater inflows at the Vasse Estuary surge barrier;
- provide recommendations for future monitoring.

Seasonal plant growth

Seasonal sampling has been valuable in understanding the growth cycle of aquatic plants, demonstrating the strong influence of environmental factors driven by annual climate patterns. The growth season commences with the onset of winter rains, rapidly reaching peak density in spring, followed by recession in summer as water levels drop. Within each region, community composition is dependent on the range of climate-related factors experienced as well as water quality and sediment characteristics. Established plant communities in spring provide the most informative data for understanding plant communities and identifying change over time.

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[1] https://rgw.dwer.wa.gov.au/applying-science/vasse-wonnerup-science/

Regional distribution

Although the system experiences inter-annual variation in aquatic plant growth within regions, plant communities do show high site affinity. Distinct plant communities occur in the Vasse Estuary regions, characterised by *Ruppia polycarpa* and *Althenia cylindrocarpa* (both species adapted to seasonal drying) in the upper Vasse; and *Ulva, Stuckenia pectinata* and *Ruppia* spp. in the Lower Vasse. The upper Wonnerup estuary is characterised by *Lamprothamnium*, but this extends into the lower Wonnerup sites so that the two regions overlap in terms of plant assemblages. The Lower Wonnerup Estuary is morphologically diverse and characterised by *Ruppia megacarpa* and *Stuckenia pectinata* in deeper channel habitats, while Lamprothamnium extends from the upper region into shallow habitats.

Change over time

Inter-annual variation in aquatic plant community assemblages were observed throughout the system, however there is no indication of long-term change in the Wonnerup Estuary or the lower Vasse Estuary.

A key finding of the aquatic plant component of the IEM program was the higher and widespread growth of *Ulva* which has developed in the upper Vasse Estuary since 2018. Future monitoring is needed to keep watch on this situation. Additional seawater inflow during summer and autumn has increased autumn water levels and reduced the extent of drying in this region, which may be contributing to this change.

Excessive growth of macroalgae has the potential to cause future loss of macrophytes (*Ruppia polycarpa* and *Althenia cylindrocarpa*) from the upper Vasse Estuary, which has historically been the highest quality seagrass assemblage in the Vasse-Wonnerup system. While the upper Vasse is still environmentally distinct, the plant community is now more similar to the lower Vasse Estuary, which has historically exhibited poor health.

Future manipulation of seawater exchange at the surge barrier will need to consider the long-term risks to the aquatic plant community and ecological condition in this region, including potential undesirable macroalgal growth.

Relationship to environmental variables

Investigation of relationships between plant communities and environmental variables using spring data found salinity, depth, nitrogen and sediment organic content to be important determinants of community assemblage. Key species within regions were associated with environmental factors as follows:

- *Ruppia polycarpa* in the upper Vasse was associated with lower TN, lower sediment organic content and shallower waters.

- *Ulva* and *Stuckenia pectinata* in lower Vasse were associated with lower TN and salinity, and higher sediment organic content.
- *Lamprothamnium* in the Wonnerup Estuary was associated with higher salinity and higher TN.

As mentioned above, the outcomes of this monitoring program indicate an increase in growth of macroalgae in the upper Vasse Estuary. Higher autumn water levels arising from increased seawater inflow has translated to reduced drying of the upper region. This may contribute to increased macroalgal growth by limiting consolidation of sediments, creating flocculent, organic sediments and deeper conditions more favorable to macroalgal growth.

Management implications

Manipulation of the surge barrier is a key tool in management of water quality in the Vasse Estuary, however if this continues to prevent drying of the upper region it may exacerbate macroalgal growth and negatively impact the seagrass meadows there. Therefore continued monitoring of both plant communities and environmental factors is critical to inform decision-making in this complex system. Additional research to understand mechanisms contributing to observed changes in the plant community in this region should also be considered. Some extent of seasonal drying in the upper Vasse may be necessary to maintain a healthy ecosystem.

Introduction

The Vasse-Wonnerup wetlands are an extensive, shallow, nutrient enriched system with wide ranging salinities. The wetlands support tens of thousands of resident and migrant waterbirds of a wide variety of species and the largest regular breeding colony of Black Swan in south-western Australia, and as such are listed as under the Ramsar Convention as having international significance (Lane et al. 2007). The system has been modified by installation of surge barriers that restrict tidal intrusion, altered catchment hydrology and increased nutrient loads.

The Vasse-Wonnerup Wetlands are highly valued by the community as a conservation estate and there is understandable concern about the severe nutrient problems the wetlands have had for many years including sudden mass fish deaths, blooms of macroalgae, toxic phytoplankton, nuisance odour and mosquito problems (Department of Water, 2010). Despite the nutrient problems, the Vasse-Wonnerup wetlands remain an important habitat for waterbirds, however the health of the system and risk to its value as waterbird habitat are an ongoing management concern.

Aquatic plant communities in the Vasse-Wonnerup Wetland System are a critical element of the ecosystem, reflecting and influencing water quality and supporting aquatic invertebrates, fish, and birds. Herbivorous and omnivorous waterbirds such as swans and ducks consume aquatic flora directly; and by supporting higher diversity and abundance of aquatic invertebrates (Heck and Crowder 1991, Paice et al. 2016), which provide additional food resources.

Nutrient enrichment generally leads to the decline of submerged macrophytes, and a corresponding loss of ecological functions of habitat and food provision and maintenance of water quality and clarity. In coastal lagoons such as the Vasse-Wonnerup wetlands, where seasonal macroalgae that thrive in nitrogen-rich environments such as *Ulva* spp. occur, a transition to a phytoplankton-dominated regime often occurs via a pathway of macroalgal blooms (Viaroli et al. 2008, Pasqualini et al. 2017).

The presence of stable, submerged macrophyte communities is indicative of a healthy ecosystem, while the transition to macroalgal blooms and subsequently phytoplankton dominance reflects an undesirable shift in ecological regime. Given the importance of aquatic plants in supporting the waterbird values for which the wetlands are Ramsar-listed, and their maintenance of and response to water quality, conservation of submerged macrophytes is vital to maintain the ecological health of the Vasse Wonnerup Wetland System.

A seasonal integrated ecological monitoring (IEM) program developed by the Department of Water and Environmental Regulation (DWER) commenced in March 2017. The program includes concurrent sampling of water quality, aquatic plants, fish, macroinvertebrates, and birds on four occasions throughout the year. The aim of this

monitoring program is to better understand the relationship between seasonal water regime (water quality and water levels) and the ecological values that support waterbirds on the Vasse Wonnerup wetlands.

This report presents the results of aquatic plant monitoring for the IEM Program over four complete years, from winter 2017 to autumn 2021, provides a summary of water quality and water level data, and examines the relationships between plants and these environmental variables.
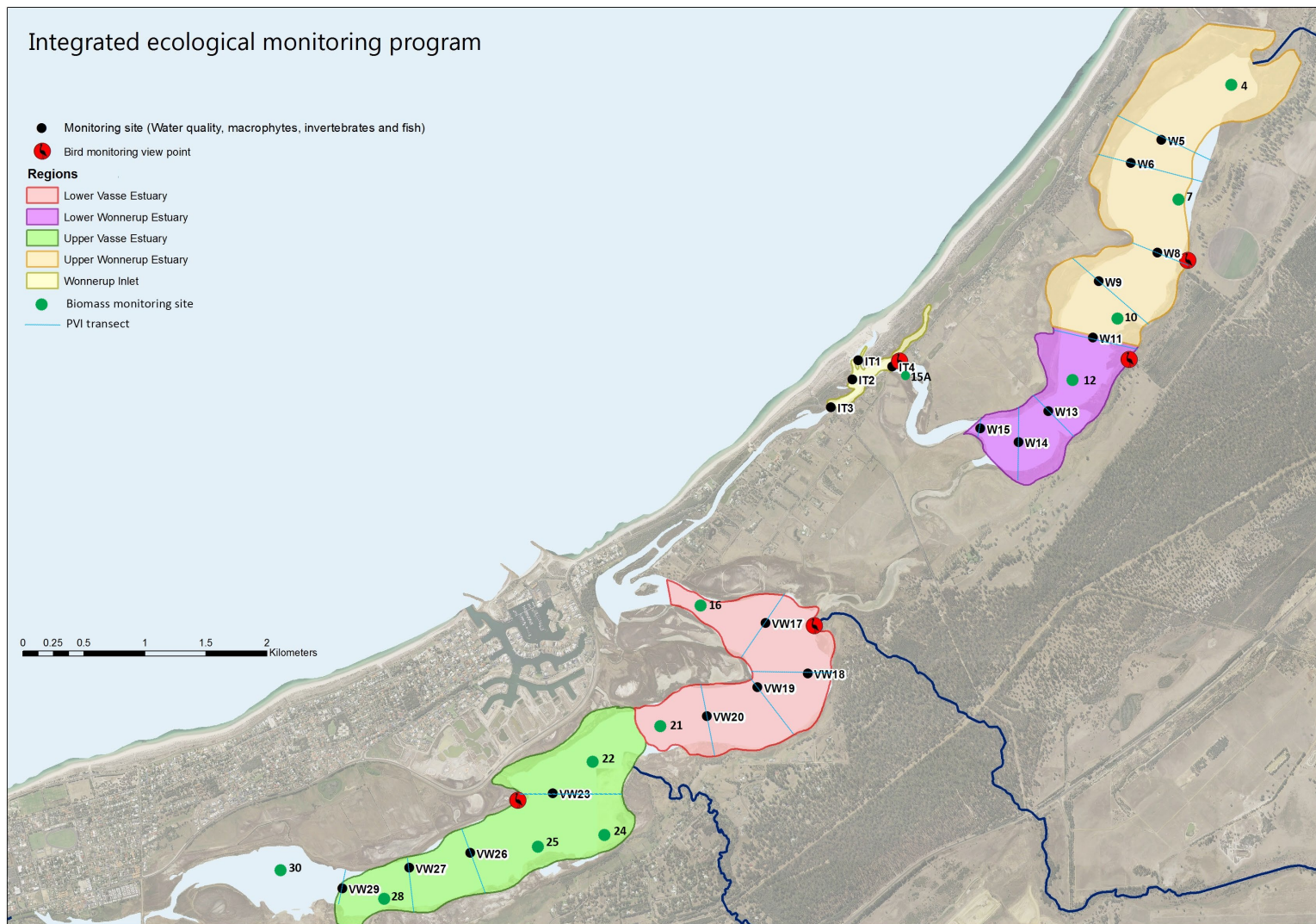
Figure 1. Sample site locations in the Vasse and Wonnerup Estuaries and the Wonnerup Inlet, showing a priori designated ecological regions.

Methods

Sampling sites and timing

Aquatic plants were sampled at 16 sites in the Vasse-Wonnerup System, four in each of five 'ecological regions' (Figure 1). These ecological regions were defined a priori for congruent sampling of multiple ecological indicators: aquatic plants, fish, macroinvertebrates, birds and water quality. They were:

- − Lower Vasse Estuary
- − Upper Vasse Estuary
- − Lower Wonnerup Estuary
- − Upper Wonnerup Estuary

The Wonnerup Inlet was also included as a region for sampling from March 2017 to March 2019 and was ceased due to the consistent absence of plants. This region is not included in this report.

The study area is located in southwestern Western Australia, which experiences a mediterranean climate with a typical seasonal pattern of hot dry summers and cool wet winters (Figure 2). This climate understandably drives plant growth patterns, however active management of water levels via surge barriers also influences depth and water quality.

To capture the seasonal changes in plant communities and density throughout the system, sampling was undertaken seasonally from March 2017 to March 2021, with additional monthly sampling from October to January in 2019-2020 and 2020-2021 (Table 1). The main growing season for aquatic plants is winter and spring with subsequent senescence in summer and autumn as water levels decline. Data is presented in this report to align with this growing season. March 2017 represents the end of the previous growing season, which was not sampled, and included some different sites, and is excluded from this analysis.

Table 1. Aquatic plant sampling dates during the IEM program

Year	Winter	Spring	Summer	Autumn
2017-2018	24-25 Jul 2917	18-20 Oct 2017	17-18 Jan 2018	19-21 Mar 2018
2018-2019	30-31 Jul 2018	30 Oct – 1 Nov 2018	17 Jan 2019	20-21 Mar 2019
2019-2020	25-26 Jul 2019	29-31 Oct 2019	20-21 Dec 2019	26 Mar 2020
		26-28 Nov 2019	20-22 Jan 2020	
2020-2021		15-16 Oct 2020	21-22 Dec 2020	16-29 Mar 2021
		27 Nov – 1 Dec 2020	28 Jan 2021	
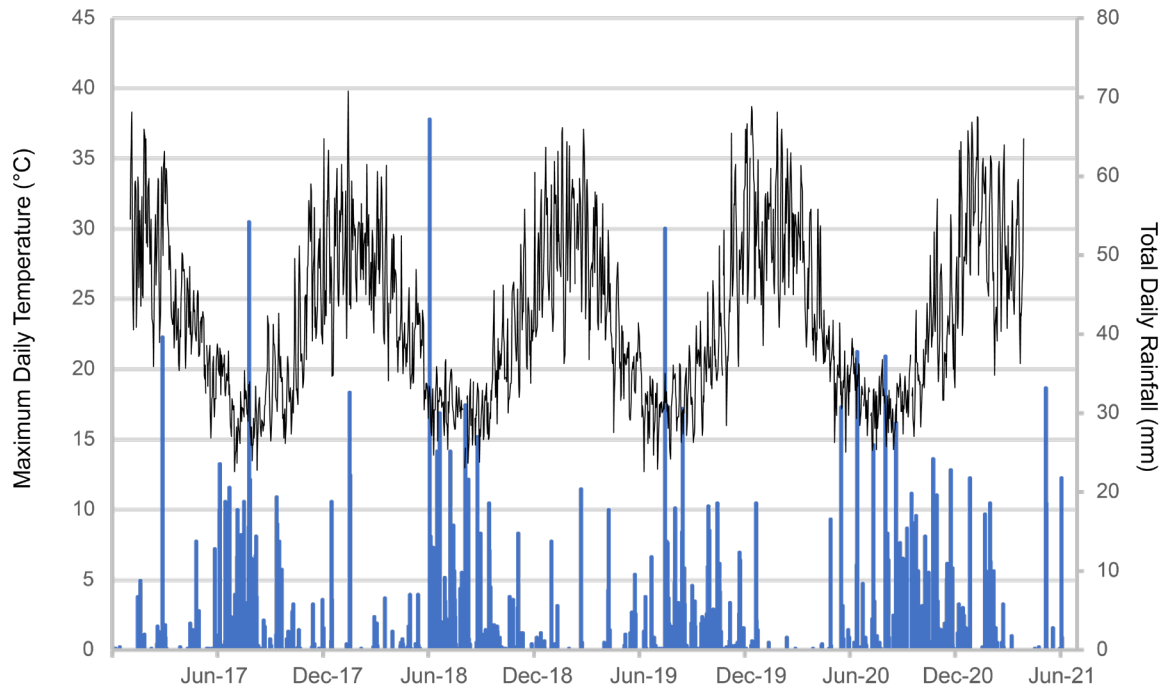
Figure 2. Rainfall and temperature data during the sampling period (BoM 2021).

Aquatic plant sampling method

For seasonal sampling, of plant density was assessed as 'percent volume inhabited' (PVI)[2]. This is a measure of plant density in terms of the proportion of a body of water taken up by plant material. In addition, biomass sampling was undertaken concurrently during spring to provide continuity of data with historic monitoring since 2006.

PVI was selected for seasonal sampling to avoid problems identified with core sampling for biomass, including: difficulty in identifying species from defrosted sample fragments; underestimation of density for canopy-forming species during coring; and time-intensive sample processing, which increases costs and delays data availability. In addition, small core diameter may lead to missed species and underestimation of density when cover is very low, as occurs at the beginning and end of the growing season. Furthermore, biomass sampling is a destructive method, and to implement this multiple times per year at the same sites may influence site characteristics (Wood et al 2012).

PVI does require expertise of field staff, but entails far less time than laboratory processing and data is available within days. Data is easily interpreted, as it allows an intuitive understanding of plant density.

The use of PVI as a measure of the standing crop of submerged vegetation has been applied in studies of large lakes using depth sounders (Maceina and Shireman 1980, Canfield *et al* 1984); and in shallower systems using observed cover and plant height (Wood *et al* 2012). It has been adapted for use in many studies of submerged aquatic vegetation to investigate growth dynamics and relationships with biotic and abiotic factors at different, for example:
- comparison of phytoplankton, zooplankton, fish and plant species richness in between lakes with turbid and clear states (Kruk et al. 2009);
- investigating the refuge function of macrophytes for zooplankton (Jensen et al. 2010);
- large scale comparisons across nutrient and latitudinal gradients (Jeppesen et al. 2000, Bachmann et al. 2002, Kosten et al. 2011);
- comparisons with fossil records (Lev et al. 2014);
- experimental effects on water quality (Nakamura et al. 2008);
- effects on sediment and phosphorus resuspension (Horpilla et al. 2005);
- allelopathy investigations (e.g. Nemoto et al. 2012);
- impacts of waterbird grazing (Chaichana et al. 2011); and
- influence of climate-induced abiotic changes on macrophyte growth (Ersoy *et al* 2020).

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[2] Also referred to in studies as 'percent volume infested', 'plant volume inhabited', 'plant volume index'.

Plant volume inhabited (PVI)

PVI was determined at five points (replicate site samples) along a transect from bank to bank across the estuary at four sites in each ecological region (Figure 1). These five transect points were located approximately equidistant along the each transect, with one of these points being the specific site location used for other sampling.

Each transect point consisted of a circular area 5m in diameter, determined using a stake with a 2.5m length of rope held to limit the area observed. At each transect point, PVI was determined using visual estimation of plant cover, combined with measurement of water depth and plant height (Figure 3). Due to the shallow nature of the Vasse-Wonnerup Wetlands, PVI was able to be determined for individual species, providing a clear indication of the extent and diversity of plant habitat relative to open water. This is valuable in an ecological sense as it describes how much of the body of water in the wetland is filled with plant material, both in total and for each species.

A bathyscope was used to observe plants and percentage cover was determined as total cover (up to 100%) and independently for each species (Figure 4a). Due to the growth of different species as layers, total of species' cover may be greater than 100%. Cover was estimated as 1%, 5%, 10% increments to 90%, or 95%. Height for each species was measured directly by viewing a marked pole through the bathyscope (Figure 4b). The most common plant height for each species was measured to the nearest 5cm for most macrophytes and charophytes, and at 1cm intervals for very small plants and filamentous algae.

The following measurements were recorded for subsequent calculation of PVI for each species:

- D = water depth (m)
- C_{total} = total cover (%)
- $PC_{species}$ = proportion of total cover for each species (%)
- $H_{species}$ = height of each species (m)

For each species, cover $C_{species}$ was determined by $PC_{species}/100$ x C_{total}. PVI was then calculated as:

$$PVI = \frac{C_{species} \times H}{D}$$

(Canfield *et al*., 1984).

PVI for each site was calculated as the average PVI of all transect points. The shallow nature of the system enabled accurate cover estimates from observations from above the water or using a bathyscope at most sites. Plant height was measured for the most common size plants of each species, using a measuring pole and bathyscope, For deeper and more turbid sites, five one-metre rake samples were pulled at each transect point. Cover for each species estimated based on the amount of rake teeth filled (i.e. all rake teeth filled on all rake pulls = 100%) and plant length measured to

provide height. This approach was only required for the deeper transect points at sites 15 and 17.

Biomass

Biomass samples were taken during spring in each year, with samples processed to determine dry weight per square metre for each species, as described in Chambers et al. (2017). Biomass sampling included additional sites (as sampled historically, Figure 1), with 5 replicate cores collected at a random location in close vicinity of each site. A perspex corer (9 cm diameter x 50 cm length) was pushed into the sediment over the benthic flora and sealed with a rubber stopper, allowing an intact core with plants and sediment to be extracted (Figure 4c). Extracted plant material was sieved to remove excess sediment and the samples bagged for transport to the laboratory, and frozen for later processing. In the laboratory each sample was sorted to separate species, and species samples then dried at 70 °C for 48 hours. Dry weights were determined to 0.0001g and species biomass was converted to grams per m^2 based on the area of the corer.
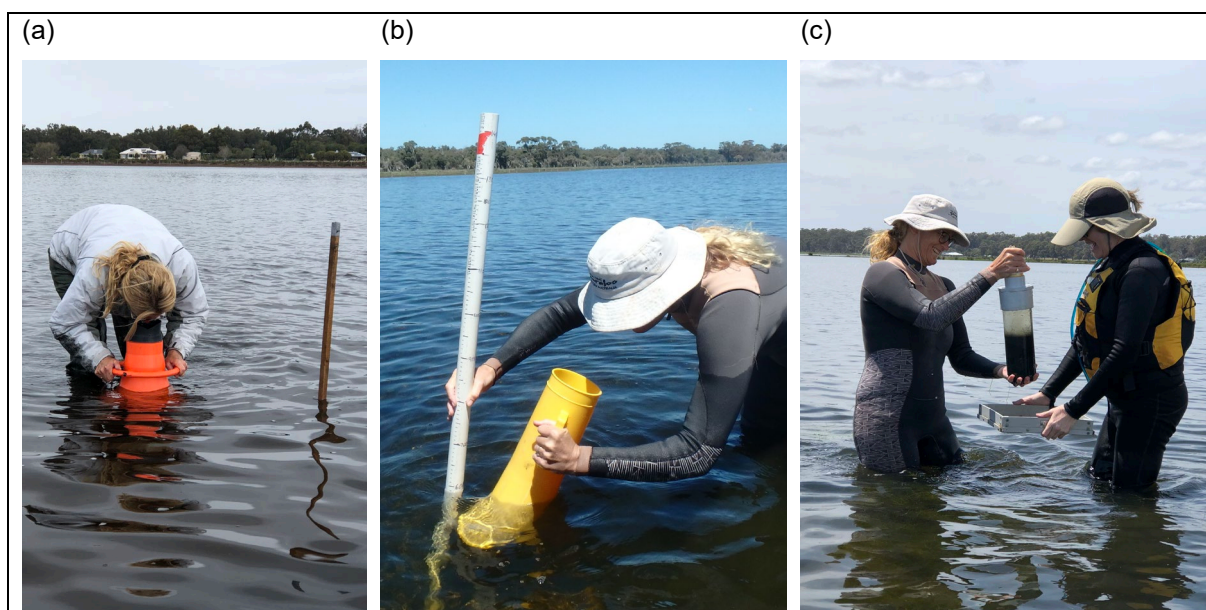
Figure 3. Sampling aquatic plants in the Vasse-Wonnerup Wetlands: (c) Estimating plant cover within a 2.5m radius; (b) Measuring plant height; (c) biomass core sampling.

Data analysis

Aquatic plant communities

Data from transect points for each site were averaged to provide site PVI values. Face-value plots and exploratory statistics were used initially to examine this data, with transect data points referred to for assessment of species distribution (i.e. isolated patches or consistent occurrence).

Differences in plant community assemblage between regions were examined using constrained (CAP) ordination in PRIMER-E v6 and tested using ANOSIM (analysis of similarity), with factors of region and year. Spring data (October) was used for analysis to allow comparisons of established communities. For these analyses, pairwise R values differences as follows: >0.75 well-separated; >0.5 overlapping but clearly different; <0.25 barely separable (Clarke and Gorley 2001). SIMPER (similarity percentages) was used to further identify the species responsible for differences identified. For testing of single species of groups repeated measures ANOVA and linear contrast analysis was performed to test significance of inter-year plant differences and trends over time.

Water levels and water quality

Long term continuous water level monitoring at the Vasse and Wonnerup surge barriers was provided by Department of Water and Environmental Regulation (DWER). Relationships between these water levels and extent of water area in different regions of the estuaries was also provided by DWER, which were applied to mean daily water levels for the period 2006-2021 to assess changes in water extent over time.

Water quality sampling was undertaken, and data provided, by DWER. Nutrients and physicochemical variables were sampled at the same sites as aquatic plants at weekly to monthly intervals, coinciding with plant monitoring. Exploration of descriptive statistics and plotting of variables was used to examine spatial and temporal variation. Repeated measures ANOVA was used to test significance of notable observed differences in SPSS (IBM).

Multivariate analysis of differences in water quality between ecological regions (*a priori* groups) included both principal components analysis (PCA) and constrained ordination via canonical analysis of principal components (CAP) using procedures in PRIMER-E v6 (Plymouth Routines in Multivariate Ecological Research, Clarke and Warwick 2001). This was done for spring (October) data only, due to strong seasonal variation making analysis of annual data problematic. Variables included those most likely to influence aquatic plants: nutrients, salinity, depth, and sediment organic content (loss on ignition [LOI], sampled during spring PVI monitoring). Dissolved organic nitrogen was excluded due to high correlation with TN ($r=0.911$, $p<0.001$). Environmental data was transformed (Log x+1) and normalised prior to analysis.

Relationships between plants and environmental variables

The BIO-ENV procedure in PRIMER-e v6 was initially used to assess overall correlation of plant species assemblage patterns with environmental variables. CAP was applied to environmental data and the similarity matrix of plant density data to identify relationships between water quality variables and plant community assemblages and investigate how these separated region groups (Clarke and Gorley 2001). Spearman's rank correlation coefficient was also used as a robust test of correlation between different water quality variables and between these and plant species. It was not practical to undertake statistical analysis of relationships between environmental data and plant community assemblages using data from January to March due to the lack of plants during this time of year. Inclusion of all data from July to December in analysis was problematic due to strong seasonal signals masking relationships.

Plant sampling methodology comparison

Comparison of plant density and biomass methods was completed using the four years of data from the IEM program and corresponding years of biomass data. This included face-value comparison of outcomes of the two methods and correlation and regression analysis. Pearson correlation and linear regression was completed for comparison of biomass data with both corresponding site points and transect data, with zero-value results excluded. Biomass and PVI data were transformed ($Log_{10}x+1$) for these analyses to achieve normality.

Figure 4. Aquatic plants occurring in the Vasse-Wonnerup Wetland System: (a) Ruppia megacarpa, (b) Ruppia polycarpa, (c) Althenia cylindrocarpa, (d) Stuckenia pectinata, (e) Lamprothamnium macropogon, (f) Ulva spp., (g) Cladophora vagabunda, (h) Rhizoclonium tomentosum.

Results

General seasonal pattern of aquatic plant growth

Key findings:

- Aquatic plants in the Vasse-Wonnerup wetlands include three species of macrophytes, one species of charophyte and three genera of macroalgae.
- Aquatic plant communities demonstrate an annual growth cycle corresponding to climate variation, commencing with the onset of winter rains, and reaching peak density in spring, and senescing in summer as water levels drop.
- The highest plant density occurred in spring; in the two years in which monthly sampling was conducted this was during November in the Vasse Estuary, and during November to December in the Wonnerup Estuary
- A macroalgal bloom, consisting mainly of *Cladophora*, occurred in the lower Vasse in summer to 2018 and extending into the upper Vasse in autumn.

Aquatic plants occurring in the Vasse-Wonnerup are (Figure 5):

1. Macrophytes - rooted submerged angiosperms including *Ruppia polycarpa, Ruppia megacarpa, Stuckenia pectinata,* and *Althenia cylindrocarpa.*
2. Charophytes - rooted plant-like green algae, *Lamprothamnium macropogon.*
3. Macroalgae - large multi-cellular green algae with sheet, tubular or filamentous morphology, often free-floating including species of *Ulva, Rhizoclonium* and *Cladophora.*

Aquatic plant growth followed a pattern of increasing biomass from winter through to spring, and subsequent summer and autumn decline, evident in results of plant density (PVI), plant cover and plant height (Figure 6, Figure 7, Figure 8). This growth pattern is expected in association with the annual pattern of climate (Figure 2) and water levels (Figure 9).

Winter plant growth varied year to year in each region in terms of total density and community composition, with both macrophytes and macroalgae present. Macroalgal growth was extensive throughout the system in winter 2017, whereas macrophyte growth was dominant in most regions in summer to winter 2018 (Figure 6). The exception was in the lower Vasse, where very little macrophyte growth occurred following the macroalgal (*Cladophora*) bloom in Autumn. Winter plant growth was lower in 2019 compared with previous years (Figure 6a), following lower than average autumn rainfall (March to May total 81.2mm, compared with a long-term average of 155.1mm (Figure 2)). Mean total plant density was 5% in the upper Vasse, where mainly macrophytes and charophytes grew; and 5-6% throughout the Wonnerup, where macroalgae (*Ulva)* occurred at similar levels to macrophytes (lower region) and charophytes (upper region).

Substantial plant growth occurred between winter and spring, when peak biomass was found in all years. In 2019 and 2020, when monthly sampling was undertaken from October to January, the greatest PVIs were observed during November (Figure 6). The Wonnerup Estuary was dominated by macrophytes in spring and had limited macroalgal growth, with respective macrophyte and macroalgae means of 40% and 2% in the lower Wonnerup and 28% and 2% in the upper Wonnerup. Mean spring density of macrophytes was greater than macroalgae in the Vasse, and contributed substantially to total density, with respective means of 28% and 10% in the lower Vasse and 36% and 11% in the upper Vasse. In these years with monthly sampling, high macrophyte density was maintained in the Wonnerup Estuary during December, but declined in the Vasse Estuary, particularly the lower Vasse.

Additional sampling during December 2019 and 2020 captured the initial seasonal decline in aquatic plants in the Vasse Estuary, while in the Wonnerup relatively high macrophyte density was maintained (Figure 6). By January, plant density declined substantially in the Wonnerup in 2018 and throughout the system in 2020 and 2021 (Figure 6Figure 7). In analysing summer data, it is important to note the effect of low water levels on PVI in the upper regions of the estuaries, where shallow waters can lead to high PVI relative to biomass. Macrophytes remained in the upper Vasse in January 2018, while macroalgae dominated the lower Vasse (Figure 6a). Macroalgae was also present at low levels throughout the Vasse Estuary during summer in 2019 and 2021, while the Wonnerup retained macrophytes.

Very little plant growth was observed throughout the system in Autumn. Macrophytes in the lower Wonnerup persisted in some years but co-occurred with macroalgae. In March 2018, macroalgal growth increased substantially in the lower Vasse during summer and autumn, and extended into the upper Vasse, while macrophytes were absent (Figure 6a). This was due to a bloom of filamentous macroalgae *Cladophora* (primarily) and *Rhizoclonium* throughout the lower Vasse Estuary (Figure 10), extending into the more downstream sites of the upper estuary. Macroalgal blooms have not been observed in either estuary in Autumn since this time.
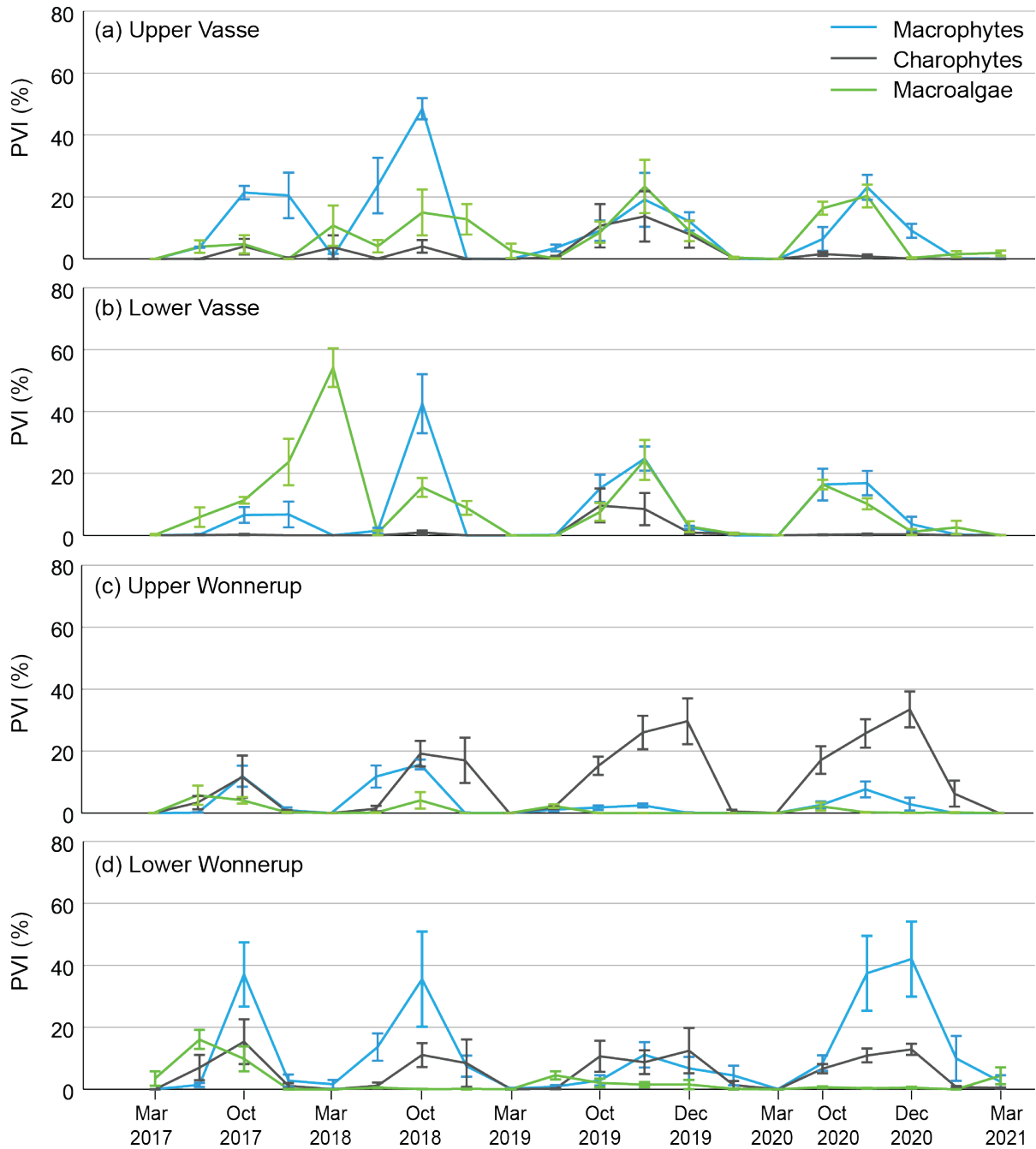
*Figure 5. Mean plant density of macrophytes, charophytes (*Lamprothamnium*) and macroalgae for each ecological region in the Vasse-Wonnerup system from March 2017 to March 2021. Error bars are ± standard error.*
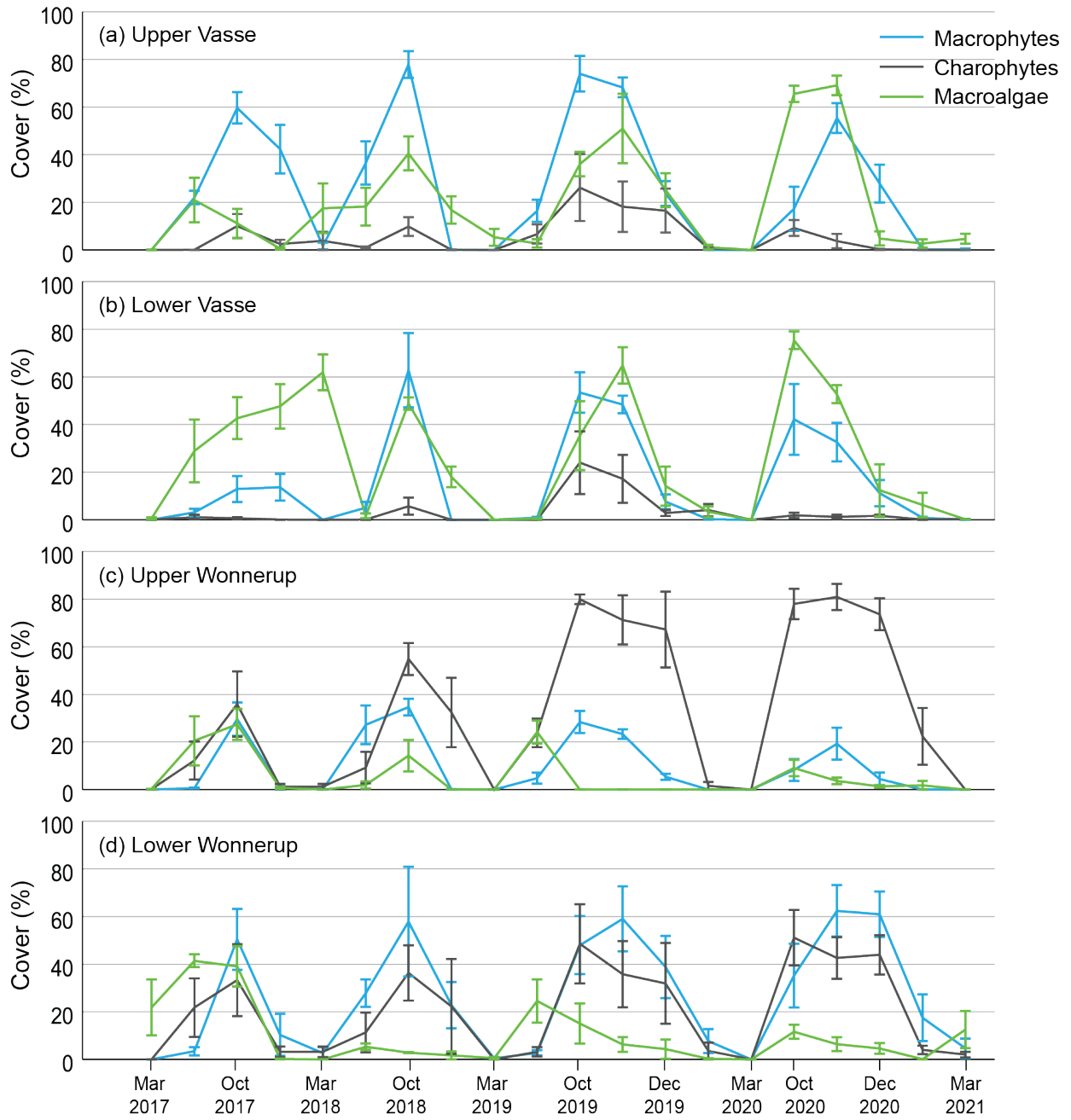
*Figure 6. Mean plant cover of macrophytes, charophytes (*Lamprothamnium*) and macroalgae for each ecological region in the Vasse-Wonnerup system from March 2017 to March 2021. Error bars are ± standard error.*
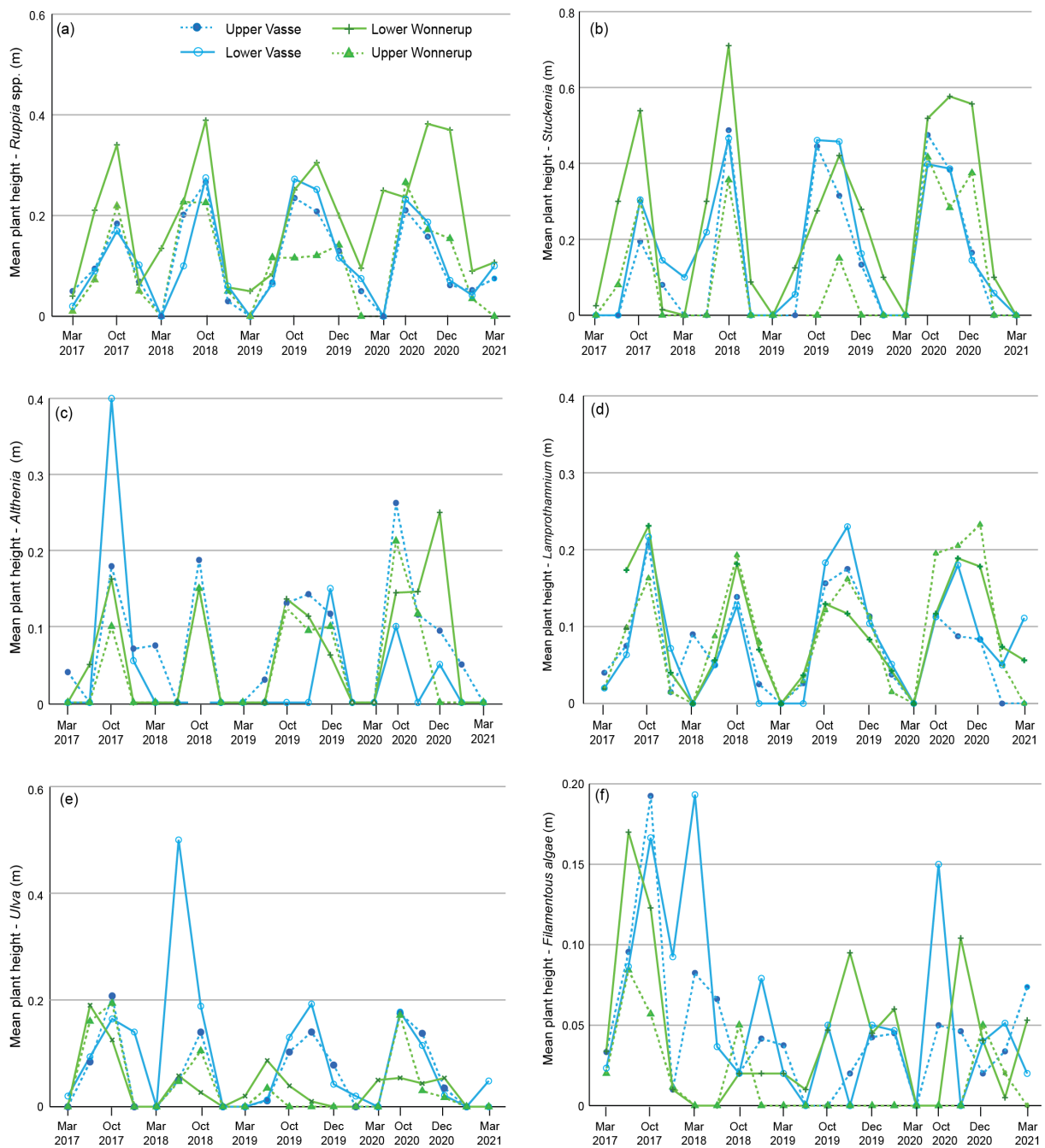
Figure 7. Mean height of each aquatic plant species/group in the ecological regions Vasse and Wonnerup Estuary from March 2017 to March 2021. Values represent actual plant heights when present, excluding transect points of zero to avoid under-representing height; values of zero indicate total absence of species throughout region.
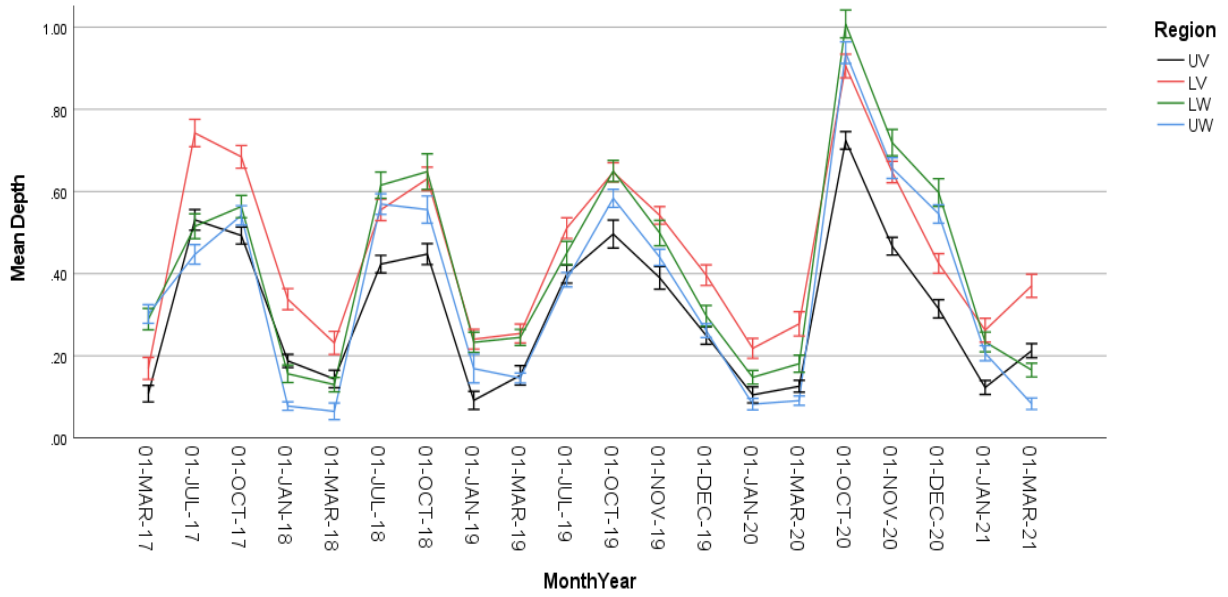
Figure 8. Mean water depth in regions from Autumn 2017 to Autumn 2021, from transect point depth measurements. Error bars show +/- standard error.
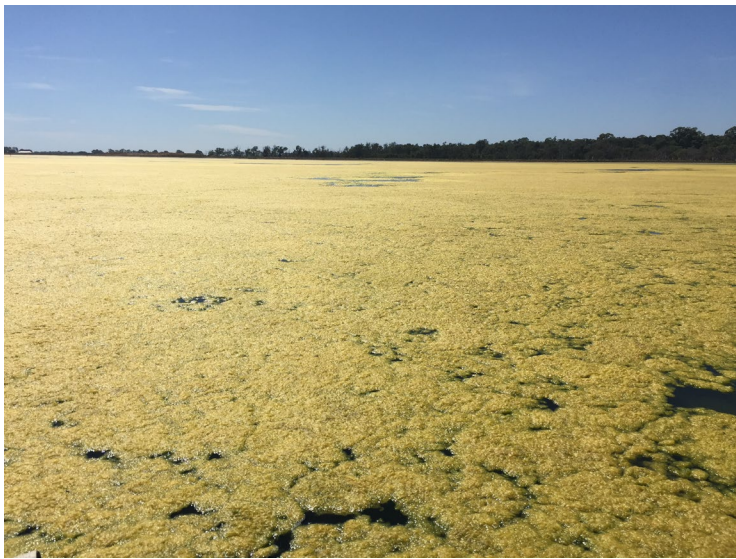
Figure 9. Extensive Cladophora growth in the lower Vasse Estuary in March 2018 – an example of a "macroalgal bloom". Average PVI was 54% and cover was up to 100%, with a thick mat floating on the surface.

Water levels

Key findings:

- Consistently higher autumn water levels relative to historical levels have occurred in the Vasse Estuary since 2018, corresponding to increased seawater inflows at the Vasse surge barrier.

- Comparison of mean values before and after this management change suggest the extent of water has increased by 15ha in March and 17ha in April in the Upper Vasse.

Water levels in the Vasse-Wonnerup Wetlands fluctuate seasonally (Figure 9), driven by rainfall and evaporation. There is an annual cycle of winter filling, decline over summer and lowest water levels in autumn, when large areas dry out. Water levels in the system tend to be highly variable during winter, but more stable during spring prior to a gradual decline (Figure 11). Historical water level data shows a high level of inter-annual variation in water levels in the system.

In addition to climate, water levels are strongly influenced by the presence of surge barriers at the exit channels of the Vasse and Wonnerup Estuaries. These surge barriers are necessary for flood protection during storm surge events, allowing outflow from the system but preventing tidal inflow. The Wonnerup Estuary water level is generally around 0.2m AHD lower than the Vasse Estuary.

The surge barrier gates have been actively managed during summer for many years to allow for fish movement during times of poor water quality. Since December 2017, increased opening of fish gates during summer has been undertaken through the Seawater Inflow Trial (DWER 2021) to increase seawater inflow to the Vasse Estuary to address extremely poor water quality in the exit channel (Table 2). This has corresponded to high water levels during March and April since 2018, relative to historical levels (Figure 11a). During this time there has been partial opening of gates at the Wonnerup Estuary with only small exchanges (Table 2), however recent March-April water levels (2019-2020) have also been higher than historically (Figure 11b).

Changes in water level translate to altered wetting and drying extent in the wetlands, particularly the case in the upper regions of the estuaries, which have extensive dry areas during late summer and autumn. In the upper Vasse, water coverage during the driest months of March and April since the increased seawater inflows commenced in the 2017-2018 summer in comparison with previous years (Figure 12). Comparison of mean values before and after this management change suggest the extent of water has increased by 15ha in March and 17ha in April in the Upper Vasse. Extent of water cover in autumn has been highly variable in the Wonnerup since 2015 but has also been relatively high since 2019. This estimate extends beyond the upper Wonnerup ecological region to about site 13, which may contribute to greater variation.
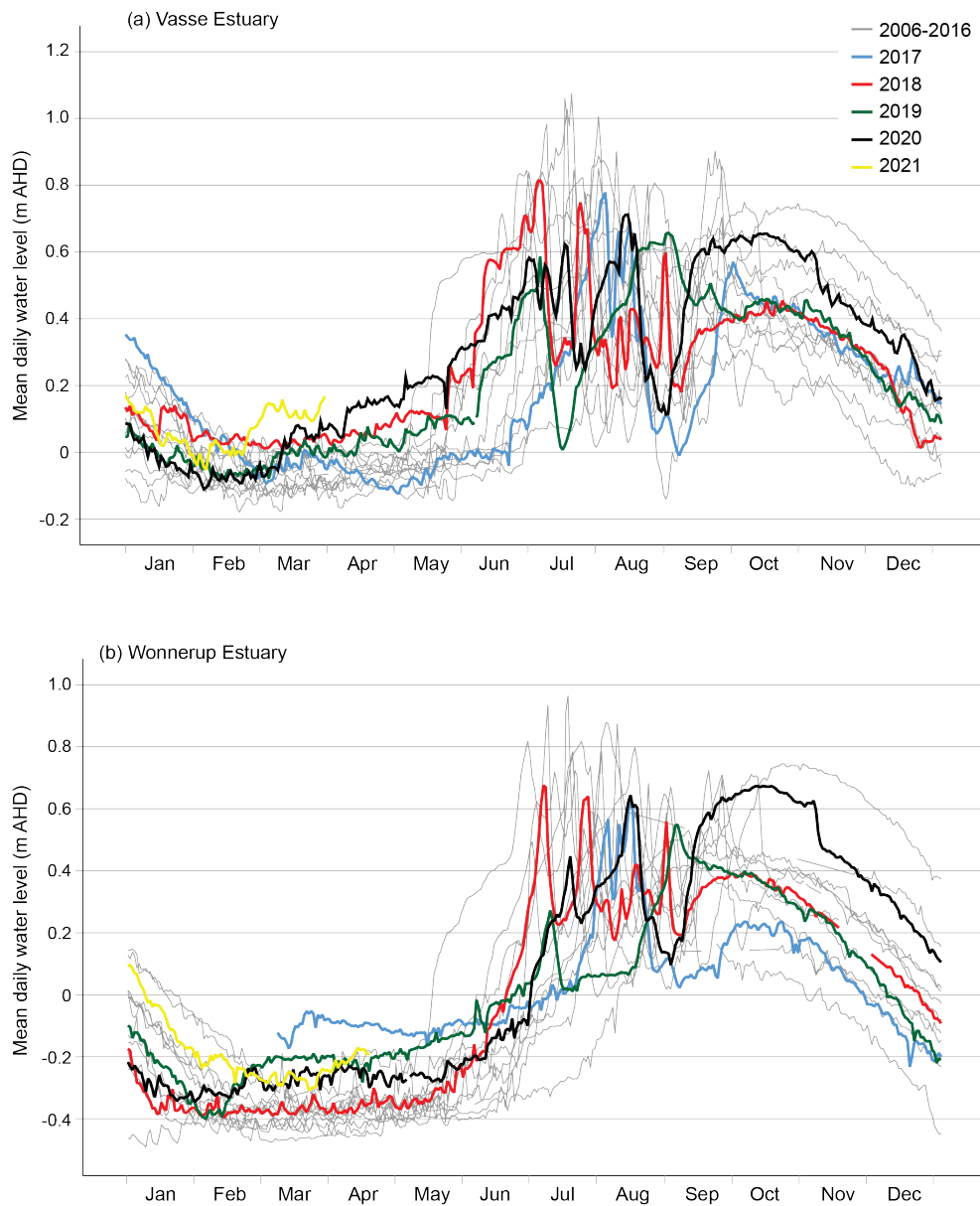
Figure 10. Mean daily water levels at the Vasse (a) and Wonnerup (b) surge barriers (upstream), highlighting levels during the Integrated Ecological Monitoring (coloured lines) against historical data (grey lines). Data from Water Corporation and Department of Water and Environmental Regulation continuous monitoring.

Table 2. Summary of fish gate opening regime at the Vasse and Wonnerup Surge Barriers during the seawater inflow trial (information provided by Department of Water and Environmental Regulation).

Summer – Autumn period	Vasse Surge Barrier	Wonnerup Surge Barrier
2016-2017	Closed until March, then 100% open for 2 weeks	Closed until March, 100% open then periodic partial opening for 2 weeks
2017-2018	100% open from early December	5-7% open from late January
2018-2019	100% open from early December	80-100% open for 2 weeks in February, then reduced to 6-14%
2019-2020	Open several times during December; then 100% open from Dec 28th.	10% open from late January
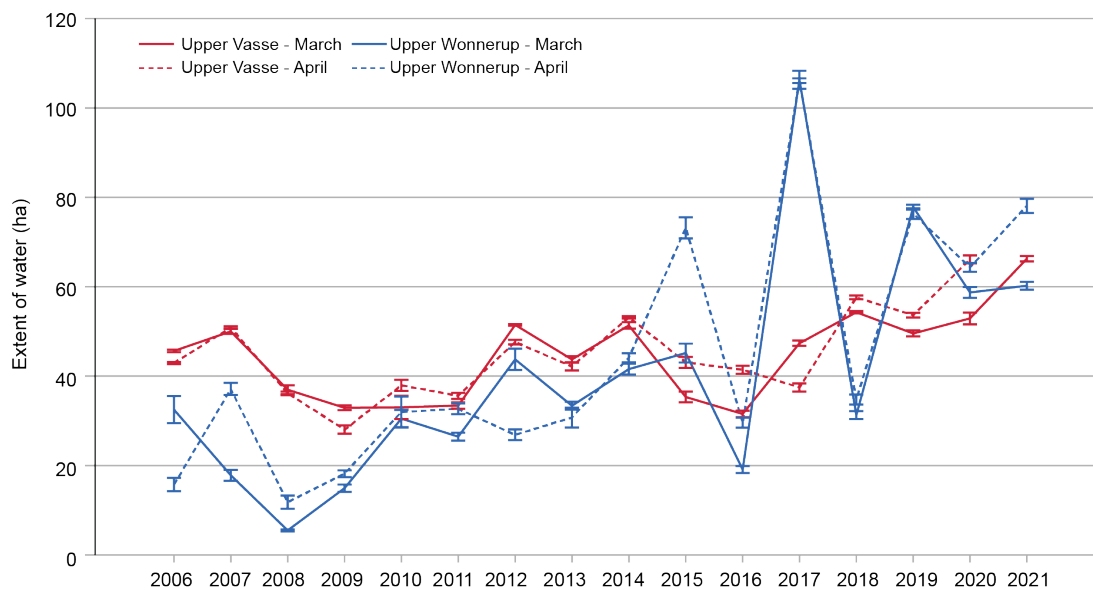
Figure 11. Area covered by water in the upper Vasse and upper Wonnerup regions in March and April since 2006, estimated from surge barrier water levels using relationships provided by DWER. Error bars are ± standard error.

Water quality

Key findings:

- Nutrient and chlorophyll *a* concentrations were consistently higher than ecosystem protection guidelines throughout the system, and extremely high in summer and autumn.

- Salinity varies greatly between seasons, with fresh to brackish conditions in winter and spring and saline to hypersaline conditions in summer and autumn.

- The upper and lower Vasse were associated with fresher conditions, lower phosphorus and lower organic nitrogen than the Wonnerup. The lower Vasse was strongly associated with high sediment organic content.

- Dissolved oxygen was high throughout the system in all seasons, but higher and more variable from spring to autumn when photosynthesis by plants and algae create diurnal fluctuations. Lower extremes were observed in summer and autumn.

- pH values were extremely high in spring throughout the system (overall mean 9.75).

Nutrients and Chlorophyll

Total phosphorus (TP) concentrations in the system are higher than the ecosystem protection guideline (30μg/L) in both estuaries throughout the year, and extremely high and variable during summer and autumn in all regions (Figure 13a). TP decreased throughout the winter months with lowest concentrations and less variation within regions in late winter and spring. The upper Wonnerup had the highest TP from summer to winter and highest annual mean of 254μg/L. TP was relatively similar in other regions from summer to winter. The upper Vasse had the lowest annual mean of 162μg/L, while annual means the lower regions were 181μg/L in the Wonnerup and 186μg/L in the Vasse. Lower concentrations occurred in all regions during spring, and the upper Vasse had the lowest spring TP concentrations (mean 45μg/L). Long term data (since 2006) also indicates lower spring TP in the upper Vasse region (Figure 14a). ANOVA demonstrated that upper Vasse TP was significantly lower than all other regions (pairwise comparisons $p<0.001$, region effect $p=0.25$, no interaction effect of year). There was no significant difference between other regions. TP in the upper Vasse has increased consistently over the last four years, but in the context of historical values has remained within the range of previous variation. This may highlight an issue in this region if the trend continues.

Filterable reactive phosphorus (FRP) was generally higher in the Wonnerup Estuary (upper region mean 62μg/L; lower region mean 38μg/L) than the Vasse Estuary (upper region mean 17μg/L; lower region mean 23μg/L), and was particularly high in the upper Wonnerup in summer and autumn (Figure 13b). There was no distinct seasonal variation in other regions. While uptake of FRP by phytoplankton can maintain lower levels in the warmer months, as observed in other regions, FRP remained high in the

upper Wonnerup despite high chlorophyll *a* (Figure 13f), suggesting another limiting factor. The upper Vasse had significantly lower spring FRP than the upper and lower Wonnerup regions (pairwise comparisons: p<0.001 and p=0.03 respectively) but did not differ significantly from the lower Vasse. There was an interaction effect of region and year (p=0.03), and mean FRP was higher in the upper Vasse than the lower Vasse in spring 2020.

Total nitrogen (TN) also exhibited seasonal variation with highest concentrations in summer and autumn, followed by a decline in winter and spring (Figure 13c). As for phosphorus, concentrations were well in excess of the ecosystem protection guideline (750μg/L). The upper Vasse and the upper Wonnerup had particularly high TN in autumn (season means 5,319 μg/L and 6,850 μg/L respectively, and the upper Wonnerup had the highest mean concentration in each season. However, spring TN was significantly lower in the upper Vasse than other regions (all pairwise comparisons : p<0.001) and long-term data indicates this has been the case since 2012 (Figure 14b).

Dissolved organic nitrogen (DON) made up a substantial fraction of TN, contributing 60-70% of mean annual nitrogen and 82-91% in spring. DON also followed a similar seasonal pattern to TN, and the two variables were highly correlated (r=0.911, p<0.001), although DON was less variable (Figure 13d). As for TN, the upper regions of both estuaries had higher DON than the lower regions in summer and autumn. In winter and spring, concentrations were lower and similar throughout the system. In contrast, dissolved inorganic nitrogen (DIN: sum of ammonium, nitrate and nitrite) contributed less than 5% of TN and was much higher in winter than other seasons (Figure 13e). High inorganic nitrogen in winter may reflect catchment loads, subsequently incorporated into the plant community in spring.

Oxidised nitrogen (nitrate + nitrite; NO_x) was much lower than guideline (45μg/L) from spring to summer (system mean <10 μg/L), but exceeded the guideline in all regions in winter and was highest in the lower Vasse (52μg/L). Ammonium (NH_4^+) concentrations were higher than NO_x^-. Concentrations of ammonium in spring were below the guideline (40μg/L) in all regions other than the Lower Vasse (57μg/L). Winter NH_4^+ was substantially higher in winter, when mean values were higher in the lower regions (673 μg/L in lower Vasse, 444 μg/L in lower Wonnerup) than the upper regions (292 μg/L upper Vasse, 218 μg/L upper Wonnerup).

Chlorophyll *a* was highest in summer and autumn, reflecting the warm conditions conducive to phytoplankton growth, and concentrations at this time were well above guideline values throughout the system (Figure 13f). The upper Wonnerup had the highest summer and autumn concentrations (seasonal means 65 μg/L and 58 μg/L respectively) and the upper Vasse had the lowest (36-37 μg/L). In winter, chlorophyll *a* was much lower in the Wonnerup estuary (mean 11 μg/L) than the Vasse (mean 35

µg/L), and in spring, concentrations declined further for all regions except the lower Wonnerup.
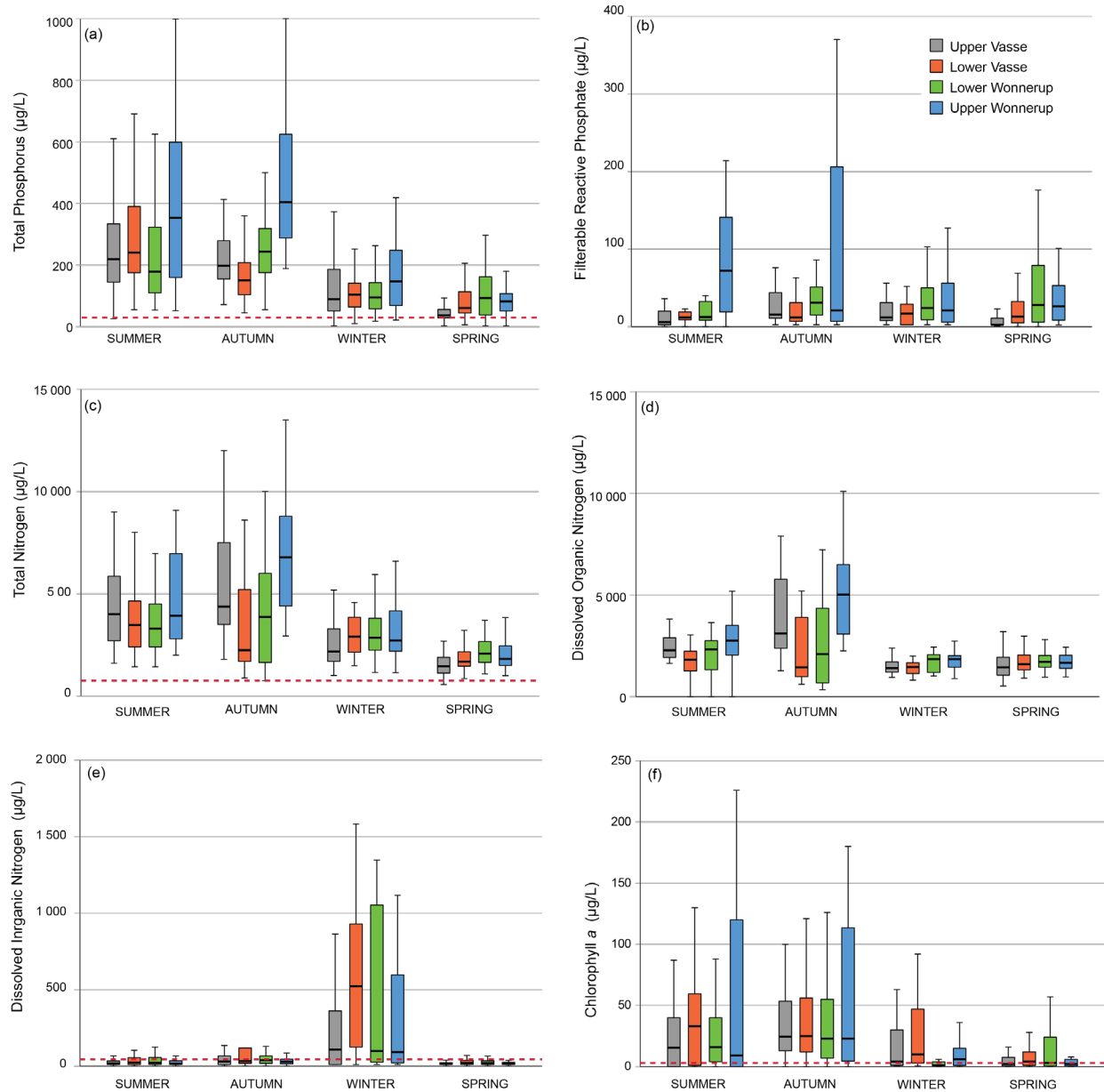
Figure 12. Seasonal nutrient (a-e) and Chlorophyll a (f) concentrations in the ecological regions of the Vasse-Wonnerup wetlands. Boxplots show median and 25-50 percentiles as box and 5-95 percentiles as bars. Red lines are ecosystem protection guidelines for southwest WA estuaries (ANZECC and ARMCANZ (2000). 2017-2020 data, provided by DWER.
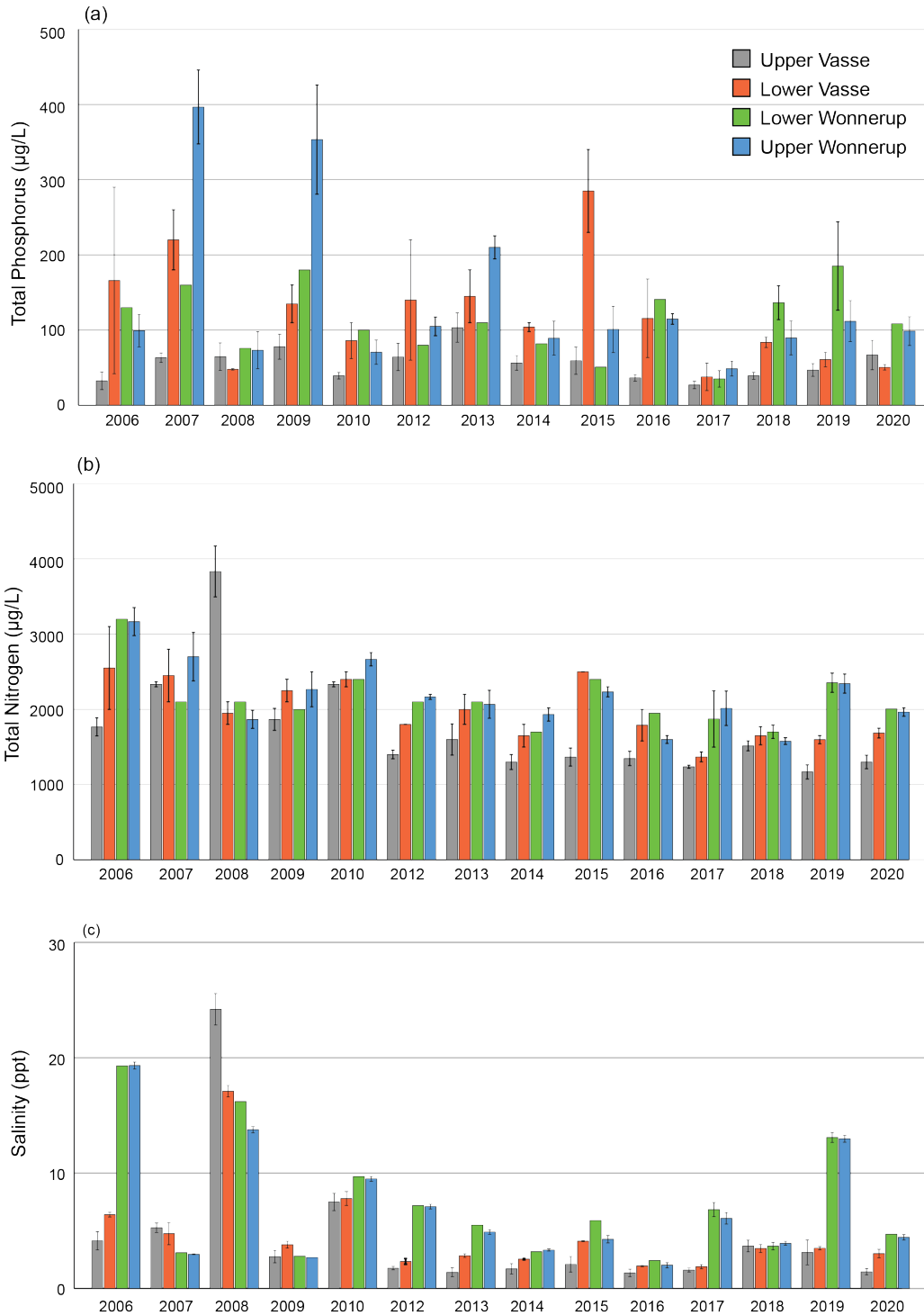
Figure 13. Long term mean spring values for (a) total phosphorus, (b) total nitrogen and (c) salinity in the ecological regions of the Vasse-Wonnerup Wetlands 2006-2020. Error bars are ± standard error. Data from Murdoch University and DWER.

Physicochemical

In relation to physico-chemical variables, plant growth is influenced by temperature and salinity but does not alter these variables; whereas the presence of plants has a strong influence on pH and oxygen levels.

Water temperature in the system corresponds to seasonal variation in air temperature and are fairly consistent across regions. The warmest conditions occur during summer, and warm conditions extend into March, with conditions then cooling through winter, and subsequently warming again in spring (Figure 15a).

Salinity varies greatly in the wetlands throughout the year, with fresh to brackish conditions in winter and spring and saline to hypersaline conditions in summer and autumn (Figure 15b). Monthly average salinity ranges were from 0.6 - 59.6ppt in the upper Vasse; 1.9 - 44.8ppt in the lower Vasse; 5.0 - 46.8ppt in the upper Wonnerup and 5.3 - 46.4ppt in the lower Wonnerup. Summer-autumn salinity has been higher in the last three summer-autumn periods compared to 2016-2017 in all regions. Hypersaline conditions occur due to evapoconcentration, particularly in the upper regions during the during periods of low water levels.

Median levels of dissolved oxygen (DO) were high for all months in all regions, with whole-system medians ranging from 94% in June to 152% in March (Figure 15c). More extreme ranges were observed from December to March, likely due to photosynthesis and respiration by greater biomass of phytoplankton during this period (indicated by chlorophyll *a*). This diurnal cycle strongly influences DO and can result in high variation linked to the time of sampling. In winter and spring more stable DO was observed, generally increasing from July to November. It is interesting to note that high plant biomass in late spring was associated with less DO variation than observed during summer when plants were scarce and phytoplankton or benthic algae would be the main primary producers. Higher median DO was observed during summer for all regions except for the lower Wonnerup, where medians were similar or lower than winter medians and the lowest values in the system were observed.

A seasonal cycle was also observed for pH, with lowest values recorded in late autumn and early winter, increasing over the spring months and declining in summer (Figure 15d). Similar levels of pH occurred throughout the system, with overall median values ranging from 8.9 in May to 10.5 in September to November. Fluctuations in pH reflect photosynthesis by plants during this period of maximum growth and biomass. Plants remove carbon dioxide (CO_2) from the water for use in photosynthesis. Hydrated CO_2 is acidic, so removing it from the water increases the pH, if the system is not buffered (Boulton et al 2014). Photosynthesis increases DO and removes CO_2 in equal proportions, so the extremely high DO is indicative of significant removal of CO_2 and concomitant pH change. Extremely high pH values occurred in spring throughout the system
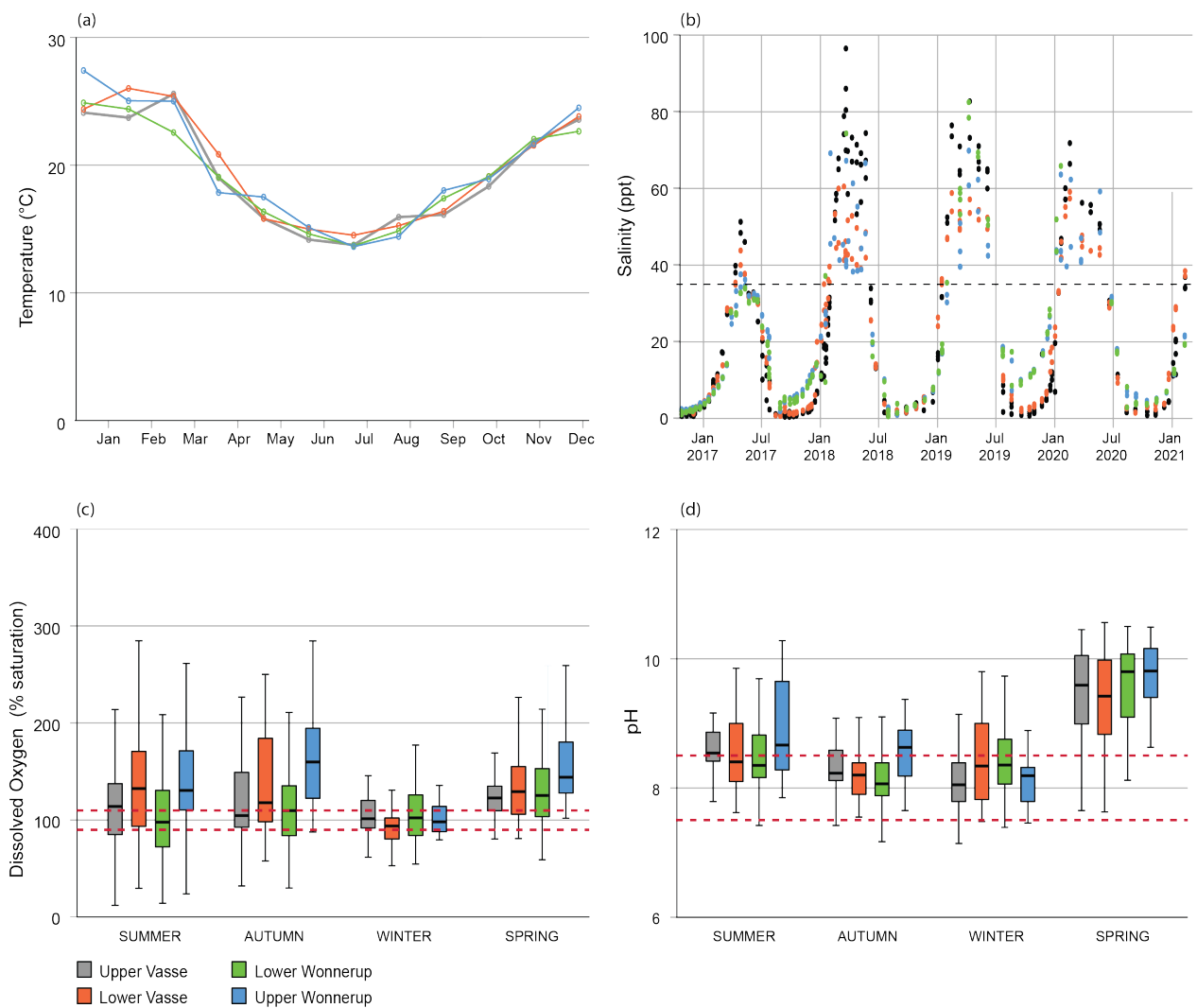
Figure 14. Summary data plots for physicochemical variables during Integrated Ecological Monitoring in the Vasse-Wonnerup Wetlands: (a) mean monthly surface water temperature; (b) Salinity values; and value ranges for (c) dissolved oxygen and (d) pH. Boxplots show median and 25-50 percentiles as box and 5-95 percentiles as bars. Red lines are ecosystem protection guidelines for southwest WA estuaries (ANZECC and ARMCANZ (2000). 2017-2020 data, provided by DWER.

Sediment quality

Loss on ignition (LOI) has been monitored as part of the biomass sampling program as an indicator of sediment organic content. The lower Vasse Estuary has consistently had the highest LOI of all the ecological regions with mean value of 8.3% since 2013 and high variation (Figure 16). Other regions have been lower and less variable. Owing to only two sample points per year in each region, it is not feasible to test for presence of trends, however none appear evident from available data. There has been no recent increase in LOI in the Upper Vasse, which has been identified as a future risk to macrophyte communities there, however this should be closely and more thoroughly monitored.

Earlier sampling has identified the lower sites in the Vasse Estuary, not subject to drying, as having the highest sediment organic and highest nutrient content, with high levels also found in the mid-lower Wonnerup (Wilson et al 2008, Chambers et al 2017). However a recent study has found higher organic content in sediments of the Wonnerup Estuary (Conway 2016). All previous sampling has indicated the upper Vasse as having the lowest levels of organic matter and nutrients, and also lowest depth of unconsolidated sediment
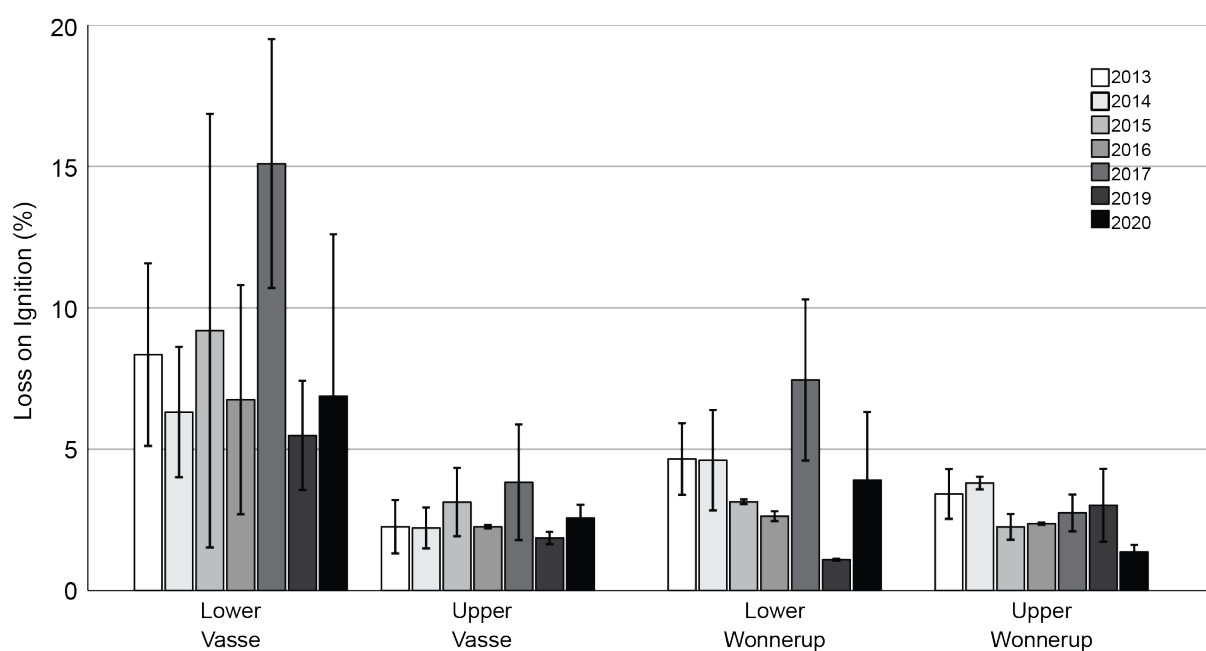
Figure 15. Average sediment organic content over time in each ecological region as measured by loss on ignition. Error bars are standard error (n=2).

Multivariate comparison of regions

Principal components analysis (PCA) for spring data provided reasonable separation of the upper Vasse Estuary from other regions based on lower phosphorus, organic nitrogen and salinity, however this only captured 37% of variability between regions overall. Constrained analysis (CAP) found reasonably strong separation between

region groups with eigenvalue correlations of 0.82 for the CAP1 axis and 0.67 for the CAP2. However, while there was differentiation of the upper and lower Vasse, the upper and lower regions of the Wonnerup were less distinguishable (Figure 17). Spearman correlations for these axes suggested the upper and lower Vasse were associated with fresher conditions, lower phosphorus and lower organic nitrogen than the Wonnerup. Within the Vasse estuary, the lower Vasse was strongly associated with high sediment organic content (LOI), which was significantly higher than other regions (pairwise comparisons, p<0.001). Lower ammonium in the upper Vasse is also indicative of lower sediment nutrient content: historical data has found correlation of ammonium with sediment TN (r=0.61, p=0.009) and TP (r=0.62, p=0.006).
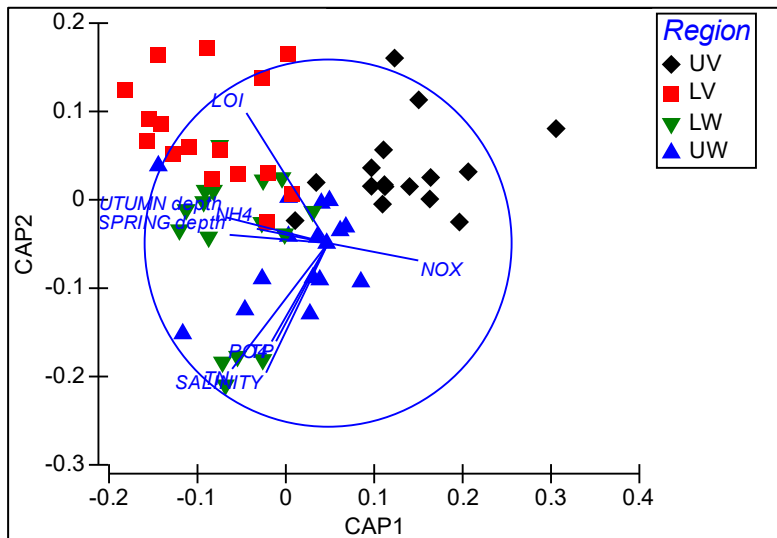
Figure 16. Constrained ordination of spring water quality data from the Vasse-Wonnerup 2017 to 2020 with vector overlay of Spearman rank correlations of water quality variables with the CAP axes (r>0.3).
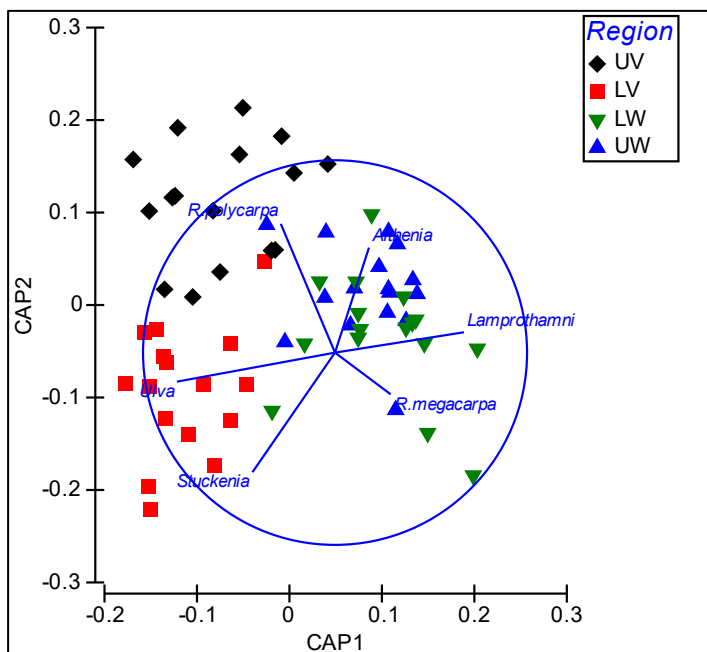
Figure 17. Constrained ordination of spring plant density (PVI) data from the Vasse-Wonnerup 2017 to 2020 with vector overlay of Spearman rank correlations of plant species with the CAP axes (r>0.3).

Aquatic plant communities in ecological regions

Key findings:

- Distinct plant communities occur in the Vasse Estuary regions, characterised by *Ruppia polycarpa* and *Althenia cylindrocarpa* (both species adapted to drying) in the upper Vasse; and *Ulva, Stuckenia pectinata* and *Ruppia spp.* in the Lower Vasse.
- Spring growth of *Ulva* has increased in the upper Vasse since 2018, resulting in a significant difference in plant assemblage to 2017. The upper and lower regions of the Vasse have become more similar since 2018 due to increased growth of *Ulva* Vasse Estuary, with similar *Ulva density* in the two regions from 2018 to 2020. This change is also evident from long-term biomass monitoring.
- The upper Wonnerup estuary is characterised by *Lamprothamnium*, but this extends into the lower Wonnerup sites so that the two regions overlap in terms of plant assemblages. Sites within the lower Wonnerup differed due to varying morphology, with the more downstream deeper sites in the channel dominated by *Ruppia megacarpa.*
- Plant communities have remained consistent the Wonnerup Estuary.

Detailed assessment of macrophyte communities is most appropriately done using spring data to correspond with peak biomass, when macrophyte communities are established. This section describes the spring aquatic plant communities within each region over the duration of the Integrated Monitoring Program (2017-2020) and presents analysis of comparisons between regions and over time. An overview of long-term biomass is also provided to give historical context to the results of the last four years.

Constrained ordination showed distinctly separate groupings of the lower and upper Vasse from the Wonnerup and overlap between the two Wonnerup regions (Figure 18). The canonical correlations of the CAP axes were strong (eigenvalue correlations 0.87 and 0.78 for CAP axes 1 and 2 respectively). Spearman correlations with the CAP axes indicated the upper Vasse was associated with *Ruppia polycarpa* and the lower Vasse with *Stuckenia* and *Ulva.* The Wonnerup Estuary (both regions) was associated with *Lamprothamnium.*

In the Vasse Estuary, ANOSIM indicated strong separation between the upper and lower regions in 2017 (R=0.68, p<0.001), due mainly to higher *R. polycarpa* in the upper Vasse and higher *Ulva* in the lower Vasse, accounting for 61% dissimilarity (SIMPER). However, in subsequent years plant communities have been less distinct (R=0.29, 0.13, 0.19 in 2018-2020 respectively). Over the 2018-2020, these regions differed mainly *R. polycarpa* and *Stuckenia* density, while average Ulva was similar.

ANOSIM indicated high similarity between the upper and lower Wonnerup regions in most years, likely due to consistent occurrence of *Lamprothamnium* in both regions.

Only in 2020 could the upper and lower regions be separated (R=0.549, p=0.001). SIMPER revealed that this dissimilarity was due to higher *Lamprothamnium* and *R. polycarpa* in the Upper Wonnerup and higher *R. megacarpa* in the lower Wonnerup, accounting for 79% of dissimilarity.

Upper Vasse Estuary

In 2017, *Ruppia polycarpa* was the dominant species present throughout the upper Vasse (Figure 19a). *Althenia* and *Lamprothamnium* were also found throughout, but with very little at site 29. Conversely, relatively high density of *Rhizoclonium* was present at site 29, while low density of *Cladophora* and *Ulva* were found at other sites. Isolated *Stuckenia* was observed. Total plant density was generally higher in the upper Vasse from 2018 onwards and, while *Ruppia polycarpa* continued to be the most dominant macrophyte species, density of *Ulva* was consistently higher throughout all sites (Figure 19a). *Stuckenia* and *Ruppia megacarpa* were also more widespread than in 2017. Relatively high density of *Lamprothamnium* occurred in sites 23 and 26 in 2019.

Results of ANOSIM indicated a significant difference in plant community composition in the upper Vasse in spring of 2017 compared with other years (R>0.5), but plant communities were similar from 2018-2020. In comparison to 2017, the community in 2018 differed due to higher density of both *Ulva and R. polycarpa* (47%, SIMPER); and in 2019 due to higher *Ulva* and *Lamprothamnium* density (44%). There was a substantial difference between 2017 and 2020 (R=0.813, p=0.03), when higher *Ulva* density contributed greatly to dissimilarity (43%).

Growth of *Ulva* in the upper Vasse was significantly higher in 2018-2020 compared with 2017 in terms of both plant density (PVI, Figure 19a) and cover (Figure 7a). Average density in 2017 was less than 1%, but was 10-16% in 2018-2020. ANOVA multiple comparisons found a significant difference between 2017 and each other year (p<0.01), but no significant difference within years 2018-2020. Increased plant density has been due to greater cover rather than increased height of plants, which has remained similar across the monitoring period at around 0.2m (Figure 8e). Observed cover of *Ulva* has increased significantly (p<0.01) from an average of 1.7% in 2017 to 40-63% in subsequent years and up to 100% observed at transect points in 2020. Despite this increase in *Ulva* growth, cover and density of macrophytes has not declined, with *R. polycarpa* and *A. cylindrocarpa* growing through the *Ulva*.

Lower Vasse Estuary

In the lower Vasse, total density was also higher from 2018-2020 than in 2017 (Figure 19b), due to higher density of macrophytes, particularly *Stuckenia*. *Stuckenia* and *Ulva* were the dominant species throughout in 2017, 2018 and 2020, and additionally *Ruppia (both species)* in 2018 and 2019. *Lamprothamnium* had relatively high density

at the more upstream sites 19 and 20 in 2019 (Figure 19b); adjacent to upper Vasse sites 23 and 26 which also had peak *Lamprothamnium* density at this time.

ANOSIM found spring plant communities in the lower Vasse differed in 2017 compared with all other years (R≥0.46), with some separation between 2019 and 2020 (R=0.37) and no significant differences between 2018 and 2019 or 2020. Year to year variation was not related to any consistent change in species composition but was related mainly to variation in growth of *Stuckenia*. High separation between spring plant communities of 2017 and 2018 (R=0.97) was due to much higher density of *Stuckenia* in 2018, accounting for 60% of dissimilarity (SIMPER, Figure 19b). Difference in 2019 and 2020 was due to lower *R. polycarpa* and higher *Stuckenia* density (50%).

Upper Wonnerup Estuary

Lamprothamnium is an important species throughout the upper Wonnerup Estuary, also extending into the lower region (Figure 19c). It forms widespread dense meadows of approximately 30cm height and often with 100% cover (Figure Figure 20). Both *Ruppia* species occur in this region, usually at low densities. Higher Ruppia density in 2017 and 2018 was mainly due to more widespread growth of *R. polycarpa* in the most upper sites 5 and 6. *Althenia* is usually patchy in the upper Wonnerup, but it was more widespread in 2019, both in this region and in parts of the lower Wonnerup. Stuckenia is present in the upper Wonnerup, generally found as small isolated patches. Macroalgae did not growth consistently in the upper Wonnerup during the monitoring period: *Cladophora* was widespread in 2017 and *Ulva* had relatively high density at site 8 in 2018 and 2020, while no macroalgae was observed in 2019 in this region.

Plant community composition was relatively stable in the upper Wonnerup, with most year-to-year comparisons showing insignificant dissimilarity. 2019 differed to 2018 (R=0.57, p=0.03) and, to a lesser extent, 2017 2(R=0.38, p=0.03). SIMPER attributed this dissimilarity to lower *R. polycarpa* density in 2019 but also to varying density of *Lamprothamnium* between years. Lamprothamnium density was higher in 2019 and 2020, owing to greater cover, while cover of macroalgae was lower (Figure 7).

Lower Wonnerup

Mean plant densities in the lower Wonnerup indicate a dominance of *Ruppia* and consistent occurrence of *Lamprothamnium* and *Stuckenia* in the lower Wonnerup estuary (Figure 19c). However, this region is morphologically variable, which influences plant communities: site 15 occurs in a narrow channel area, with a deep thalweg on the eastern side that retains permanent water. Sites 13 and 14 also retain a permanently wet channel but are shallower and include larger extents of seasonally dry shallow sediments. Site 11 is located on a wider part of the estuary without a distinct channel.

Deeper channel environments in this region are dominated by *R. megacarpa* and *Stuckenia*, often with extremely high density as they are canopy-forming species reaching the water surface (Figure 21). These species, and also *R. polycarpa,* do extend upstream but generally with decreasing density. *Lamprothamnium* is more common in shallower parts and increases in density moving upstream. *Althenia* occurs mainly in the eastern sections of sites 13 and 14 in most years but is rarely found at other sites.

These observable differences were reflected in ANOSIM analysis, with the lower Wonnerup the only ecological region to have a significant difference in plant community composition among sites (R=0.64, p=0.001). Pairwise tests showed this was due to very high dissimilarity between site 11 and 14 (R=0.88, p=0.03); 11 and 15 (R=0.98, p=0.03) and 13 and 15 (R=0.92, p=0.03). SIMPER attributed dissimilarity mainly to differing densities of filamentous algae, *Lamprothamnium* and *R. megacarpa* between sites Site 11 had higher *Lamprothamnium* and lower Filamentous algae than site 13 (accounting for 51% dissimilarity) and 14 (57%). Higher filamentous algae and *R. megacarpa* in site 15 accounted for most dissimilarity with both site 11 and 13 (67, 68%). Also in contrast to other regions, overall dissimilarity between years was insignificant in the lower Wonnerup (R=-0.026, p=0.5).
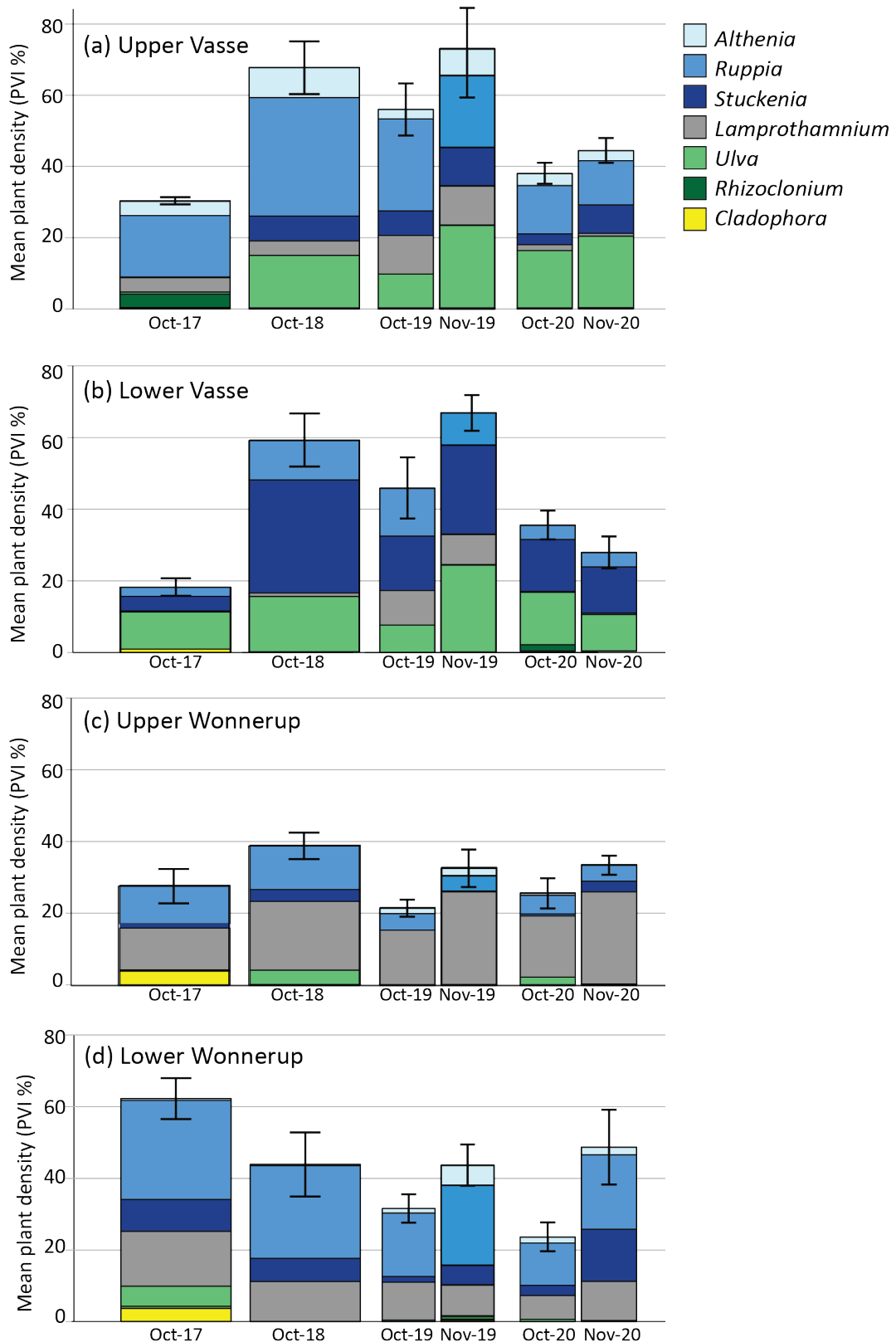
Figure 18. Aquatic plant community composition in spring for each year of sampling (mean PVI for each species, error bars +/- standard error total PVI). Note two spring sampling occasions in 2019 and 2020.

Figure 19. Extensive Meadow of Lamprothamnium macropogon in the upper Wonnerup Estuary with 100% cover (R. Paice).
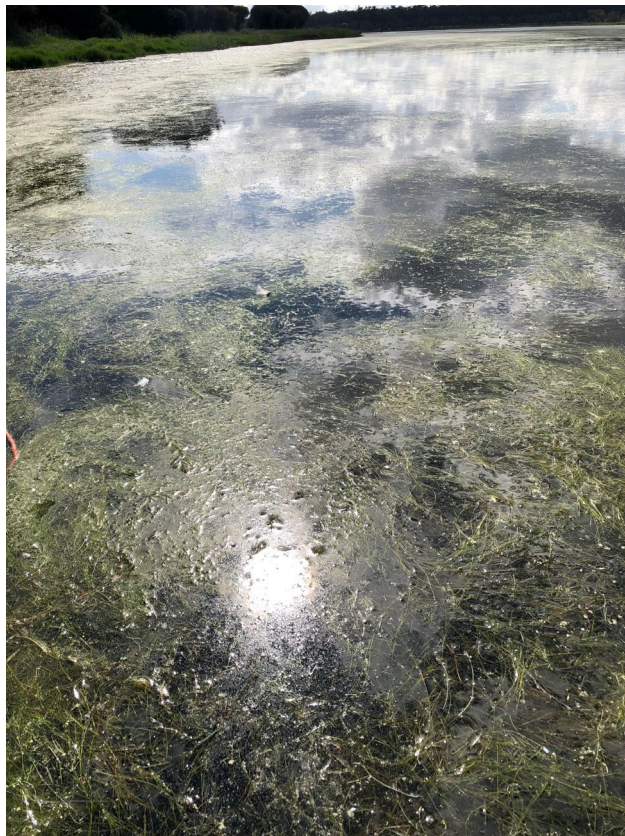
Figure 20. Large dense meadow of Ruppia megacarpa with Stuckenia pectinata in the deep channel at lower Wonnerup site 15, demonstrating 100% PVI.

Context of long-term monitoring

Aquatic plant monitoring in the Vasse-Wonnerup wetland during spring since 2006 provides a good historical context for assessing changes that have been observed during the IEM program (Figure 22). This sampling has included the same sites as IEM (though not transects) plus additional sites. Analysis of biomass data from 2006-2016 found high site fidelity, with each region characterized by different plant communities of aquatic plant communities as follows (Chambers et al. 2017):

- Upper Vasse Estuary: *Ruppia polycarpa* and *Althenia cylindrocarpa* dominance.
- Lower Vasse Estuary: *Ulva* dominance with *Ruppia megacarpa* and *Stuckenia pectinata* (since 2014) also present.
- Upper Wonnerup Estuary: *Lamprothamnium* and *Ruppia polycarpa* dominance.
- Lower Wonnerup Estuary: *Ruppia megacarpa* dominance in the very lower Wonnerup; consistent but small amounts of *Althenia*. Macroalgal growth in some years.

In the upper Vasse, the increase in *Ulva* PVI since 2018 is also evident from biomass sampling (Figure 22). While there has been growth of macroalgae in this region in some previous years (e.g. 2007), it has not been recorded at the scale and consistency seen from 2018 to 2020. Prior to this, the upper Vasse was variably dominated by *R. polycarpa* and *Althenia cylindrocarpa* with macroalgae in some years only, and at much lower biomass than macrophytes. Linear contrast analysis found a significant ($p<0.001$) increasing trend in *Ulva* biomass in the upper Vasse region with post hoc contrasts demonstrating a significant difference in mean *Ulva* biomass between each of years 2018-2020 with each other year except 2007. Importantly for conservation of macrophytes, although *Ulva* has increased in this region, this has not coincided with a decrease in macrophyte density (Figure 22). Observations during sampling were that the *Ulva* grows as a layer within the macrophytes.

Higher growth of *Stuckenia* in the lower Vasse region since 2014 has continued in recent years, apparent from both PVI and biomass results. While previously described as having Ulva dominance, this region appears to be in a status of co-dominance of macrophytes and macroalgae, with generally lower biomass of Ulva since 2012. In the Wonnerup Estuary, macroalgae was more common in spring from 2006 to 2010 and has since occurred inconsistently at low biomass and also at low PVI.
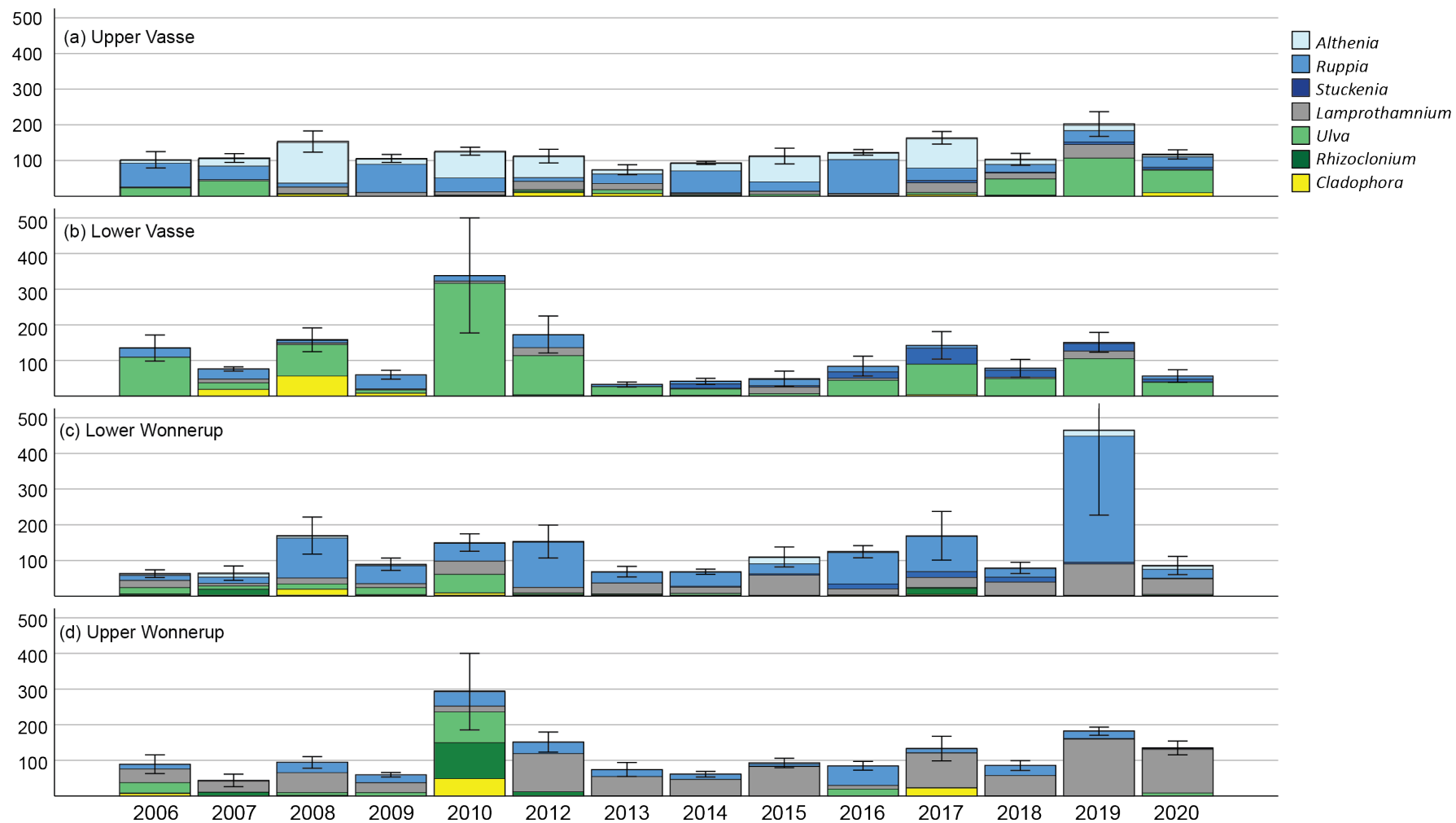
Figure 21. Aquatic plant community species composition from long-term biomass sampling in the Vasse-Wonnerup wetlands from 2006 to 2020. Data from Murdoch University spring sampling.
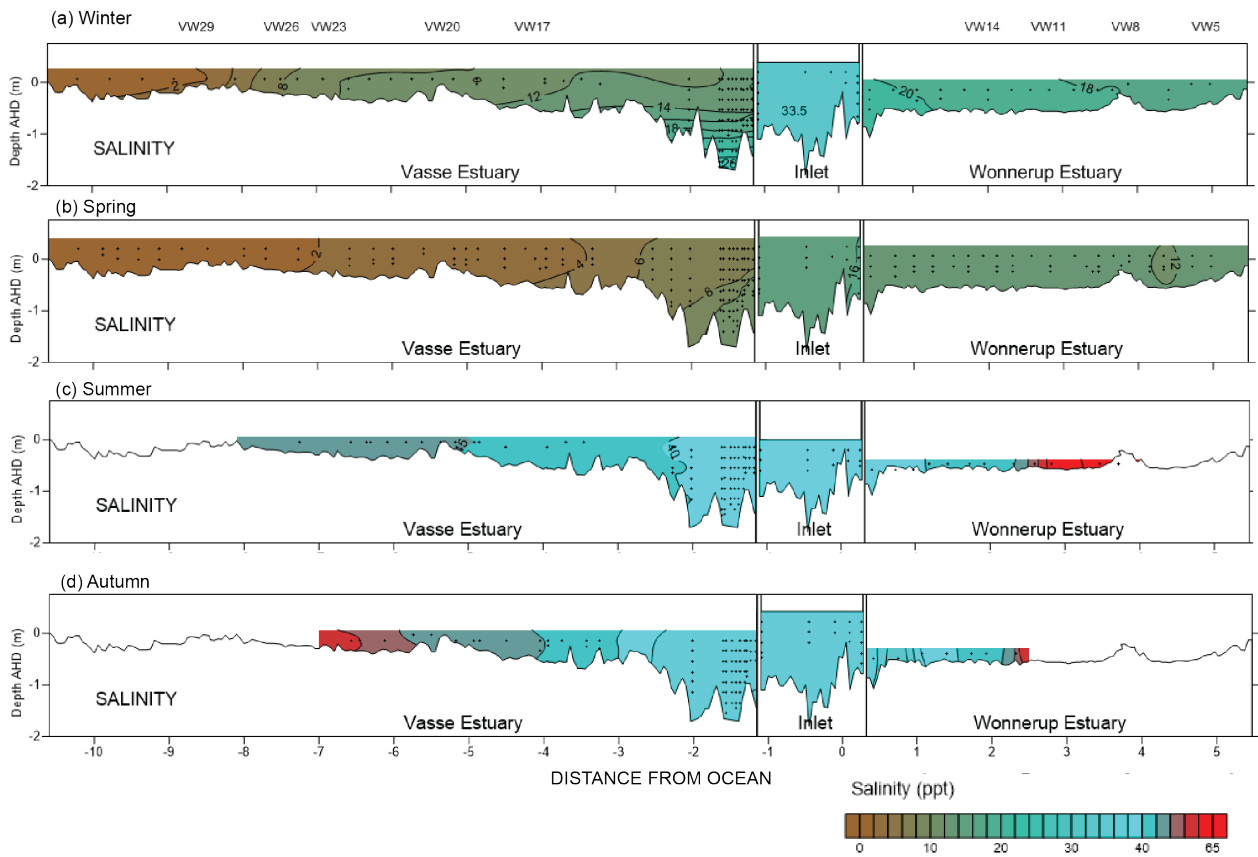
Figure 22. Salinity profile contour plots and water levels for the Vasse-Wonnerup wetlands throughout an annual cycle from July 2019 to March 2020. Plots provided by DWER.

Relationships between plant communities and environmental variables

Key findings:

Dominant species within the ecological regions correlated with environmental variables as follows:

- *Ruppia polycarpa* in the upper Vasse was associated with lower TN, lower sediment organic content and shallower waters.
- *Ulva* and *Stuckenia pectinata* in lower Vasse were associated with lower TN and salinity, and higher sediment organic content.
- *Lamprothamnium* in the Wonnerup Estuary was associated with higher salinity and higher TN.

On a seasonal basis, aquatic plant communities are clearly linked to natural changes in abiotic factors of water level, salinity, and temperature. This annual cycle of environmental conditions is shown clearly by the depth and salinity contour plots in Figure 23. This drives the seasonal growth pattern of aquatic plants: commencing in winter, peaking in spring, and senescing in summer. Within each region, the range in these conditions and magnitude of seasonal change varies, contributing to the aquatic plant community structure, depending on the tolerances of different species.

In the lower regions there is less drying and deeper waters, supporting canopy-forming macrophyte species such as *Stuckenia pectinata* and *Ruppia megacarpa* with potential for perennial occurrence (as seen in the lower Wonnerup). Both these species are less tolerant of elevated salinity and drying (Brock 1982, Borgnis and Boyer 2016). *Stuckenia pectinata* was first observed in the system in 2014 when conditions had been deeper and fresher for several years. The upper regions are shallower, the wide flat bathymetry leads to greater areas of drying and evapoconcentration of salts in summer and autumn, and seasonal drying also allows for consolidation of sediments. Smaller annual species are more prevalent in these upper regions and different salinity ranges in the two estuaries further influences characteristic species. In the upper Vasse, macrophytes *Althenia cylindrocarpa* and *Ruppia polycarpa* are well adapted to conditions extensive drying and extreme fluctuations in salinity, and germinate most effectively in freshwater conditions (Vollebergh and Congon 1986) which are experienced in winter and spring. The charophyte *Lamprothamnium macropogon* is adapted to salinity (Casanova 2013) and dominates shallow areas of the Wonnerup Estuary. Although germination of *Lamprothamnium* is initiated by rainfall, subsequent growth requires saline conditions (Garcia and Chivas 2004), which are present throughout the year in the Wonnerup while the Vasse remains fresh until late spring.

Changes in hydrology and catchment development also influence plant communities, most importantly: alterations to tidal connectivity from installation of floodgates; and nutrient enrichment from diffuse rural and urban catchment sources. These

anthropogenic factors affect the hydrologic and salinity regime, nutrient availability and sediment characteristics, potentially leading to a change in dominant species, with nutrient enrichment favouring less desirable macroalgal species (Cloern 2001, Viaroli et al. 2008).

BIO-ENV performed on spring data resulted in low overall matching between species assemblage patterns and environmental variables: the maximum correlation was 0.315, using variables of salinity, sediment organic content, and depth. The CAP routine applied to plant density and water quality data was successful at determining axes from water quality variables that best explained the plant community data; and applying the region factor to the ordination plot resulted in reasonable separation (Figure 24). Strong correlations were found of 0.81 for the CAP1 axis, most strongly influenced by TN (eigenvector value = -0.665); and 0.76 for the CAP2 axis, most strongly influenced by decreasing LOI. Spearman correlation vectors indicated separation of regional plant communities in the two estuaries by lower TN in the Vasse; the upper Vasse by shallower conditions and the lower Vasse by higher sediment organic content (Figure 24).

This outcome is consistent with previous regional CAP separation for water quality and plant density: higher salinity and TN in the Wonnerup Estuary and *Lamprothamnium* density; lower TN and salinity, and higher sediment organic content in lower Vasse and *Ulva* and *Stuckenia* density; and lower TN, salinity, sediment organic content and depth in the upper Vasse and *R. polycarpa* density.
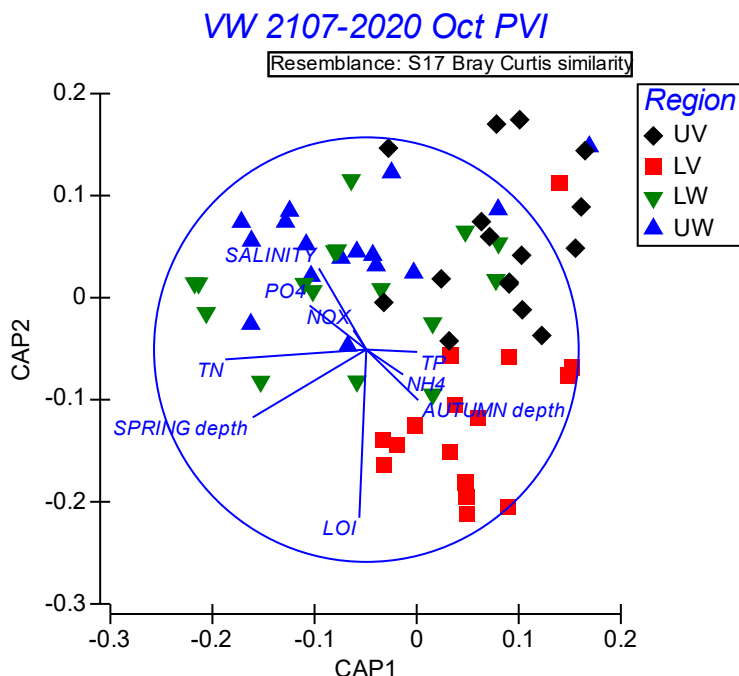

Figure 23. CAP ordination plot relating aquatic plant community assemblage to environmental variables.

Comparison of plant density and biomass methods

Spring monitoring of plant density for the IEM program corresponded with annual biomass sampling in 2018-2020 (late October - early November). In 2017, plant density monitoring occurred in late October and biomass sampling was done in late November; with additional sampling on each occasion with the alternative method to provide additional data for comparison of the results.

Face value comparison of data from the two methods (Figure 25) shows similarity in plant species composition in the defined ecological regions, but some differences in proportional contribution to total PVI and biomass. This is not surprising for two main reasons. Firstly, the methods use different spatial extents: with PVI sampling extending across transects the width of the estuary at each of 16 sites; and biomass replicates (5) at a greater number of site points (26) distributed longitudinally in the system. Secondly, plant species vary in water content and growth habit, hence dry weight does not equate to plant density similarly for different taxa.

There was generally a lower proportion of *Ulva* reflected in PVI data compared to biomass results in the Vasse Estuary, where it was more common (Figure 25). *Ulva* tends to occur in dense layers (author observation), with a greater amount of plant material per unit volume than macrophytes, so it follows that biomass of a similar PVI would be greater for *Ulva*. Proportion of macrophytes and charophytes in the Wonnerup Estuary were more consistent between the two methods.

Correlation and regression analysis compared biomass results with PVI from both corresponding site points data and transect means, using data pooled across the four years of sampling. Regression was performed for each main taxa, with *Rhizoclonium* and *Cladophora* grouped as filamentous algae (owing to less data), and for macrophyte and macroalgae groupings. For site point data, regression was repeated using biomass and PVI as both dependent and predictor variables. For transect data comparisons, PVI was selected as the dependent variable in consideration of the potential for conversion of long-term biomass data to PVI.

Site-based PVI and biomass data had significant positive correlations (R) for all taxa and for macrophyte and macroalgae groups and regression showed significant linear relationships between the data sets (Table 3). The amount of variation explained by the linear model (R^2: coefficient of determination) was over 30% for Ruppia and Stuckenia, 58% for Althenia, 65% for Lamprothamnium and total macrophytes and 43% for Ulva. Scatterplots did indicate linear relationships, but with considerable variation around the regression line (Figure 26) and very broad 95% confidence intervals in most cases (Table 3).

Correlation was strongest and the linear model most reliable for Lamprothamnium. This species is generally low growing and robust and biomass cores usually capture whole plants, providing a good reflection of plant density. Filamentous algae had a

weaker positive correlation than other groups, although significant (R=0.36, P=0.05), and PVI explained only 13% of variation in biomass (Table 3).This may reflect its presence both dispersed throughout the water column and as dense accumulations on the surface or bottom, resulting in more variable biomass per volume inhabited. For macroalgae combined, correlation was moderate and significant, reflecting the stronger relationship for Ulva and the weaker relationship for filamentous algae.

Regression of PVI transect data against biomass data found significant correlations for Ruppia, total macrophytes, *Lamprothamnium*, *Ulva* and total macroalgae (Table 4). Interestingly, stronger relationships were identified for *Ulva* and total macroalgae. For *Stuckenia* and *Althenia*, the lack of correlation may reflect the patchy distribution of these species in the system, while *Ruppia*, *Lamprothamnium* and *Ulva* are more widespread within regions. Confidence intervals were also more constrained for all significant relationships than for site point analysis and scatterplots showed reasonable fit (Figure 27), providing more confidence for prediction of transect PVI from biomass for these taxa and groups.
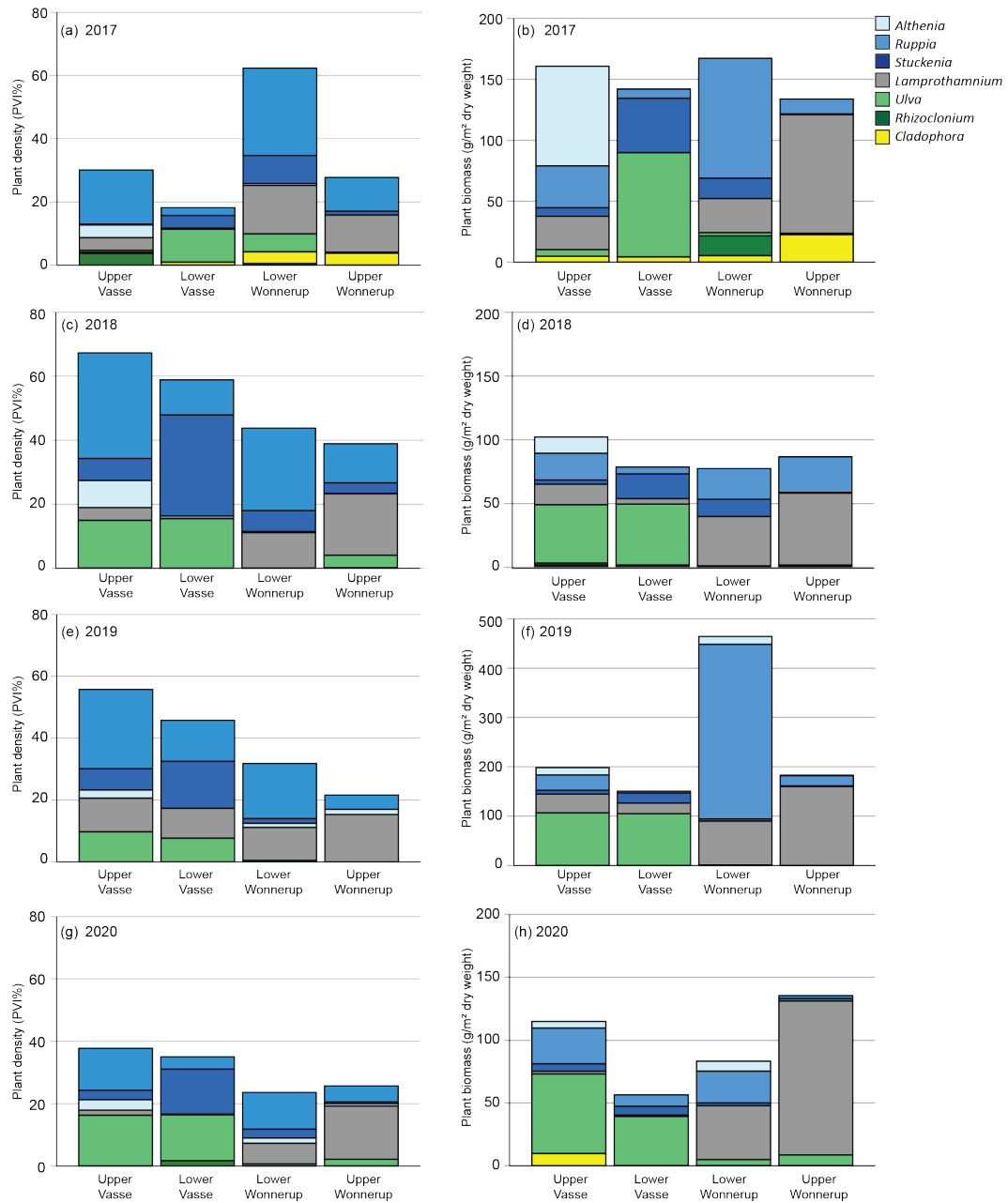
Figure 24. Comparison of results for PVI sampling of transects (transect means) and biomass sampling (site means) in spring in 2017 (a, b), 2018 (c, d), 2019 (e, f) and 2020 (g, h). In 2017, plant density was measured in October biomass in November; in other years density and biomass were sampled on the same day.

Table 3. Results of correlation and regression analysis for PVI and biomass from spring 2017-2020, for corresponding site data.

Species	R	R^2	F-ratio	P	Model parameter	Biomass as dependent		PVI as dependent	
						Value	95% CI	Value	95% CI
Ruppia spp.	0.570	0.324	38.4	<0.001	Intercept	0.210	-0.11-0.53	0.68	0.51-0.84
					Slope	0.819	0.56-1.08	0.40	0.27-0.52
Stuckenia. pectinata	0.595	0.355	18.7	<0.001	Intercept	0.234	-0.16-0.33	0.674	0.73-0.98
					Slope	0.630	0.33-0.93	0.563	0.30-0.83
Althenia cylindrocarpa	0.689	0.474	15.3	0.001	Intercept	0.416	-0.08-0.90	0.222	-0.19-0.64
					Slope	0.859	0.40-1.32	0.552	0.26-0.85
Total macrophytes	0.637	0.65	90.1	<0.001	Intercept	0.142	-0.18-0.46	0.751	0.58-0.92
					Slope	0.876	0.66-1.10	0.464	0.35-0.58
Lamprothamnium macropogon	0.763	0.65	90.3	<0.001	Intercept	0.340	-0.10-0.58	0.211	0.03-0.39
					Slope	1.051	0.83-1.27	0.553	0.44-0.67
Ulva spp.	0.657	0.43	42.4	<0.001	Intercept	0.544	-0.31-0.77	0.110	-0.10-0.32
					Slope	0.860	0.60-1.12	0.501	0.35-0.66
Filamentous algae	0.356	0.13	4.2	0.050	Intercept	0.639	0.32-0.96	0.264	-0.01-0.53
					Slope	0.496	0.001-0.99	0.255	0.001-0.51
Total macroalgae	0.530	0.28	27.3	<0.001	Intercept	0.709	0.47-0.95	0.232	0.02-0.44
					Slope	0.721	0.45-1.00	0.389	0.24-0.54

Table 4. Results of correlation and regression analyses for PVI and biomass data from spring 2017 to 2020, for site point biomass data and transect-based PVI data. (Dependent variable = PVI).

Species	R	R^2	F-ratio	P	Model parameter	Value	95% CI
Ruppia spp.	0.594	0.353	38.7	<0.001	Intercept	0.694	0.56-0.83
					Slope	0.353	0.24-0.47
Stuckenia. pectinata	0.268	0.072	2.6	0.115	Intercept		
					Slope		
Althenia cylindrocarpa	0.299	0.089	2.5	0.130	Intercept		
					Slope		
Total macrophytes	0.570	0.324	36.0	<0.001	Intercept	0.938	0.80-1.07
					Slope	0.310	0.21-0.41
Lamprothamnium macropogon	0.757	0.574	76.7	<0.001	Intercept	0.154	-0.04-0.24
					Slope	0.535	0.41-0.66
Ulva spp.	0.724	0.525	57.4	<0.001	Intercept	0.241	0.04-0.44
					Slope	0.501	0.37-0.63
Filamentous algae	0.408	0.166	3.39	0.083	Intercept		
					Slope		
Total macroalgae	0.735	0.541	68.3	<0.001	Intercept	0.277	0.10-0.45
					Slope	0.485	0.37-0.60
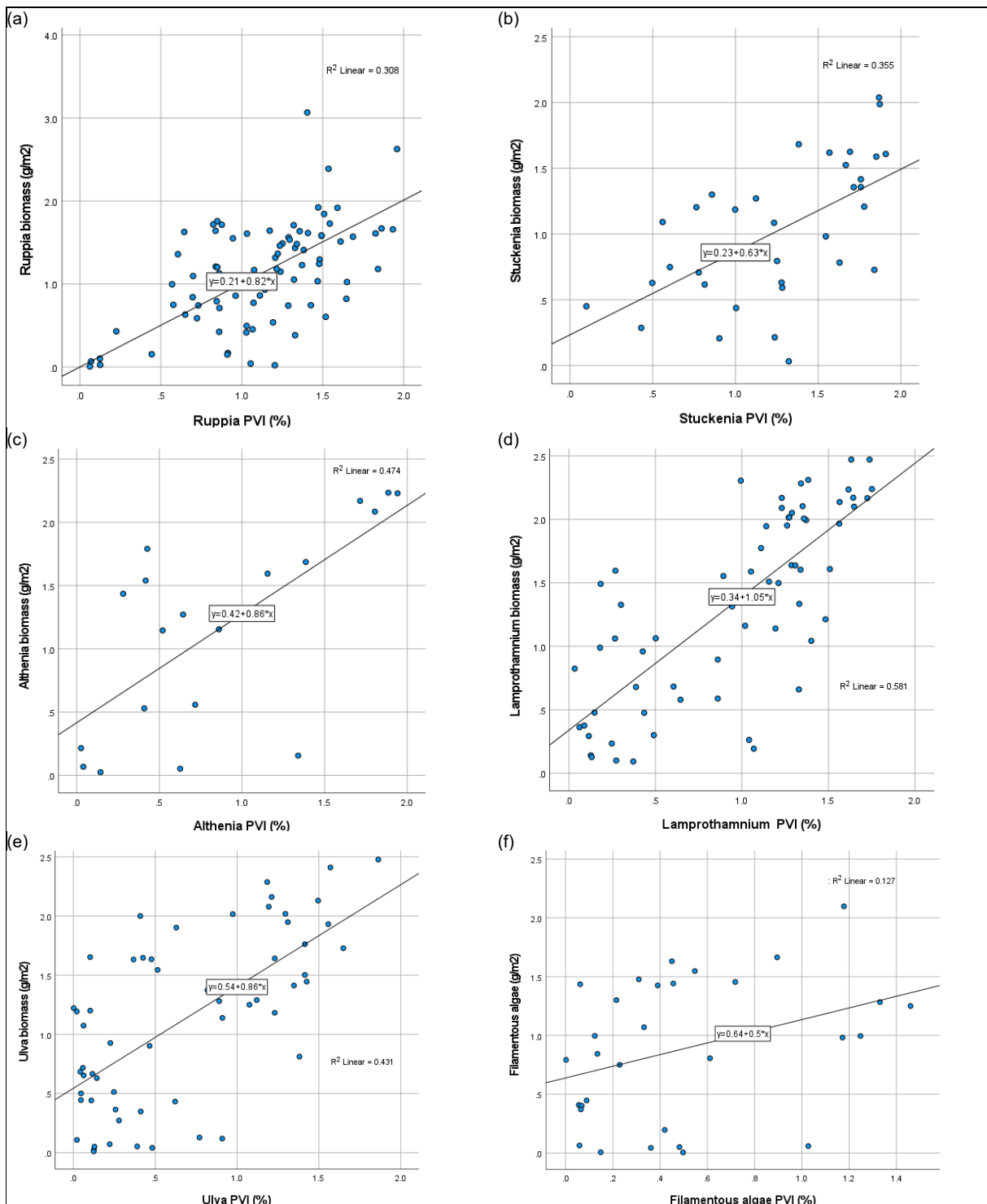
Figure 25. Scatterplots of biomass and PVI data from site points for main plant taxa in the Vasse-Wonnerup system 2017-2020 showing linear regression line and coefficient of determination (R^2). PVI data corresponds to the transect point where biomass cores were collected. Transformed data presented ($log_{10}(x+1)$.
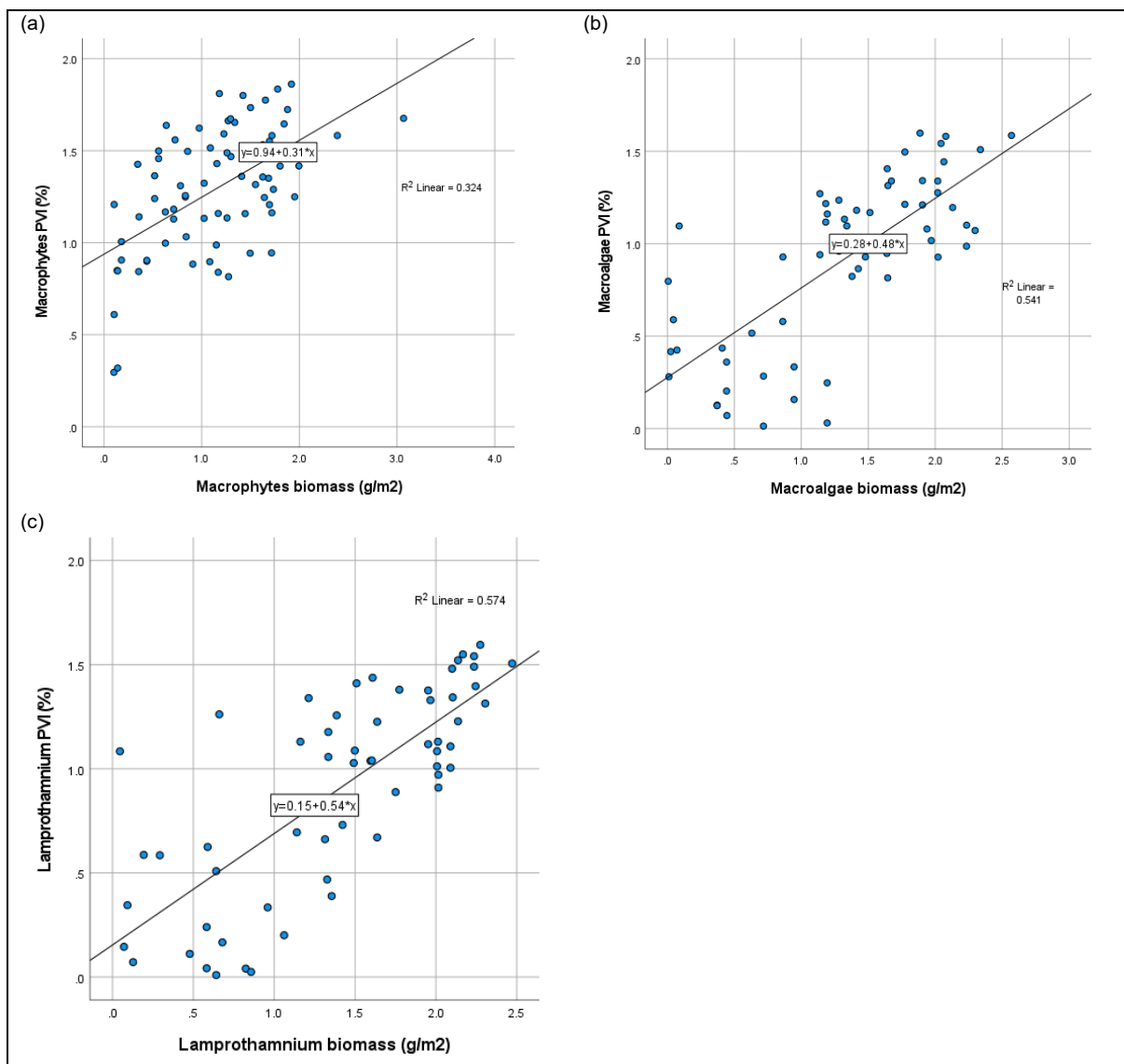
Figure 26. Scatterplots of transect PVI against site point biomass for key aquatic plant groups in the Vasse-Wonnerup system 2017-2020 showing linear regression line and coefficient of determination (R^2).

Discussion

This report presents the results of aquatic plant monitoring for the IEM Program over four complete years, from winter 2017 to autumn 2021, provides a summary of water quality and water level data, and examines the relationships between plants and environmental variables.

Importantly, the IEM program was developed to better understand the relationship between seasonal water regime (water quality and water levels) and the ecological values that support waterbirds on the Vasse Wonnerup wetlands. Aquatic plant monitoring has indicated a change in conditions in the upper Vasse Estuary that favour growth of the macroalga *Ulva,* and a continuation of increased growth of macrophytes in the Lower Vasse observed prior to the IEM program.

Spatial and temporal variation of plant communities

Submerged aquatic plants throughout the Vasse-Wonnerup wetlands exhibit strongly seasonal growth patterns, with plant communities forming highest density and biomass in spring and senescing in summer as water levels drop. Seven key taxa occur in the system, including four species of macrophytes, one species of charophyte and three genera of macroalgae. Distinct plant assemblages occur in four regions, although there is overlap between the upper and lower Wonnerup, with the following dominant species apparent from both IEM and long-term monitoring (Chambers et al. 2017). These can be considered a baseline for future assessment of changes in aquatic plant communities in the Vasse-Wonnerup:

- Upper Vasse Estuary: *Ruppia polycarpa* and *Althenia cylindrocarpa* dominance.
- Lower Vasse Estuary: Co-dominance of macroalgae *Ulva* and macrophytes *Ruppia megacarpa* and *Stuckenia pectinata* (since 2014) also present.
- Upper Wonnerup Estuary: *Lamprothamnium* and *Ruppia polycarpa* dominance.
- Lower Wonnerup Estuary: *Ruppia megacarpa* dominance in the channel of the Wonnerup to the gates; *Ruppia* dominance in deeper channel habitat more downstream; *Lamprothamnium* and *Ruppia polycarpa* in the mid-estuary, similar to the upper region.

The upper Vasse Estuary has historically been in a macrophyte-dominated state, dominated by *Ruppia polycarpa* and *Althenia cylindrocarpa,* and *Lamprothamnium* is also common throughout and *Stuckenia* is distributed patchily. *R. polycarpa* had the highest density of all species from 2017 to 2019, and its occurrence associated with lower organic nitrogen, lower sediment organic content and shallower waters. *Ulva* had greater density than *R. polycarpa* in 2020.

The IEM program has identified significantly higher and widespread growth of *Ulva* in the upper Vasse since 2017. This change is also evident from long-term biomass monitoring, which found growth of macroalgae in the upper Vasse in some years, but

not at the scale and consistency seen from 2018 to 2020 (Chambers et al. 2017). The upper and lower regions of the Vasse have become statistically more similar, with comparable *Ulva* density in the two regions from 2018 to 2020. Fortunately, although macroalgal growth was higher, there was not a concomitant decrease in growth of key species *R. polycarpa* and *A. cylindrocarpa*. Field observations in this region were of a layer of *Ulva* growing over the sediments, with macrophytes growing through this (Figure 28).

There has been variation in plant assemblages in the lower Vasse region in terms of relative density of different species, but no consistent changes over time. Historical biomass monitoring suggested this region was in a transitional state with co-dominance of macrophytes and macroalgae, which has continued in recent years. There has been with variable density of key macrophyte species and greater proportion of macrophytes than macroalgae by density, but a greater proportion of macroalgae by biomass.

Plant communities in the Wonnerup Estuary were relatively stable for the duration of IEM monitoring, with variation in overall density but similarity in proportions of species. *Lamprothamnium* is the dominant species in most of the Wonnerup Estuary, forming widespread dense meadows in spring. This species was associated with high salinity and high organic nitrogen, and is tolerant of generally higher nutrient concentrations in the Wonnerup. It is a charophyte algae that functions ecologically as a macrophyte, providing important structural habitat and a food source for Black Swans (Kissane 2019), and its ongoing dominance in the Wonnerup is desirable.

The Wonnerup ecological regions defined for the IEM program did not reflect species distribution owing to variation in site morphology within the lower region. L*amprothamnium* extended downstream of the upper region boundary and the lower two sites included deep channel habitat Wonnerup dominated by *R. megacarpa*, with *S. pectinata* and L*amprothamnium* also common. This channel habitat supports extremely dense macrophyte canopies in spring and perennial growth in some years.

Environmental variables related to plant community changes

The principal factor that has changed since 2017 and likely to contribute to the increase in *Ulva* in the upper Vasse region is the management of water levels in the Vasse Estuary through additional seawater inflow during summer and autumn. This has led to consistently higher autumn water levels relative to historical levels in the Vasse Estuary since 2018, which has translated to reduced drying of the upper region. DWER has reported an increase in autumn water levels of approximately 10cm, resulting in wetting of upper estuary areas that have previously been dry after mid-January (Kalnejais 2020). The seawater inflow trial has substantially altered water quality in the Vasse Estuary exit channel, but water quality in the main estuary waters has not changed significantly. However, this change in water level was sufficient to drive change in plant assemblages in the upper Vasse.

Seasonal drying has transformative effects on plant life cycles and sediment characteristics (Boulton et al. 2014). Many wetlands on the Swan Coastal Plain naturally experience seasonal drying, which results in consolidation of sediments. A change to permanent inundation removes potential for consolidation and the very high nutrient levels and warm temperatures are conducive to algal growth throughout summer and autumn (Davis et al. 2010). Greater algal growth and decay and lack of consolidation contribute to organic, flocculent sediments (Volkman et al. 2008) that are less favourable to aquatic plants due to poorer anchorage (Schutten et al. 2005) and growth inhibition (Barko and Smart 1983, Conway 2016). Lack of drying would also result in deeper conditions earlier in the year, as the estuary does not need to saturate prior to filling, which may also favour macroalgae and affect germination and growth of macrophytes.

The presence of *Ulva* can induce a feedback mechanism for its increasing dominance, as its decomposition can exacerbate build-up of poor-quality organic sediments and anoxia, creating more hostile conditions for macrophytes (de Wit et al. 2001, Viaroli et al. 2008, Raun et al. 2010). Increased growth of macroalgae can indicate a transition to a phytoplankton-dominated regime, often occurs via a pathway of macroalgal blooms in coastal lagoons (Viaroli et al. 2008, Pasqualini et al. 2017). High sediment organic content was associated with *Ulva* in the lower Vasse and, although monitoring has not indicated an increase in sediment organic content in the upper Vasse to date, there is a risk of increasing macroalgal growth in the upper region leading to a deterioration of sediment quality. While spring macrophyte species in the upper Vasse have been maintained despite the increase in growth of Ulva, there is a risk that they will be lost from this region.

Future sampling

Future monitoring should align with requirements for assessment of "Limits of Acceptable Change" for the Vasse-Wonnerup system, which are under review (DBCA 2019). The outcomes of both the IEM program and long-term biomass monitoring (Chambers et al. 2017) should inform this review.

The use of the percent volume inhabited (PVI) for seasonal sampling was an efficient method, providing data within days of sampling at a much lower cost than biomass sampling. Communication of PVI results conveys an easily relatable image of plant density, and data appears to be less variable than biomass. Furthermore, the sparse plant communities encountered during summer and autumn may not have been well described by the biomass method. Notwithstanding the usefulness of the PVI method, particularly for more frequent sampling, the long-term (15-year) dataset from annual biomass sampling in spring is a more quantitative method and provides a critical and comparable historical record of estuarine health from an aquatic plant perspective and

would be valuable to continue. Further consideration could also be given to species cover as a useful rapid-assessment metric.

Monitoring aquatic plants on a seasonal basis has provided important information on plant life cycles throughout the system. However, as this study confirmed, spring sampling during peak biomass when plant communities are established provides the most valuable information on distribution and assemblage patterns and the greatest potential to detect change. Future monitoring should include both methods (PVI and biomass) during the same sampling trip in spring only each year, preferably in November when peak biomass occurs. This will provide both rapid assessment (PVI) and add to the long-term quantitative biomass for rigorous future analysis of trends.

During summer and autumn when plant growth is low and the wetlands support large numbers of birds, it would be interesting to understand the food web in the absence of large plants. The microphytobenthos (microalgae and other microorganisms) can form extensive mats on the surface of the sediments during these times (Figure 29) and may support bird diets. Sampling and analysis of this component would therefore be useful.
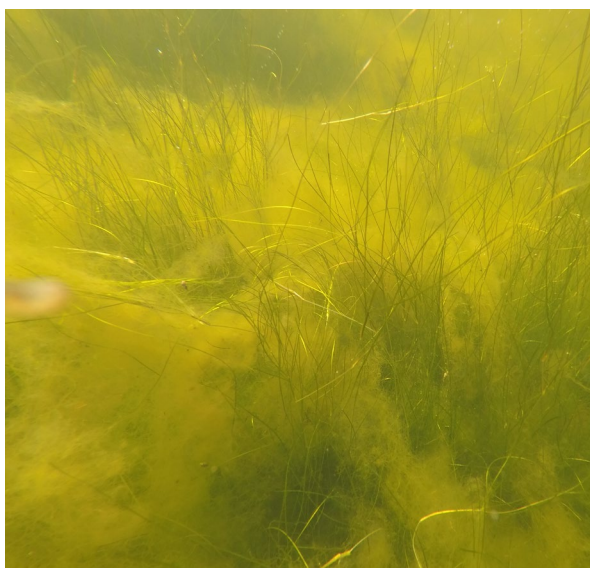

Figure 27. Co-occurrence of Ruppia polycarpa *and* Ulva *in the upper Vasse region.*
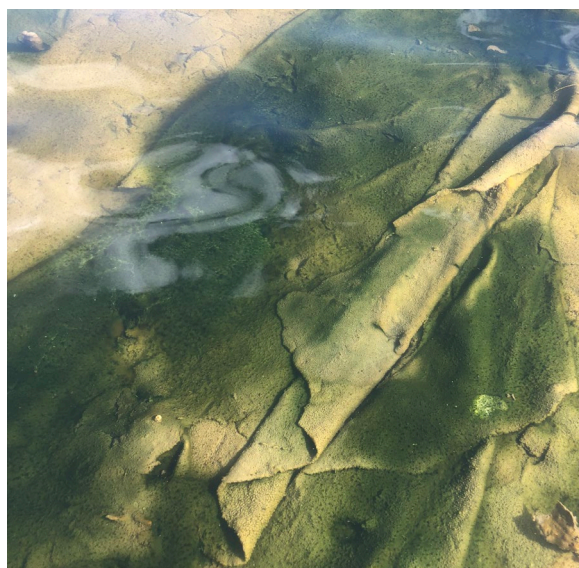

Figure 28. Benthic mat in the Vasse Estuary. Photo: R. Paice.

Management implications

The relationships between aquatic plants and environmental conditions are complex, but monitoring of species distribution and community composition is useful in tracking the effects of multiple interacting factors on ecological health. The diversity and abundance of waterbirds on which the Ramsar nomination for the Vasse-Wonnerup wetlands is supported by aquatic plant communities through the provision of direct and indirect (via food web) food resources, habitat and nesting materials, and so maintaining these communities is critical.

This monitoring and assessment approach has identified a deterioration in the ecological health of the upper Vasse Estuary, indicated by significantly higher and more widespread growth of the macroalga *Ulva* since 2018. This indicates a risk of a shift in this region from what has historically been macrophyte-dominated, supporting healthy meadows of *R. polycarpa and A. cylindrocarpa,* to macroalgae dominance.

Additional seawater inflow during summer and autumn creating consistently higher autumn water levels and reducing the extent of drying is likely to be a key factor contributing to this change. While the upper Vasse is still environmentally distinct, the dissimilarity in plant communities has decreased. This change has the capacity to cause future loss of seagrasses, *Ruppia polycarpa* and *Althenia cylindrocarpa* (which are adapted to seasonal drying) which has comprised the highest quality seagrass ecosystem in the Vasse Wonnerup wetland complex. If the new management conditions are maintained, the likely outcome is that biodiversity will be lost, and the upper and lower Vasse Estuary will share the poor condition exhibited in the lower Vasse for over a decade.

Other plant communities may be sensitive to depth and salinity changes, and also need to be considered in ongoing management of the surge barriers. For example, the establishment of *Stuckenia* in the lower Vasse has been linked to deeper, fresher conditions in spring since 2014; while these conditions may restrict *Lamprothamnium* growth in the Wonnerup Estuary (Chambers et al. 2017).

It is also essential to recognise that while environmental factors are important determinants of plant distribution and community composition, the plants themselves can influence these conditions. The presence of macrophytes can buffer nutrient enrichment, maintain oxygen balance, limit turbidity and stabilise sediments; while macroalgae have lower potential for nutrient retention and can exacerbate nutrient enrichment through build-up of poor-quality organic sediments and anoxia (de Wit et al. 2001, Viaroli et al. 2008). In the Vasse-Wonnerup, macrophyte communities appear tolerant of the nutrient-enriched conditions and may be buffering the system against a shift to macroalgal and phytoplankton dominance (Cloern 2001, Viaroli et al. 2008).

Increased summer seawater inflows have helped to address severe water quality issues in the Vasse Estuary exit channel but sustainability of healthy aquatic plant communities needs careful consideration, ss these provide resources for aquatic fauna

and waterbirds and help maintain water quality. The upper Vasse has, up to 2017, supported the highest quality macrophyte meadows in the wetland system. Continuation of this management approach and associated lack of drying in the upper Vasse has potential to increase organic content and flocculant nature of sediments with undesirable outcomes. Management is recommended to enable seasonal drying in the upper Vasse if we are to maintain a healthy ecosystem. Additional research into how altered water regime contributes to changes in the aquatic plant community would further inform decision-making in this complex system. Surge barrier trials allowing drying of the Upper Vasse perhaps every second year could be attempted.

Recommendations

- Ongoing annual spring monitoring of biomass and PVI of aquatic plant communities throughout the Vasse-Wonnerup wetlands at existing sites.

- Ongoing monitoring of environmental factors with increased sampling of sediment characteristics.

- Consideration of analysis of the microphytobenthos in summer-autumn to further understand its ecological role in the system.

- Support further research into the impacts of altered water regimes in the upper Vasse Estuary on the composition of aquatic plant communities there.

- Consideration of surge barrier trials allowing drying of the upper Vasse in some years.

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