Post-flood Intervention Monitoring of Wetland and Floodplain Vegetation & Carp Exclusion Pilot Study Gunbower Forest

August 2015

Fire Flood & Flora

Prepared for the North Central Catchment Management Authority

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NOTE: This report is accompanied by a Technical Addendum which provides further details of data and analyses performed as part of the monitoring program.

EXECUTIVE SUMMARY

Fire, Flood and Flora was engaged by the North Central Catchment Authority to survey and report on sentinel wetland and understorey vegetation and the results of a carp exclusion pilot study in the Gunbower Forest Icon Site autumn 2015.

Wetland and Understorey Vegetation Intervention Monitoring

- The primary aim of surveying the wetland and understorey flora was to assess the impact of 98 GL of environmental water delivered (eFlow) to the forest through the newly constructed Hipwell Regulator during the 2014 winter-spring period. The secondary aim was to assess the condition of the vegetation and report on progress toward Icon Site ecological objectives.
- The full cohort of 15 wetland transects and 110 understorey quadrats that were established in Gunbower Forest were sampled in autumn 2015, and the data were analysed using univariate and multivariate techniques, as well as vegetation condition indicators that were developed in 2014.

Wetlands

- All eleven wetland monitoring sites were inundated by the 2014 eFlow, and classified in the 'Receding' wetland phase class, based on their hydrological condition at sampling in 2015.
- 83 flora species were observed in the wetland sites in 2015. 34% of these species are aquatic (i.e. establish and grow while inundated and do not tolerate sustained drying) and are therefore unlikely to have been observed in the wetlands had the forest not been inundated by the 2014 eFlow.
- Five rare and threatened flora species were also observed in the wetland sites, including large swords of the nationally listed River Swamp Wallaby-grass (*Amphibromus fluitans*) in Reedy Lagoon.
- Detailed analysis of the data suggests the wetland vegetation responded in a similar manner to the 2014 eFlow as it did to previous inundation events between 2005 and 2014. For example, the mean diversity of native species in the Receding wetlands that were surveyed after the 2014 eFlow (2015) was not significantly different to that recorded in Receding wetlands surveyed after earlier inundation events (2005-2014).
- Multivariate analysis of the wetland data highlighted an association between Receding wetlands that were inundated via a regulator and/or small natural inflows and higher covers of native aquatic flora.
- Wetlands with a healthy richness of characteristic Plant Functional Group (PFG) species were only recorded in 2010 (3/6 Receding wetland and 3/5 Dry wetlands) and 2015 (2/11 Receding wetlands) during monitoring (2005-2015).
- The lower number of healthy wetlands in the latter survey suggests that wetland health has not improved between 2005 and 2015 when assessed using the characteristic PFG species richness Index. This implies that the overarching objective for the wetlands has not yet been achieved.
- In summary, it appears the 2014 eFlow provided the appropriate hydrological conditions for aquatic flora species to establish in the wetlands, yet that factors other than hydrology, such as turbidity and carp, maybe influencing the site's species richness.

Red Gum Forests and Woodlands

- A total of 124 plants identified to species level were recorded at the 77 Red Gum monitoring sites in autumn 2015. This list includes seven rare and threatened, and 81 native species.
- Just over a third of the Red Gum monitoring sites were inundated (28/77) to some degree by the 2014 eFlow, yet only two of these sites were mapped as Red Gum with Flood Tolerant Understorey (the rest were Red Gum with Flood Dependent Understorey).
- Red Gum sites that were inundated in 2014 by water delivered through the Hipwell regulator were found to have significantly higher mean native species diversity, and were associated with higher covers of aquatic, amphibious and mudflat species in autumn 2015 than sites that remained dry. This suggests that a diversity of understorey flora characteristic of Red Gum floodplain vegetation were stimulated in areas that were inundated by the 2014 eFlow.
- In 2015, 15.6% of the Red Gum sites were calculated with a healthy richness of characteristic PFGs species, and 16.9% of sites were calculated with a healthy tree canopy cover.
- The 2015 vegetation condition Index results suggest that Red Gum vegetation condition has been enhanced by the 2014 eFlow, and therefore, meets the overarching objective for maintenance of the vegetation type. The average of the two Index scores (16.2%) does not however meet the objective target of 21% of sites in healthy condition.
- In summary, it is therefore reasonable to assume, based on the above results, that the 2014 eFlow at least temporarily increased habitat complexity on the floodplain by triggering understorey flora to germinate, grow and flower.

Box Woodlands

- A total of 100 plant species were observed in the Black and Grey Box woodland monitoring sites. 81 of these species are native and six are listed as rare and threatened in Victoria.
- Only one Box Woodland site was inundated in the 2014 eFlow. Therefore only the vegetation condition Index results for the broad vegetation type are presented here.
- In 2015, 12.1% of the Box sites were calculated with a healthy richness of characteristic PFGs species, and 15.2% of sites were calculated with a healthy tree canopy cover. This represents an increase in health from monitoring inception for species richness (2005, 0%), but a decline in tree canopy health (2005, 36.0%).
- The Box vegetation Index results are mixed. However, when the averages of the two indices are compared (2005, 18.0% vs 2015, 13.6%), it could be concluded that there has been a decline in overall vegetation condition in the broad vegetation type over the monitoring period. This implies that the overarching ecological objective for the Box woodlands has not been achieved in 2015.

Conclusions

- The presence of aquatic and amphibious flora species in areas that were inundated suggests the 2014 eFlow was appropriately timed and of sufficient duration to stimulate the floodplain vegetation.

- The watering event is likely to have, in particular, buffered the Red Gum vegetation and its associated species from the effects of the arid conditions recorded during 2014-2015.
- Notwithstanding the above, the majority of the forest monitoring sites remained dry in 2014, which indicates a large portion of floodplain did not directly benefit from the eFlow. This is particularly true for areas upstream of the Hipwell Regulator.
- It is likely vegetation condition in Gunbower Forest reflects the effects of the dry weather recorded recently and during the Millennium Drought (1996-2010).

Recommendations

→ Continue the environmental watering program to wetlands in Gunbower Forest.

→ Devise strategies that help control turbidity and carp.

→ Develop a second vegetation condition indicator for wetland health.

→ Continue the environmental watering program to Gunbower Forest in order to further improve the condition of the Red Gum vegetation and achieve the ecological objective target.

→ Develop and implement management actions aimed at improving Box woodland canopy condition, such as delivery of environmental water, removing artificial levees preventing natural inflows and ecological thinning.

CARP EXCLUSION PILOT STUDY

- Carp (*Cyprinus carpio*) have been clearly demonstrated to have a significant negative impact on aquatic ecosystems around the world (Vilizzi *et al.* 2015). They affect submerged aquatic vegetation indirectly through changes to water quality (turbidity), (Weber & Brown 2009, Vilizzi *et al.* 2014), and to a lesser degree, directly through physical uprooting and fragmentation (García-Berthou 2001). Shallow wetlands with a soft or silty base, such as found in Gunbower Forest, are the most vulnerable to degradation by carp (Weber & Brown 2009).
- The aim of the carp exclusion pilot study was to summarise current literature and investigate the effect of carp on aquatic macrophyte vegetation in Gunbower Forest.
- A total of 24 carp pilot study plots were established prior to inundation with the 2014 eFlow. The experimental design included a wetland level treatment (two wetlands, one fenced and one unfenced) and three plot treatments (fully fenced, partially fenced and control). The goal of fencing was to exclude large carp.
- Very few carp were recorded in the experimental wetlands over the period of sampling, making it difficult to determine whether treatment effects could be attributed to the impact of carp.
- Multivariate analysis of the data suggests that the cover and richness of aquatic and amphibious species (PFGs 1-3) was more closely correlated with water depth than wetland or plot treatment. The

lack of strong evidence of a treatment effect is, however, possibly due to the small sample size and compounding factors such as turbidity.

- Notwithstanding the above four of the eight fully fenced plots had considerably higher covers of aquatic and amphibious species than the partially fenced and control plots at the same sites. This suggests that the exclosures promoted the aquatic vegetation in some but not all sites.
- The plots in which fencing appeared to have an effect on vegetation density were situated in the shallow end of Little Reedy Lagoon (LR2) and in the deep end in Reedy Lagoon (RL1). This effect was potentially due to the exclusion of carp and possibly water birds.
- The absence of a plot treatment effect in the deep end of Little Reedy Lagoon (LR1) was potentially due to turbidity, as turbidity levels recorded in the lagoon were around four times that in Reedy Lagoon and increased as water levels dropped. This meant that vegetation in all plot types at this site was light-limited, regardless of the fence type.
- Turbidity was most likely, but perhaps not exclusively caused by carp bioturbation, given that Biosis (2015) reported three times the number of and larger carp in Little Reedy Lagoon compared to Reedy Lagoon. While carp abundances were low in both wetlands when sampled in March 2015, it is possible that Little Reedy Lagoon was affected by more individuals than recorded, as it is connected by flood runners to Greens Lagoon, where both the highest abundance of carp and most turbid water in the forest were recorded (Biosis 2015).
- In addition to the inter-wetland differences in carp abundance and turbidity, Reedy Lagoon anecdotally supported a higher cover of aquatic and amphibious flora, which lends support to the hypothesis that the fences constructed across the wetland’s flood runners limited carp access to Reedy Lagoon during inundation. (Particularly large carp).
- The pilot study produced a number of other learnings, including that the fencing material trapped the floating fern Azolla, causing it to cover the surface of fenced plots, which may have influenced the results. Furthermore, the height of the exclosures (3m) may have excluded waterbirds and other fauna from the plots, and consequently, it is not possible to distinguish the impact of excluding these species from that of carp.
- Finally, the study was designed as a pilot, and was not sufficiently replicated to produce statistically significant results. This means that the results are only applicable to Reedy and Little Reedy lagoons. The study did, however, provide valuable background data on the effectiveness of the exclosures, the interaction between the exclosures and aquatic vegetation, and baseline composition and level of variability in the vegetation.
- Given the potential serious impact carp are having on wetland flora diversity in Gunbower Forest, it is recommended the pilot study be refined, expanded and continued into the future in order to fully understand the complexities and create a framework that guides effective management.

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Table A: Definitions of terms and acronyms referenced in the current report

Term/Acronym	Definition in this report
CMA	Catchment Management Authority
Component	An ecological attribute monitored to report on condition (e.g. threatened flora species richness).
Diameter at Breast Height (CPUE)	Is the number of adult fish collected per trap net or gill net night or per hour of electrofishing
Ecological objective	The stated reason for including this ecological component in the program. Comparable with Management objective.
eFlow	Environmental water delivered to ecological assets.
Extent	Short for 'spatial extent', the distribution of the organism in the environment. Where it occurs. Similar to occupancy, but differs because it geo-references or maps the distribution (e.g. species X occurs in this (mapped) portion of the Icon Site).
Ecological Vegetation Classes (EVCs)	Ecological Vegetation Classes are the standard unit for classifying vegetation types in Victoria (DEPI 2015)
Healthy	Site - Meeting or exceeding the PoR for PFG species richness (wetland and understorey sites) and the PoR for tree canopy health (understorey sites). Broad vegetation type – Meeting the target for percentage of sites meeting or exceeding the PoRs.
Icon Site Reference	Compliance of quadrat/wetland level indices at an Icon Site level.
Index	The reported measure, usually against a point of reference. For example, weediness may be an ecological indicator of condition and % native species by biomass may be the Index.
Indicator	The component or aspect of the ecosystem being evaluated.
Linear Mixed Effects Modelling (LMEM)	Univariate analysis
Maintain	Retain similar PFG species richness, appropriate to the stage of wetting and drying, and canopy condition over time, to that sampled at the inception of the monitoring program (2005).
MDBA	Murray Darling Basin Authority
NMDS	Non-metric multidimensional scaled ordinations used in multivariate analysis
Plant Functional Groups (PFGs)	Groups pf plants based on common ecological, morphological and functional responses to inundation based on a system adapted from Brock and Casanova (1997) (See Table 6 in the current report.)
Point of Reference (PoR)	Target value indicating 'healthy' condition of a component (this definition incorporates sampling strategy used).
Power	Measure of the effect size that can be detected with statistical confidence.
Predicted species	Could occur at site (site is within the theoretical/known range of the species).
Promote	Undertake actions that facilitate vegetation processes important for ecosystem function (e.g. nutrient cycling, energy flow, interactions).
Red Gum FDU	River Red Gum with Flood Dependent Understorey (FDU) WRC
Red Gum FTU	River Red Gum with Flood Tolerant Understorey (FTU) WRC
Receding (R)	Receding wetland phase class

Term/Acronym	**Definition in this report**
Recently Inundated (RI)	Recently inundated wetland phase class
Resilient	Able to 'bounce back' from disturbance. Regenerate appropriate richness of species following climatic and/or hydrological disturbance events.
Sampling protocol	The procedure that is followed within a sampling site, including the population to count or quadrats to place; whether it is species, length or height abundance that is recorded; and the instruments required to collect the sample data).
Sampling strategy	The method used to select the sites to be sampled. For example stratification, random placement, distance between sites, frequency of sampling, and so on.
Sensitivity	Evidence of how the Index responds to change in condition as a result of TLM operations. Supported by the results of a statistical sensitivity analysis.
Tree Crown Health	The tree crown health system (also referred to as 'crown condition index' and 'crown index score') is based on six tree crown health categories. Tree canopies are scored by comparison with the photographs and in reference to descriptive text that reflect the health category.
Water regime class (WRC)	WRCs are a classification system that describes broad vegetation communities in Gunbower Forest based on forest stand-class and hydrological mapping (Crome 2004a).
Wetland Phase Classes	Classes of wetland (i.e. receding, dry, recently inundated) based on the stage of the hydrological cycle at which they were sampled.

1 INTRODUCTION

Wetland and floodplain flora was surveyed in Gunbower Forest, a Murray Darling Basin Icon Site, in autumn 2015. The main aim of the survey was to assess the impact of 98 GL of environmental water that was delivered to the forest through the newly constructed Hipwell Regulator during the 2014 winter-spring period. The 2015 intervention results are interpreted in reference to a decade (2005-2014) of vegetation monitoring data collected in Gunbower Forest. A secondary aim was to assess progress towards Icon Site ecological objectives quantified using vegetation condition indicators that were developed in 2014. This report also documents a review of scientific literature on the impact of carp (*Cyprinus carpio*) on wetland flora and the preliminary findings from a carp exclusion pilot study established in 2014.

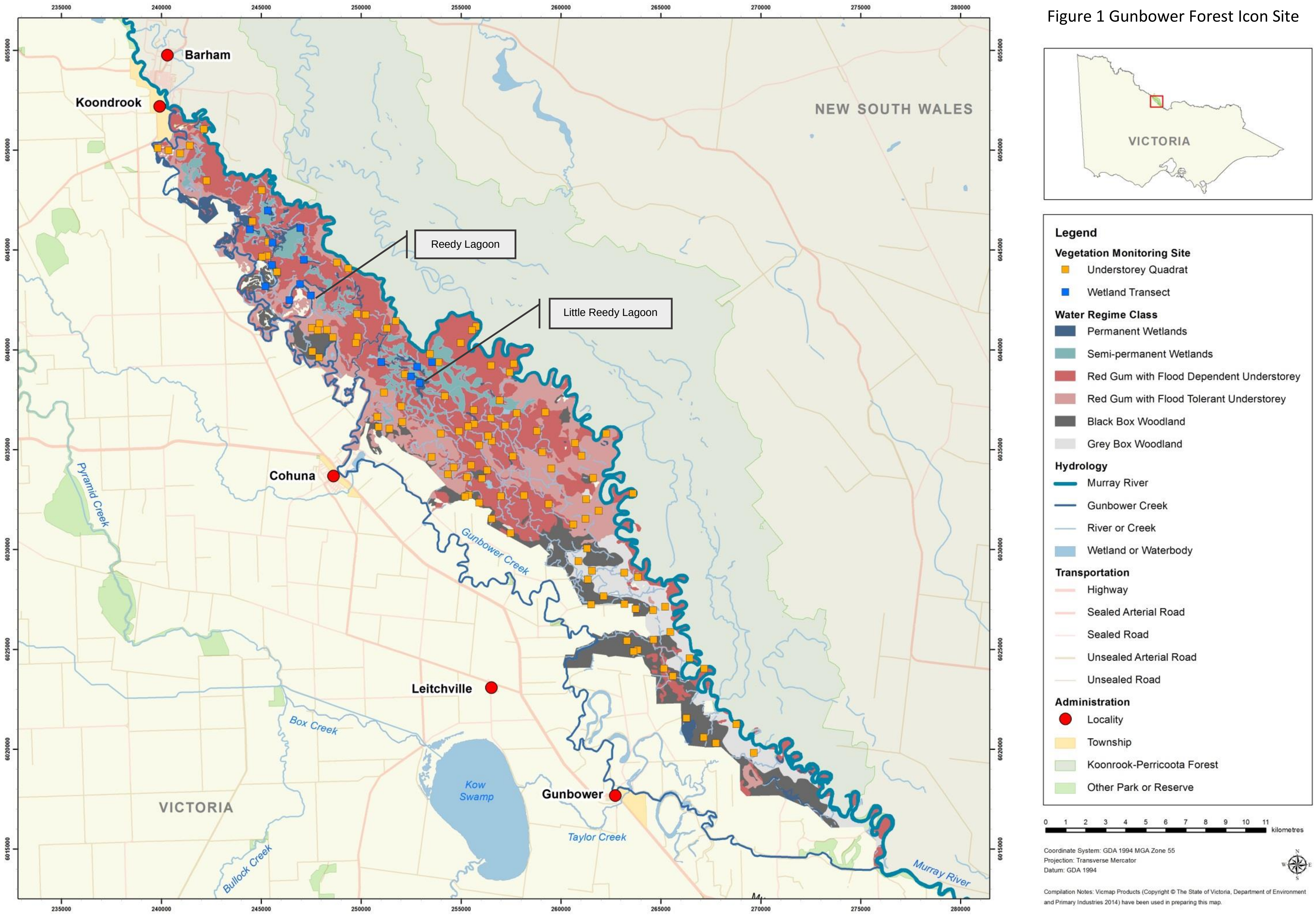
The report presents the outcome of the intervention monitoring and carp exclusion pilot study undertaken by Fire Flood & Flora and Dr Lien Sim for the North Central Catchment Management Authority (CMA). Finally, the report makes recommendations that are designed to advance our understanding of the ecology of the floodplain flora, the impact of carp on wetland flora and proposed management interventions.

1.1 Study area

Gunbower Forest is a large (19,450 ha), narrow parcel of public land located on the River Murray floodplain between the Victorian towns Koondrook and Torrumbarry (Figure 1). The forest's characteristically flat landscape is punctuated by small sandy rises and dissected by a diversity of wetlands and waterways. Soils alternate between clay loams at lower elevations and sandy loams at higher elevations. The region's climate is typically hot and dry in summer and cold and wet in winter.

Vegetation across the floodplain is mapped into six Water Regime Classes (WRCs) based on tree species distribution and modelled hydrological regimes. 110 understorey and 15 wetland permanent monitoring sites are surveyed. These are randomly distributed within the WRCs. The 24 carp exclusion study plots are split between Reedy and Little Reedy lagoons (Figure 1).

Figure 1 Gunbower Forest Icon Site



Coordinate System: GDA 1994 MGA Zone 55
Projection: Transverse Mercator
Datum: GDA 1994

Compilation Notes: Vicmap Products (Copyright © The State of Victoria, Department of Environment and Primary Industries 2014) have been used in preparing this map.

1.2 Rainfall and Flooding

The hydrological regime in Gunbower Forest and its wetlands is the product of high flows in the Murray River and targeted environmental flows delivered by regulators on the Gunbower Creek. Water initially enters the floodplain through effluents in the lower, north-west end of the forest, at river flows of 13,700 ML/day (Ecological Associates 2003). Consequently, many of the wetlands surveyed were inundated to some degree by natural flows in 2003, 2004, 2005, 2010, 2011, 2012, 2013 and 2014 (Figure 2). The forest, however, requires river flows over 30,000 ML/day for 'worthwhile' flooding and flows of 55,000 ML/day for at least a month of major flooding (O'Bryan, 1977). River flows over 30,000 ML/day were recorded at the Torrumbarry Weir between September 2010 and September 2012 (Figure 2). During this period, wetlands in the lower landscape were continuously inundated, and considerable areas of Red Gum and some areas of Black Box vegetation were flooded more than once. Small natural inflows also occurred in September-October 2013 and August 2014.

Prior to 2014, environmental water (referred to as 'eFlow' from here on) was delivered almost annually to wetlands in the lower landscape by four independent regulators on the Gunbower Creek (Yarran, Reedy, Blacks and Little Gunbower), (Figure 2). (Refer to Figure 8 in Section 2.21 for additional detail.) Water delivery targeted particular wetlands or complexes of wetlands, thereby creating a mosaic of wet and dry conditions at the monitored sites.

In 2014 the Hipwell Regulator was commissioned with the release of 98 GL of water between May and December into Gunbower Forest via Spur Creek. Over this period, water flowed north-west through the forest, inundating all downstream wetlands and an estimated 20% of Red Gum communities (North Central CMA 2015a, Figures 4 and 5). Approximately 70% of the water was returned to Gunbower Creek through Chinamen's Bend near Koondrook (A. Chatfield, 2015, North Central CMA, pers. comm. 19 January). Limited areas of Black Box woodland were also flooded.

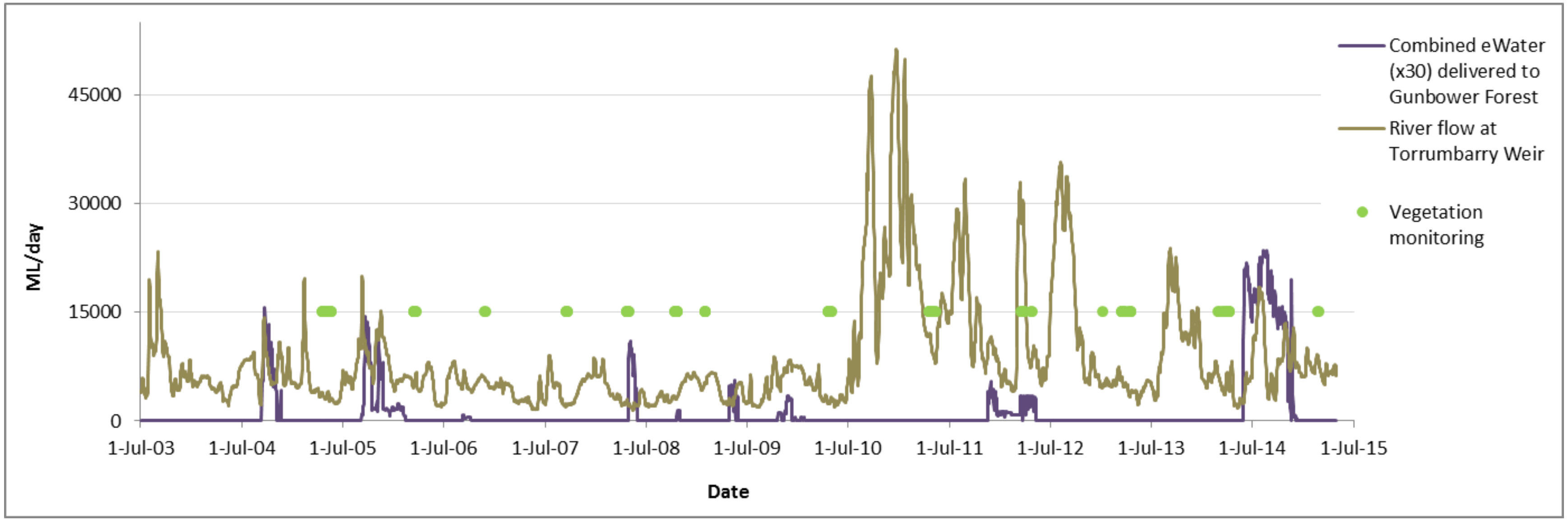


Figure 2 Murray River flow at Torrumbarry Weir (Source: MDBA 2015), combined environmental water delivered (source: G-MW 2014 & North Central CMA 2015b) and timing of vegetation monitoring events, Gunbower Forest, 2003-2015.

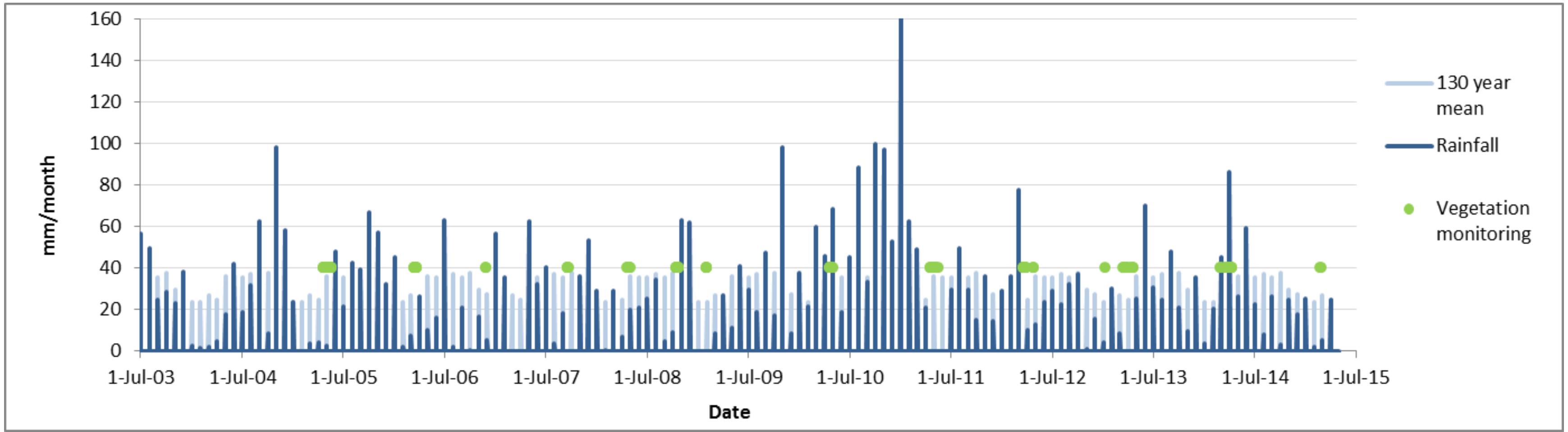


Figure 3 Monthly rainfall (recorded and 130 year mean) Kerang Victoria (Source: BoM 2015) and timing of vegetation monitoring events Gunbower Forest, July 2003-2015.

Figure 4 Red Gum Forest during flooding in January 2015, Gunbower Forest.

Figure 5 Reedy Lagoon inundated with eFlow water, January 2015, Gunbower Forest.

Areas of the forest beyond the reach of regular flooding depend on rainfall as their key source of moisture. This dependence increases with elevation and a corresponding decrease in flooding frequency. Rainfall has been logged at record lows (e.g. during the millennium drought 1996-2010) and highs (e.g. spring 2010) over the period that monitoring has been undertaken in Gunbower Forest. (See Technical Addendum). In the twelve months between the 2014 and 2015 survey, rainfall was well below the 30 year average (i.e. 9 out of 12 months, as per Figure 6) and temperatures were above the average in spring 2014 (Figure 7). Consequently, areas of the floodplain that did not benefit from eFlow in 2014 exhibited exceptionally low soil moisture.

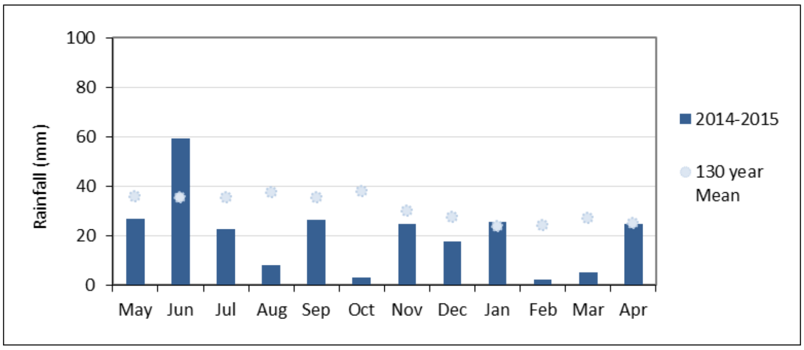


Figure 6 Monthly rainfall (recorded and 130 year mean), Kerang Victoria, May 2014 to April 2015 (Source: BoM 2015).

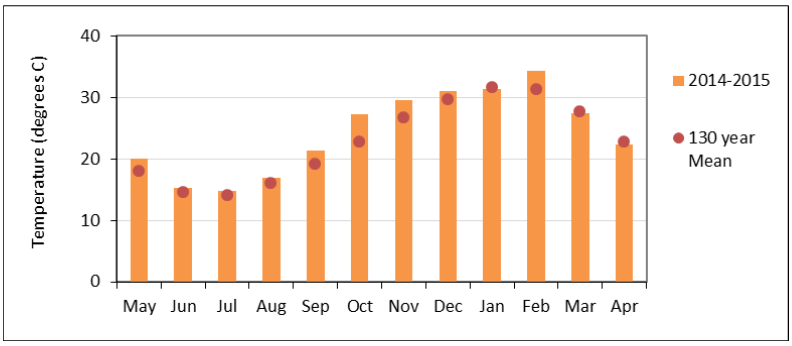


Figure 7 Mean monthly maximum temperature (recorded and 130 year mean), Kerang Victoria, May 2014 to April 2015 (Source: BoM 2015).

1.3 Icon Site Objectives

Ecological objectives have been developed to guide environmental water delivery in Murray Darling Basin Icon Sites such as Gunbower Forest. (Refer to MDBA 2012.) Monitoring of biotic components of the floodplain is undertaken in order to determine progress towards these objectives.

In 2014, the Murray Darling Basin Authority (MDBA) initiated a project that was aimed at refining the process for quantifying progress towards Icon Site ecological objectives based on condition indicators. The statistical power of three Gunbower Forest vegetation condition indicators, focusing on species richness and tree canopy health, were analysed. (Refer to Sim & Bennetts 2014.) It became apparent during this process that there was also a need to refine the Icon Site's vegetation ecological objectives and targets in order to ensure that they were measurable under the sentinel wetland and understorey monitoring program implemented in Gunbower Forest.

Accurate reporting on condition/health depends on clearly defined benchmarks that link analysis to objectives. Table 1 below details the current vegetation objectives for Gunbower Forest and suggested refinements that are designed to improve assessment and accountability. The current project trials the assessment of these suggested objectives and targets.

Table 1 Current (grey text, source MDBA 2012) and suggested (black text) vegetation objectives, targets and Points of Reference for Gunbower Forest

Vision: *To maintain and improve Gunbower Island by enabling native plants and animals to flourish, restoring the floodplain's health for future generations.*			
Icon Site Ecological Objectives		**Targets**	**Points of Reference (PoR) for 'healthy' vegetation** (source: Sim & Bennetts 2014)
Overarching Objectives	**Specific Objectives**		
Current: Increase area of healthy permanent and semi-permanent wetlands. **Suggested**: Continue to increase health of permanent and semi-permanent wetlands.	**Current**: Promote functioning floodplain and wetland ecosystems that are resilient under a range of climate conditions. Successful recruitment of wetland and floodplain vegetation resulting in a structurally diverse landscape. Provide suitable habitat for wetland and floodplain dependent fauna (e.g. waterbirds, macroinvertebrates, frogs and fish). Facilitate an increase in abundance of threatened flora species.	**Current**: 80% of wetlands in healthy condition by 2025 (sustainable intact floristic assemblage). **Suggested:** 100%[1] of monitored wetland sites in healthy condition by 2025.	**Suggested**: Plant Functional Group (PFG) Species Richness Index For each Wetland Phase Class, PFG species richness (comprising native species from characteristic PFGs) at a site is considered healthy if it is on or above the PoR curve (based on the 90th percentile of residuals) since 2005. For each Wetland Phase Class these 90th percentile residual values are: Dry phase wetlands - 2.506 Receding phase wetlands - 6.870 Recently inundated wetlands - not calculated due to insufficient data. For each Red Gum and Box (forest) WRC, PFG species richness (comprising native species from characteristic PFGs) at a site is considered 'appropriate' if it is on or above the 90th percentile of all records since 2005. For the treed WRCs these values are: Red Gum Flood Dependent Understorey (FDU) - 9 species from PFG 3-5 Red Gum Flood Tolerant Understorey (FTU) - 11 species from PFG 4-6 Black Box - 5.9 species from PFG 4-6 Grey Box - 14 species from PFG 5-7. Tree Canopy Health Index For each treed WRC, tree canopy health at a site is considered healthy if it is on or above the 90th percentile of all index values between 2005 and 2014. Each index value represents the proportion of trees with a tree crown health category of 4 or 5 at each site on each date. For each treed WRC these values are: Red Gum FDU - 0.80 Red Gum FTU - 0.815 Black Box - 0.90 Grey Box - 0.95
Current: Ensure maintenance of healthy River Red Gum communities. **Suggested**: Maintain River Red Gum vegetation health.		**Current**: 30% of River Red Gum forests in healthy condition by 2025 (sustainable intact floristic assemblage and tree canopy cover >60%). **Suggested**: At least 21%[1] of monitored River Red Gum sites in healthy condition by 2025.	
Current: Maintain Black Box and Grey Box communities. **Suggested**: Maintain Black Box and Grey Box vegetation health.		**Suggested**: Maintain the 2005 number of healthy Black and Grey Box sites.	

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[1] Predicted number of monitoring sites able to be inundated based on a modelled extent of 1650ML/day delivered via the Hipwell Regulator (pre-testing).

2 WETLAND & FLOODPLAIN UNDERSTOREY INTERVENTION MONITORING

Section Two of the current document reports on the response of wetland and floodplain vegetation to inundation with environmental water in 2014. Included is a summary of the field survey methods undertaken, data analysis applied, results based on both the key research question and application of the vegetation indicators, and lastly, a discussion of the findings. The results and subsequent discussion focus on a) wetlands in the Receding wetland phase class and b) Red Gum vegetation, given that *all* wetlands that were inundated in the 2014 eFlow fell into this wetland phase class and only one Box Woodland monitoring site was inundated during this event.

2.1 Methods

2.1.1 Field Survey

Flora Data

A field survey of Gunbower Forest permanent monitoring sites was undertaken between 24 March and 16 April 2015. The full cohort of the 125 permanent monitoring sites established within Gunbower Forest (refer to Technical Addendum) were sampled in accordance with the Manual of Field Procedures for Monitoring in Gunbower Forest (Crome 2004b), regardless of whether they were inundated in the 2014 eFlow. The number of sampling units and sampling frequencies are outlined in Table 2 and Table 3 (on following page).

All ground flora species that occurred either within each of the 110 10^2 metre understorey quadrats, or within each two metre wide vegetation zone along the 15 wetland transects, were identified to a specific level, and projected foliage cover (percentage) was estimated. The plant taxonomy that was employed follows the Victorian Plant Name Index (DELPW 2015), with consideration to the Census of Victoria Vascular Plants (Walsh & Stajsic 2015).

The canopy condition of trees, both in the understorey sites and along the wetland transects, was scored with reference to both tree crown health categories (Table 4) and images contained within the monitoring manual (Crome 2004b). Other tree attributes such as diameter at breast height were recorded for eucalypts present in the monitoring sites, although they have not been analysed in the current project.

Table 2 Total number of wetland sites sampled autumn 2005-2015, Gunbower Forest.

Wetland Phase Classes analysed (R = Receding, D = Dry, RI = Recently Inundated, NA = not assessed, refer to Table 6) is included in the table for comparison purposes.

Wetland Sites	Wetland code	No. of transects	Sampling year & Wetland Phase Class								
			2005	2006	2008	2010	2011	2012*	2013	2014	2015
Black Swamp	BLS	2	R	R	D	R	R	RI	R	D	R
Corduroy Swamp	COS	1	NA	R	D	D	R	RI	D	D	R
Green Swamp	GS	1	R	R	D	D	R	RI	R	R	R
Little Gunbower Complex	LG1	1	R	R	D	R	R	RI	D	D	R
Little Gunbower Creek**	LG2	1	R	R	D	R	R	RI	R	R	R
Reedy Lagoon	RL	3	R	R	D	R	R	RI	R	D	R
Permanent wetland total		*9*	*5*	*6*	*6*	*6*	*6*	*6*	*6*	*6*	*6*
Charcoal Swamp	CS	1	D	D	D	D	R	RI	D	D	R
Football Grounds	FG	1	D	R	D	D	R	RI	D	D	R
Iron Punt Lagoon	IPL	1	R	R	D	D	R	NA	R	D	R
Little Reedy Lagoon	LRL	2	R	R	D	D	R	RI	R	R	R
Long Lagoon	LL	1	D	R	D	R	R	NA	D	D	R
Semi-permanent wetland total		*6*	*5*	*5*	*5*	*5*	*5*	*3*	*5*	*5*	*5*
Total for all wetlands		***15***	***10***	***11***	***11***	***11***	***11***	***9***	***11***	***11***	***11***
Total for Dry wetlands			*3*	*1*	*11*	*6*	*0*	*0*	*5*	*8*	*0*
Total for Receding wetlands			*7*	*10*	*0*	*5*	*11*	*0*	*6*	*3*	*11*
Total for recently inundated wetlands			*0*	*0*	*0*	*0*	*0*	*9*	*0*	*0*	*0*

* Two sites were in accessible due to flooding

** Different to 2014, the Little Gunbower Creek site has been analysed independently to the Little Gunbower Complex site, as they are governed by significantly different water regimes (i.e. at capacity the creek site is over 8 m deep, whereas the complex site is less than 3 m deep), which has a direct effect on the flora.

Table 3 Total number of understorey sites sampled autumn 2005-2015, Gunbower Forest.

Numbers in each column indicate the quantity of quadrats sampled in each WRC in each year.

Water Regime Class	Sampling year								
	2005	2006	2008	2010	2011	2012*	2013	2014	2015
Red Gum with flood dependent understorey	42	42	50	50	50	48	50	50	50
Red Gum with flood tolerant understory	23	23	27	27	27	27	27	27	27
Black Box	14	14	19	19	19	19	19	19	19
Grey Box	11	11	14	14	14	14	14	14	14
Total No. quadrats sampled	*90*	*90*	*110*	*110*	*110*	*108*	*110*	*110*	*110*

* Two sites were in accessible due to flooding

Table 4 Tree crown health categories and descriptions (Source: Crome 2004b)

Tree Crown Health Categories		Health Description
0	Dead Tree	Dead tree with no original canopy. All main branches dead No epicormic growth
1	Unhealthy Tree	Tree with no original/intact canopy Most main branches dead All epicormic growth
2	Unhealthy Tree	Tree with <25% of the original/intact canopy present Some main branches dead (<50% canopy) Predominantly epicormic growth (>50% of remaining canopy)
3	Tree	Tree with 25-50% of the original/intact canopy present Some small dead branches Some epicormic growth (<50% of remaining canopy)
4	Healthy Tree	Tree with 50-75% of the original/intact canopy present Some dead branchlets (<50% of canopy) <10% epicormic growth
5	Healthy Tree	Tree with >75% of the original/intact canopy present May include some dead branchlets and leaves <5% epicormic growth

Other variables that were recorded at each site during the field survey included evidence of disturbance (e.g. inundation, timber harvesting, grazing, fire and tree fall) and incidental observations.

Hydrological Data

Water depth at the time of survey was recorded along the wetland transects. Without the benefit of precise flood extent data, the probability of flooding at each understorey site over the previous 12 months was estimated on the basis of anecdotal observations made during the field surveys. Evidence of flooding included ponded water (at or nearby the site), recent water marks on tree trunks, fresh silt coating over ground flora, deposits of water-borne litter, and/or the presence of water dependent species such as *Ludwigia peploides ssp. montevidensis*, *Nymphoides crenulata*, *Myriophyllum* spp. and *Juncus* spp. (excluding *J. subsecundus*).

2.1.2 Data Preparation

Field data were aggregated (Table 5) to allow the description and analysis of quadrat and transect floristics, canopy health, and vegetation condition indicators. The key aims of these analyses were to:

- Identify potential effects of the 2014 eFlow
- Identify potential patterns in floristics and vegetation health in relation to the eFlow and other factors such as flooding in the past 12 months (for all years of data), or sampling year
- Compare 2015 data to that collected in autumn of previous years (2005-2014)
- Calculate the health of the survey sites in 2015 based on vegetation indicators.

Table 5 Framework for data analysis

Data Grouping	Purpose
Broad vegetation types	Collate data and results in line with Icon Site ecological objectives: Wetlands River Red Gum Forest & Woodlands Black and Grey Box Woodlands
Water Regime Classes	To delineate vegetation types and establish how they are influenced by their landscape position (Landscape Logic). Permanent Wetlands Semi-permanent Wetlands Red Gum with Flood Dependent Understorey (Red Gum FDU) Red Gum with Flood Tolerant Understorey (Red Gum FTU) Black Box Woodlands (Black Box) Grey Box Woodlands (Grey Box)
Wetland Phase Classes	To group wetlands based on the stage of the hydrological cycle at which they were sampled
Plant Functional Groups	To group plants based on common ecological, morphological and functional responses to inundation.

Wetland sites were classified into wetland phase classes (see Table 2) based on the stage of the hydrological cycle at which they were sampled, in accordance with categories in Table 6. Wetland phases were found to reflect more similar floristic composition than WRCs and, therefore, produce more meaningful results (see Bennetts 2014).

Table 6 Phase of the wetland cycle observed at wetland monitoring sites in Gunbower Forest in autumn 2005 - 2015.

Water depth	Wetland phase	Wetland phase class analysed
0 cm	Dry	Dry
> 0 cm	Recently inundated (i.e. within last month)	Not included in analysis[2]
< 10 cm	Drying	Receding
10 - 100 cm	Receding, shallowly inundated	
> 100 cm	Receding, deeply inundated	

The primary source of inundation of Receding class wetlands in the 12 months preceding sampling, was also determined, by reference to the hydrological conditions outlined in Table 7.

Table 7 Water source rules for Receding wetland categorisation in Gunbower Forest data.

Primary water source	Wetlands	Hydrological conditions
Regulators	Yarran regulator (Corduroy, Charcoal and Greens swamps and Little Reedy Lagoon) Little Gunbower regulator (Black Swamp, Football Grounds, Little Gunbower Complex/Creek, and Long and Reedy lagoons) Blacks Swamp regulator (Black Swamp, Football Grounds, Little Gunbower Complex/Creek and Long Lagoon) Reedy Lagoon regulator (Reedy Lagoon)	Any volume of flow intentionally delivered through a regulator

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[2] 'Recently Inundated' Wetland Phase Class were not analysed because all data were recorded in 2012, making it irrelevant to the 2015 intervention monitoring. The dataset was also small and strongly confounded with year of sampling.

Primary water source	Wetlands	Hydrological conditions
Natural flooding (small inflow)	Wetlands inundated via Yarran Creek/Shillinglaws (Corduroy, Charcoal and Greens swamps and Little Reedy Lagoon)	Flows between 13,700 and 25,000 ML/day for over 1 month at Torrumbarry Weir (source: Ecological Associates 2003)
	Wetlands inundated via Barham Cut (Black Swamp, Football Grounds, Little Gunbower Complex/Creek, and Long and Reedy lagoons)	Flows between 15,300 and 25,000 ML/day for over 1 month at Torrumbarry Weir (source: Ecological Associates 2003)
Natural flooding (widespread)	All wetlands surveyed	Flows of > 25,000ML/day at Torrumbarry Weir (Anna Chatfield, 2012, North Central CMA, pers comm. 18 July)
Rainfall	All wetlands surveyed	No natural inflows/flooding or delivered water in 12 months prior to sampling

Flora species were classified into Plant Functional Groups (PFGs), employing a system adapted from Brock and Casanova (1997) which groups species in terms of their response to both inundation and drying (Table 8).

Table 8 Plant Functional Groups applied in Gunbower Forest flora data analysis.

PFG Code	PFG Name	Description
1	Submerged & Free floating Flora	S - Submerged (including strictly aquatic floaters) Adult plants do not survive prolonged exposure of the wetland substrate (drying) and lack perpetuating rootstocks. Seed or spores may persist in soil during dry times.
2	Floating Amphibious Flora	ARf - Amphibious Fluctuation - Responders Floating Amphibious species that produce floating foliage when inundation. Aerial parts of plants survive exposure of the wetland substrate (drying) for sustained periods of time. Plants survive drying by dying back to rootstocks.
3	Adaptive Amphibious Flora	ARp - Amphibious Fluctuation - Responders Plastic Amphibious species that alter their growth pattern or morphology in response to water conditions. Can actively grow when substrate exposed but still moist, but may die back to rootstocks or seed during sustained dry periods.
4a	Perennial Mudflat Flora	ATl - Amphibious Fluctuation - Tolerators Low Growing Perennial amphibious species that tolerate changes in water conditions and maintain same general growth form during brief periods of inundation, but may die back to rootstocks if unable to develop emergent growth during sustained inundation.
4b	Annual Mudflat Flora	ATl - Amphibious Fluctuation - Tolerators Low Growing Annual (or functionally so) amphibious species that may tolerate very brief periods of shallow flooding during growth phase, but essentially short-lived plants which germinate following flood water recession and produce inundation-tolerant seed during the drying phase.
5	Emergent Amphibious Flora	ATe - Amphibious Fluctuation - Tolerators Emergent Amphibious flora that tolerates changes in water conditions, typically with emergent habit. Rootstocks tolerant of shallow inundation but plants intolerant of sustained total immersion. Recruitment and/or long-term maintenance of populations are generally dependent on at least occasional inundation events.
6	Terrestrial Damp	Tda - Terrestrial Damp Rootstocks intolerant of more than superficial inundation, but occurring in areas of good soil moisture conditions which may be influenced by proximity to river and water seepage through soil
7	Terrestrial Dry	Tdr - Terrestrial Dry Dry-land plants (i.e. flood intolerant and going through life cycles independently of flooding regime)
0	Not-vegetated	Bare ground, litter, logs, water etc.
NA	Not Assigned	Species for which there is insufficient information to be assigned to a PFG

2.1.3 Intervention Monitoring Analysis

As part of the intervention monitoring, a subset of ground flora monitoring data (wetland and understorey) was collected in Gunbower Forest and analysed. Research questions guiding the analysis included:

- How did species diversity differ between sites sampled before and after the 2014 eFlow, and sites that had been flooded or not flooded in the past 12 months?
- Were there clear groupings of sites within each wetland phase class or broad vegetation type based on species composition?
- If so, do these groupings correspond with factors such as year of sampling or flooding in the past 12 months?

Native species diversity and flora composition data were analysed for the 'Receding' wetland phase class and Red Gum broad vegetation type. Samples were categorised based on treatment ('B' before the 2014 eFlow; 'A' after the 2014 eFlow) and impact ('yes' flooded in preceding 12 months to sampling; 'no' not flooded in preceding 12 months to sampling).

Numbers of sampling units and frequencies are outlined in Tables 2 and 3. Data from wetlands with multiple transects per site were averaged. A number of samples were removed from the analysis due to little or no native flora cover. The wetland samples removed from the modelling were BS_2005, LG1_2013, LG1_2014 and IPL_2013. In addition to the samples above, CS_2012, IPL_2008, LL_2013, LL_2008 and COS_2008 were also removed from the wetland ordination. Thirty five Red Gum samples were excluded from the modelling (2005, 6, 42, 58; 2006, 6, 15, 42, 57, 58, 78, 79; 2010, 6, 13, 21, 22, 24, 28, 35, 42, 46, 48, 65; 2012, 6; 2013, 6, 11, 67, 68, 127; 2014, 6, 22, 24; and 2015, 22).

Calculating Diversity

The 'effective number of species' or 'true diversity' – henceforth known as 'diversity' – for each sample (i.e. wetlands or quadrats in a particular sampling year) was calculated as exponential Shannon diversity, using EstimateS (EstimateS, Colwell 2014, Connecticut, USA). Percentage cover of all native ground flora data (excluding exotics, bare ground, leaf litter and water) was used.

Univariate Analyses

Univariate analyses (Linear Mixed Effects Modelling, LMEM) were used in order to test relationships between the species diversity of sites (wetlands or Red Gum quadrats) before and after the 2014 eFlow ('treatment'), and between sites that were flooded ('impact') or not flooded in each year. LMEM was also run in order to test the interaction between treatment and impact, with 'site' and 'sampling year' as random variables. Separate analyses were run for wetlands in the 'Receding' phase class and the 'Red Gum' broad vegetation type.

Univariate analyses were performed using the open-source statistical package R (version 3.0.2, R Core Team 2013) and the interface RStudio (version 0.98.501, RStudio 2012), while the LMEM were run using the `lme` function in the `nlme` package. As part of the process of analysis, diagnostic tests were run on the dataset in order to ensure that it conformed to the assumptions for linear models (Zuur *et al.* 2009, Logan 2010). Several `lme` models were created in order to check the effects of inclusion of the fixed factors (treatment, impact) on the residuals. As well, a model selection process (`model.sel` in the `MuMIn` package) was run in order to determine which was the best model fit (employing Akaike's Information Criterion for small sample sizes, AICc), (Zuur *et al.* 2009).

Multivariate Analyses

Non-metric multidimensional scaling (NMDS) ordinations were used in order to explore similarities in wetlands or quadrats native flora composition in a particular sampling year. In addition, factors such as 'sample year,' 'site name,' 'Water Regime Class' (or 'Wetland Phase Class') and 'Treatment' and 'Impact' were overlaid on the ordination plots to allow a visual interpretation of the similarities between sampling units. A measure of the dominance of aquatic, amphibious and mudflat species at each site (total % cover of PFGs 1-4) and the maximum water depth that was recorded during the previous financial year were both overlaid as bubbles on the ordination plots in order to illustrate the correlation of these variables with observed species composition.

The analyses were performed in the PRIMER software package (version 6; PRIMER-E, Plymouth, UK). A table of site date versus native flora species was produced for Receding phase wetlands and for Red Gum vegetation, each using percentage cover data. Percentage cover data were square-root transformed in order to reduce the influence of dominant species (and increase the influence of rare species) on the ordination result. Both datasets (a and b) were used to generate a Bray–Curtis similarity matrix. Ordination by NMDS was then performed on these data with the aim of examining site date groupings based on species composition.

Note that ordinations for Red Gum only include data from 2014 and 2015, since the dataset from the entire monitoring period is so large as to make it difficult to interpret plots that include the whole data set.

Once NMDS analyses had been run (25 restarts, minimum stress 0.01), factors including site, survey year, wetland phase class or WRC, Treatment (B/A) and Impact (yes/no) were overlaid on the plots to see if the factors corresponded with the observed unit groupings.

2.1.4 Vegetation Condition Indicator Analysis

Vegetation condition indicators have been developed in order to assist reporting on ecological objectives for Gunbower Forest (see Sim & Bennetts 2014). Three indicators, focusing on species richness and canopy health, are considered in the current report, as outlined in the following section.

Wetland PFG Species Richness Index

Species richness is considered 'healthy' for each wetland site if it is on or above the wetland phase class's PoR curve. Wetland phase class PoRs were calculated for each site/year sampled from non-linear regression curves fitted to the 2005-2014 raw data (Sim & Bennetts 2014). PoR curves were used instead of a single PoR value in order to determine wetland index scores, as the weighted values more accurately reflect the relationship between area sampled and number of species recorded. This approach was applied as the wetland area sampled varied between years due to water depth affecting the vertical position and therefore the length of wetland transects.

PoR values, based on this approach, were derived by adding the predicted number of native characteristic PFG species for each site/year sample (based on the regression curve) and the 90th percentile residual values. The residual values were calculated as the 'actual' minus 'predicted' number of native characteristic PFG species across the 2005-2014 sampling period.

The 90th percentile residual values for the wetland phase classes analysed were:

Dry phase wetlands = 2.506
Receding phase wetlands = 6.870

The method for calculating wetland species richness index scores was as follows:

- Wetland sites were classified by wetland phase class in each year that was sampled (see Table 6). The wetland phases 'Drying', 'Receding, shallowly inundated' and 'Receding, deeply inundated' were grouped into one class called 'Receding'. Wetlands classified as 'Recently Inundated' were not analysed as all data were from 2012, meaning that the dataset was small and strongly confounded with sampling year.
- Both the total number and total cover (m^2) of native characteristic PFG species were calculated for each site/year sample. Data from wetland with multiple transects were combined to produce wetland site values.
- The predicted numbers of native characteristic PFG species for each site/year sample were calculated using the following non-linear regression curve equations:

  Dry wetlands equation: $y = -3.5810 + 7.2744 \times C^{0.1936}$

  Receding wetland equation: $y = 0.6853 + 4.5272 \times C^{0.2696}$

  Where y = Predicted number of native characteristic PFG species, and C = total cover of native characteristic PFG species.
- Index scores for each site/year sample were then calculated according to the formula:

  Index = Sqrt(native characteristic PFG richness) ÷ Sqrt(PoR)
- Scores were corrected so that any values >1 were recorded as 1, as the index lies between 0 and 1.

- Sample native characteristic PFG richness that is greater than or equal to the PoR value results in an index of 1 (meaning that it is compliant), and richness less than the PoR results in an index of <1 (meaning that it is not compliant).
- WRC index scores were calculated as the proportion of compliant sites in each wetland phase class.

Understorey Site PFG Species Richness Index

For each understorey site, species richness is considered 'healthy' if it is on or above the WRC's PoR value. PoR values have been determined as the 90th percentile of native characteristic PFG species richness across the 2005-2014 sampling period in each WRC.

The understorey site PFG species richness PoR values are:

- Red Gum FDU - 9 species from PFGs 3-5
- Red Gum FTU - 11 species from PFGs 4-6
- Black Box - 5.9 species from PFGs 4-6
- Grey Box - 14 species from PFGs 5-7

The method for calculating understorey species richness index scores was as follows:

- Richness of native characteristic PFG species was calculated for each site/year sampled in the WRCs.
- Species richness data was converted to an index using the formula:

  Index = Sqrt(characteristic PFG richness) ÷ Sqrt(PoR)
- Scores were corrected so that any values >1 are recorded as 1, as the index lies between 0 and 1.
- Sample native characteristic PFG richness that is greater than or equal to the PoR results in an index of 1 (meaning that it is compliant), and richness less than the PoR results in an index of <1 (meaning that it is not compliant).
- The WRC index score was based on the proportion of compliant sites in each WRC.

Tree Canopy Health Index

For each treed WRC (Red Gum FDU, Red Gum FTU, Black Box and Grey Box), tree canopy at a site is considered 'healthy' if it is on or above the WRCs' PoR value (90th percentile of all index values across the 2005-2014 sampling period). PoR index values represent the proportion of trees with a crown health category of 4 or 5 at each site on each date.

The Tree Canopy Health PoR index values are:

Red Gum FDU - 0.80
Red Gum FTU - 0.815
Black Box - 0.90
Grey Box - 0.95

The method for calculating tree canopy health index scores was as follows:

- The site index score was calculated as the proportion of healthy trees (crown health category > 3) in each site/year sampled.
- Index scores lie between 0 and 1.
- Site/year index scores greater than or equal to the PoR index value for each WRC indicate compliant sites.
- The WRC index score was calculated as the proportion of compliant sites in each WRC.

2.1.5 Limitations

Sample size, pattern and frequency all influence the utility of a dataset. Due to the cryptic nature and seasonal growth cycles of certain species, ecological surveys are often unable to detect all taxa present at a particular site. It should be recognised, therefore, that the sample data are, at best, indicative of the total species richness supported by the forest, and are skewed towards reporting a lower than actual level of richness.

Overall limitations with the study and analysis include:

- Suitable environmental conditions did not exist for all species in all years.
- The monitoring program is principally undertaken in autumn and, therefore, does not represent the full annual diversity of flora.
- Sentinel wetland monitoring sites were subjectively located at known wetlands. The results may therefore be biased and may not reflect the diversity and/or trends of wetlands as a whole in Gunbower Forest.
- Wetland transects were re-established using a hand-held compass. While care was taken to overlap the sampled transect with previous years, this was not always possible, particularly at the longer (i.e. >100m) and/or densely treed sites. Consequently, it is likely there is some data mismatch, namely in the sapling sample in Black Swamp. This limitation is, however, unlikely to substantially affect the other results.
- Wetland transects change in length each year, depending on degree of inundation. We would expect more species to be recorded at a larger transect. To correct for this, weighting by area has been performed using a species vs abundance curve for the wetland PFG species richness indicator. (See details under 'Wetland PFG Species Richness Index' in Section 2.1.4.)
- Wetland data are highly variable due to intrinsic differences in size, condition and flooding regime between wetlands, plus the inability to sample at the same stage of inundation each year, which dramatically affects which species are recorded. Summarising wetland data into a single index value for each WRC is likely to incorporate significant error.
- For the analysis of wetland data, we have assumed spatial independence of sites (although sites are located close to each other and are likely to be connected when inundated).

- Ground flora data is analysed within WRCs based on pre-determined 'characteristic PFGs'. This approach implies that distinct groups of species occur in discrete WRCs. While this approach offers a practical method for analysing the data, it does not account for the broad ecotones between communities that are created by the subtle environmental gradient across the floodplain. Consequently, naturally occurring 'non-characteristic species' can contribute to a poor health score.

Additionally, as the monitoring program and the 2014 eFlow (intervention) is not measuring response under controlled conditions (i.e. there are no control or impact sites), causality is not demonstrated. Rather inferences and anecdotal observations can be made in relation to driving factors that affect floristic composition and ecological condition.

2.2 Results

The intervention monitoring results are structurally based on the broad vegetation types, Wetlands, Red Gums Forests and Woodlands, and Box Woodlands. Species diversity (univariate analysis), flora composition (multivariate analysis) and vegetation condition indicator results are included.

2.2.1 Wetlands

Eleven wetland monitoring sites were re-surveyed in autumn 2015. These sites comprise a range of wetland types including a deeply incised creek (i.e. Little Gunbower Creek), paleo-river lagoons (e.g. Black Swamp and Iron Punt Lagoon) and low lying, open areas (e.g. Corduroy Swamp and Little Reedy Lagoon). The wetlands are mapped as either permanent or semi-permanent WRCs, based on their landscape position and modelled water regime. While semi-permanent wetlands are thought to flood less frequently and at lower depths than permanent wetlands (URS 2001), previous analysis (e.g. Bennetts 2014; Bennetts & Sim 2014) has shown a greater floristic similarity between sites grouped by wetland phase than by WRC. The wetlands have hence been treated as one broad vegetation type and analysed by wetland phase classes in the current project.

The following section presents the diversity results for wetlands in the Receding wetland phase class ('Receding wetlands'). Data gathered since the 2014 eFlow (sampled 2015) is compared to that collected before the watering event (sampled 2005-2014). The analysis aims to provide an insight into the impact of the 2014 eFlow on wetland flora relative to previous wetland-based eFlows and natural flooding. It commences with a brief account of the general condition of the wetlands in autumn 2015.

General Condition

All lower landscape wetlands that were monitored in Gunbower Forest were inundated by the 2014 eFlow (Figure 8). Maximum water depths recorded at the time of the autumn 2015 survey were between 5 cm

(Corduroy Swamp) and over 6 m (Little Gunbower Creek), with an average depth of 40 cm (excluding Little Gunbower Creek). All wetlands were therefore classified as 'Receding' for analysis. (See Table 6).

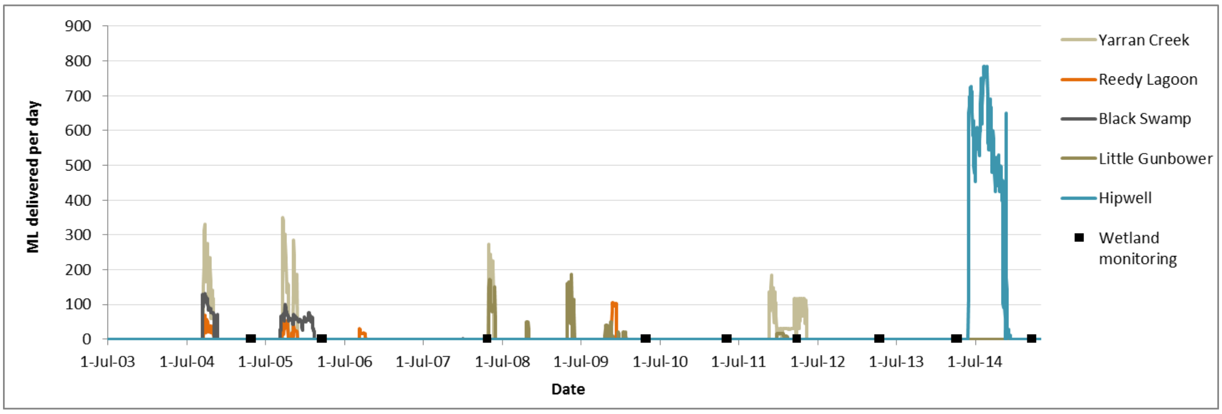


Figure 8 Environmental water delivered via regulators to wetlands in Gunbower Forest, 2004-2014 (Source G-MW 2015 & North Central CMA 2015b).

A total of 83 flora species that were identified to species level were recorded within the wetland transects in autumn 2015. 81% of these species were native. This represents the second highest annual species richness (i.e. less than 2010 when 109 species were identified). Included in the list is the:

- Nationally vulnerable River Swamp Wallaby-grass (*Amphibromus fluitans*),
- State listed Wavy Marshwort (*Nymphoides crenata* – see front cover of report), and
- Rare Dwarf Bitter-cress (*Rorippa eustylis*), Riverina Bitter-cress (*Cardamine moirensis*) and Water Nymph (*Najas tenuifolia*).

Of these species the River Swamp Wallaby-grass was most commonly recorded, notably as dense swards at the shallow ends of Reedy Lagoon.

34% (28/83) of the flora species recorded in 2015 along the wetland transect are aquatic (i.e. establish and grow while inundated and do not tolerate sustained drying, PFGs 1-3). It is unlikely these species would have been present in the forest if the wetlands had not been inundated by the 2014 eFlow. The 2015 species list however also included two new weeds for the wetland monitoring dataset, Water Crassula (**Crassula natans var. minus*) and Burr Medic (**Medicago polymorpha*).

The following panorama images (Figures 9-19) provide a visual account of the wetlands (ordered alphabetically) when sampled in 2015.

Figure 9 Black Swamp (BLS2A) Gunbower Forest, autumn 2015.

Figure 10 Corduroy Swamp (COS1A) Gunbower Forest, autumn 2015.

Figure 11 Charcoal Swamp (CS1B) Gunbower Forest, autumn 2015.

Figure 12 Football Grounds (FB1B) Gunbower Forest, autumn 2015.

Figure 13 Greens Swamp (GS1A) Gunbower Forest, autumn 2015.

Figure 14 Iron Punt (IPL1B) Gunbower Forest, autumn 2015.

Figure 15 Little Gunbower Creek (LG1B) Gunbower Forest, autumn 2015.

Figure 16 Little Gunbower Complex (LG2B) Gunbower Forest, autumn 2015.

Figure 17 Long Lagoon (LG1A) Gunbower Forest, autumn 2015.

Figure 18 Little Reedy Lagoon (LR2A) Gunbower Forest, autumn 2015.

Figure 19 Reedy Lagoon (RL3A) Gunbower Forest, autumn 2015.

Species Diversity

Native flora species diversity that is calculated for Receding wetlands, and sampled before the 2014 eFlow (B, i.e. sampled between 2005 and 2014) and after the 2014 eFlow (A, i.e. sampled in 2015), is presented in Figure 20. The analysis suggests that the median and minimum diversities recorded in Receding wetlands were slightly higher after the 2014 eFlow (2015) compared to before the eFlow (2005-2014). The maximum diversity score was, however, recorded before the eFlow (B, e.g. Black Swamp_2013). There was also a broader range of diversities recorded before the eFlow than after in 2015. The difference between the means of the groups Receding wetlands (i.e. before the eFlow (2005-2014) and after the 2014 eFlow (2015)) was not however statistically significant (Table 9).

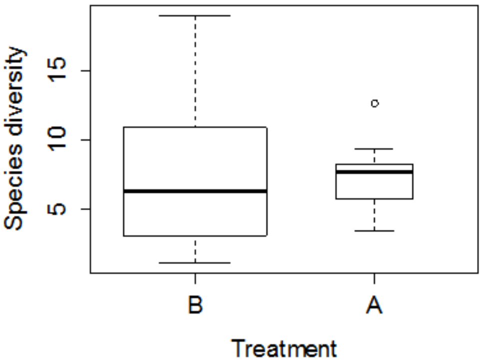


Figure 20 Boxplot of autumnal species diversity (Shannon Exponential Diversity) for Receding phase class wetland with samples grouped by treatment (B = Before 2014 eFlow (2005-2014); A = After 2014 eFlow (2015)), Gunbower Forest.

Note that that the 'box' contains the middle 50% of samples and the box width indicates the sample size it represents. The median is represented by the black line inside the boxes. The top and bottom 'whisker' represents the top and bottom 25% of samples and outliers samples are shown as dots.

Table 9 LMEM output for autumnal Receding phase wetlands (significant results in bold), Gunbower Forest, 2005-2015.

	Value	Std.Error	DF	t-value	p-value
(Intercept)	3.380573	1.298109	47	2.6042285	**0.0123**
Treatment (Before/After 2014 eFlow)	0.069407	1.391652	47	0.0498741	0.9604

Plotting the native species diversity in Receding wetland across a temporal scale provides a different insight into the effect of eFlows, compared to natural flooding/inflow on wetland flora (Figure 21). Wetland-targeted eFlows were delivered prior to surveying in 2005, 2006, 2010 and 2015. Widespread natural flooding occurred prior to surveying in 2011, and smaller natural inflows predated the 2013 and 2014 surveys. While the annual sample sizes for the Receding wetland phase class are unbalanced (e.g. 2015 n = 11 vs 2014 n = 3), the analysis suggests that the four eFlows were associated with higher median species diversity in the Receding wetlands than the large natural flood in 2011. It also suggests that wetlands sampled in 2013 and 2014 had the highest median species diversity in the Receding wetland dataset. This latter outcome was potentially driven by fringing terrestrial flora diversity, rather than aquatic and amphibious flora, given the above average temperatures and below average rainfall recorded preceding these surveys. (See Technical Addendum.)

The highest diversity of native species in the Receding wetlands was recorded at Black Swamp in 2013. In 2015, Reedy Lagoon was recognised as the most diverse wetland and considerably more so than the remaining wetlands (outlier dot above the 2015 box).

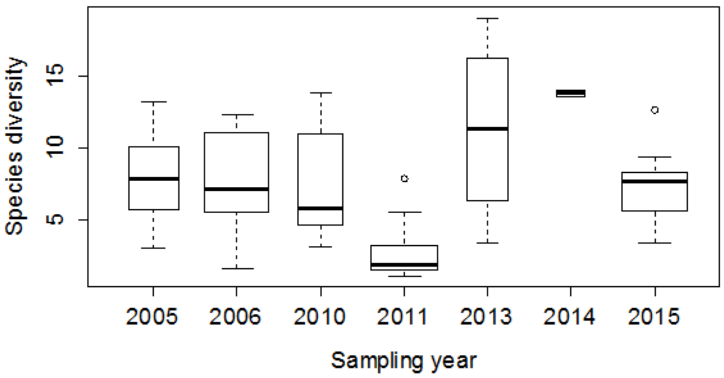


Figure 21 Boxplot of autumnal species diversity (Shannon Exponential Diversity) for Receding phase class wetland samples grouped by year, Gunbower Forest, 2005-2015.

In addition to the potential influences of water sources that are discussed above, site-based factors also appear to affect wetland species diversity. Figure 22 below ranks the Receding wetlands according to diversity. Reedy Lagoon (RL) exhibited the highest median and minimum diversity of all wetlands surveyed, while Iron Punt Lagoon (IPL) and Little Gunbower Creek (LG1) had the lowest median diversity. Note that these numbers are not corrected for wetland area. This data suggests that Reedy Lagoon has more than three times the flora diversity in any given sample year than has Iron Punt Lagoon. While care should be taken when interpreting this data, due to unbalanced sample sizes (i.e. RL, *n* = 6 vs ILP, *n* = 2) and differences in wetland area, the ranking aligns with field observations.

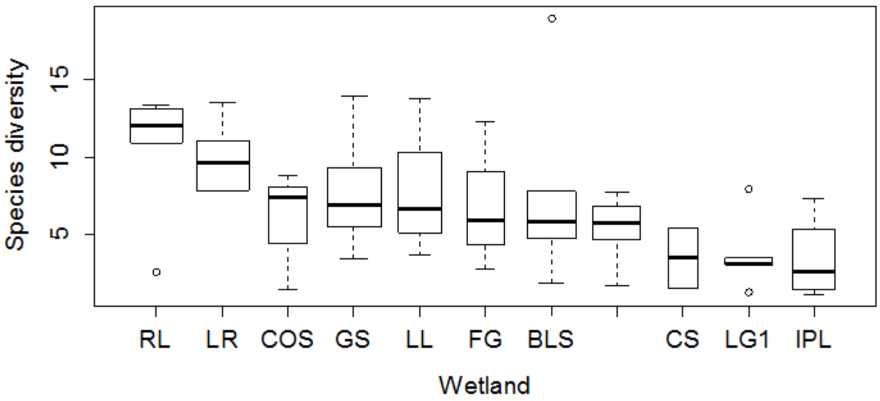


Figure 22 Boxplot of autumnal species diversity (Shannon Exponential Diversity) for Receding phase class wetland samples, ordered by median diversity, Gunbower Forest, 2005-2015.

Flora Composition

Multivariate analyses (NMDS ordinations) were used to explore similarities in native flora composition in the wetlands, surveyed across the 11 years (Figure 23). Samples were coded by factors such as 'sample year', 'wetland phase class', 'water source', 'Treatment' (2014 eFlow) and 'Impact' (i.e. flooded in the preceding 12 months) in order to help visualise each factor's influence on the flora composition. The aim was to identify groupings of wetlands in the ordination plots that corresponded with the factors listed above in order to assist the interpretation of the floristic data. A measure of the dominance of aquatic-amphibious species in each wetland sample (total % cover of PFGs 1-4) was also overlaid as bubbles on the ordination plots in a way that reveals any correlation of these species with the wetland grouping (Figures 23f).

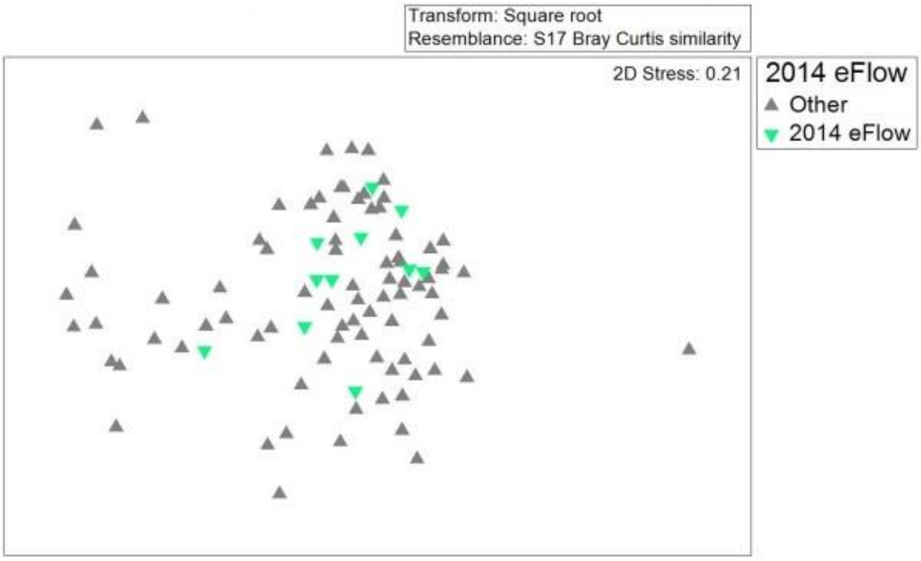
Note: the 'stress' of NMDS plots should be <0.2 in order to be reliable. The lower the stress, the better the two dimensional plot represents the high dimensional assemblage structure of the dataset (Clarke & Gorley 2006). Two dimensional plots have been included in the current report for ease of interpretation, even though they have a stress of 0.21. The documented explanation of these plots was, however, checked against three dimensional plots, with a stress of 0.15, in order to ensure that their interpretation is reliable.

Treatment (samples surveyed before or after the 2014 eFlow) did not appear to result in any distinct groupings of wetlands (Figure 23a). This suggests that the flora composition of the wetlands sampled after the 2014 eFlow (2015) was not distinctly different to that within wetlands sampled in previous years (2005-2015). This outcome reflects the species diversity result in Table 9, in which there was no significant difference between wetlands sampled before and after the eFlow was delivered.

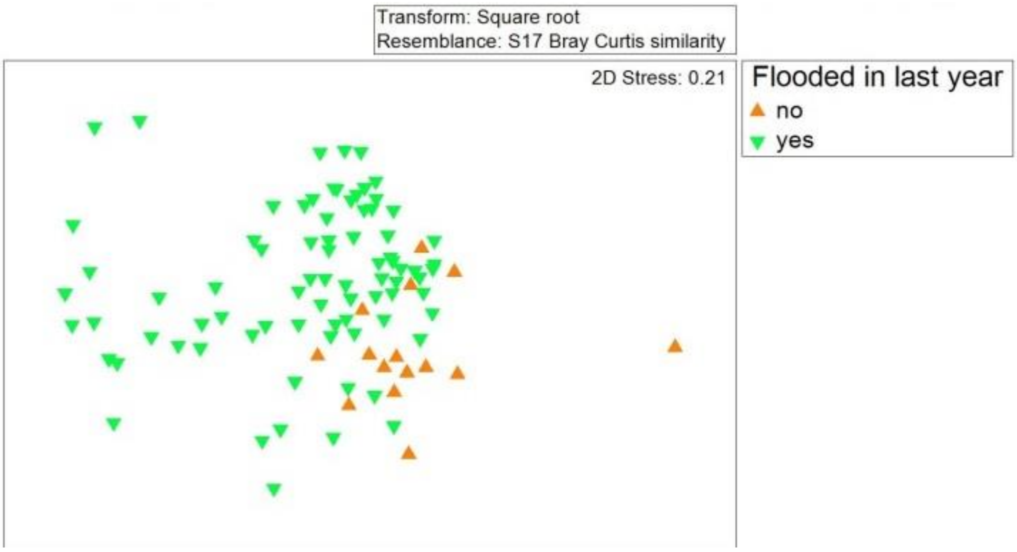
Factors that did appear to stratify the wetland samples to some degree included Impact ('yes' wetlands were flooded in the 12 months prior to sampling, or 'no' they were dry during this period) (Figure 23b), sample year (Figure 23c), water source (Figure 23d) and wetland phase class (Figure 23e), although the samples were at times widely spread (indicating floristic dissimilarity) and overlapping (indicating floristic similarity). Analysis of these factors enabled the detection of broad groups of wetlands.

The first group of wetlands includes the widely spread samples on the left side of plot, which were surveyed in 2011 and 2012, naturally flooded and classed as Recently Inundated or Receding wetlands (Figures 23e-d). The distance between these samples indicates that the flora composition of the wetlands were both dissimilar to each other *and* dissimilar to the remaining wetlands. The second group of wetlands are loosely clustered just below the centre of the plot and included samples that were typically surveyed after drier periods (e.g. 2008 and 2013, as per Figure 3), *or* were dry when surveyed, *or* had not been flooded in the previous 12 months (Figures 23c, 23b & 23e). The third group are clustered just above the centre of the plot and were predominantly surveyed after more mesic conditions (e.g. 2005, 2006, 2010 and 2014, as per Figure 3). They were classed as Receding wetlands that had been inundated via regulators and/or small natural inflows

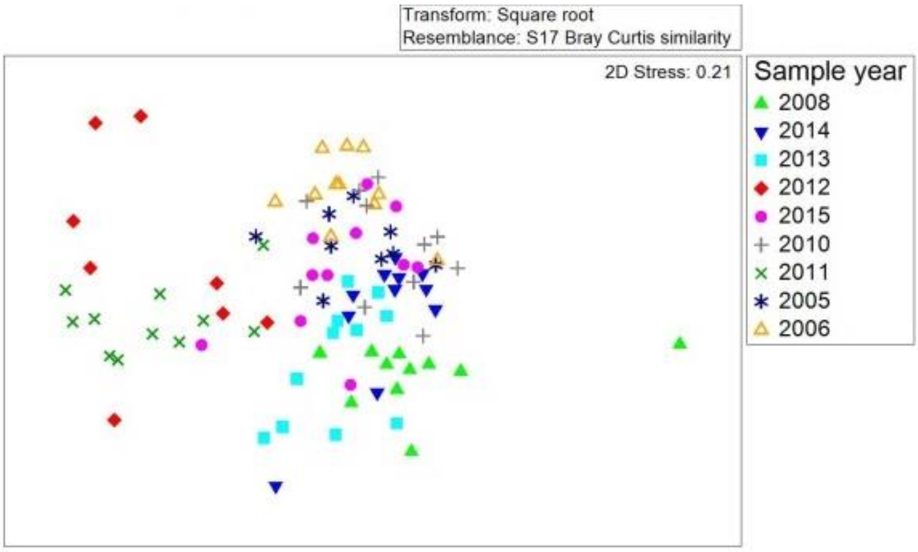
(Figures 23c-e). There is, however, some overlap between the second and third groups, indicating that they represent part of a gradient between wet and dry conditions.



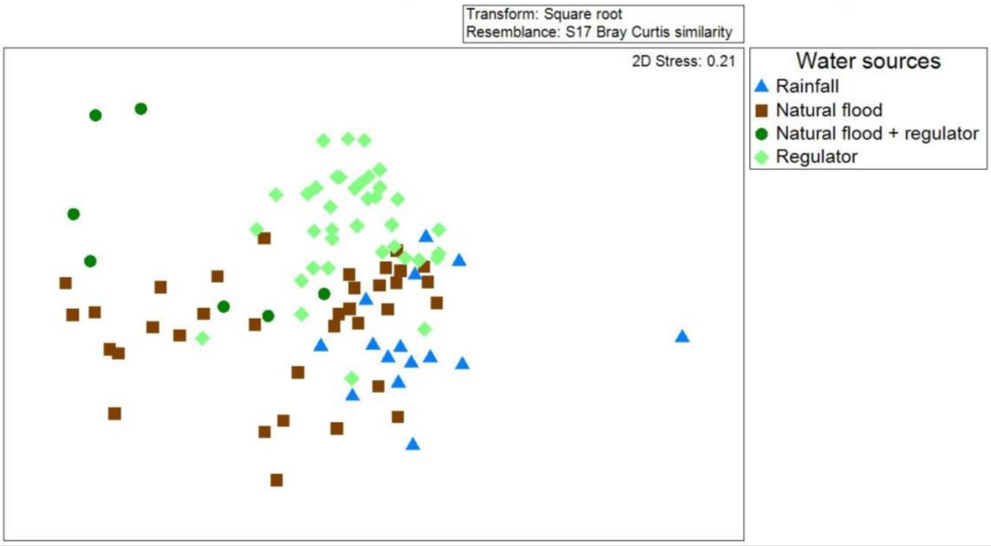
(a)



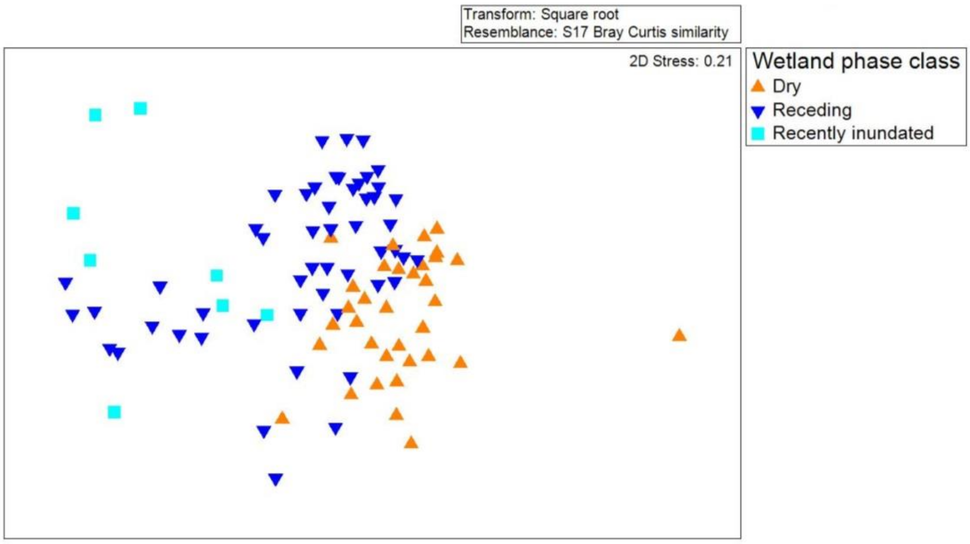
(b)


Transform: Square root
Resemblance: S17 Bray Curtis similarity
2D Stress: 0.21
Sample year
2008
2014
2013
2012
2015
2010
2011
2005
2006

(c)


Transform: Square root
Resemblance: S17 Bray Curtis similarity
2D Stress: 0.21
Water sources
Rainfall
Natural flood
Natural flood + regulator
Regulator

(d)


Transform: Square root
Resemblance: S17 Bray Curtis similarity
2D Stress: 0.21
Wetland phase class
Dry
Receding
Recently inundated

(e)

(f)

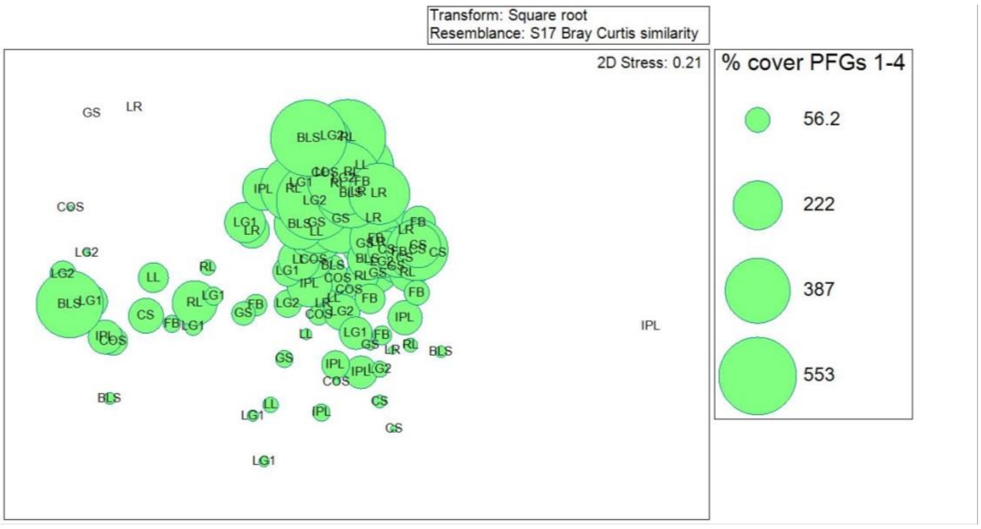


Figure 23 NMDS ordination plots showing all Gunbower Forest wetland sites (autumn 2005-2015), based on native flora cover and arranged by Bray-Curtis similarity. Sampling units are coded by (a) Flooded/not flooded in 2014 eFlow ('other' 2014 and 2015 sites not flooded by 2014 eFlow; 2015 sites flooded by 2014 eFlow), (b) Impact (Yes - flooded in last 12 months or No – not flooded in last 12 months), (c) sampling year, (d) source of water, (e) wetland phase class, (f) bubble plot with scaled bubbles representing total % cover of aquatic and amphibious species (PFGs 1-4).

The cover of aquatic and amphibious species (i.e. bubbles of PFGs 1-4) appears to correspond to this moisture gradient (Figures 23e and 23f) with larger total cover of characteristic wetland species above the centre of the plot and lower cover below the centre of the plot. Higher covers of aquatic and amphibious species also overlapped wetlands that were sampled in 2011 (after widespread natural flooding), although previous analysis of this data suggests that this result was driven by the free floating *Azolla* fern and does not reflect a diverse wetland flora (Bennetts & Jolly 2011).

PFG Species Richness Indicator

The proportions of Receding and Dry wetland sites that complied with the PoRs for the characteristic PFG species richness Index between 2005 and 2015 are presented in Figure 24. For each site, area-weighted species richness is considered 'healthy' (i.e. compliant) if it is on or above the wetland phase class's PoR curve. (Refer to Section 2.1.4.) Species that are considered characteristic include submerged, floating, amphibious and, in semi-permanent wetlands, mudflat flora (PFGs 1-4, Table 8).

Receding wetlands supported sites with healthy species richness in two of the eight years in which they were sampled (Figure 24). (Note that no wetlands were no classified as Receding in 2012.) Compliant Receding wetlands included Black Swamp, Long Lagoon and Reedy Lagoon in 2010 (*n* = 6), and Greens Swamp and Reedy Lagoon in 2015 (*n* = 11).

Dry wetlands only supported sites with healthy species richness in 2010 (Figure 24). Compliant Dry wetlands comprised Football Grounds, Greens Swamp and Little Reedy Lagoon (*n* = 5). No wetlands, however, were classified as Dry in 2012 and 2015.

The conditions preceding the 2010 survey included above average spring rainfall in 2009 (see Technical Addendum) and water delivery to wetlands inundated via the Reedy Lagoon and the Little Gunbower regulators (Figure 8, Table 7). Inundated wetlands include compliant sites (Black Swamp, Football Ground, Long Lagoon and Reedy Lagoon) and non-compliant sites (Little Gunbower Creek, Little Gunbower Complex and Iron Punt Lagoon). Moreover, these wetlands possibly received up to four separate eFlows, delivered through the Little Gunbower regulator, in the two years preceding the 2010 survey (Figure 8). Similar conditions proceeded the 2005 and 2006 samples (i.e. high spring-summer rainfall and the delivery of at least one eFlow, Figures 2 and 8), yet there were no compliant sites.

The conditions preceding the 2015 survey were typically hot and dry (i.e. above average summer temperatures and below average rainfall, as per Figures 6 and 7). The wetlands, however, were flooded with environmental water from mid-2014 until early 2015. There was also a small natural inflow around spring 2014 (Figure 2).

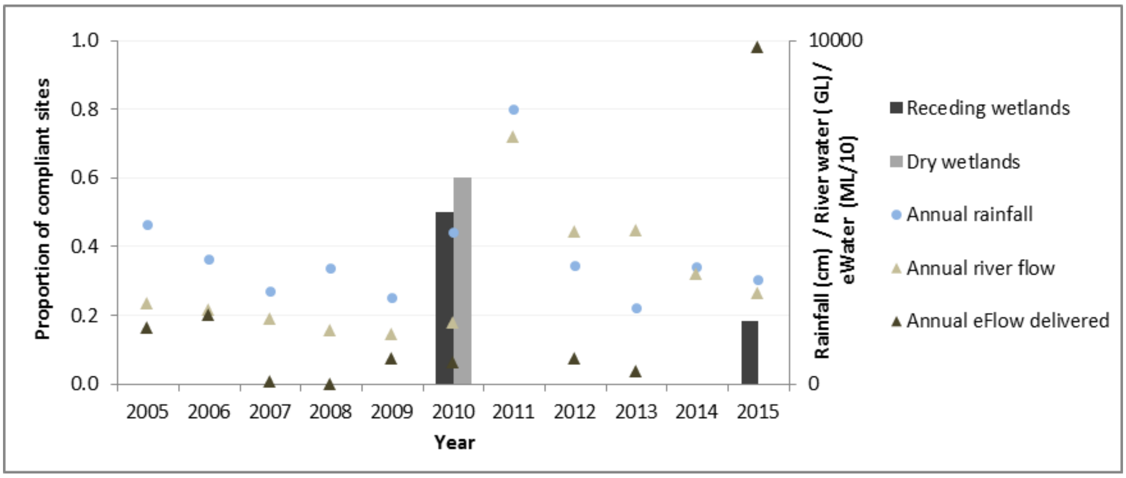


Figure 24 Number of sites that complied with the Species Richness Index PoR in Receding (*n* = 61) and Dry (*n* = 26) wetland sites; annual rainfall (autumn-autumn); annual river flow recorded at Torrumbarry Weir; and annual environmental water delivered to the wetlands, Gunbower Forest, autumn 2005-2015.

Note that years 2007 and 2009 were not sampled and 2012 wetlands were classified as recently inundated and not analysed.

Reedy Lagoon Case Study

Reedy Lagoon is a striking place, with giant pre-European River Red Gums marking the high water line and providing hollows and perches for fauna species that frequent the horseshoe-shaped wetland. The wetland is somewhat of an outlier, as it is species rich and offers a diversity of habitats, making it a priority for management.

Reedy Lagoon maintains the highest richness of native flora species of the wetlands sampled in the Gunbower Forest (e.g. see Figure 22). It is also one of the most species rich wetlands in the region (Damien Cook, 2015, Rakali Consulting, pers. comm. 3 August). Furthermore the lagoon has supported high covers of aquatic macrophytes, often considerably more so than other wetlands monitored. Reedy Lagoon consequently regularly looks lush. This was particularly true in when sampled in 2005, 2006 and 2015 (e.g. Figure 23f), after the wetland had been inundated with environmental water and/or small natural inflows in the two years prior to sampling.

Possibly coincidental, but worth noting, the River Murray carp population was at its lowest in 2005 and 2006, when sampled between 1999 and 2013 (Koehn 2015). Exclusion fencing was also installed in prior to the 2015 survey to prevent large carp from entering the wetland when flooded in 2014-2015. It is possible the reduction in local carp numbers allowed aquatic vegetation to flourish in the wetland.

The diversity of species in Reedy Lagoon is reflected in the array of Ecological Vegetation Classes (EVCs) recognised at the wetland, which highlights its habitat values. While inundated in 2015, seven EVCs were recognised, including (listed from the terrestrial to aquatic zones):

- Floodway Pond Herbland (EVC 810) - characterised by herbs that germinated in wet mud as flood water recedes
- Spike Sedge Wetland (EVC 819) - typified by swards of straw-like Spike-sedge (*Eleocharis acuta*)
- Tall Marsh (EVC 821) - indicated by thickets of Giant Rush (*Juncus ingens*) and/or Common Reed (*Phragmites australis*)
- Floodplain Grassy Wetland (EVC 809) - recognised by a dense cover of the River Swamp Wallaby-grass (*Amphibromus fluitans*)
- Aquatic Herbland (EVC 653) - that includes soft stemmed plants that grow both above and below the water's surface
- Submerged Aquatic Herbland (EVC 918) - composed of flora species that complete their entire life cycle under water
- Dwarf Floating Aquatic Herbland (EVC 949) – represented by small floating species including ferns and liverworts

There is a range of possible reasons why Reedy Lagoon is more diverse than other wetlands. For example, it was set aside as a 'sanctuary' in the 1930s (Jenny Bottcher 2013, local resident, pers. comm. 23rd April), which presumably limited timber harvesting, but not other activities such as shooting and soil disturbance (i.e. digging up earth ovens). The wetland was fenced off from stock in the 1970s (Dick Tresize 2013, local resident, pers. comm., 24th April) but so were other less diverse areas in Gunbower Forest. There is some mention of Reedy Lagoon been watered historically in compensation for hydrological changes caused by river regulation (Eddie Munroe, 2015, local resident, pers. comm. 25th July). There is, however, limited documented evidence of such, other than rain rejection flows which were diverted into various areas in the forest (VEAC 2006).

The density of Aboriginal earth ovens and birthing trees around the wetland, indicate it was an important cultural site well before Europeans settlement. This suggests the lagoon was a reliable source of food and other resources and was, potentially, managed for such. It is also possible the lagoon's location beside Gunbower Creek (increasing its chances of inundation prior to river regulation) and/or the lagoon's bathymetry have promoted the wetland's floristic diversity.

The flora diversity observed in Reedy Lagoon indicates the wetland retains a rich seedbank which contains species uncommon in other wetlands. Included in the seed bank is the nationally vulnerable River Swamp Wallaby-grass (*Amphibromus fluitans,* see case study photograph below) and state threatened Wavy Marshwort (*Nymphoides crenata*, see report cover page).

Reedy Lagoon is clearly both an important ecological and cultural asset. The site should therefore be actively managed to ensure the lagoon, not to mention Gunbower Forest, retains these unique values.

Case study photograph: Reedy Lagoon with a green carpet of the nationally vulnerable River Swamp Wallaby-grass (*Amphibromus fluitans*) between the deep water in the foreground and the trees and rushes in the background.

2.2.2 Red Gum Forest and Woodlands

Figure 25: Red Gum FDU vegetation during the recession of the 2014 eFlow, Gunbower Forest, January 2015.

Gunbower Forest supports an area of 13,020 ha that is dominated by River Red Gum (*Eucalyptus camaldulensis*). The Red Gum broad vegetation type is mapped into two WRCs - Red Gum with Flood Dependent Understorey (Red Gum FDU, Figure 25 above) and Red Gum with Flood Tolerant Understorey (Red Gum FTU). The two WRCs form part of a continuum along a hydro-geographical gradient with Red Gum FDU typically in more frequently inundated areas and Red Gum FTU in less frequently inundated areas.

The following section presents the results from the univariate and multivariate analyses for the Red Gum vegetation type. It commences with a brief overview of the general condition of Red Gum vegetation in 2015, followed by the species diversity, flora composition and vegetation condition indicator results.

General Condition

When sampled in 2015, there was a visible difference in the cover and composition of the Red Gum understorey vegetation between areas that were inundated in the 2014 eFlow and areas that remained dry (Figures 26-31). Evidence of inundation was recorded at 36.4% (28/77) of Red Gum monitoring sites (as per Section 2.1.1) and an additional three sites possibly benefited from nearby flooding (i.e. <20 m). The majority (28) of these sites are mapped as Red Gum FDU vegetation. All sites, however, were dry when sampled.

A total of 124 plants identified to species level were recorded at the 77 monitoring sites in autumn 2015. 66% of these were native. This represents the third highest annual species richness after the 2011 and 2012 samples, which were surveyed after widespread natural flooding. The list includes five new species, although all of these were exotic and/or uncharacteristic terrestrial species. Also included are seven previously observed species which are of conservation significance, including the:

- Nationally vulnerable River Swamp Wallaby-grass (*Amphibromus fluitans*);
- Rare Blue Burr-daisy (*Calotis cuneifolia*) and Smooth Minuria (*Minuria integerrima*);
- Poorly known Smooth Plains Joyweed (*Alternanthera* sp. 1 (Plains)), Blue-rod (*Stemodia glabella s.s.*), Native Couch (*Cynodon dactylon var. pulchellus*) and Native Peppercress (*Lepidium pseudohyssopifolium*).

Panoramic photographs on the following pages provide a visual account of the Red Gum monitoring sites in 2015 and 2011. Sites 25-26, 51-52 and 66-68 have been paired to contrast the striking differences in the understorey vegetation between nearby sites that were flooded in the 2014 eFlow and those that remained dry. (See Figures 26-31.) Sites 5, 42 and 60 have been included in order to highlight differences in the understorey vegetation after both natural flooding (2011) and inundation resulting from the 2014 eFlow (2015). (See Figures 32-34).

Figure 26 Grassy Riverine Forest (Red Gum FDU) site 25 after flooding with 2014 eFlow, Gunbower Forest, autumn 2015.

Figure 27 Grassy Riverine Forest (Red Gum FDU) site 26, not flooded in 2014, Gunbower Forest, autumn 2015.

Figure 28 Red Gum FDU site 51, flood runner after inundation with 2014 eFlow (note the cover of the red *Myriophyllum papillosum*), Gunbower Forest, autumn 2015.

Figure 29 Red Gum FDU site 52, flood runner not inundated in 2014, Gunbower Forest, autumn 2015.

Figure 30 Grassy Riverine Forest (Red Gum FDU) site 66 after flooding with the 2014 eFlow, Gunbower Forest, autumn 2015.

Figure 31 Sedgy Riverine Forest (Red Gum FTU) site 68, not flooded in 2014, Gunbower Forest, autumn 2015.

Figure 32 Riverine Swampy Woodland (Red Gum FTU) site 5, after natural flooding in 2010 (2011, top) and no flooding in 2014 (2015, bottom), Gunbower Forest, autumn 2015.

Figure 33 Grassy Riverine Forest (Red Gum FDU) site 17 after natural flooding in 2010 (2011, top) and flooding with 2014 eFlow (2015, bottom) Gunbower Forest, autumn.

Figure 34 Riverine Swamp Forest (Red Gum FDU) site 42, after natural flooding in 2010 (2011, top) and flooding with 2014 eFlow (2015, bottom) Gunbower Forest, autumn.

Species Diversity

Median native flora species diversity calculated for Red Gum sites sampled between 2005 and 2015, is presented in Figure 35. Sites are grouped by Treatment, as 'B' before the 2014 eFlow (sampled 2005-2014) or 'A' after the 2014 eFlow (sampled 2015); and Impact with 'yes' for flooded or 'no' for not flooded in the last 12 months. The majority (98%) of 100 flooded sites surveyed before the 2014 eFlow (i.e. yes_B) were sampled in 2011 and 2012. Hence, this group is thought to reflect the outcome of widespread natural flooding between 2010 and 2012.

The sites sampled in 2015 that were flooded in the 2014 eFlow (yes_A) had a slightly higher median diversity than the 2015 samples that did not flood (no_A). A similar effect can be seen in the 2005-2014 samples (yes_B vs no_B), suggesting that flooding in the previous 12 months was correlated with higher median diversity in both cases. Median diversities recorded in 2015 (yes_A and no_A) were, however, marginally lower than the median 2015-2014 values (yes_B and no_B), which were influenced by the very high diversity year of 2011 (outlier dots above 'B' boxes).

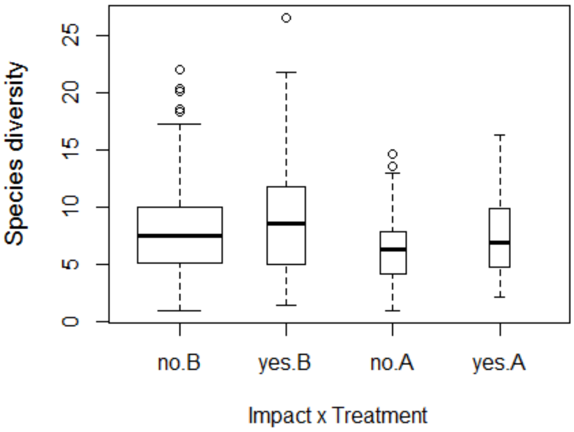


Figure 35 Boxplot of autumnal species diversity (Shannon Exponential Diversity) for Red Gum vegetation grouped by the Treatment (B, before/A, after 2014 eFlow) x Impact (yes/no flooded in 2014 eFlow) interaction, Gunbower Forest, 2005-2015.

Note the 'box' contains the middle 50% of samples and the box width indicates the sample size it represents. The median is represented by the black line inside the boxes. The top and bottom 'whisker' represents the top and bottom 25% of samples and outliers samples are shown as dots.

Further analysis of the data using LMEMs, suggests that there were significant differences in native species diversity between sites thate flooded and those that did not flood (Impact), and in the interaction between eFlow Treatment (B/A) and Impact (flooding), (Table 10). Therefore, it can be inferred that, across the monitoring period (2005-2015), sites which flooded in the previous 12 months had significantly higher mean species diversities than sites which remained dry (Figure 35).

Table 10 LMEM output for transformed Red Gum data (significant results in bold), Gunbower Forest, autumn 2005-2015.

	Value	Std. Error	DF	t-value	p-value
(Intercept)	2.4883	0.5688	8.2000	4.374	**0.00226**
Treatment	0.6708	0.5946	7.9000	1.122	0.29242
Impact	0.7112	0.2199	592.6000	3.235	**0.00129**
Treatment x Impact	-1.0434	0.2617	584.7000	-3.987	**7.53e-05**

A post-hoc Tukey's test found that the significant difference in the interaction term (Treatment x Impact) lay between no_A and yes_A, implying that sites sampled in 2015 that were flooded in the 2014 eFlow had significantly higher species diversity than sites sampled in 2015 that were not flooded (Table 11, Figure 35). There were also no significant differences between B_yes and A_yes samples, which indicates the mean diversity of species in sites that were flooded in the 2014 eFlow was similar to those flooded in the 2010-2012 natural flood.

Table 11 Post-hoc Tukey's test for Red Gum LMEM (significant results in bold), Gunbower Forest, autumn 2005-2015.

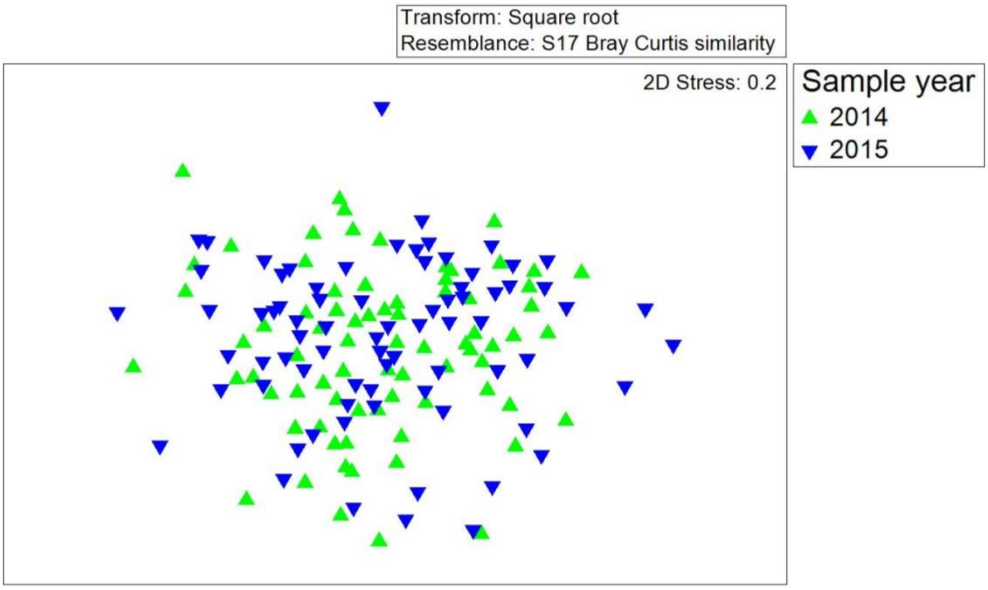
Interaction	Estimate	Std. Error	z value	Pr(>|z|)
B_no - A_no	0.67079	0.59458	1.128	0.62614
A_yes - A_no	0.71123	0.21988	3.235	**0.00534**
B_yes - A_no	0.33859	0.61095	0.554	0.93416
A_yes - B_no	0.04044	0.60588	0.067	0.99987
B_yes - B_no	0.33220	0.15895	-2.090	0.12561
B_yes - A_yes	0.37265	0.61782	-0.603	0.91717

Flora composition

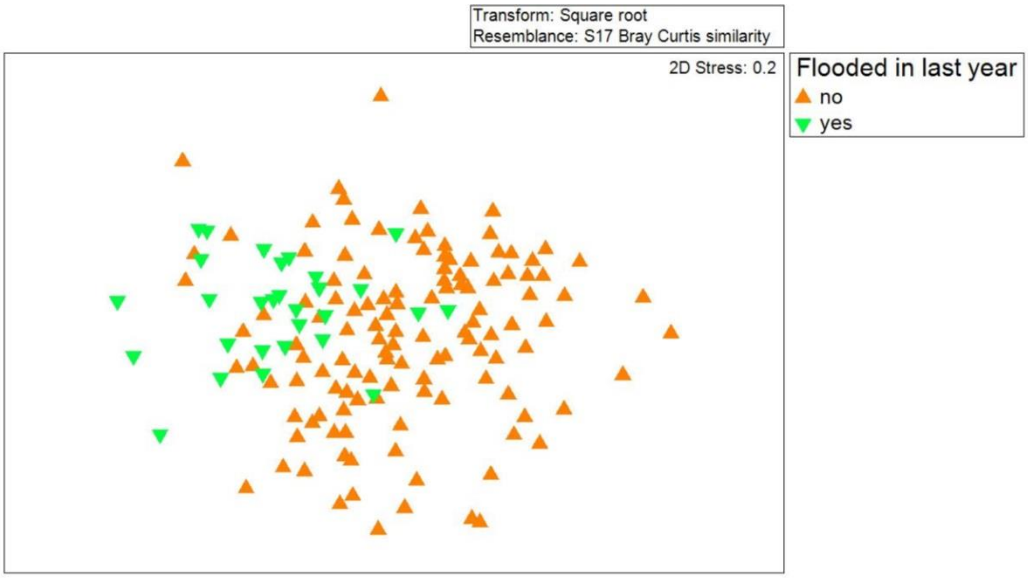
Similarities in flora composition between the 2014 and 2015 Red Gum sites were explored using multivariate analyses (NMDS ordinations) (Figure 36). Samples were coded by 'sampling year' and 'Impact' (flooding) to help visualise the influence of these factors on the flora composition. Estimated maximum water depth from the 2014 eFlow and a measure of aquatic, amphibious and mudflat species dominance (total % cover of PFGs 1-4) at each site were overlaid as bubbles on the plots to illustrate the correlation of these variables.

The ordination of the data suggests that there was minimal difference in flora composition between the 2014 and 2015 samples (Figure 36a). When sites were classified by Impact, flooded and non-flooded sites formed loose, overlapping groups (Figure 36b), indicating that the sites inundated by the 2014 eFlow supported similar but slightly different compositions of species to the sites that remained dry in 2014.

However, a distinct pattern emerges in the ordinations when water depth and cover of aquatic, amphibious and mudflat species (PFGs 1-4) are overlaid on the data (Figures 36b-d). There appears to be a correlation between the sites that were flooded by the 2014 eFlow to estimated maximum inundation depth, and higher cover of these desirable floodplain species. The grouping of these sites to the left of the plot suggest that they supported different species composition, compared to the dry sites on the right, which had little or no evidence of inundation at the time of sampling and limited or no cover of aquatic, amphibious and mudflat species.



(a)



(b)

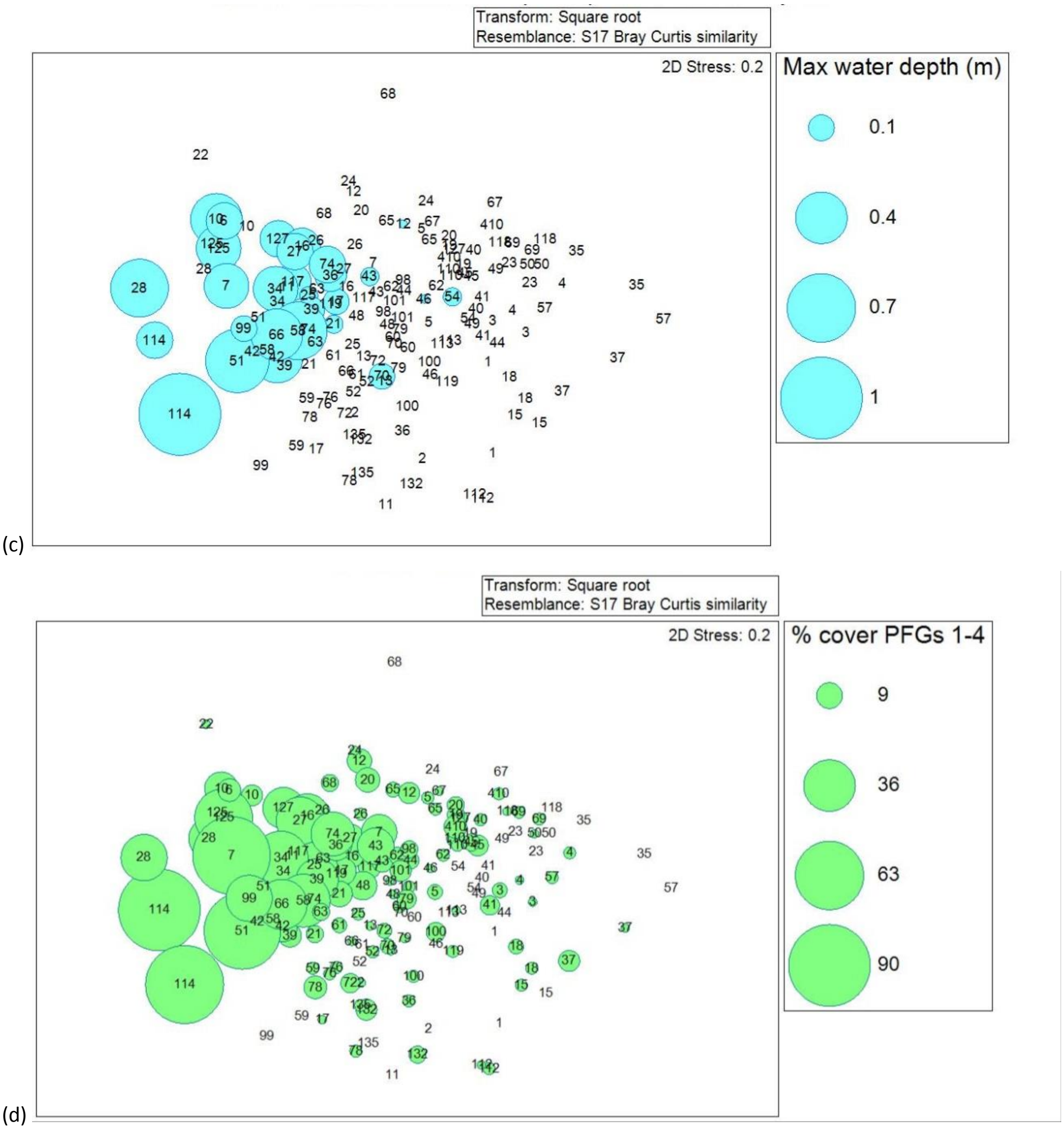


Figure 36 NMDS ordination plots showing Gunbower Red Gum vegetation monitoring sites (each quadrat on each sampling date) from 2014 and 2015, based on native flora cover and arranged by Bray-Curtis similarity. Sampling units are coded by (c) sampling year, (b) Impact (Yes - flooded in last 12 months (2014 eFlow) or No – not flooded in last 12 months), (c) bubble plot with scaled bubbles representing maximum depths recorded in the past financial year (m) and (d) bubble plot with scaled bubbles representing % cover of aquatic species (PFGs 1-4).

PFG Species Richness Indicator

For each WRC, understorey sites that are considered ‘healthy’ (i.e. ‘compliant sites’) have a total species richness equal to or above the 90th percentile of all native characteristic PFG species richness recorded across the 2005-2014 sampling period (PoR). The PoR for the Red Gum FDU WRC is nine adaptive and emergent amphibious (PFGs 3 and 5) and mudflat (PFGs 4a and 4b) species. Whereas the PoR for the Red Gum FTU WRC is 11 mudflat, emergent amphibious and terrestrial damp (PFG 6) flora species. The proportions of Red Gum FDU and FTU sites that complied with the PoRs for the characteristic PFG species richness Index between 2005 and 2015 are presented in Figure 37.

The pattern in compliant Red Gum sites for the characteristic PFG species richness Index appears to correspond to the pattern of rainfall and river flow at Torrumbarry Weir (which is a surrogate measure for natural flooding) (Figure 37). For example, there was a considerable increase in the number of sites credited with a healthy PFG species richness Index score in 2011, after a peak in rainfall and river flow, followed by a decline that mirrored the combined hydrological conditions between 2012 and 2014. In 2015, after the 2014 eFlow, there was a three-fold increase in the proportion Red Gum FDU compliant sites compared to that recorded in 2014. The pattern in proportion of healthy Red Gum FTU sites was similar to the pattern calculated for Red Gum FDU site, the exception being 2015 when there were no compliant Red Gum FTU sites.

When the WRC Index results are combined, 15.6% of the 2015 Red Gum sample were determined to have healthy species richness, an increase of 4.6% in 2005, but less than the 50.6% and 26.7% recorded in years 2011 and 2012 respectively. (See Technical Addendum.)

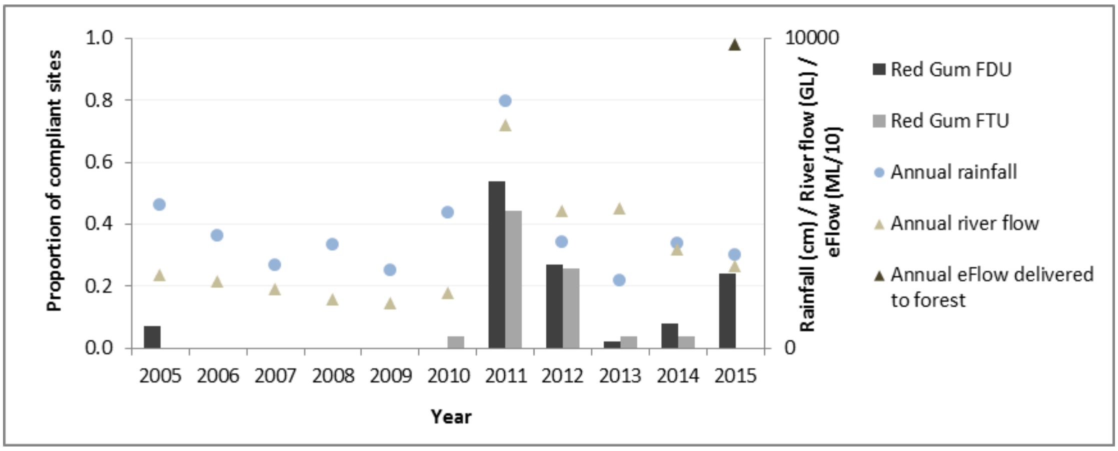


Figure 37 Proportion of sites that complied with the Species Richness Index PoR in Red Gum FDU (*n* = 42, 2005 - 2006; *n* = 50, 2008–2011 and 2013-2015, and *n* = 48 2012) and Red Gum FTU (*n* = 23, 2005-2006; *n* = 27, 2008-2014) WRCs, annual rainfall (autumn-autumn), annual river flow recorded at Torrumbarry Weir and annual environmental water delivered, Gunbower Forest, autumn 2005-2015.

Note years 2007 and 2009 were not sampled.

Tree Canopy Indicator

For each treed WRC, tree canopy health is considered ‘healthy’ if it is on or above the 90th percentile of all Index values between 2005 and 2014 (PoR). The PoR for the Red Gum FDU WRC is 80% of trees per sites that have at least 50% intact canopy (i.e. tree crown health category over 3, Table 4). For Red Gum FTU the PoR is 81.5% of trees per site in this health range. The tree canopy Index results for the Red Gum FDU and FTU WRCs between 2005 and 2015 are presented in Figure 38.

The proportion of Red Gum sites with healthy canopy Index scores was below 20% in all years apart from 2005. The decline in the proportion of compliant sites between 2005 and 2006 is substantial, and although there was a decrease in rainfall between the two years, the difference is thought to be more likely due to different observers than a significant change in canopy condition. Between 2006 and 2015 (sampled by a consistent team of observers), there appears to be a gradual increase in the proportion of sites with healthy canopies in both WRCs. Consequently, it is assumed that the current percentage of sites with a healthy tree canopy actually represents an increase in tree canopy health based on the 2006 value. Interestingly, the proportion of compliant sites was higher for Red Gum FTU than Red Gum FDU between 2005 and 2008, but that the reverse was true from 2010 onwards.

Combining the WRCs' Index results suggests that 16.9% of the 2015 Red Gum sites had healthy tree canopies, which was the highest figure since 2005 (43.1%). (See Technical Addendum.)

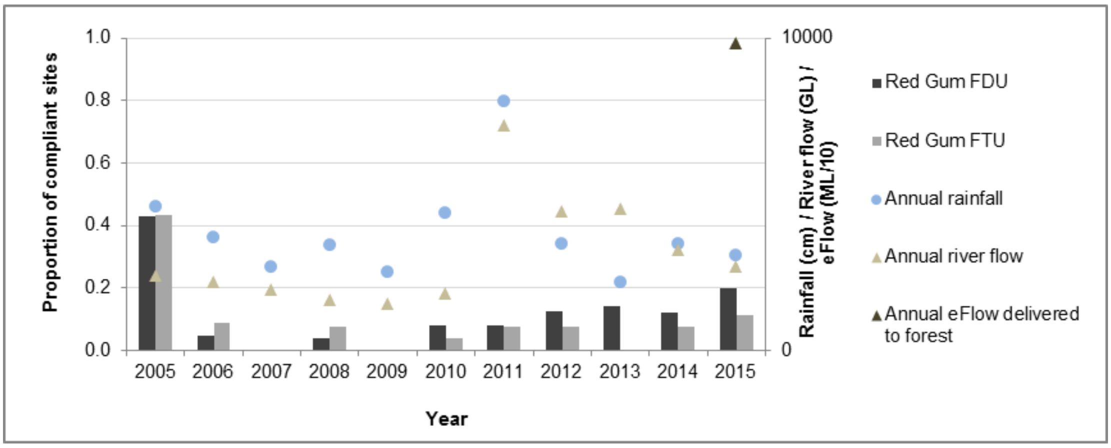


Figure 38 Proportion of sites that comply with the Tree Canopy Health Index PoR in Red Gum FDU (*n* = 42, 2005-2006; *n* = 50, 2008–2011 and 2013-2015, and *n* = 48 2012) and Red Gum FTU (*n* = 23, 2005-2006; *n* = 27, 2008-2015) WRCs, annual rainfall (autumn-autumn), annual river flow recorded at Torrumbarry Weir and annual environmental water delivered, Gunbower Forest, autumn 2005 – 2015.

Note that years 2007 and 2009 were not sampled.

2.2.3 Box Woodlands

Gunbower Forest is mapped as supporting 4894 ha of Box woodland that are characterised by the presence of Black Box (*E largiflorens*) and Grey Box (*E. microcarpa*) trees. Both species span a hydro-geographical gradient from frequently inundated (i.e. at least two years over a three year period, for example, Riverine Swampy Woodland to rarely inundated (i.e. once every 10 – 25 years e.g. Riverine Chenopod Woodland (Fitzsimons *et al.* 2011)) to almost never inundated (e.g. Plains Woodland). They are, however, mapped as discrete WRCs, based on the dominant tree species.

Only one of the 33 Box woodland sites was inundated by the 2014 eFlow (Black Box site 77, Figure 39), and hence, only a brief summary of the results for this vegetation type is presented in the current report. What is included, however, is an overview of the general condition, panoramic images of the monitoring sites over time and under different conditions, and the vegetation condition indicator results.

General Condition

When surveyed in autumn 2015, the Black Box and Grey Box woodlands were notably dry with scattered hardy perennial understorey species. A total of 100 plants were recorded to species level, 81 of which were native. The 2015 richness is the third highest score for the woodlands (after 2011 and 2012) since monitoring began.

Six species of conservation significance were observed in the Box monitoring plots in the current project. These included the:

- Rare Blue Burr-daisy (*Calotis cuneifolia*) and Smooth Minuria (*Minuria integerrima*),
- Poorly known Native Couch (*Cynodon dactylon var. pulchellus*), Native Peppercress (*Lepidium pseudohyssopifolium*), Plains Joyweed (*Alternanthera* sp. 1 (Plains)) and Smooth Blue-rod (*Stemodia glabella* s.s.).

The native Fuzzy New Holland Daisy (*Vittadinia cuneata var. hirsuta*) is new to the Box woodland monitoring species list.

Panoramic photographs of a number of the sites are provided on the following pages in order to illustrate the condition of the Black and Grey Box woodland. Figure 39 highlights the influence of flooding in understorey site 77 in 2011 (after natural flooding), in 2014 (no flooding) and in 2015 (after 2014 eFlow). Figure 40 also contrasts flooded (2011) and non-flooded (2015) images of the same site, but in Grey Box woodland. Figures 41-44 include examples of Black and Grey box sites, at either end of the hydro-geographical gradient, from 2005 (monitoring inception) and 2015.

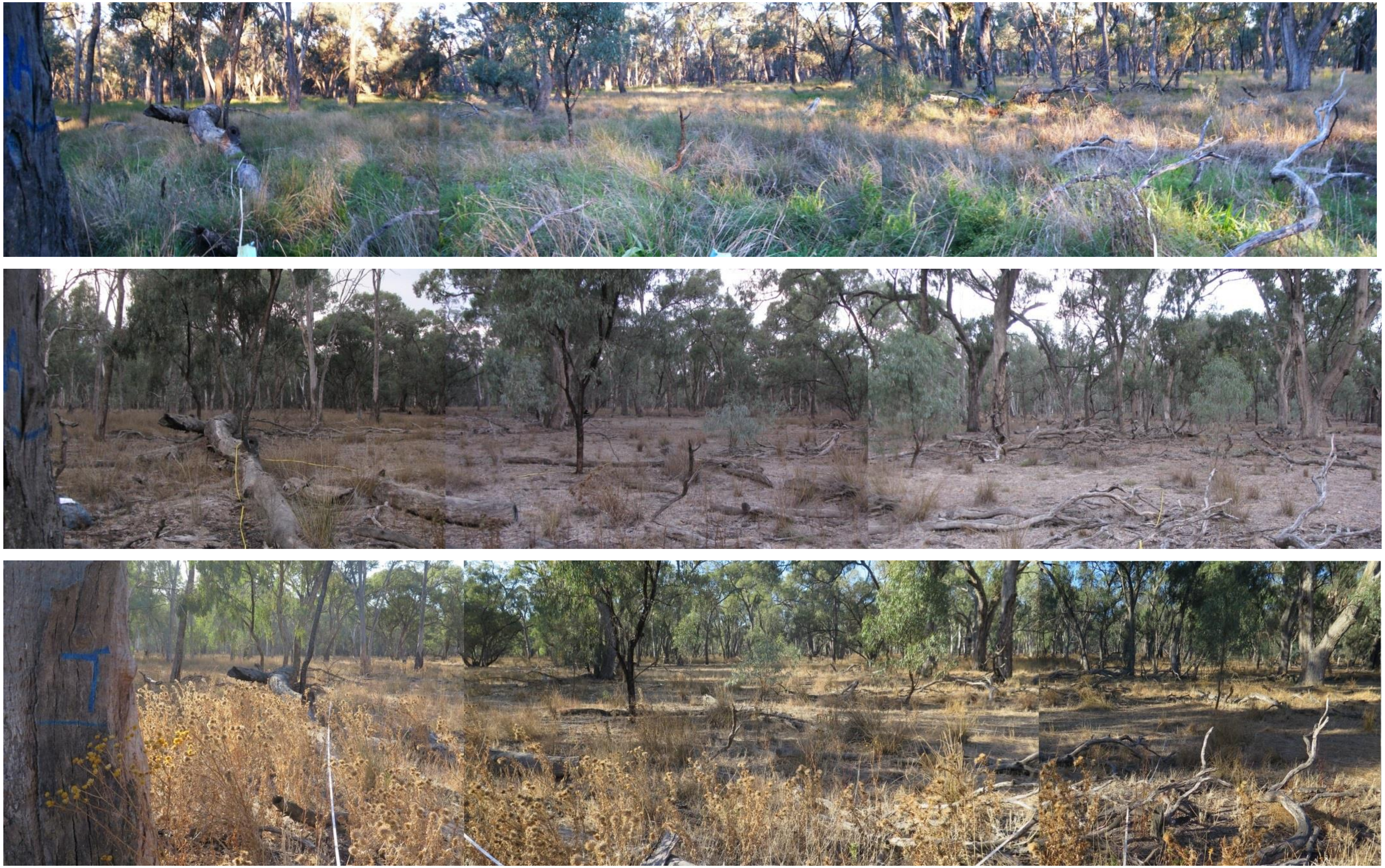


Figure 39 Riverine Swampy Woodland (Black Box) site 77, in order, after flooding in 2010 (2011), no flooding (2014) and after flooding with 2014 eFlow (2015) Gunbower Forest, autumn.

Figure 40 Riverine Swampy Woodland (Grey Box) site 75, after flooding in 2010, (2011, top) and no flooding (2015, bottom), Gunbower Forest, autumn 2015.

Figure 41 Riverine Chenopod Woodland (Black Box) site 29, not flooded, 2005 (top) and 2015 (bottom), Gunbower Forest, autumn 2015.

Figure 42 Riverine Swampy Woodland (Black Box) site 85, not flooded, 2005 (top) and 2015 (bottom), Gunbower Forest, autumn 2015.

Figure 43 Plains Woodland (Grey Box) site 88, not flooded, 2005 (top) and 2015 (bottom), Gunbower Forest, autumn 2015.

Figure 44 Riverine Swampy Woodland (Grey Box) site 93, not flooded (2005, top), after flooding in 2010 (2011, middle) and not flooded (2015, bottom), Gunbower Forest, autumn.

PFG Species Richness Indicator

Understorey sites that are considered ‘healthy’ have a species richness total equal to or above the 90th percentile of all native characteristic PFG species richness across the 2005-2014 sampling period in each WRC (PoR). The PoR for the Black Box WRC is 5.9 mudflat (PFGs 4a and 4b), emergent amphibious (PFG 5) and terrestrial damp (PFG 6) flora species. The Grey Box WRC PoR is 14 emergent amphibious, terrestrial damp and terrestrial dry flora (PFG 7) species per site. The proportion of Box woodlands sites that complied with the PoRs for the characteristic PFG species richness Index between 2005 and 2015 is presented in Figure 45.

The number of Black and Grey Box sites with a healthy richness of characteristic PFG species was greatest in 2011 and 2012, after above average rainfall in 2010-2011 (Figure 45). In the subsequent years, only one Black Box site (77, Figure 39) achieved the Index PoR. This site is situation on an ecotone between Red Gum and Black Box vegetation and was flooded in 2010 (sampled 2011) and 2014 (sampled 2015). The number of compliant Grey Box sites appears to have increased from zero in 2013 to four in 2015. The Grey Box result is intriguing given that only one of the compliant sites was flooded (87 sampled 2011). This suggests that other factors such as rainfall were the key drivers in the 2011 and 2012 outcomes. Rainfall prior to the 2014 and 2015 surveys was not remarkably high, but it was greater than the rainfall recorded in the 12 months prior to the 2013 survey.

When Black and Grey Box Index results are combined, 12.1% of the 2015 sample was determined to have healthy species richness, up from 9.1% in 2014 and 0% in 2005, but down from 36.4% and 30.3% in years 2011 and 2012. (See Technical Addendum.)

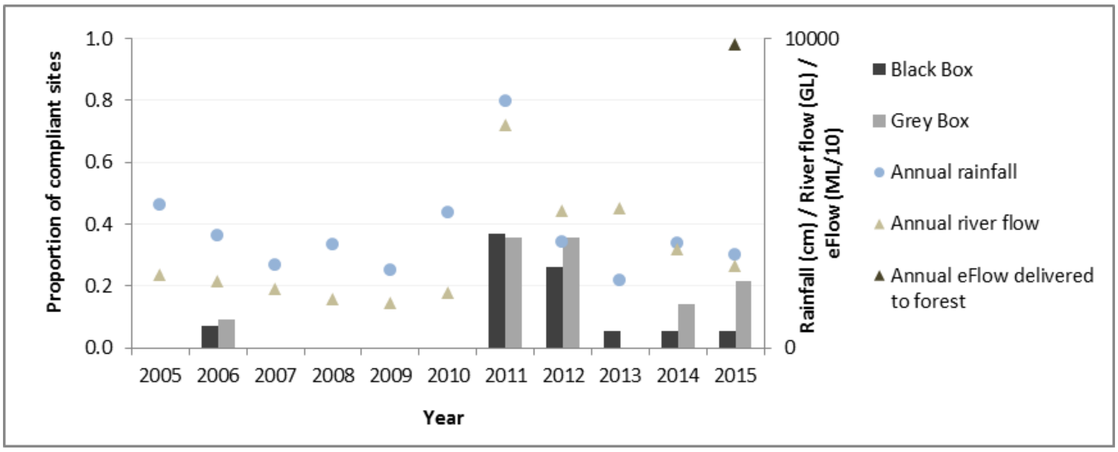


Figure 45 Proportion of sites that complied with the Species Richness Index PoR in Black Box Woodland (*n* = 14, 2005-2006; *n* = 19, 2008-2015) and Grey Box Woodland (*n* = 11, 2005-2006; *n* = 14, 2008 – 2015) WRCs, annual rainfall (autumn-autumn), annual river flow recorded at Torrumbarry Weir, and annual environmental water delivered, autumn 2005 – 2015 Gunbower Forest.

Note years 2007 and 2009 were not sampled.

Tree Canopy Indicator

For each treed WRC, tree canopy health is considered 'healthy' if it is on or above the 90th percentile of all Index values between 2005 and 2014 (PoR). The PoR for the Black Box WRC is 90% of trees per site with at least 50% intact canopy (i.e. tree crown health category over 3, Table 4). For Grey Box the PoR is 95% of trees per site in this health range. The proportion of Black and Grey Box sites that complied with the PoRs for the tree canopy Index between 2005 and 2015 are presented in Figure 46.

The number of Black Box sites with healthy tree canopies declined between 2005 and 2012, but improved in 2013 and 2015 (Figure 46). There was also a pattern of decline in the number of Grey Box sites with healthy tree canopies in the first eight years of monitoring, that is, before the numbers of sites stabilised in 2013 and 2015. Note that, unlike the Red Gum tree canopy results, the 2005 Box woodland tree canopy health Index scores are assumed to be an accurate representation of canopy condition at sampling, as the subsequent year's score was not dramatically different (2006, 32.0%). Given that there were no compliant Black or Grey Box sites in 2014, it is difficult to determine whether the decline in tree health has halted.

When the WRCs' Index results are combined, 15.2% of the Box sites were calculated with healthy tree canopies in 2015, down from 36.0% in 2005. (See Technical Addendum.)

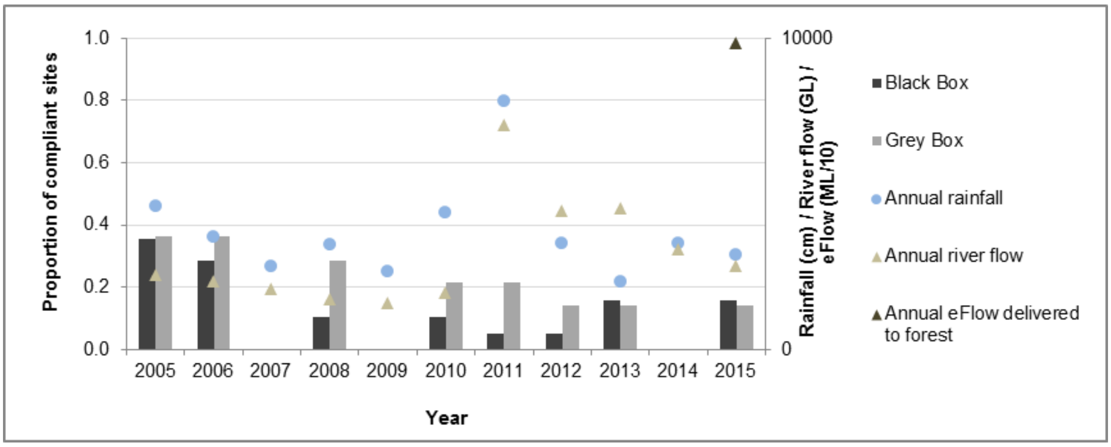


Figure 46 Proportion of sites that comply with the Tree Canopy Health Index PoR in Black Box (*n* = 280, 2005-2006; *n* = 380, 2008-2015) and Grey Box (*n* = 220, 2005-2006; *n* = 280, 2008-2015) WRCs, annual rainfall (autumn-autumn), annual river flow recorded at Torrumbarry Weir and annual environmental water delivered, autumn 2005 – 2015 Gunbower Forest.

Note years 2007 and 2009 were not sampled.

2.3 Discussion

Data from 125 monitoring sites collected in autumn 2015 were compared to a decadal dataset in order to investigate the effect of a large environmental flow delivered to Gunbower Forest in 2014. Specifically, wetland and Red Gum data were analysed to determine if species diversity differed before and after the watering event in the sampled sites. Data were also analysed to explore similarities in native flora composition in the sampled sites, considering factors such as 2014 eFlow (before and after the treatment), water source and cover of native aquatic and amphibious species. The following section discusses the results of this analysis, as well as progress towards the Icon Site's ecological objectives, based on vegetation condition indicator scores for the wetlands, Red Gum and Box woodland vegetation types.

2.3.1 Wetlands

All eleven wetland monitoring sites were inundated by the 2014 eFlow and classified in the 'receding' wetland phase class, based on their hydrological condition at sampling in 2015. The following discussion focuses on the results for Receding wetlands, with consideration of Dry and Recently Inundated wetlands surveyed in previous years, where appropriate.

The 2014 eFlow created hydrological conditions that triggered a welcome resurgence of a range of submerged, floating, amphibious and other wetland flora species in the monitored wetlands. Detailed analysis of the data suggests that there was a similar response to the 2014 eFlow as to previous inundation events. For example, the mean diversity of native species in the Receding wetlands that were surveyed after the 2014 eFlow (2015) was marginally higher, yet not significantly different, to that recorded in Receding wetlands surveyed between 2005 and 2014. Multivariate analysis of the wetland data also indicates that the species composition in the wetlands which were inundated with the 2014 eFlow was similar to other Receding wetlands sampled. This outcome is not surprising, given the range in diversity scores calculated for and the similar hydrological conditions (e.g. maximum water depth and inundation season) recorded in the Receding wetlands between 2005 and 2015.

Multivariate analysis of the wetland data also, however, highlighted an association between Receding wetlands inundated with eFlows and higher cover of native aquatic and amphibious flora (Figures 23 d-f). A similar correlation is evident in wetlands inundated by small natural floods (i.e. in 2013 and 2014) and to some degree the large natural flood (2011). Previous analysis of the 2011 data, however, indicated that the latter outcome was driven by the floating Azolla fern, rather than by a diversity of species (Bennetts & Jolly 2011). These results suggest that eFlows and small natural floods promoted a greater abundance of desirable wetland flora than have large natural floods in the last 11 years.

Not surprisingly, Dry wetlands were not associated with high covers of aquatic and amphibious species in the multivariate analysis. Given the severity of the Millennium drought (1996-2010), it is more than likely that the majority of wetlands sampled over the last 11 years would have remained in this state had eFlows not been delivered. Water delivered to the forest's wetlands during this dry period ensured aquatic habitat for flora, which in turn, is likely to have also offered refuge for fauna.

Plotting native species diversity annually for the Receding wetlands makes it possible to see the effects that different hydrological conditions have made in previous sampling years. The pattern in median species diversity appears to be driven by both the size of the inundation event and prevailing climatic conditions. From an ecological point of view it makes sense that high levels of inundation lead to dominance of fewer favoured aquatic species with lower overall diversity, whereas intermediate flooding promotes greater diversity.

The data analysis also highlights considerable differences in native species diversity between the different Receding wetlands sampled. While not corrected for sample area, these results suggest that Reedy Lagoon had the highest median species diversity, followed by Little Reedy Lagoon, and that Iron Punt Lagoon, Little Gunbower Creek and Charcoal Swamp had the lowest median species diversity (Figure 22). The median species diversities of the remaining six wetlands were relatively similar. While not directly related to the 2014 eFlow, such information might prove useful for prioritising management actions.

Progress Towards Icon Site Ecological Objectives

The overarching objective for permanent and semi-permanent wetlands in Gunbower Forest (as assessed in the current project) is to secure an increase in wetland health. The suggested target for this objective is 100%[3] of monitored wetland sites in healthy condition by 2025 (Table 1). The characteristic PFG species richness Index has been applied to determine health in the wetland samples.

Wetlands with a healthy richness of characteristic PFG species were only recorded in two years (2010 and 2015) for Receding wetlands and only one year (2010) for Dry wetlands. The lower number of healthy wetlands sampled in 2015 (2/11) compared to 2010 (6/11), and the general absence of healthy wetlands in other years surveyed, suggests that wetland health has not tangibly improved between 2005 and 2015 when assessed using the characteristic PFG species richness Index.

Notwithstanding the above, the multivariate analysis suggests that higher covers of aquatic macrophytes were associated with wetlands that received environmental water. The presence of healthy Receding wetlands sampled in 2015 after the 2014 eFlow also suggests that the delivery of environmental water to the wetlands was beneficial. Water delivered in four separate events prior to the 2010 sample (Figure 8) which, along with high spring rainfall in 2009 (see Technical Addendum), seems to have also improved the wetlands' health even when dry. The fact that there were no healthy wetlands identified from 2005-2006 (after eFlow deliveries), 2011-2012 (after widespread natural flooding) or 2013-2014 (after natural inflows) indicates that factors other than water are affecting species richness in the wetlands.

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[3] Percentage of wetland monitoring sites within the modelled Hipwell regulator 1650 ML/day inundation extent.

2.3.2 Red Gum Forest and Woodlands

Just over a third of the 77 Red Gum monitoring sites were inundated to some degree by the 2014 eFlow. Only two of the 28 flooded sites were, however, mapped as the Red Gum FTU WRC, and hence the following text largely refers to the Red Gum FDU WRC.

Red Gum sites inundated in 2014 by water delivered through the Hipwell regulator were found to have significantly higher mean native species diversity (Table 11), and were associated with higher covers of aquatic, amphibious and mudflat species in autumn 2015 than sites which remained dry (Figures 36b and 36d). This suggests that the 2014 eFlow stimulated a diversity of understorey flora characteristic of Red Gum floodplain vegetation in areas that were inundated.

Furthermore, while the median diversity in the flooded 2015 quadrats (yes_A) was lower than that of the quadrats flooded in 2011 and 2012 (yes_B), the difference was not significant (Table 11). This indicates that the engineered flood encouraged a similar level of native flora diversity to the 2010-2012 natural flood. It is, however, likely that the period of active growth associated with the 2014 eFlow was shorter than that associated with the natural floods, given that all sites sampled in 2015 were dry, as opposed to the 2011 and 2012 surveys. (See Figures 31-34.)

Climatic conditions preceding the 2015 survey were hot and dry (Figures 6 and 7). Monitoring sites that were not inundated in the previous 12 months had low covers of (if any) aquatic, amphibious and mudflat species (Figures 36b and 36d). It is consequently unlikely that the forest would have supported the lush floodplain and aquatic vegetation observed in areas that were flooded if the eFlow had not been delivered (Figures 4 and 5). It is, therefore, reasonable to assume that the 2014 eFlow at least temporarily increased habitat complexity on the floodplain, and importantly, buffered the Red Gum vegetation and its associated fauna from the effects of the arid conditions.

Progress Towards Icon Site Ecological Objectives

The overarching objective for Red Gum vegetation in Gunbower Forest (as assessed in the current project) is to maintain vegetation health. For the purpose of evaluating the degree to which this objective is being met, we have defined the term ‘maintain’ to mean - *retain similar PFG species richness, appropriate to the stage of wetting and drying and canopy condition over time to that sampled at the inception of the monitoring program (2005).* The suggested target for this objective is at least 21%[4] of monitored sites in healthy condition by 2025 (Table 1).

The 2015 vegetation condition Index results suggest that Red Gum health has been maintained since monitoring inception and, therefore, meets the overarching objective for the broad vegetation type. The

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[4] Percentage of Red Gum sites within the modelled Hipwell regulator 1650 ML/day inundation extent.

average of the two Index scores (16.2% sites) does not, however, meet the objective target of 21% of sites in healthy condition. When interpreting these results, though, it should, be acknowledged that this outcome averages the results from both frequently inundated Red Gum FDU sites and rarely inundated Red Gum FTU sites, and that vegetation health is likely to vary across this hydro-geological gradient.

2.3.3 Box Woodland

Only one Box Woodland site was inundated in the 2014 eFlow. The following discussion therefore focuses on the vegetation condition Index results.

In summary, the proportion of Box woodlands sites with a healthy richness of characteristic PFG species (i.e. sites achieving the Index PoR) appears to mimic the annual rainfall pattern, peaking in 2011 and 2012, declining in 2013, and in Grey Box woodland, rising again in 2014 and 2015 (Figure 45). It is likely, though, that these results have been influenced by other factors such as flooding and grazing. It is also possible that the increase in Grey Box sites with healthy richness of characteristic PFGs between 2013 and 2015 reflects an increase in the number of terrestrial flora species (PFG 7), which are characteristic of the WRC, but not characteristic of the Black Box WRC.

As seen in the Red Gum vegetation, the Box woodland tree canopy health results appear to follow a different pattern to the understorey strata. Over the 11 year monitoring period, the number of sites supporting healthy tree canopies (i.e. sites that achieved the tree canopy health Index PoR) has actually halved in both Box WRCs. Notwithstanding the absence of sites with healthy canopy in 2014, there does appear to be some plateauing in tree health and even an increase in Box woodland after the mesic conditions recorded in 2010-2012. The earlier decline in tree health was presumably caused by moisture stress experienced during the Millennium drought (1996-2010). High tree densities, resulting from over 130 years of timber harvesting in Gunbower Forest, are likely to have increased competition among trees for moisture and possibly exacerbated the situation.

Progress Towards Icon Site Ecological Objectives

The overarching objective for Black and Grey Box vegetation in Gunbower Forest (as assessed in the current project) is to maintain vegetation health (i.e. retain similar PFG species richness, appropriate to the stage of wetting and drying, and canopy condition over time to that sampled at the inception of the monitoring program in 2005). The suggested target for this objective is to maintain the 2005 number of healthy Black and Grey Box sites (Table 1).

The 2015 Box vegetation Index results are mixed, suggesting either an increase or decrease in vegetation health from monitoring inception, depending on which strata is considered. However, when the averages of the two indices are compared (2005, 18.0% vs 2015, 13.6%), it could be concluded that there has been a decline in overall vegetation health in the broad vegetation type over the monitoring period. This implies that the overarching ecological objective for the Box woodlands has not yet been achieved.

2.4 Conclusions

The presence of aquatic and amphibious flora species in areas that were inundated suggests the 2014 eFlow was appropriately timed and of sufficient duration to stimulate the floodplain vegetation. Having said that, the majority of Red Gum and Box Woodland monitoring sites remained dry in 2014, which indicates a large portion of the forest did not directly benefit from the eFlow. This is particularly true for areas upstream of the Hipwell Regulator. It is, however, likely that some dry areas indirectly benefited from the eFlow, as the lower landscape was a mosaic of aquatic and terrestrial conditions created by the network of flood runners gravitating water through the forest.

2.5 Recommendations

- → Continue the environmental watering program to wetlands in Gunbower Forest.
- → Devise strategies that help control turbidity and carp.
- → Develop a second vegetation condition indicator for wetland health.
- → Continue the environmental watering program to Gunbower Forest in order to further improve the condition of the Red Gum vegetation and achieve the ecological objective target.
- → Develop and implement management actions aimed at improving Box woodland canopy condition, such as delivery of environmental water, removing artificial levees preventing natural inflows and ecological thinning.

3 CARP EXCLUSION PILOT STUDY

Section three focuses on the carp exclusion pilot study that was undertaken in Gunbower Forest between 2014 and 2015. It commences with a summary of the scientific literature on the common carp (hereafter 'carp') and the effect that carp have on aquatic vegetation. This is followed by the survey and analysis methods applied and a summary of the floristic results. The aim of the vegetation component of the pilot study is to determine if carp exclusion increases aquatic plant cover and richness (i.e. is there a net benefit to the wetland ecosystem?). Additional questions were postulated, such as whether carp can be excluded at a wetland scale with physical barriers and whether any barriers would affect the wetland fish community. These questions, however, are outside the scope of the current project.

3.1 Literature Review

The purpose of this literature review is to summarise what is known about the direct and indirect impacts of carp (*Cyprinus carpio*) on aquatic plants, including the effects of feeding, body size and water depth. Additionally, the review documents what is known about the impacts of waterbirds on aquatic vegetation, although this was a secondary focus. For a detailed analysis of the effect of carp on aquatic ecosystems, we refer the reader to Vilizzi *et al* (2015).

Note: for the purposes of this document, the terms 'aquatic plant' and 'aquatic vegetation' refer to both higher plants and charophyte algae (large plant-like algal species), since both groups of flora are likely to be susceptible to damage by carp.

3.1.1 Carp

Direct effects of carp on aquatic vegetation

- Carp consume benthic (bottom) detritus and invertebrates by sucking in mouthfuls of substrate, ingesting food particles and expelling non-food items (Koehn 2004, Vilizzi *et al.* 2014).
- In some cases aquatic plants may be ingested incidentally, due to the strong sucking action of carp feeding, which allows them to ingest items up to one head length in front of them (Roberts *et al.* 1995).
- Carp tend to have a negative impact on aquatic plants through uprooting or damaging structures, rather than grazing (García-Berthou 2001).
- Where substrates are soft (e.g. in the case of a silty bottom), carp can displace roots or rhizomes as they disturb the sediment during feeding activity (Weber & Brown 2009).
- Different species and growth forms of aquatic plants vary in their susceptibility to carp. This depends on factors such as the length and strength of their roots or rhizomes and the strength of the vegetative parts of the plant (Weber & Brown 2009).

- In general, submerged aquatic plant species (e.g. *Potamogeton*, *Vallisneria*) are more vulnerable to the impacts of carp disturbance than emergent aquatic plant species (e.g. rushes and reeds), due to their softer structural material and less developed root systems (Roberts *et al.* 1995, Weber & Brown 2009).
- In addition, Roberts *et al.* (1995) also found that the sediments near more robust plants (*Juncus*, *Schoenoplectus* and *Myriophyllum*) were less 'worked over' than the sediments near the more readily uprooted taxa (*Vallisneria* and *Chara*).
- Only soft-leaved macrophytes tend to be directly ingested, probably because these are easily fragmented by physical disturbance (Vilizzi *et al.* 2014).

Indirect effects of carp on aquatic vegetation

- Carp have a rapid, but lasting negative impact on water clarity (evident within less than one day), especially where substrates are soft (Roberts *et al.* 1995, Weber & Brown 2009).
- The increase in turbidity is caused by the gulping of sediment and expulsion of non-food items, leading to a re-suspension of solids in the water column (Roberts *et al.* 1995, Weber & Brown 2009).
- In some cases, depending on the nutrient content of the sediments, the bioturbation of sediments can also lead to increases in the level of water column nutrients, and a decrease in water clarity, due to high concentrations of phytoplankton (Weber & Brown 2009, Vilizzi *et al.* 2014).
- Importantly, the decrease in water clarity that is caused by carp and consumption of large bodied zooplankton are the main drivers in the loss of aquatic plants from carp-dominated wetlands, and not direct removal of the plants (Weber & Brown 2009, Vilizzi *et al.* 2014).
- Increased levels of turbidity negatively affect aquatic macrophytes by both shading and smothering foliage (Vilizzi *et al.* 2015). Therefore, carp have a significant negative, albeit indirect impact on aquatic plants, and are considered major ecosystem modifiers (Vilizzi *et al.* 2014).
- Increased turbidity can lead to the loss of relatively robust aquatic plants that are not as susceptible to physical damage or uprooting (e.g. *Myriophyllum*.) (Weber & Brown 2009, Vilizzi *et al.* 2014).
- Depending on the types of aquatic vegetation present, and their vulnerability to carp disturbance, the presence of aquatic plants can inhibit any increase in turbidity by restricting carp foraging or movement (Roberts *et al.* 1995).
- A carp density of 198± 40 kg/ha has been proposed as the threshold for negative physical impacts on aquatic plant communities (e.g. plant removal), (Vilizzi *et. al* 2015), however, much lower densities (i.e. below 100 kg/ha) have been found to have both a significant negative impact on water clarity and consequent detrimental effects on aquatic vegetation (Vilizzi *et al.* 2014).
- Lakes in the north west of the USA, where the relative abundance carp exceeded 0.6 catch per unit effort (CPUE), have been associated with lower native fish abundance and impaired water quality (i.e. higher dissolved solids and lower secchi depth, which are measures of turbidity) (Weber & Brown 2011).

- Low carp densities can still result in a shift from clear to turbid water, but this may just take longer than at higher densities of carp (Weber & Brown 2009).
- The effect of decreased water transparency and macrophyte coverage associated with carp generally increase with fish biomass, but have been reported from fish less than one year old at densities of 175 kg/ha and above (Weber & Brown 2015).
- Adult carp tend to have a greater effect on vegetation than juveniles (Weber & Brown 2009).

Reproduction and growth

- Carp population dynamics operate at large spatial scales Vilizzi *et al.* (2015).
- Carp have highly successful reproductive strategies that allow them to reproduce and mature rapidly (Koehn 2004, Weber & Brown 2009).
- Carp reach sexual maturity early: males at 1 year, females at 2 years, and they have a rapid generation time of 2–4 years (Koehn 2004).
- Eggs hatch quickly (2 days at 25 °C) and the new hatchlings grow fast, allowing them to soon grow beyond the reach of most predators (Koehn 2004).
- The growth of young carp has been measured in North America at an average of 163 mm by one year of growth, 279 mm by two years, and 366 mm by three years (Weber & Brown 2009).

Differences in feeding ecology between juvenile and adult carp

- Small, young carp feed on zooplankton, and large numbers of these small individuals may contribute to the reduction in size and abundance of zooplankton (Weber & Brown 2009).
- The literature varies in relation to the size of carp when they switch from water column (planktivorous) to bottom (benthic) feeding occurs. Weber & Brown (2009) cite 100 mm in length, but Vilizzi *et al.* (2014) cite a smaller length of 25 mm. In either case, the switch occurs at a small size, and carp are known to grow rapidly (Weber & Brown 2009).
- Young carp leave their nursery-grounds after reaching critical a size/development stage.

Interactions between effects of carp on aquatic vegetation and water depth

- It is somewhat difficult to assess the effect of 'depth' on carp behaviour and impact, since what is considered 'deep' varies greatly between both studies and habitat types.
- The conventional boundary between 'deep' and 'shallow' standing waters is three metres (Boulton *et al.* 2014), however, in some Australian waters, particularly if they are turbid, the boundary between deep and shallow is likely to be less than this (e.g. Vilizzi *et al.* 2014 'deep habitat' = >1.7 m).
- There is also evidence of seasonal differential in carp's habitat preference, with shallow (often temporary) vegetated wetlands and literal edges preferred during the warmer months (for feeding and reproducing) and deeper, permanent waters preferred during the cooler months (García-Berthou 2001, Smith *et al.* 2009, Vilizzi *et al.* 2015).

- During spring and autumn surveys of Brenda Park Wetland in South Australia, carp were common in the open water (at a depth 0.7-1.4 m) and in low numbers in the deeper areas (>1.7 m) where there was little or no aquatic vegetation, (Vilizzi *et al.* 2013).
- Part of the reason for carp preferring the shallower waters during the warmer months is that spawning takes place in aquatic plant beds (usually <0.5 m in depth) (Weber & Brown 2009).
- Carp numbers are usually highest in shallow lakes/wetlands and may partly be restricted by the shape of the basin (Weber & Brown 2009).
- The most vulnerable ecosystems to carp damage are shallow wetlands with fine sediments and a low diversity and cover of aquatic vegetation (Weber & Brown 2009).

3.1.2 Waterbirds

Direct effect of waterbird grazing of aquatic vegetation

- Herbivorous Australian waterbirds include the Black Swan (*Cygnus atratus*), the Australian Shelduck (*Tadorna tadornoides*), the Eurasian Coot (*Fulica atra*) and the Australian Wood Duck (*Chetonetta jubata*).
- Black Swans are almost entirely herbivorous and feed mainly on the leaves and shoots of aquatic plants, usually while swimming, sometimes with their heads below water (Smith *et al.* 2012).
- Australian Shelduck graze in shallow water or terrestrial areas, preferring grass, algae, insects and molluscs (Bird Life Australia 2015a).
- In Australia, Eurasian Coots are mainly herbivorous, and eat only a small amount of animal prey (Bird Life Australia 2015b).
- Australian Wood Ducks forage in shallow water or on land. When in the water they are either on the surface or only their heads are submerged (Evans 2013). They do not dive for food. Their main foods are grasses, clover and other herbs (Evans 2013).
- Of these four species, Black Swans are the largest (weighing 5–6 kg) and can have a significant impact on the ecology of the water bodies in which they graze (Mitchell & Wass 1996, Smith *et al.* 2012).
- In order to harvest emergent macrophytes, Black Swans have been observed snapping off stems above water level (Smith *et al.* 2012). They are believed to prefer the actively growing parts of the plants, however, they will feed on seagrass rhizomes as well as leaves and stems (Smith *et al.* 2012). They are not believed to feed on tubers.
- Black Swan grazing can lead to the removal of aquatic plants from particular areas, but does not directly result in increased turbidity (Smith *et al.* 2012).
- Waterbird grazing can lead to a change in plant morphology and growth (Blanch and Brock 1994), and can also allow the establishment of new aquatic plant species in areas previously dominated by a monoculture (Smith *et al.* 2012).

- Black Swan numbers have been observed to be linked to the abundance of submerged aquatic plants – swan numbers increasing in response to an increase in plants – however, grazing consumption does not appear to lead to a decline in plant abundance (Mitchell & Wass 1996).
- Grazing by Black Swans may help to regulate aquatic plant populations and may have a cumulative impact on plant survival if other impacts such as low light are also present, however, such grazing does not significantly reduce plant stands (Mitchell & Wass 1996).

Indirect effects of waterbirds on aquatic vegetation

- Waterbirds may have an indirect influence of aquatic plants through their role in contributing to wetland nutrient loading, and therefore, to the occurrence of phytoplankton blooms. Waterbirds may significantly contribute to the nutrient loads of water bodies, however, the degree of influence depends on season, whether the site is used for nesting, whether it is closed or gets flushed, and the level of retention of nutrients in the sediments (Baxter & Fairweather 1994, Hahn *et al.* 2008).

Interactions between effects of waterbirds on aquatic vegetation and water depth

- The presence of aquatic vegetation in wetlands is related to the depth to which light can penetrate the water column, which in turn, is an interaction between depth and water clarity (Boulton *et al.* 2014).
- In deeper water, particularly deeper turbid water, aquatic plants may not be present for waterbirds to graze.
- Different herbivorous waterbirds have different foraging habits. Both the Australian Shelduck and Australian Wood Duck feed in shallow wet areas or adjacent terrestrial areas (such as crops and pasture) (Evans 2013, Bird Life Australia 2015a). When congregating in large numbers, the Australian Wood Duck has been known to damage crops and pastures (Evans 2013) However, due to the shallowness of both species' feeding areas, these two ducks are unlikely to have a significant negative effect on natural wetland vegetation in areas that are also affected by carp.
- Eurasian Coots mainly forage for aquatic plants underwater. They can dive up to 7m (Bird Life Australia 2015b). They also feed on the surface of the water and on land. It is unclear to what degree these birds might have an impact on submerged plant communities.
- Black Swans tend to occur in water that is deep enough to allow them to graze while swimming, which is less energetically demanding than walking (Smith *et al.* 2012).
- Deeper water also allows some protection from terrestrial predators (Smith *et al.* 2012).
- In a study on the Clarence River Floodplain (NSW, Australia), Black Swans were found in greatest numbers on wetlands of 'intermediate' water depth (20-50 cm), followed by lower numbers on wetlands deeper than 1 m, and lowest numbers on very shallow wetlands (<20 cm) (Smith *et al.* 2012).

3.1.3 Additional information

Additional research that would be helpful for understanding and managing the impact of carp on aquatic flora in Gunbower Forest includes:

- measuring the precise carp densities that trigger an increase in turbidity specific to the Murray River floodplain,
- identifying critical turbidity levels (again specific to the Murray River floodplain) and their interaction with water depths in terms of the loss of aquatic plants,
- assessing the combined effect of waterbirds and carp on aquatic plants,
- identifying specific impacts of Eurasian Coots on submerged plant communities.

3.2 Methods

3.2.1 Experimental Design

In 2014, 24 3x3 m pilot study plots were established at two wetlands in Gunbower Forest, namely, Reedy and Little Reedy Lagoons (Figures 18 and 19, respectively), as part of a pilot study examining the effect of excluding carp from aquatic plant beds (Table 12). (See Technical Addendum for locations.) The wetland treatments were fenced and unfenced (one replicate of each). Reedy Lagoon had an exclusion fence constructed across flood runners leading in and out the wetland (Figure 47), while flood runners leading into Little Reedy Lagoon were uninhibited. Within each wetland, plot treatments were fully fenced, partially fenced (allowing animal access to the plot vegetation) and controls (unfenced) (Figure 48). There were a total of eight of each plot treatments (four at each wetland). All plots were marked with a tagged star picket in the north-west corner of the quadrat. The fully fenced plots were completely enclosed to a height of 3 m, with a 0.2 m lip of wire pinned to the ground on the outside to prevent animals from burrowing under the fence into the plot. The partially fenced plots were enclosed on three sides (with the opening facing the middle of the wetland) to a height of 1 m. Exclusion fencing used for flood runners and plots were constructed from wire netting with 40 mm gaps. Three-wire fences were constructed on the outside of the flood runner exclusion fences in order to reduce debris that might affect the stability of the exclusion fences.

Figure 47 Exclusion fence (left) and accompanying debris fence (right) constructed on a major flood runner leading into Reedy Lagoon, May 2014.

Figure 48 carp pilot study plots, fully fenced (RL1A C2, left), partially fenced (LR2B C1, middle), and unfenced (LR2A C1, right), Gunbower Forest, February 2015.

Table 12 Exclosures used in the Gunbower Forest carp exclusion experiment and their allocated water depth category (D=Dry, Di=Drying, SI=Shallowly Inundated, and MI=Moderately Inundated, refer to Table 13). Cases with bold border were outliers from the NMDS ordination.

Wetland	Site & side	Plot	Treatment	2014	2015	2015
				May	February	April
Little Reedy Lagoon	LR1A	C1	Partially fenced	D	SI	SI
		C2	Not fenced	D	SI	SI
		C3	Fully fenced	D	SI	SI
	LR1B	C1	Partially fenced	D	SI	SI
		C2	Fully fenced	D	SI	Di
		C3	Not fenced	D	SI	Di
	LR2A	C1	Not fenced	D	SI	D
		C2	Partially fenced	D	SI	D
		C3	Fully fenced	D	SI	D
	LR2B	C1	Partially fenced	D	Di	D
		C2	Fully fenced	D	SI	D
		C3	Not fenced	D	Di	D
Reedy Lagoon	RL1A	C1	Not fenced	D	MI	MI
		C2	Fully fenced	D	MI	MI
		C3	Partially fenced	D	MI	MI
	RL1B	C1	Partially fenced	D	MI	MI
		C2	Not fenced	D	MI	MI
		C3	Fully fenced	D	MI	MI
	RL3A	C1	Partially fenced	D	SI	D
		C2	Not fenced	D	SI	D
		C3	Fully fenced	D	SI	D
	RL3B	C1	Partially fenced	D	SI	D
		C2	Fully fenced	D	SI	D
		C3	Not fenced	D	SI	D

3.2.2 Data Collection

The projected foliage cover of flora species (vascular species and charophytes) was estimated within each pilot study plot prior to inundation (30th May 2014), during inundation (24-25th February) and on draw down (28th April 2015). Taxonomy of plants recorded follows the Victorian Plant Name Index (DELWP 2015), with consideration to the Census of Victoria Vascular Plants (Walsh & Stajsic 2015). Photographs of the vegetation in the plot and of the whole plot were also taken during sampling.

Water depth was recorded at each plot at the time of sampling, along with an estimate of maximum inundation depth that was based on water marks on the plot fences. Water samples were collected at each wetland site and sent to Envirolab Services Pty Ltd for analysis. The turbidity of the water samples was measured nephelometrically, using a turbidimeter in accordance with the latest edition of the American Public Health Association standards (2130 B).

3.2.3 Data Analysis

Species diversity and cover of aquatic plants was compared between wetlands, sampling dates, water depth category (Table 13) and plot treatments (within and between wetlands, Table 12). Exploratory data analysis is presented in the forms of bar graphs and NMDS ordination plots in order to view a visual assessment of differences between treatments. (Refer to Multivariate Analysis description on Section 2.1.3 of the current report.)

Table 13 Water depth categories for carp pilot study, Gunbower Forest.

Water depth	Water depth category
0 cm	Dry
0 - <10cm	Drying
10 - <50cm	Shallowly inundated
>50cm	Moderately inundated

Note that the study does not include sufficient replication of control and treatment plots to facilitate formal statistical analysis. The experiment was designed to help understand potential effects of carp exclusion, and the likely magnitude of differences in flora species cover and richness. The conclusions are therefore only relevant to the two wetlands tested and statistical significance has not been calculated. The results may not, therefore, be applicable to other wetlands.

These background data are, however, valuable for their contribution towards our understanding of the effectiveness of the exclosures, the potential interaction between the exclosure structures and measures of cover and richness (e.g. overgrowth of Azolla within the fenced plots), and the baseline composition and level of variability in the vegetation. This measure of variability will also enable a more statistically powerful design, should this exclosure experiment be extended in the future.

3.2.4 Limitations

The carp exclusion pilot study was designed to trial measures for excluding the fish species from areas of aquatic habitat, at both wetland and local scales. However, the following limitations apply to this dataset:

- The barrier material selected to exclude carp may have limited the activity of other species (e.g. large bodied native fish and/or water birds) in the monitoring plots and thereby influenced the results.
- The cryptic nature and seasonal growth cycles of certain species may have hindered the detection of these taxa at the monitoring sites. In particular, submerged species were at times concealed, and possibly inhibited by a thick cover of floating species. Therefore, when interpreting the results, it should be noted that the data are skewed towards reporting a lower than actual level of richness.
- A lack of water during sampling periods meant that aquatic species richness and cover could not be compared between all sampling occasions (Table 12).
- Low numbers of carp recorded in the March 2015 fish survey (Reedy Lagoon: 2 small carp; Little Reedy Lagoon: 6 carp including 2 large individuals) mean that it is more difficult to know what influence carp have had on the results (Biosis 2015).
- The fish surveys were generally performed in the wetland area, and not in the exclosures, and the timing of the fish surveys was different to that of the flora surveys (Biosis 2015). This means that it is difficult to explicitly link flora results to carp results for the purposes of statistical analysis. The degree to which carp numbers fluctuated during the sampling period is also unclear.
- The effect of other species with potential impacts on aquatic vegetation (e.g. black swans) is not known and was not measured.

3.3 Results

General Condition

All plots in the carp exclusion pilot study were inundated from May 2014 through to 2015 (Table 12). On average, plots in Reedy Lagoon were more deeply inundated (with a maximum depth 0.85 m in February 2015) than those in Little Reedy Lagoon (which had a maximum depth 0.3 m. also in February 2015) (Figure 49). Each wetland included both shallow and deep sites. Water was observed flowing over the top of the 3 m exclusion plots in the deeper sites in Reedy Lagoon (RL1) in August 2014, but not at any other site. Shallow sites (RL3 and LR2) at both wetlands were dry by the time of the April 2015 survey, such that half the study plots were wet and half dry.

The turbidity differed between the two wetlands (Table 14). Little Reedy Lagoon, with no carp exclusion fencing, was around four times as turbid as Reedy Lagoon when sampled in both February (Figure 49) and April. The level of turbidity also increased considerably between the February and April samples in Little Reedy Lagoon.

Figure 49 Difference in turbidity in Little Reedy Lagoon (LR1A C1, right photo, note the plume of turbidity from walking to the plot) and Reedy Lagoon, (RL1A C3, left photo), Gunbower Forest February 2015.

Based on the Australian and New Zealand guidelines for fresh and marine water quality (ANZECC & ARMCANZ 2000), the turbidity levels in Reedy Lagoon would be considered low (undisturbed) on both sampling occasions, while the turbidity levels in Little Reedy Lagoon, may suggest some disturbance. The ANZECC guidelines do not specifically provide trigger levels for floodplain wetlands. However, it would be expected that the turbidity levels sampled would lie in the range appropriate for lowland rivers in well vegetated catchments (ANZECC & ARMCANZ 2000). Anecdotal evidence suggests that the dispersible sediments found in the Gunbower Forest wetlands could cause the turbidity to rise rapidly if disturbed (Figure 49).

Table 14 Turbidity results for wetland sites, Gunbower Forest, February and April 2015

Wetland	Site	Turbidity (NTU PQL=0.1)	
		February 2015	April 2015
Little Reedy Lagoon	LR1	19	27
Little Reedy Lagoon	LR2	19	NA (dry)
Reedy Lagoon	RL1	6	7
Reedy Lagoon	RL3	5.3	NA (dry)

In March 2015, Biosis sampled carp in the wetlands around the plots, but not within them. Two juvenile carp (i.e. 95 mm and 123 mm long, respectively) were recorded in Reedy Lagoon, while six carp of varying ages and sizes were recorded in Little Reedy Lagoon. One of these carp was approximately three years old (i.e. 330 mm in length) and another estimated over ten years of age (i.e. 670 mm) (Biosis 2015). These results indicate there were fewer and smaller carp in Reedy Lagoon, which had exclusion fencing preventing large fish (i.e. >30 mm in width) from accessing the wetland.

Inflow and/or churning debris at one of the Reedy Lagoon exclusion fences appears to have carved a hole approximately 30 cm in diameter under the fence, which may have allowed large carp into the wetland. It is, however, conceivable that the two individuals sampled in Reedy Lagoon could have passed through the barrier fence when they were <30 mm in width and subsequently increased in size once in the wetland. This was expected given the gap size in the fencing wire used.

A total of 68 plants were recorded in the plots, 76% of which were native. All bar one of the exotic species were observed when the wetlands were dry. Four species of conservation significance were also included in this list: namely, the nationally vulnerable River Swamp Wallaby-grass (*Amphibromus fluitans*) and the rare Water Nymph (*Najas tenuifolia*), Riverina Bitter-cress (*Cardamine moirensis*) and the Dwarf Bitter-cress (*Rorippa eustylis*). The River Swamp Wallaby-grass and Water Nymph were only observed in Reedy Lagoon; the Riverina Bitter-cress was only observed in Little Reedy Lagoon; while the Dwarf Bitter-cress was seen in both wetlands. The cover and distribution of River Swamp Wallaby-grass in Reedy Lagoon was the greatest the author has observed in 11 years monitoring the wetland. Section 2.2.1, and in particular Figure 22, in the current report provide more detail about the floristic character of the two wetlands.

Flora composition

Multivariate analyses (NMDS ordinations) were used to investigate similarities and patterns in floristic composition between the pilot study plots. Samples were coded by factors such as 'Treatment' (fully fenced, partially fenced and not fenced), 'water depth category,' and 'wetland,' in order to help visualise the factor's influence on flora composition (Figures 50a-c). Measures of aquatic and amphibious flora species (PFGs 1-3) richness and dominance (total % cover), along with water depth, were overlaid as bubbles on the ordination plots in order to highlight any correlation of these variables with data groupings (Figures 51a-b).

Figure 50a shows that the two wetlands supported different compositions of species (i.e. separately grouped in the ordination plot), and that one group of samples from Reedy Lagoon (RL3, shallow plots, sampled in May 2015) was very different to all others recorded at this wetland. When the samples were categorised by depth (Figure 50b), dry samples were separated from inundated samples, highlighting their different compositions. The moderately inundated samples were the least like dry samples (i.e. positioned further apart on the plot), and formed a group that was distinct from drying and shallowly inundated samples (Figure 50b). There was, however, some overlap in the flora composition of samples in the drying and shallowly drying categories (Figure 50b), and the spread of samples within these two categories suggested variability in species compositions within the two categories. In contrast to the above, though, there was no clear grouping of samples by experimental treatment, indicating that species composition in this pilot study was not determined by treatment (i.e. exclosure type, as per Figure 50c).

When variables such as water depth, and richness and cover of aquatic and amphibious species are overlaid on the plots, it is clear that patterns in species composition related more to the presence of water at the time of sampling than factors explored above (Figure 51a-c). The cover of aquatic and amphibious species appears to correlate well with water depth (i.e. overlap in Figures 51a and 51b). Higher species richness also correlates to water depth, although not as strongly in deeper samples and recently dried samples (Figures 51a and 51c).

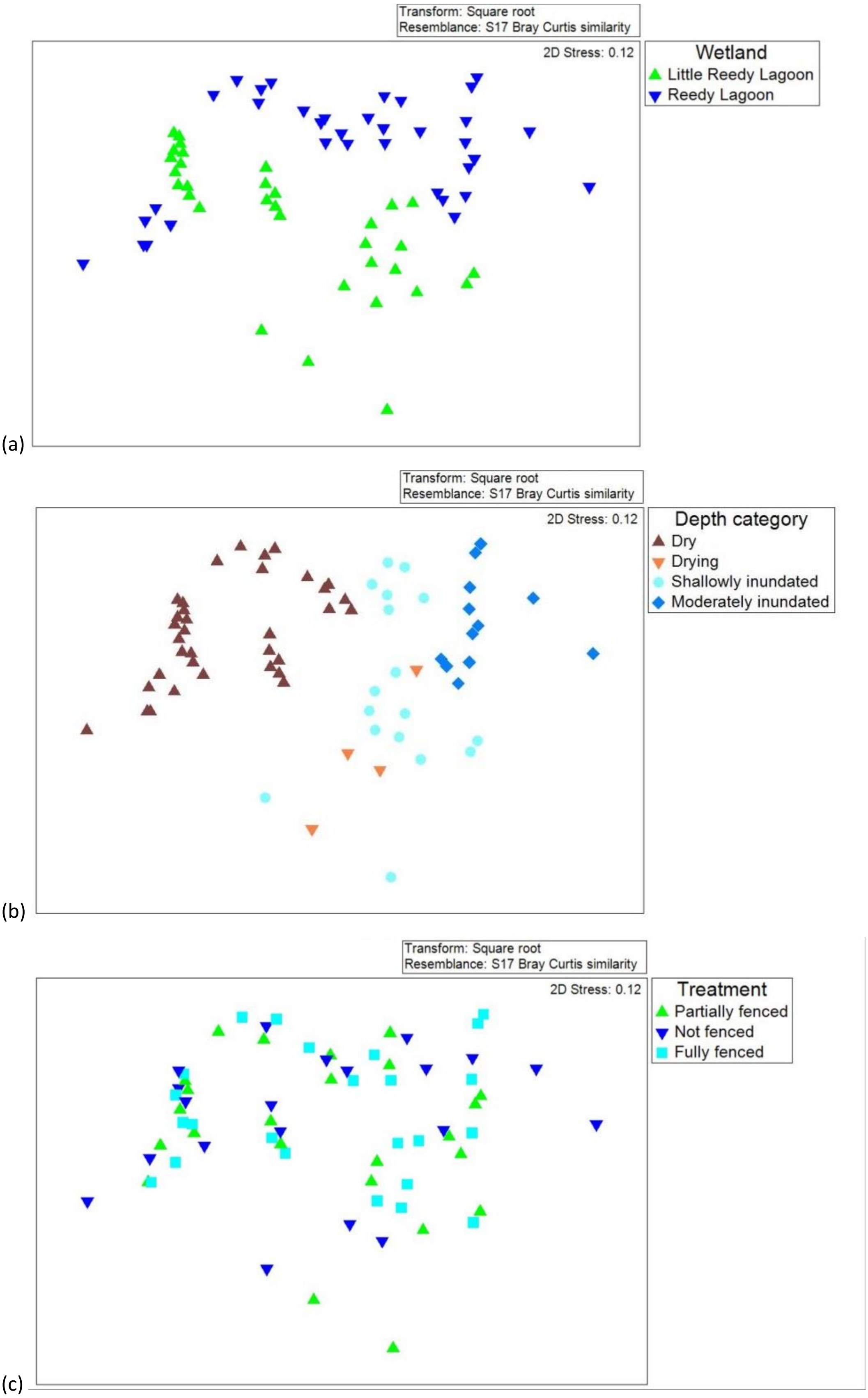


Figure 50 NMDS plot showing all native flora percent cover data for the carp exclusion pilot study, coded for (a) wetland, (b) water depth category (c) 'treatment' (exclosure type), (four outliers removed), 2014-2015, Gunbower Forest.

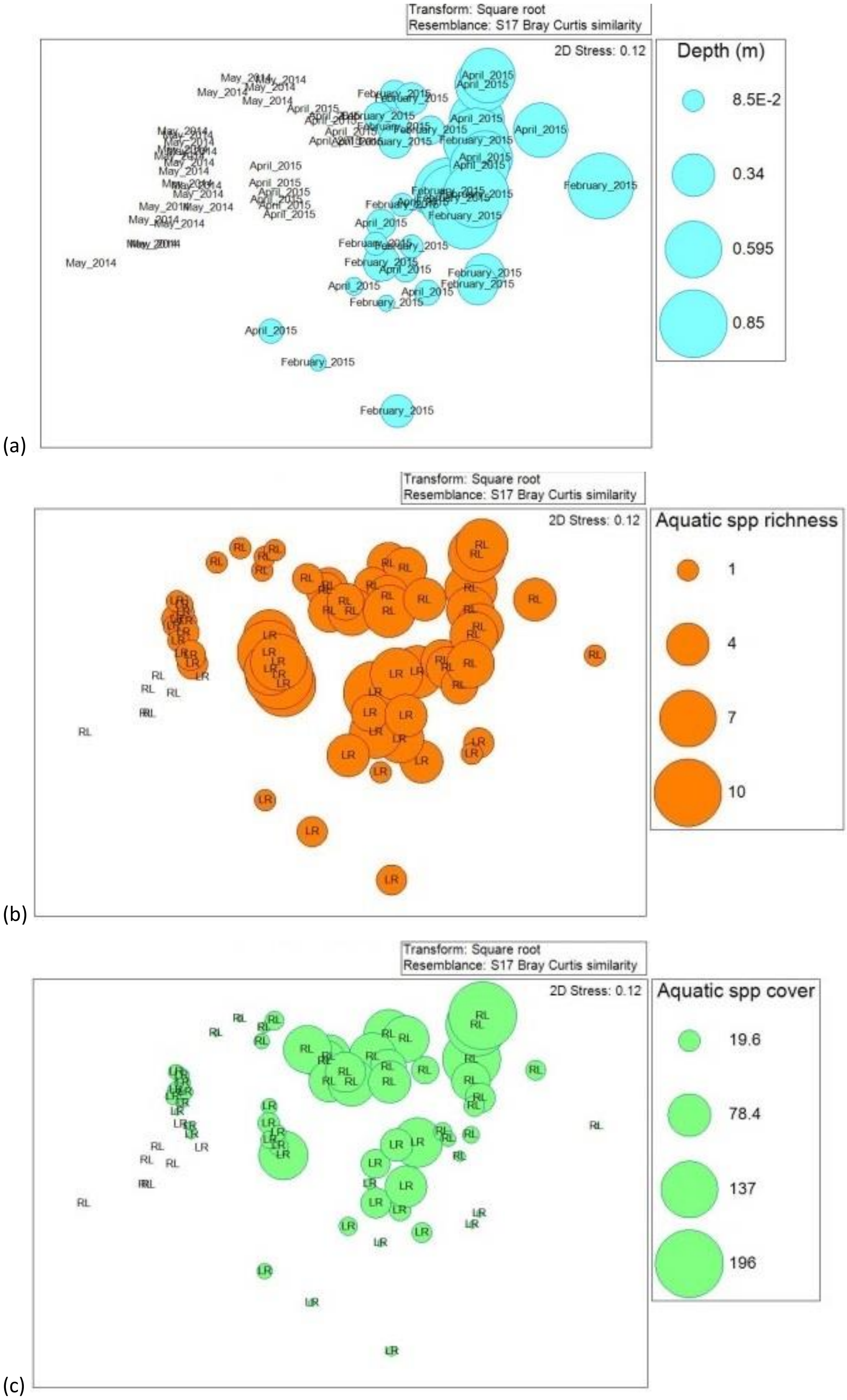


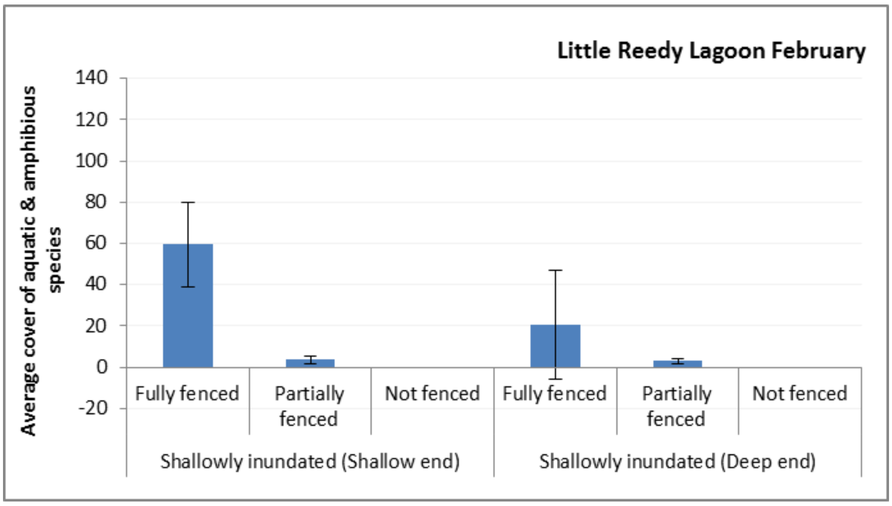
Figure 51 NMDS plot showing all native flora % cover data for the carp exclusion pilot study, with scaled bubbles showing (a) the relative magnitude of water depths recorded at each exclosure, (b) the numbers of aquatic species recorded at each exclosure, and (c) total cover of aquatic species recorded at each exclosure. Site names have been superimposed, 2014-2015, Gunbower Forest.

Flora Cover

While multivariate analysis of the data did not differentiate samples by treatment (i.e. exclosure type), anecdotal observations suggest that there were considerable differences in four of the fully fenced plots, compared to other treatments at the same sites. The following section presents the 2015 results for the two most distinct of these cases. Caution should be applied when interpreting these results, as they are based on small sample sizes (i.e. two plots per treatment per wetland). (Note that the study is not replicated at the wetland level).

In February, fully fenced plots in the shallow end of Little Reedy Lagoon (i.e. shallowly inundated to a depth of 0.1 m) supported on average 60% cover of aquatic and amphibious species (PFGs 1-3, Figures 52a, 53 and 55). In contrast, the partially fenced and control plots that were accessible to fish and birds, and sampled at the same site and time, had a maximum cover 5% of these species. Figure 52a also suggests an exclusion effect in the deep end of the wetland, but this was largely due to a 30% cover of Azolla in one of the plots when sampled. (Note the large error bars.)

(a)



(b)

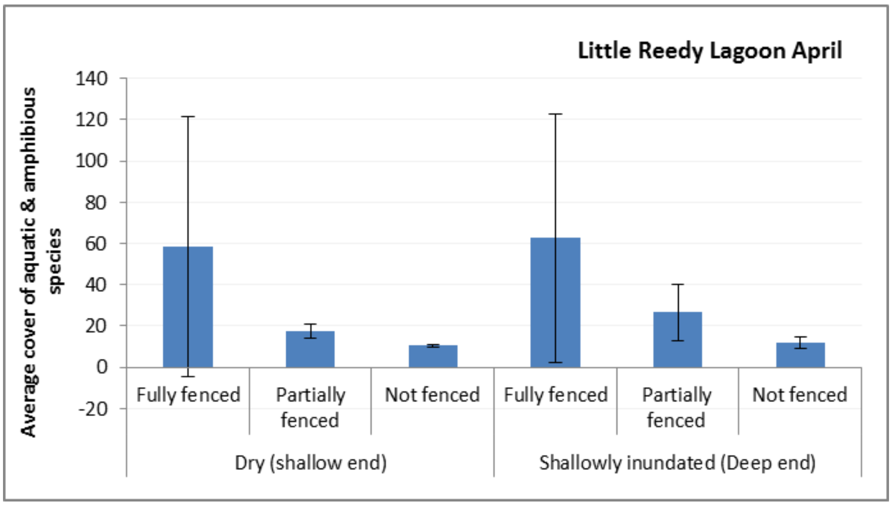


Figure 52 Average percentage cover (± std dev) of native aquatic and amphibious flora (PFGs 1-3) in the carp exclusion pilot study plots in Little Reedy Lagoon in (a) February (b) April, 2015, Gunbower Forest. Note each treatment represents two plots.

The difference in the cover of aquatic and amphibious flora between fully fenced and other treatments was still apparent when the Little Reedy Lagoon plots were sampled in April, even though the shallow end of the wetland had dried out (Figure 52b). High Azolla covers inside the fully fenced plots in the deep end again give a false impression that the exclusion of fish and birds caused an effect (Figure 54).

Figure 53 Aquatic and amphibious flora inside the fully fenced carp exclusion pilot study plots, at the shallow end of Little Reedy Lagoon, Gunbower Forest, January (left) and March (right) 2015.

Figure 54 High cover of Azolla (PFG 1) inside a fully fenced carp exclusion pilot study plot, at the deep end of Little Reedy Lagoon, Gunbower Forest, April 2015.

Little Reedy Lagoon (shallow end), February 2015

Control

LR2A C1

LR2B C1

Partially fenced

LR2A C2

LR2B C1

Fully fenced

LR2A C3

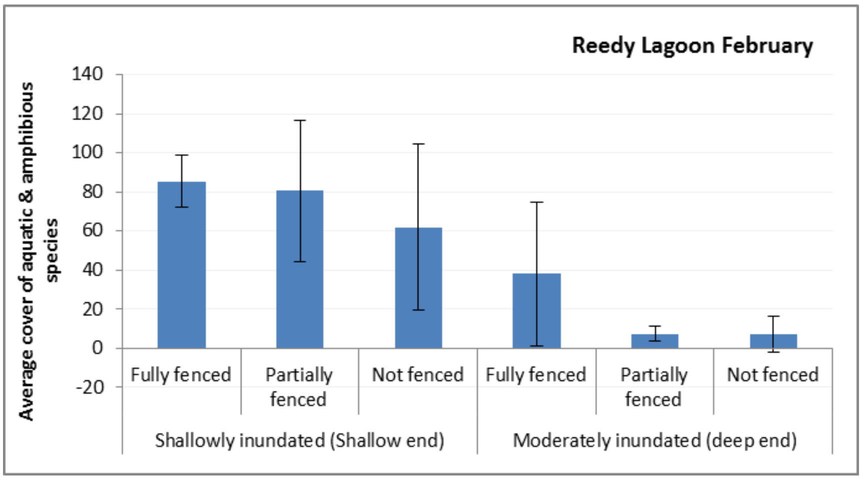
LR2B C2

Figure 55 Carp exclusion pilot study plots, ordered by treatment, in Little Reedy Lagoon, Gunbower Forest, February 2015.

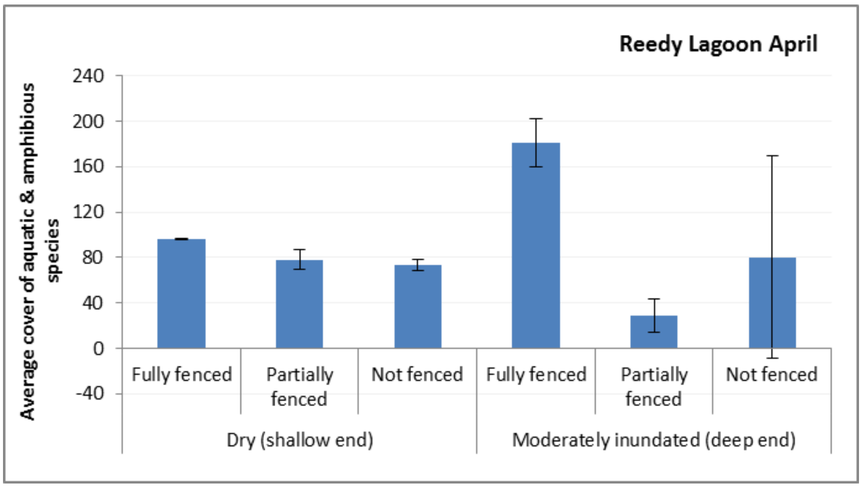
In Reedy Lagoon, the deeper inundated plots showed contrasting covers of aquatic and amphibious species (PFGs 1-3) between treatments (Figures 56-58). A difference in plant cover between the fully fenced and other treatments, particularly in one of the two fully fenced plots, was beginning to emerge in February, (Figure 56a). At this stage, it was already evident that delicate species such as *Potamogeton* spp. were only rooted and flowering inside the fully fenced plots (Figure 58).

However in April, when the water levels had drawn down from 0.8 m to 0.55 m, the effect was more obvious (Figure 56b, 57 and 58). In this sample month, the average cover of aquatic and amphibious species in the fully fenced plot was 181%, whereas the covers of these species in the other two treatments were on average 28% (partially fenced) and 80% (no fence). Some cover figures at Reedy Lagoon were in excess of 100%, as they included between 85% and 100% of Azolla floating above the typically submerged species. Care was hence taken to identify and estimate the cover of the submerged species. (See Figure 58.)

There was no obvious effect of the exclosure treatment in the shallow end of Reedy Lagoon in either February or April (Figure 56a and 56b). Aquatic vegetation at this end of the lagoon was particularly thick across all treatments.



(a)



(b)

Figure 56 Average percentage cover (± std dev) of native aquatic and amphibious flora (PFGs 1-3) in the carp exclusion pilot study plots in Reedy Lagoon in (a) February (b) April, 2015, Gunbower Forest. Note each treatment represents two plots.

Reedy Lagoon (deep end) April 2015

Control

RL1A C1 **RL1B C2**

Partially fenced

RL1A C3 **RL1B C1**

Fully fenced

RL1A C2 **RL1B C3**

Figure 57 Carp exclusion pilot study plots, ordered by treatment, in Reedy Lagoon, Gunbower Forest, April 2015.

Figure 58 Native aquatic and amphibious flora inside the fully fenced carp exclusion pilot study plots, in the deep end of Reedy Lagoon, Gunbower Forest, March (top, left and right, close up of flora) and April (bottom, left and right) 2015.

3.4 Discussion

Carp are known to have a significant negative impact on aquatic ecosystems around the world (Vilizzi *et al.* 2015). They affect submerged aquatic vegetation indirectly through changes to water quality (turbidity, Weber & Brown 2009, Vilizzi *et al.* 2014), and to a lesser degree, through physical uprooting and fragmentation (García-Berthou 2001). Carp grow rapidly and switch from being water column (planktivorous) to bottom (benthic) feeders, and can hence start to affect turbidity when less than one year of age (Weber & Brown 2015). Even at low densities carp increase turbidity (Vilizzi *et al.* 2014). Shallow wetlands with a soft or silty base, such as found in Gunbower Forest, are the most vulnerable to degradation by carp (Weber & Brown 2009).

Carp have been observed in Gunbower Forest since the 1970s (D. Tresize 2013, local resident, pers. comm. 24 April). While their numbers have varied spatially and temporarily, when surveyed over the last decade (Biosis 2015), it is hypothesised that carp have had a significant impact on the forest's wetland flora.

The current pilot study into the effect of carp on wetland flora in Gunbower Forest found that the cover and richness of aquatic and amphibious species (PFGs 1-3) was more closely correlated with water depth than wetland treatment (flood runners fenced off or not fenced) or plot treatment (i.e. fully fenced, partially fenced and not fenced (Figures 50 and 51). The lack of evidence of a treatment effect in the multivariate analysis is, however, potentially due to both the small sample size (e.g. 24 sample plots) and compounding factors, given that four of the eight fully fenced plots had considerably higher covers of aquatic and amphibious species than the partially fenced and control plots at the same sites (Figures 52 and 56).

The sites that demonstrated anecdotal evidence of an effect of plot fencing were situated in the shallow end of Little Reedy Lagoon (LR2) and in the deep end in Reedy Lagoon (RL1). The effect was also more obvious in the Little Reedy Lagoon sites in the February survey when inundated to a depth of 0.1 m and before species that germinate in moist mud (i.e. PFG 4) appeared in the April survey (Figures 52). In contrast the effect of treatment was more obvious in Reedy Lagoon in the latter survey when water levels had drawn down from 0.8 m (February) to 0.55 m (April) (Figures 56). These results align with those from the multivariate analysis, in that water depth appears to be a driving factor in the establishment of the aquatic macrophytes. The results also indicate that, in fifty per cent of cases, plots with exclusion fencing were associated with higher covers of aquatic and amphibious species compared to plots which carp and other large animals could freely access (i.e. partially fenced and control plots).

The absence of a plot treatment effect in the deep end of Little Reedy Lagoon (LR1) was possibly due to the effect of turbidity, as availability of light is a major driver of aquatic plant growth and survival (Boulton *et al.* 2014). Turbidity levels recorded in Little Reedy lagoon were around four times those recorded in Reedy Lagoon in both February and April, and increased from 19 NTU to 27 NTU as water levels dropped between the surveys (Table 14, Figure 49). Water depth possibly compounded the effect of turbidity in the deep end of the Little Reedy Lagoon.

In a South Australian study on the ecological effects of carp, turbidity levels of 20 NTU coincided with a dramatic decrease in aquatic macrophyte cover and biomass (Vilizzi *et al.* 2014). It is, therefore, possible that the turbid water in Little Reedy Lagoon reduced light penetration and inhibited aquatic plant establishment (other than Azolla) in all plot treatments in the deep end of the wetland.

There is strong evidence linking carp abundance to turbidity (Vilizzi *et al.* 2015). For example, when sampled in March 2015, the highest carp abundance (99 individuals) was recorded in the wetland with highest turbidity level in Gunbower Forest (Greens Lagoon, 297 NTU, sampled on a calm day, Biosis 2015). While sampled fish abundances were low in both Little Reedy and Reedy lagoons in March 2015, three times the number of carp was recorded in the former, more turbid wetland than the latter, fenced-off wetland (Biosis 2015). Furthermore, five of the six carp caught in Little Reedy Lagoon were larger (i.e. older) than the two carp caught in Reedy Lagoon, and therefore, more likely to be benthic feeders stirring up sediments. These results suggest that the greater number of mature carp sampled in Little Reedy Lagoon may have resulted in the higher turbidity levels than those recorded in Reedy Lagoon.

Carp, however, are not the only cause of reduced water clarity. For example, during fish sampling in 2015, Biosis recorded turbidity levels more than three and a half times the levels recorded in the previous and subsequent months in the current project (e.g. Reedy Lagoon, 25 NTU; Little Reedy Lagoon, 98 NTU). Weather conditions on the day that the samples were taken by Biosis were reportedly exceptionally hot and windy (A. Byrnes 2015, Biosis, pers. comm. 18 June). Vilizzi *et al.* (2015) also note that wind and substratum characteristics are possible causes of turbidity. The fact that turbidity returned to the previous level after the March survey suggests that the effect of wind on water clarity was only temporary and unlikely to have any on-going influence on aquatic macrophyte establishment.

By way of contrast, the lack of treatment effect in the shallow end of Reedy Lagoon (RL3) is unlikely to be due to turbidity, given that the NTU was recorded at 5.6, and all treatment plots supported high covers of aquatic and amphibious species when inundated in February 2015 (Figure 56a). It is, however, possible that the aquatic macrophytes became established at the shallow end of the wetland in the absence of mature carp, due the exclusion fence limiting their access to Reedy Lagoon. It is also likely that, once established, the high cover of submerged flora (authors' observation) limited the effect of any carp present, as dense aquatic vegetation has been found to curb both carp movement and foraging (Roberts *et al.* 1995), particularly since these carp were relatively small.

The inter-wetland differences in turbidity and higher covers of aquatic and amphibious species in the partially fenced and control plots (i.e. that carp could potentially access) in Reedy Lagoon compared to the same treatment plots in Little Reedy Lagoon (Figures 52 and 56), add support to the hypothesis that the exclusion fence at Reedy Lagoon limited carp access to the wetland (particularly large carp).

However, Biosis (2015) rightfully state that it is difficult to draw conclusions on the effectiveness of the exclusion fence on the carp population, given the low numbers recorded in both lagoons in 2015 and previous years. Furthermore, with only one fish survey conducted in the wetlands during the 2014 eFlow, it is not possible to determine if Little Reedy Lagoon had supported a greater number of carp in previous seasons. Intrinsic differences between the Reedy and Little Reedy lagoons are also likely to have influenced the inter-wetland floristic results.

It should be added that the pilot study identified a number of limitations with the experimental design. For example, the fencing material used on the exclusion plots appears to have trapped Azolla (see Figure 54), which at high cover is known to blocked light and potentially affected both nutrient levels and consequently the cover of submerged species (Bailey *et al.* 2002). One of the exclusion fences leading into Reedy Lagoon developed a hole, through which carp could have passed during inundation. The carp survey method, which reports carp abundances as CPUE, is not directly comparable to the majority of studies on the impact of carp (see Vilizzi *et al.* 2015), making comparisons between other systems and research difficult. It is also possible that the exclusion plots restricted waterbirds and other fauna such as turtles from accessing the flora, which make it hard to distinguish between the potential impacts of these species and fish. It is possible that the treatment effect observed in the deep end of Reedy Lagoon reflects the impact of bird grazing rather than carp. Any future experimentation would need to address this design limitation.

In summary, the pilot study on the impact of carp on aquatic vegetation in Gunbower Forest was not of sufficient scale to produce statistically significant results, and would need to be replicated across multiple fenced and unfenced wetlands in order to do this. The study did, however, provide valuable background data on the effectiveness of the exclosures, the interaction between the exclosures and aquatic vegetation, and baseline composition and level of variability in the vegetation. This measure of variability will enable a more statistically powerful design, should this exclosure experiment be extended in the future.

Moreover, the anecdotal results of the study suggest that carp were limited by the exclusion fences that were constructed at Reedy Lagoon, and that aquatic vegetation flourished in the relatively clear water in this lagoon. The anecdotal results also suggest that exclusion plots were associated with higher cover of aquatic flora in turbid wetlands at shallow water depths, yet this effect disappeared as water depth and/or turbidity increased. Mature carp appear to be the key cause of the turbidity.

3.5 Recommendations

Given the potential serious impact carp are having on wetland flora diversity in Gunbower Forest, it is recommended that the pilot study be refined, expanded and continued into the future in order to both fully understand the issue and provide a framework that guides future management.

Future research questions

- Is a treatment effect evident when sample size is increased (i.e. can we detect a difference in aquatic macrophyte diversity between fully fenced, partially fenced and control plots when more fenced and unfenced wetlands are sampled)?
- At what turbidity levels is the presence/growth of aquatic macrophytes significantly inhibited in Gunbower Forest?
- Can the relationship between carp and turbidity be quantified in wetlands in Gunbower Forest (i.e. how many/what density of carp are required to cause a noticeable change in turbidity, and how is this related to carp size).
- What is the impact on aquatic macrophytes when water birds, but not fish, are excluded from treatment (i.e. in the case of floating exclosures)?
- Does the impact of carp significantly increase in the second year of inundation?

4 REFERENCES

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