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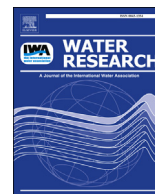
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Tree water-use strategies to improve stormwater retention performance of biofiltration systems

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ABSTRACT

Biofiltration systems are highly valued in urban landscapes as they remove pollutants from stormwater runoff whilst contributing to a reduction in runoff volumes. Integrating trees in biofilters may improve their runoff retention performance, as trees have greater transpiration than commonly used sedge or herb species. High transpiration rates will rapidly deplete retained water, creating storage capacity prior to the next runoff event. However, a tree with high transpiration rates in a biofilter system will likely be frequently exposed to drought stress. Selecting appropriate tree species therefore requires an understanding of how different trees use water and how they respond to substrate drying. We selected 20 tree species and quantified evapotranspiration (ET) and drought stress (leaf water potential; Ψ) in relation to substrate water content. To compare species, we developed metrics which describe: (i) maximum rates of ET under well-watered conditions, (ii) the sensitivity of ET and (iii) the response of Ψ to declining substrate water content. Using these three metrics, we classified species into three groups: risky, balanced or conservative. Risky and balanced species showed high maximum ET , whereas conservative species always had low ET . As substrates dried, the balanced species down-regulated ET to delay the onset of drought stress; whereas risky species did not. Therefore, balanced species with high ET are more likely to improve the retention performance of biofiltration systems without introducing significant drought risk. This classification of tree water use strategies can be easily integrated into water balance models and improve tree species selection for biofiltration systems.

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1. Introduction

Cities generate large volumes of polluted stormwater runoff that often flows untreated from impervious surfaces into natural waterways via hydraulically efficient engineered drainage networks. Urban streams that receive this runoff degrade rapidly as a result (Bernhardt and Palmer, 2007; Walsh et al., 2005b). In response, a suite of stormwater control measures (SCMs) have been developed to reduce the volume and velocity of surface runoff entering waterways (Melbourne Water, 2005; NCDEQ, 2017). Current thinking suggests that intercepting runoff from impervious surfaces can restore natural hydrological processes and improve the health of urban waterway ecosystems (Burns et al., 2012; Walsh et al., 2005a).

Biofilters, also referred to as rain gardens or bioretention cells, are SCMs which are highly efficient at reducing peak flow rates,

achieved through appropriate sizing, extended detention zones and internal water storages (Brown and Hunt, 2011b; Davis, 2008; Hatt et al., 2009; Hunt et al., 2008). The substrates used in biofilters intercept heavy metals (Davis et al., 2001) and plants play a vital role in removing some soluble nutrient pollutants (Bratières et al., 2008). Consequently, plant selection for biofiltration systems has focussed on nutrient uptake ability (Bratières et al., 2008; Hunt et al., 2015; Read et al., 2008). In contrast, relatively little attention has been given to selecting plants to improve runoff retention performance (Payne et al., 2018). Despite evapotranspiration (ET) representing the major pre-urbanisation loss of infiltrated rainfall, plants are generally considered to play a minor role in the water balance of biofilters ($ET < 5\%$ of runoff generated; Brown and Hunt (2011b)). However, several authors have recently suggested that ET losses can represent a larger proportion of the water balance in biofilters (Hess et al., 2017; Li et al., 2009; Wadzuk et al., 2015). Few studies have directly quantified ET in biofilters (Denich and Bradford, 2010; Hess et al., 2017; Wadzuk et al., 2015), as it is difficult to measure *in situ* (Hamel et al., 2014). As such, ET is

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typically estimated i) by quantifying other measured system losses (Li et al., 2009), ii) the use of standard equations (Brown and Hunt 2011a, 2011b) or iii) scaling up from leaf-level measures of stomatal conductance (Scharenbroch et al., 2016). Coupled with the perception that *ET* represents a minor component of the water balance, the difficulties in quantifying *ET* from biofilters may explain the relative lack of research focus to increase *ET* as a means of improving retention performance.

We propose that runoff retention performance of biofiltration systems can be improved by selecting trees with high *ET*. Trees, or in fact any plants, with high *ET* capacity have the potential to rapidly deplete intercepted stormwater and therefore increase storage capacity before the next runoff event (Wadzuk et al., 2015; Yuan et al., 2017). Estimations of *ET* from crop factors (K_c) and reference *ET* (ET_0) are particularly useful for comparing different vegetation types (Allen et al., 1998). Plant species commonly used in biofilters, e.g. *Carex appressa*, have a high crop factor ($K_c = 1.65$; Hamel et al. (2012)), but a small total leaf area which reduces overall transpiration. In comparison, trees can support a far greater leaf area, which can extend beyond the area of the biofilter, facilitating transpiration of large volumes of water (Litvak et al., 2012; McCarthy et al., 2011; Pataki et al., 2011) from a small 'footprint' at ground level (Berland et al., 2017; Yuan et al., 2017). Trees are also likely strong candidates for biofiltration systems, as many possess traits which have been linked with improved runoff retention and nutrient pollutant removal, including: extensive root systems, high biomass and rapid growth rates (Payne et al., 2018). Selecting tree species with a high K_c and high canopy leaf area should therefore increase runoff retention and complement, rather than jeopardise, pollutant removal performance (Denman et al., 2006; Mangangka et al., 2015; Nocco et al., 2016; Payne et al., 2018).

One significant risk of selecting trees with high *ET* is that they will rapidly deplete stored moisture, which while beneficial for retention performance, will likely expose them to drought stress (Grossiord et al., 2017). This not only puts the vegetation at risk of failure but is also likely to decrease treatment performance (Payne et al., 2018). This risk may be mitigated by designing systems with more water storage, e.g., with a deeper substrate or an internal water storage created via a raised outlet (Glaister et al., 2017; Wadzuk et al., 2015; Xiao and McPherson, 2011). However, once trees are established, they can use substantial volumes of water and are likely to encounter soil moisture deficits. Even though we expect that tree roots in unlined system will eventually extend in to the surrounding *in situ* soil, these soils will similarly experience periods of high and low water availability. Therefore, as well as having high *ET* rates when runoff water is available, trees also need a high sensitivity to drought, i.e., the ability to rapidly down-regulate *ET* as the biofilter substrates dry out. Rapid declines in *ET* as substrates dry will delay the onset of drought stress, allowing biofilter trees to survive for longer periods when substrate water is not available (Martin-StPaul et al., 2017). High sensitivity of *ET* to decreasing substrate water availability is achieved in the short term via stomatal closure. However, the level of drought stress at which trees begin closing stomata, plus how quickly and effectively they close, is highly variable among tree species (Klein, 2014; Litvak et al., 2012; Oren et al., 1999). Therefore, while it is relatively easy to select for high *ET* using traits such as a high leaf area or fast growth rates, it is difficult to select for high stomatal sensitivity, either in relation to declining moisture in the substrate or atmospheric demand (Litvak et al., 2012).

There are several ways of quantifying *ET* in relation to atmospheric demand, as well as stomatal sensitivity to drying substrates. For selecting trees with the most appropriate water use strategies for biofiltration systems, we suggest the best way to integrate these strategies is to represent trees in the water balance (e.g. Daly et al.,

2012). Stormwater management decisions at the catchment scale are underpinned by water balance models that estimate both treatment and runoff retention performance of different SCMs. Modelling an individual system, or a treatment train of systems, therefore requires accurate estimations of water inflows, outflows, fluxes and storages. Depending on the SCM type, different components receive more attention than others (Eger et al., 2017). For example, green roof water balance models focus heavily on accurate estimations of *ET* as it is the major flux driving retention performance (Poë et al., 2015). In contrast, *ET* is generally considered less important in biofiltration systems as it typically represents a minor component of the water balance (Brown and Hunt, 2011b). Improving estimates of *ET* under well-watered conditions by using species-specific crop factors, combined with functions which represent reductions in *ET* as substrates dry, will likely improve *ET* estimations in water balance models (Szota et al., 2017). Water balance models should not only quantify fluxes such as *ET*, but also provide additional outputs on vegetation status, such as drought stress (e.g. Laio et al., 2001; Porporato et al., 2001; Vico et al., 2014). Integration of a drought stress metric will aid the design process, such that retention performance can be weighed against drought risk for alternative species (Stovin et al., 2013; Szota et al., 2017; Vico et al., 2014).

The aim of this study was to identify plant water use strategies likely to improve the retention performance of biofiltration systems. Specifically, we were trying to identify species with a combination of: (i) high evapotranspiration rates under well-watered conditions to maximise retention performance, (ii) high sensitivity to developing soil moisture deficits to minimise drought stress. Our approach was to develop functions to describe these behaviours, such that they can be easily integrated into existing water balance models used to simulate performance of biofiltration systems (e.g. Daly et al., 2012).

2. Materials and methods

2.1. Species and substrate selection

To identify tree species with suitable water use strategies, we applied a habitat template approach (Lundholm, 2006) to select species which have evolved in habitats with variable access to water, but which also experience high atmospheric demand; including: ephemeral wetlands, coastal and inland riparian zones and savannas. We selected twenty tree species across these habitats. Species were selected to cover three different leaf morphologies: broadleaf (12), cladode (4) and needle (4). Occurrence data were sourced from the Atlas of Living Australia (<http://www.ala.org.au>) to summarise the climatic distribution of each species (Table 1), representing the realised niche of each species on the Australian continent (i.e., in nature, as well as in cultivation).

In spring 2015, 15 one-year old seedlings of each species were sourced from nurseries across Australia and potted in black plastic pots (diameter: 230 mm, height: 210 mm). The pots were filled by weight to achieve approximate depths of (from bottom to top): ~2 cm of 14 mm drainage gravel, a layer of geotextile (to prevent loss of substrate), ~16 cm of substrate (filter media) and ~2 cm of 7 mm gravel mulch (to minimise evaporation). The filter media was created by mixing a sandy loam soil with composted pine bark at a ratio of 3:1 (by volume). The sandy loam fraction was well-graded with a particle size distribution (by volume) of: 0.5% silt and clay (<0.05 mm), 7% very fine sand (0.05–0.15 mm), 18% fine sand (0.15–0.25 mm), 37% medium sand (0.25–0.5 mm), 28% coarse sand (0.5–1 mm), 9% very coarse sand (1–2 mm) and 0.5% fine gravel (>2 mm). Unplanted, or 'bare' pots were also included as a means of quantifying evaporation. Pots were placed outside,

Table 1

Tree species used in the experiment, described by leaf type, family, mean annual temperature (MAT) and mean annual precipitation (MAP). 'n' refers to the number of individuals in the Atlas of Living Australia database on which the climate summary data is based which includes occurrences outside their natural distribution.

Leaf type	Family	Species	n	MAT (°C)			MAP (mm)		
				Mean	Min	Max	Mean	Min	Max
Broad	Capparaceae	<i>Capparis mitchellii</i>	1703	19.6	14.2	28.6	500	162	1413
	Malvaceae	<i>Brachychiton populneus</i>	6577	16.1	3.4	26.1	726	218	2368
	Myrtaceae	<i>Agonis flexuosa</i>	814	16.4	11.4	27.0	922	293	2079
		<i>Corymbia ficifolia</i>	388	15.3	12.2	25.0	1010	405	2087
		<i>Eucalyptus camaldulensis</i>	30257	16.7	8.2	29.6	531	151	1736
		<i>Eucalyptus largiflorens</i>	7349	17.5	13.3	23.7	347	144	1321
		<i>Eucalyptus sideroxylon</i>	3053	16.3	11.6	20.3	642	255	1561
		<i>Eucalyptus torquata</i>	360	17.9	12.4	23.9	318	206	983
		<i>Melaleuca quinquenervia</i>	4836	19.3	14.1	27.6	1361	516	4150
		<i>Hymenosporum flavum</i>	1302	17.3	12.0	25.3	1213	534	4150
	Pittosporaceae	<i>Pittosporum angustifolium</i>	11057	17.4	13.0	28.0	370	142	1223
	Rutaceae	<i>Geijera parviflora</i>	6489	18.5	12.1	24.0	529	210	1195
Cladode	Cupressaceae	<i>Callitris glaucophylla</i>	14819	17.5	10.4	27.7	558	155	1744
		<i>Callitris gracilis</i>	5738	16.2	11.3	21.0	405	217	1049
		<i>Callitris rhomboidea</i>	2994	14	7.5	23.1	778	323	1953
		<i>Callitris verrucosa</i>	3664	16.6	14.1	24.1	327	171	933
Needle	Casuarinaceae	<i>Allocasuarina verticillata</i>	13948	14.8	6.4	20.3	602	187	2674
		<i>Casuarina cunninghamiana</i>	3928	17.7	10.0	27.9	867	212	3504
		<i>Casuarina obesa</i>	598	18.2	12.8	24.4	433	196	1036
		<i>Casuarina pauper</i>	4056	18.1	14.1	24.3	262	156	980

irrigated twice per day and watered with 1 g L⁻¹ liquid NPK fertiliser (20:20:20) every two weeks for 3 months. While fertilisation is avoided in the field, it was required in this glasshouse study to increase growth rates to ensure that plant roots fully explored the pot volumes prior to the start of the experiment. After 3 months, one species (*Capparis mitchellii*) was considered too small and was removed from the experiment, leaving 19 species for the experiment (plus the bare pots). After 3 months of growing outside, 15 plants of each species (including bare pots) were moved into a glasshouse and watered every 1–3 days to acclimate them to glasshouse conditions for 3 weeks. After 3 weeks of this regime, all pots were well-watered, i.e., watered to exceed capacity of the pot, every day for 1 week.

2.2. Experimental design

The 15 plants per species (including bare pots) were then allocated to one of three groups: (i) initial harvest, (ii) well-watered treatment or (iii) drought treatment, with 5 replicates per group. All pots were arranged in a randomised complete block design. The experiment started on 17/02/2016 (day 1) and ran until 30/04/2016 (day 74). Well-watered plants were watered to exceed pot capacity at dusk each evening, whereas the drought treatment plants were watered for the last time on the 16/02/2016 (day 0).

2.3. Reference evapotranspiration

We calculated reference evapotranspiration (ET_0 ; in mm d⁻¹) using the Penman-Monteith equation according to (Allen et al., 1998), using data collected from a weather station installed in the glasshouse. The weather station measured air temperature and relative humidity (HMP-60, Campbell Scientific Inc., North Logan, UT, USA), wind speed (03101-L Wind Sentry 3 cup Anemometer, R.M Young Company, Traverse City, MI, USA) and solar radiation (SP-110 Silicon Pyranometer, Apogee, Santa Monica, CA, USA). Data were recorded with a Campbell CR1000 data logger every second and stored with data averaged to 5-min resolution (CR1000, Campbell Scientific Inc., North Logan, UT, USA). During the experiment, the mean daytime temperature (0600–1800h) in the

glasshouse was 23.9 ± 0.2 °C (range: 14.5–30.2 °C) and mean day-time relative humidity was $61.5 \pm 1.1\%$ (range: 38.6–83.2%). Mean ET_0 was 1.15 ± 0.04 mm d⁻¹, ranging from 0.58 to 2.73 mm d⁻¹.

2.4. Biomass

Plants in the initial harvest group were removed from the glasshouse on day 1 and destructively harvested to determine initial total biomass for each species. Leaves were stripped and leaf area determined using a LI-3000 (LI-COR Biosciences, Lincoln, NE, USA). Stems were cut at the substrate surface, substrate was washed from roots which were then patted dry. Fresh weights were determined separately for leaves, stems and roots before drying in a 70 °C oven for one week to determine dry weight. The same procedure was followed for the final harvest. All well-watered plants were harvested on day 74, whereas droughted plants were removed from the glasshouse and harvested once leaves were desiccated.

2.5. Evapotranspiration

Daily evapotranspiration (ET) was determined from pot weights using a scale (Mettler Toledo, Melbourne, Australia, 1g resolution). Pot weights were measured three times per day; at pre-dawn (0400–0600 h), midday (1200–1400 h) and dusk (1800–2000 h). Well-watered pots were re-watered every day following the dusk measurement and no post-watering weight was captured after drainage had finished. We therefore were not able to quantify night-time evapotranspiration. To quantify ET , we first calculated the weight of water evapotranspired from the change in pot weight from pre-dawn to dusk each day. Due to differences in biomass among replicates within a species, we standardised the weight of water evapotranspired each day, such that daily evapotranspiration (ET g d⁻¹) was calculated as:

$$ET = \frac{W_{pre} - W_{dusk}}{W_{biomass[1:5]}} \times W_{final.biomass}$$

where W_{pre} and W_{dusk} represent pot weights at pre-dawn and dusk (in g), $W_{biomass[1:5]}$ represents the total dry biomass of replicates 1 to

5 on the same day (in g) and $W_{final,biomass}$ represents the mean total dry biomass for all 5 replicates as determined at the final harvest (in g). To relate ET to ET_0 , we converted the weight of water evapotranspired ($g\ d^{-1}$) to volume ($ml\ d^{-1}$), then to depth ($mm\ d^{-1}$), by dividing the volume by the surface area of the pot ($415.5\ cm^2$). We estimated the total dry biomass of each replicate ($W_{biomass[1:5]}$) for each day of the experiment using a linear interpolation between the initial and final harvests. A linear assumption is reasonable for the relatively short timeframe of this experiment and agreed with the observed linear increase in pre-dawn pot weights in the well-watered treatment over time (data not shown).

2.6. Substrate volumetric water content

Substrate volumetric water content was determined from pot weights at predawn, midday and dusk each day of the experiment. To determine water content at any given point, the dry weight of non-substrate components (pot, drainage gravel, mulch gravel and the geotextile) and the total fresh plant weight were subtracted from the pot weight. The dry weight of each non-substrate component was determined for all bare pots (droughted and well-watered; $n=10$) at the end of the experiment after drying at $105\ ^\circ C$ for 1 week. Total fresh plant weight on each day of the experiment was estimated from linear interpolation of the change in biomass between the initial and final harvests. Substrate volumetric water content (S ; in %) was then calculated as:

$$S = \frac{W_{wet} - W_{dry}}{W_{dry}} \times BD \quad (2)$$

where W_{wet} and W_{dry} are the wet and dry weights of the substrate (in g) and BD is the bulk density of the substrate (in $g\ cm^{-3}$). The bulk density of the substrate was determined from all bare pots (droughted and well-watered; $n = 10$), by dividing the dry weight of the substrate by the volume it occupied in the pots.

2.7. Plant drought stress

To determine plant drought stress, leaf water potential (Ψ) samples were taken at predawn and midday every 1–5 days. Samples were collected from both well-watered and droughted plants, with the sampling frequency designed to cover the full range of S . At each sampling, a leaf or terminal shoot was cut, sealed in a zip-lock plastic bag and stored in an insulated box on ice until it was measured. Leaf water potential was measured using a Scholander-type pressure chamber (Soilmoisture Equipment Corp, Santa Barbara, CA, USA). A wet sponge was placed in the chamber to minimise leaf dehydration during measurements.

2.8. Data and statistical analyses

Three metrics were derived to describe the water use strategy of each tree species: i) maximum ET under well-watered conditions (i.e., their crop factor; K_c), ii) sensitivity of ET to declining S (S^{ET}) and iii) sensitivity of Ψ to declining S (S^Ψ).

To determine K_c for each species, linear regression was used to relate ET and ET_0 for well-watered plants; where the slope of the relationship is equivalent to K_c . These models were forced through the origin and as such contain bias, therefore, adjusted R^2 and P -values were derived from a regression of fitted and observed values of ET . To describe sensitivity of ET to declining S , Multi-Adaptive Regression Splines (MARS; or “broken-stick models”) were fitted to the relationship between daily ET as a proportion of ET_0 (ET/ET_0) and S for the droughted plants only. We used ET/ET_0 to exclude the influence of evaporative demand and focus on ET sensitivity to

declining S . To describe the sensitivity of leaf water potential to declining S , broken-stick models were also fitted to the relationship between Ψ (predawn and midday) and S for both well-watered and droughted plants. By using broken-stick models, we could identify the critical value of S at which both ET/ET_0 and Ψ began to decline from their maximum values in response to drying of the soil substrate. We refer to the critical ET/ET_0 value as ‘ S^{ET} ’ and the critical Ψ as ‘ S^Ψ ’. As broken-stick models were non-linear, adjusted R^2 and P -values were derived from linear models of fitted and observed values of both ET/ET_0 and Ψ . Broken-stick models were fitted with R version 3.4.1 (R Core Team, 2017) using the ‘earth’ package (Milborrow, 2015) which is based on the work of Friedman (1991). We also used one-way ANOVA and Tukey’s post-hoc tests to identify any significant differences among leaf types for each metric. To determine the influence of plant size, we used linear regression to relate final total biomass (dry weight at the end of the experiment) to each of the three metrics (K_c , S^{ET} and S^Ψ). For all these analyses, residuals for each model were inspected for heteroscedasticity, data were checked for normality (Shapiro-Wilk test) and log-transformed where necessary.

To categorise species based on these three metrics (K_c , S^{ET} and S^Ψ), we used a combination of Principal Components Analysis (PCA) and K-means Cluster Analysis. Each metric was centred and standardised (mean = 0, standard deviation = 1) and initial analysis of the PCA biplot indicated three groups of species. Using the ‘cluster’ package (Maechler et al., 2017) in R (R Core Team, 2017), we used the K-means algorithm of Hartigan and Wong (1979) to formally classify species into one of three groups (i.e., $K = 3$), based on our metrics. The initial random cluster assignment of each observation can affect how the K-means algorithm assigns species to groups (James et al., 2013), therefore we ran this analysis with 1000 random starting points (i.e., $nstart = 1000$).

3. Results

3.1. Maximum rates of evapotranspiration

Crop factors (K_c) of well-watered plants, represented by the slope of the relationship between ET and ET_0 , indicate how efficiently each species can deplete substrate water. Values of K_c varied from 0.66 to 1.87 (Fig. 1), with *A. flexuosa*, *E. camaldulensis* and *M. quinquenervia* showing very high water use ($K_c > 1.5$). The bare (unplanted) treatment showed a K_c of 0.48 which indicates the contribution of evaporation to ET . The strength of the relationships between ET_0 and ET under well-watered conditions were weak ($R^2 = 0.16$ – 0.54), but all models were highly significant ($P < 0.001$). There were no significant differences in K_c among leaf types ($P = 0.516$).

3.2. Sensitivity of evapotranspiration to declining substrate water content

The sensitivity of ET (adjusted for ET_0 ; i.e., ET/ET_0) to declining substrate water content (S) indicates the value of S at which different species started reducing ET (S^{ET}). We suggest that a species with high sensitivity will show reductions in ET/ET_0 at higher values of S . Species showed decreases in ET/ET_0 at values of S between 11.3 and 19.7% (Fig. 2). *C. gracilis*, *G. parviflora* and *C. ficifolia* were the most sensitive species, showing reduced ET/ET_0 at substrate water contents $> 19\%$. Broken-stick models described the variability in ET/ET_0 in relation to S , with R^2 between 0.34 and 0.87 (Fig. 2). There were no significant differences in S^{ET} among leaf types ($P = 0.619$).

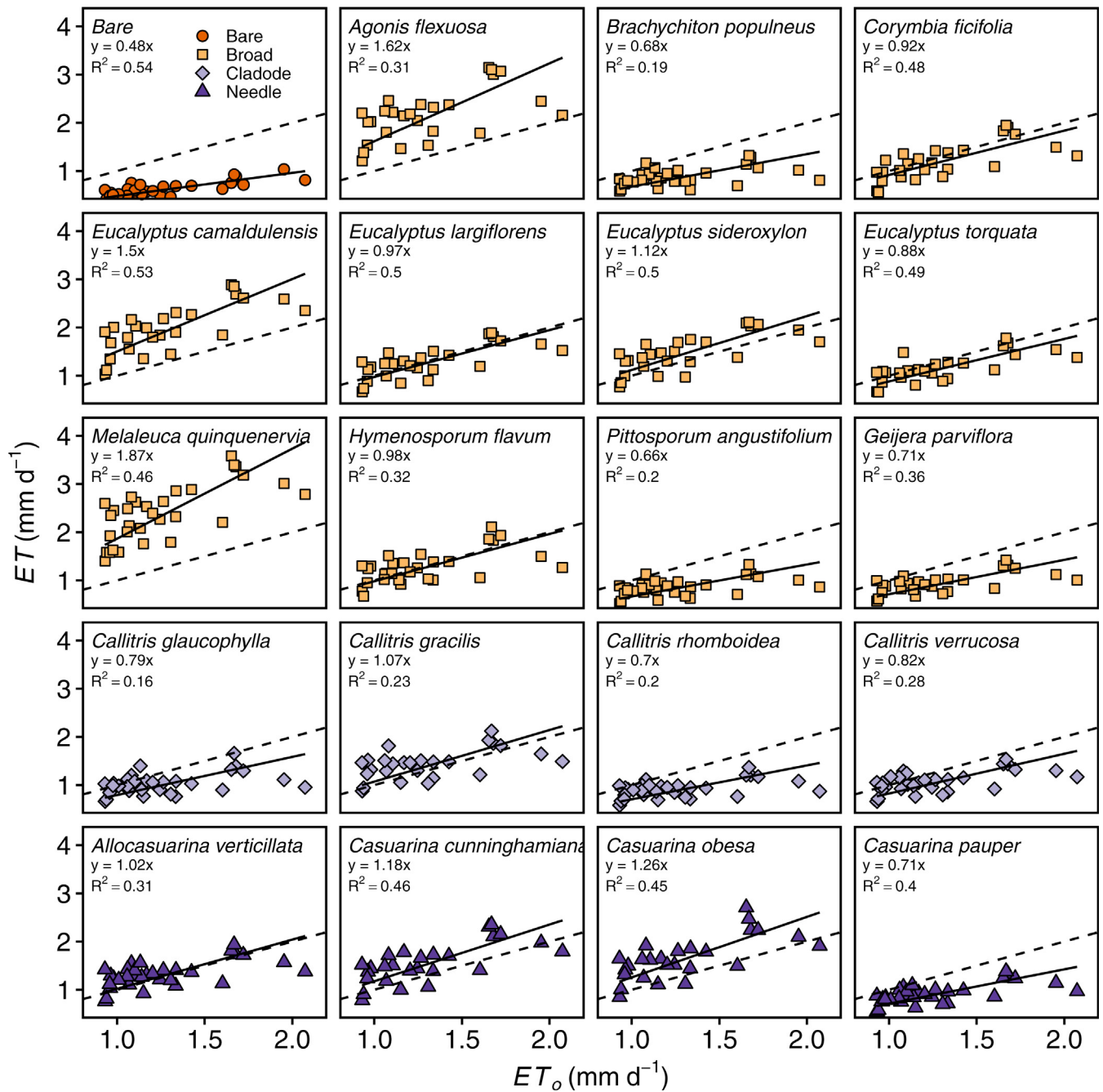


Fig. 1. Relationship between evapotranspiration (ET) and reference evapotranspiration (ET_0) for well-watered plants. Symbols indicate leaf type and the equation is equivalent to the crop factor (K_c) for each species (including bare). R^2 was derived from fitted versus observed values (as line was forced through the origin) and all $P < 0.001$. Dashed line represents the 1:1 relationship.

3.3. Sensitivity of plant drought stress to declining substrate water content

The sensitivity of drought stress (leaf water potential; Ψ) to declining S indicates the value of S where different species started to show signs of drought stress (S^Ψ). We suggest that a less sensitive species shows increased drought stress (i.e., a lower value of Ψ) at a lower value of S . Several species, including *P. angustifolium*, *A. verticillata* and *C. rhomboidea* showed the onset of drought stress at very low values of S ($S^\Psi < 9\%$; Fig. 3). Leaf water potential was strongly related to S ($R^2 = 0.39$ – 0.88) in all species except *Brachychiton populneus* ($R^2 = 0.08$); however, all models were highly

significant ($P < 0.001$). There were no significant differences in S^Ψ among leaf types ($P = 0.167$).

3.4. Categorising species according to their water use and sensitivity to declining substrate water content

The first two PCA axes explained 81.9% of the variation (Fig. 4). The first axis (PC1) explained 50.2% of the variation and summarised variation between maximum rates of ET (i.e., the K_c) and sensitivity of drought stress to declining substrate water content (S^Ψ). That is, species with a high K_c that showed the onset of drought stress at relatively high values of S showed positive PC1

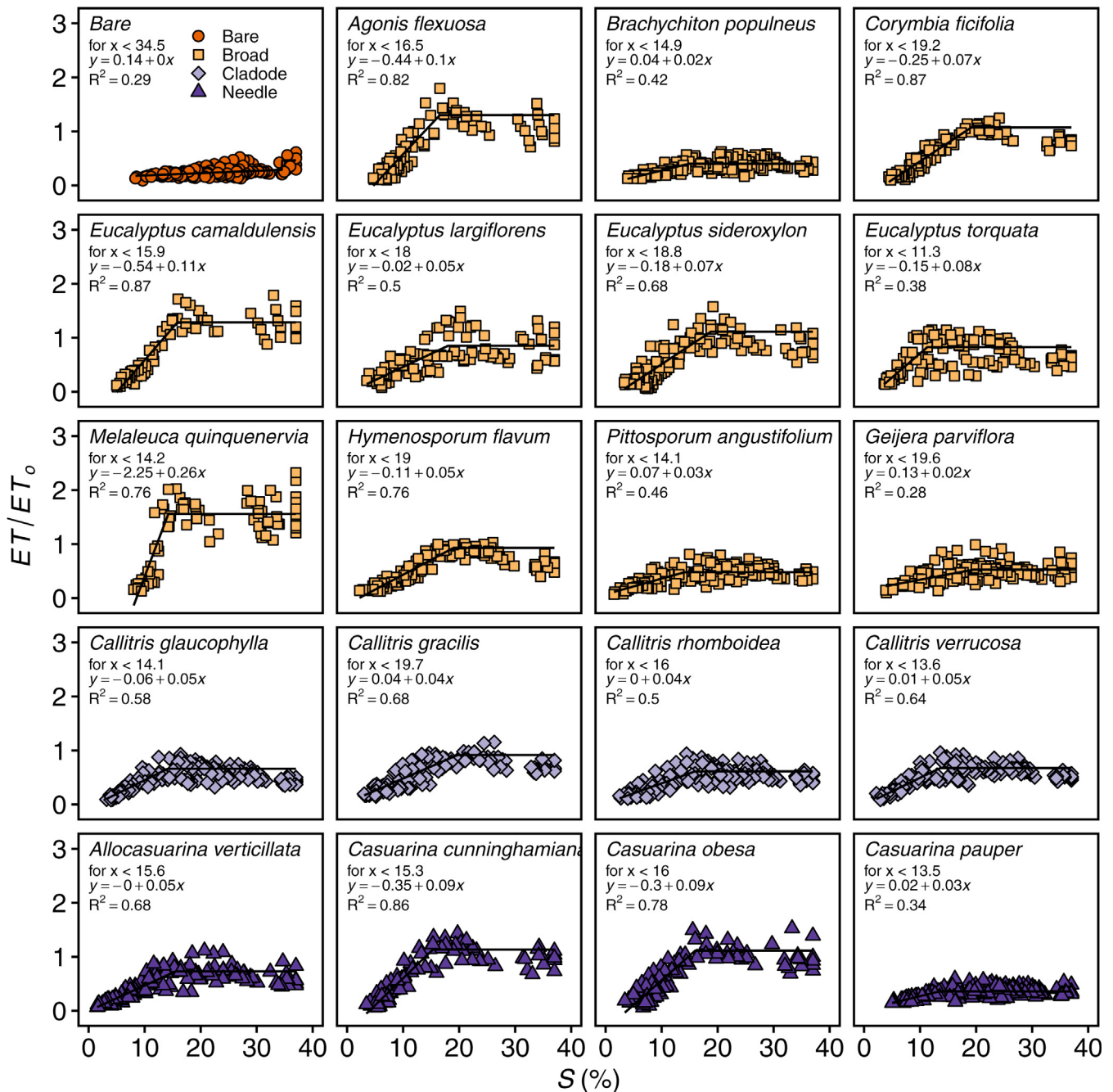


Fig. 2. Relationship between evapotranspiration (ET) as a proportion of reference evapotranspiration (ET_0) and substrate volumetric water content (S) for droughted plants. Symbols indicate leaf type and the equation applies to the range of S values below the breakpoint, where the breakpoint is equal to S^{ET} . R^2 was derived for fitted versus observed values and all $P < 0.001$.

values, whereas species with a low K_c that showed increasing drought stress at low values of S showed negative PC1 values. The second axis (PC2) was strongly correlated with sensitivity of ET (S^{ET}) to declining values of S and PC2 explained 31.7% of the variation. Species with more positive PC2 values had higher values of S^{ET} , indicating species that reduced ET at higher values of S ; i.e., they showed higher sensitivity to declining S .

Using K-means cluster analysis, we classified species into three groups: 1) *risky*, 2) *balanced* and 3) *conservative* to describe their overall water use strategy (Table 2). Risky species showed high K_c and only reduced ET after showing an increase in drought stress (e.g. *Melaleuca quinquenervia*). Balanced species also showed high

K_c , but they reduced ET prior to the onset of drought stress (e.g. *Eucalyptus sideroxylon*). The conservative species were low water users (low K_c) and only showed reduced water use and drought stress at very low substrate water contents (e.g. *Casuarina pauper*).

3.5. Relationships between plant biomass and maximum rates of water use and sensitivity to declining substrate water content

The final total dry biomass of well-watered plants was strongly and positively related to maximum rates of ET , i.e., species K_c (Fig. 5A). There were no relationships between final dry biomass and S^{ET} or S^{W} at $\alpha = 0.05$; however, final dry biomass was related to

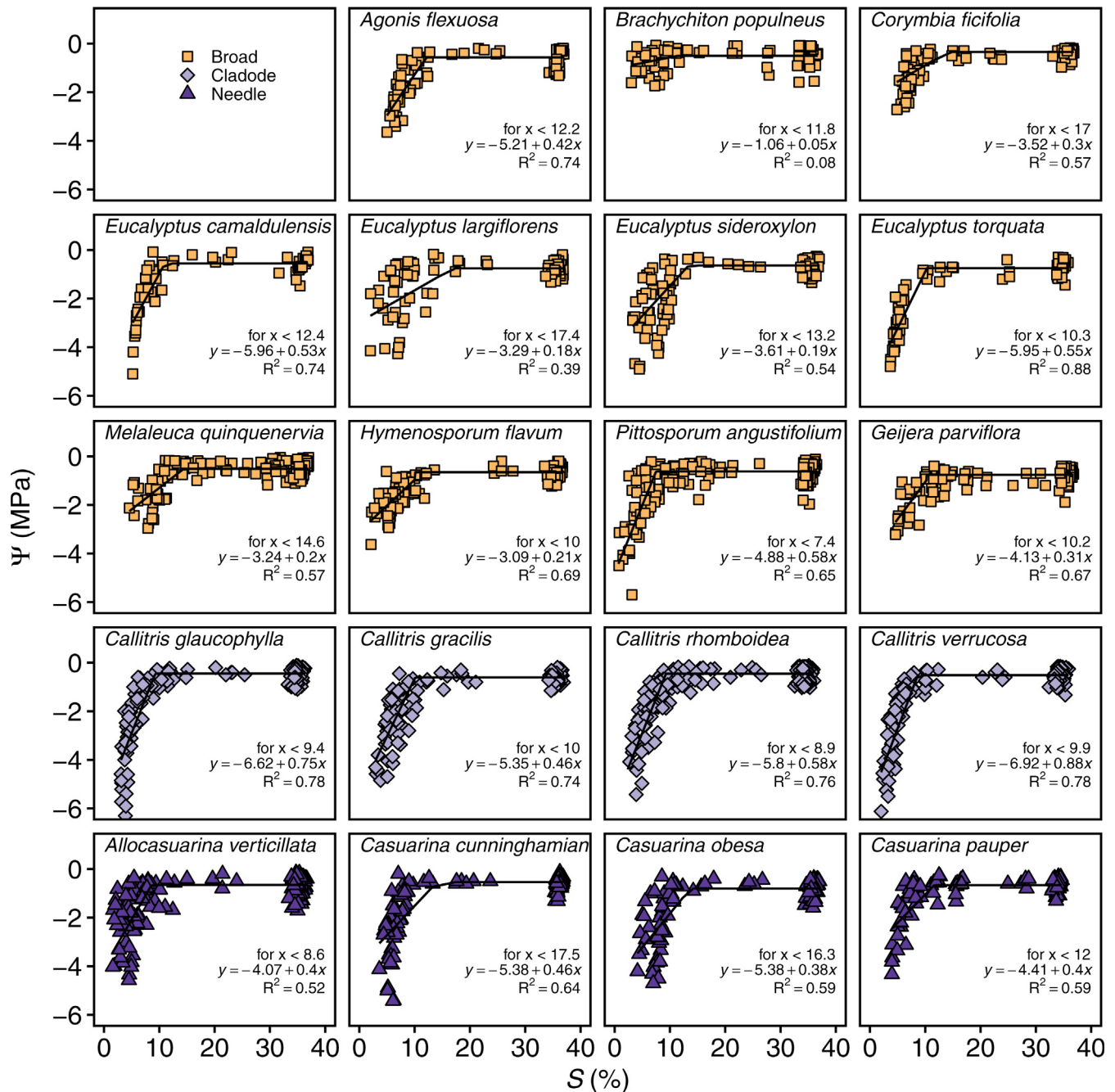


Fig. 3. Relationship between leaf water potential (Ψ) and substrate volumetric water content (S) for well-watered and droughted plants. Symbols indicate leaf type and the equation applies to the range of S values below the break point, where the break point is equal to S^b . R^2 was derived for fitted versus observed values and all $P < 0.001$.

S^b at $\alpha = 0.10$ (Fig. 5B and C).

4. Discussion

4.1. Integrating high water users into biofiltration systems

Three species showed very high ET ($K_c > 1.5$) under well-watered conditions which, when planted in a biofilter, should facilitate rapid depletion of substrate water compared with low water users ($K_c < 1$). Rapid depletion of the substrate via ET will increase storage capacity between inflow events and likely improve runoff retention performance (Hess et al., 2017; Mangangka et al., 2015; Nocco et al., 2016; Yuan et al., 2017). The high K_c values in

our study were similar to those used for sedges ($K_c = 1.65$) in previous biofiltration studies (Hamel et al., 2012). This suggests limited advantage in the use of trees as compared with commonly used species, e.g., *Carex appressa* which have also demonstrated high nitrogen removal efficiency (Bratières et al., 2008). However, we studied tree seedlings in a glasshouse and although our K_c values were appropriately scaled to the pot surface area, mature trees will likely have higher K_c values (Allen et al., 1998). Tree canopy areas in the field will exceed the area of a typical biofilter as the tree is maturing, resulting in higher ET per unit treatment area. The area used to adjust ET from volume to depth is central to this assumption and for comparing vegetation cover types. For instance, urban trees can utilise large volumes of water (up to 260 kg d^{-1} ; Pataki et al.

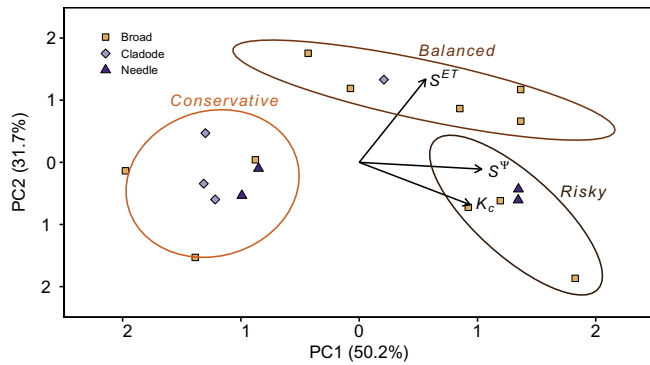


Fig. 4. Result of the Principal Components Analysis (PCA), showing the loading vectors for the three metrics: i) maximum rates of evapotranspiration (ET) under well-watered conditions (i.e., crop factor; K_c), ii) sensitivity of ET to declining substrate volumetric water content (S^{ET}) and iii) sensitivity of Ψ to declining substrate volumetric water content (S^{Ψ}). The result of the K-means cluster analysis is shown by the ellipses which categorise the species into three groups which we labelled: risky, balanced and conservative (see Table 2). The axes show the variance explained by the first (PC1) and second (PC2) principal components. The total variance explained by PC1 and PC2 was equal to 81.9%. Symbols represent leaf type.

(2011)). When this volume is converted to depth at the plot scale with an assumed tree density (e.g. 100 trees ha^{-1}); the depth of water transpired is similar to, or significantly lower than, other vegetation surfaces, including grass (Litvak et al. 2014, 2017a). However, if volumetric transpiration is expressed relative to the surface area of a biofilter, it is likely that trees with a high K_c (>1.5) will use more water than an equivalent surface area planted with sedges or rushes. This hypothesis reveals the major limitation of our study, in that it was performed with seedlings in a glasshouse, rather than trees in the field. Field studies describing ET relative to inflow events, soil moisture regimes and a wide range of ET_0 are required to support this hypothesis.

The extent to which selecting trees with a high K_c will improve runoff retention will be influenced by the biofiltration system design. It has been suggested that evapotranspiration can account for a significant proportion of generated runoff (19%), even when sized at 4.5% of their impervious catchment area (Li et al., 2009). This system/catchment area ratio is generally considered adequate

to decrease runoff volumes, peak flows and achieve regional pollutant removal targets (NCDEQ, 2017; Payne et al., 2015). However, at this size, the volume of inflow is likely to overwhelm any gains in runoff retention achieved by selecting species with higher ET . Increasing system surface area to facilitate ET is unlikely to be practicable, therefore solutions which increase storage volume without increasing system area are required. Increasing storage by increasing depth of the filter media from current recommendations of ~900 mm (e.g. NCDEQ, 2017; Payne et al., 2015) would be highly suitable for deep-rooted tree species and would return to the original design guidelines of Clar and Green (1993), which recommended 1.2 m of substrate for adequate root development of trees and shrubs (Brown and Hunt, 2011b). Storage volume can also be increased by integrating an internal water storage via a raised outflow (Brown and Hunt, 2011a; Glaister et al., 2017; NCDEQ, 2017; Winston et al., 2016), utilising sub-surface structures or engineered soils (Bartens et al., 2009; Page et al., 2015) and/or outlet control structures (Scharenbroch et al., 2016), which will provide a greater opportunity for trees to utilise captured runoff between inflow events (Hess et al., 2017; Scharenbroch et al., 2016; Wadzuk et al., 2015; Xiao and McPherson, 2011). Deeper substrates and more complicated sub-surface structures will likely increase the cost of installation, however, we suggest that integrating trees into biofiltration systems without increasing storage volume may limit the gains achieved by selecting tree species with a high K_c .

As per other stormwater control measures, climate will be a major determinant of the extent by which selecting species with a high K_c will improve retention performance (Hunt et al., 2006; Stovin et al., 2013). In the short term, ET_0 in the days following rainfall will be a major driver of ET and therefore retention of subsequent events, which is why the antecedent dry weather period is often strongly related to retention performance (Mangangka et al., 2015; Yuan et al., 2017). In very cold climates with year-round low ET_0 , selecting high water users will likely have a minimal impact on retention performance. Even in warmer climates where most rainfall occurs during winter when ET_0 is low, species with a high K_c may take a long time to dry out the substrate between events which will limit their potential benefits. In temperate cities such as Melbourne, Australia which has an even rainfall distribution (54.0 ± 1.6 mm per month; long-term (128 year) average for Melbourne Regional Office station 86071; sourced

Table 2

Summary of the three categories of species identified according to crop factor (K_c), sensitivity of evapotranspiration (S^{ET}) and leaf water potential (S^{Ψ}) to declining substrate water content. K_c values are taken as the slope between reference evapotranspiration (ET_0) and evapotranspiration (ET), both measured in $mm\ d^{-1}$ (see Fig. 1). S^{ET} and S^{Ψ} refer to the substrate volumetric water contents where ET and Ψ begin to decline as the substrate dries which indicates species sensitivity (see Figs. 2 and 3).

Category	Species	Leaf type	K_c	S^{ET} (%)	S^{Ψ} (%)	Description
Risky	<i>Agonis flexuosa</i>	Broadleaf	1.62	16.5	12.2	Very high or high water users which only reduce water use after the onset of drought stress
	<i>Eucalyptus camaldulensis</i>		1.50	15.9	12.4	
	<i>Melaleuca quinquenervia</i>		1.87	14.2	14.6	
	<i>Casuarina cunninghamiana</i>	Needle	1.18	15.3	17.5	
	<i>Casuarina obesa</i>		1.26	16.0	16.3	
Balanced	<i>Corymbia ficifolia</i>	Broadleaf	0.92	19.2	17.0	High water users which reduce water use before the onset of drought stress
	<i>Eucalyptus largiflorens</i>		0.97	18.0	17.4	
	<i>Eucalyptus sideroxylon</i>		1.12	18.8	13.2	
	<i>Geijera parviflora</i>		0.71	19.6	10.2	
	<i>Hymenosporum flavum</i>		0.98	19.0	10.0	
	<i>Callitris gracilis</i>	Cladode	1.07	19.7	10.0	
Conservative	<i>Brachychiton populneus</i>	Broadleaf	0.68	14.9	11.8	Low water users which only reduce water use and show drought stress at low substrate water contents
	<i>Eucalyptus torquata</i>		0.88	11.3	10.3	
	<i>Pittosporum angustifolium</i>		0.66	14.1	7.4	
	<i>Callitris glaucophylla</i>	Cladode	0.79	14.1	9.4	
	<i>Callitris rhomboidea</i>		0.70	16.0	8.9	
	<i>Callitris verrucosa</i>		0.82	13.6	9.9	
	<i>Allocasuarina verticillata</i>	Needle	1.02	15.6	8.6	
	<i>Casuarina pauper</i>		0.71	13.5	12.0	

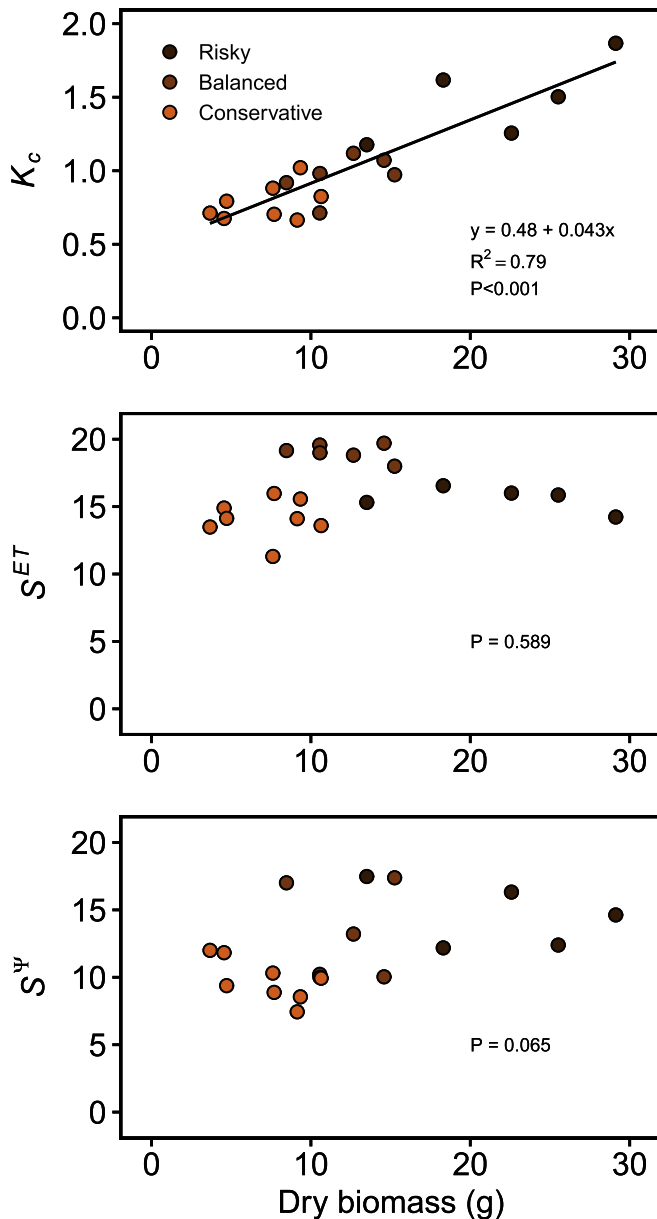


Fig. 5. Relationships between plant dry biomass at the end of the experiment and A) maximum rates of evapotranspiration under well-watered conditions (i.e., crop factor; K_c), B) sensitivity of ET to declining substrate volumetric water content (S^{ET}) and C) sensitivity of Ψ to declining substrate volumetric water content (S^{Ψ}). R^2 and P -values were derived from linear regression. Symbols indicate species groups: *risky*, *balanced* and *conservative* (see Table 2).

from SILO (Jeffrey et al., 2001)); species with a high K_c will likely have significant opportunity to deplete stored water between events in all but the coldest winter months (Wadzuk et al., 2015). We therefore suggest that species with a high K_c should only be prioritised in climates with an even distribution of rainfall or higher rainfall in the summer months.

4.2. Selecting trees with high water use and sensitivity to drying substrates

Selecting for a high K_c alone is likely to result in drought stress and potentially tree death; however, this will depend on the regularity of inflows, system storage and tree water use. For example, even in Melbourne, where rainfall is relatively evenly distributed

over the year, the 20% annual exceedance probability (AEP) antecedent dry weather period (ADWP) is 31 days, and 50 days for the 1% AEP (Melbourne Regional Office station 86071; sourced from SILO (Jeffrey et al., 2001)). A biofilter sized to 2% of a 100 m² impervious catchment, with a depth of 1 m, could store 700 L of water with an assumed field capacity of 35%. Established trees have the potential to utilise 70–260 L d⁻¹ (Litvak et al., 2012), therefore 700 L of storage could be rapidly depleted and an extended ADWP period will likely expose trees to drought stress. Therefore, our distinction between *risky* and *balanced* species is important as it describes how species responded to substrate drying. Species identified as *risky* showed increased drought stress before they down-regulated ET , putting them at greater risk of experiencing severe drought stress. Down-regulation of ET is related to stomatal sensitivity to decreasing leaf water status (i.e., Ψ) which during drought is primarily governed by substrate water content (Klein, 2014; Martin-StPaul et al., 2017; Porporato et al., 2001). As leaves dehydrate, they lose turgor which results in stomatal closure and a decrease in transpiration. Species differ in the level of drought stress at which they begin to regulate water loss (Klein, 2014; Oren et al., 1999). Our *risky* species dried out more before closing stomata. This strategy is most likely to improve retention performance as *risky* species will effectively continue to transpire until water in the substrate is depleted. However, this strategy comes with significant risk, as once soil moisture is depleted, *risky* species have already dehydrated and therefore have less water stored internally to facilitate recovery once drought stress is alleviated (Blackman et al., 2016). In contrast, *balanced* species reduced ET at higher substrate water contents, effectively delaying the onset of drought stress and reducing the risk of severe drought stress developing. Selecting *balanced* species represents a trade-off as they were not among the highest water users, however, they all showed values of $K_c > 1$. We therefore suggest that *balanced* species with high K_c and the ability to reduce water use as substrates dry will improve runoff retention performance, while minimising the risk of drought stress.

As we would expect, K_c was strongly related to plant biomass, where larger plants used more water (Lundholm et al., 2010; Nagase and Dunnett, 2012; Payne et al., 2018; Yuan et al., 2017). However, plant biomass was not related to the substrate water content at which plants down-regulated (S^{ET}) and poorly related to the S where plants showed increasing drought stress (S^{Ψ}). Therefore, while selecting tree species based on size, leaf area or growth rates may inform selection for maximum rates of ET , it will likely not describe drought response and therefore risk of drought stress and possible death. We evaluated these responses in the glasshouse, but ideally, they should be tested also in the field, e.g., by instrumenting trees with sapflow sensors and stem or leaf psychrometers to monitor diel patterns of transpiration and water potential. These measures should be combined with substrate moisture content (including water level sensors where subsurface reservoirs are installed) and meteorological measures to calculate as well as adjust ET_0 and vapour pressure deficit (D) for site-specific factors (Litvak et al., 2017b), to develop functions which describe drought response. Ideally, these functions should be related to physiological (e.g. hydraulic vulnerability; Brodribb et al. (2010)), morphological or anatomical traits (e.g., wood anatomy; Litvak et al. (2012)), such that drought response can be estimated in the absence of glasshouse or field data when selecting new species.

Several of the species we evaluated had a low K_c and only down-regulated ET and became drought stressed at very low substrate moisture contents. We classified these as *conservative* species and suggest that although they are more likely to survive extreme drought in biofiltration systems the low ET would decrease runoff retention compared with *balanced* or *risky* species. These conservative species would be highly suitable in urban plantings not

irrigated with stormwater, or in stormwater control measures in arid climates with strongly seasonal rainfall. There is currently significant focus on selecting drought tolerant urban tolerant trees for hotter, warmer future climates (Steenberg et al., 2017). Integration of tree plantings with stormwater control measures may reduce the vulnerability of currently used tree species (e.g. May et al., 2013; Nitschke et al., 2017). However, this will depend on whether tree species vulnerability is driven by soil moisture deficit, atmospheric demand or heat stress (Grossiord et al., 2017).

4.3. Implementing tree water use strategies in water balance models

The three classifications we derived can be easily incorporated into existing water balance models for biofilters, e.g. Daly et al. (2012). This will allow for comparisons of retention performance among potential tree species in relation to both climate and system design. To briefly describe one approach, we follow the assumptions of Daly et al. (2012), where ET is primarily responsible for depletion of water and will occur at a maximum rate (K_c ; or E_{max} in Daly et al. (2012)) until the critical substrate water content is reached (S^{ET} ; or S^* in Daly et al. (2012)). In contrast to Daly et al. (2012), our results suggest that below S^{ET} , ET will decline linearly towards a species-specific threshold substrate water content, rather than toward the permanent wilting point of the substrate (S_w in Daly et al. (2012)). We attribute this to differences in the ability of species to adjust leaf water potentials to facilitate transpiration at values of S below the permanent wilting point of the substrate (Sanders and Arndt, 2012). We suggest that our methods of deriving K_c and S^{ET} can be applied to any species and integrated into water balance models with relatively minor adjustments, allowing us to compare differences in ET and therefore runoff retention among alternative species.

When employing water balance models for biofilter design and species selection, it is relatively simple to include an estimate of drought stress as an output to weigh against retention performance (Szota et al., 2017; Vico et al., 2014). In the absence of functions to describe plant water status in relation to substrate moisture, incidence of drought stress can be inferred from the substrate water content, e.g. number of days below permanent wilting point (Stovin et al., 2013). However, using a species-specific value of S indicating the onset of drought stress (S^w), or the substrate permanent wilting point, only provides a relative indication of drought risk, rather than an estimate of tree mortality (e.g. lethal water potential; Brodribb et al. (2010)). It has been suggested that once substrate moisture has been depleted, the volume of water stored within various plant tissues (capacitance) may predict how long the plant will 'survive', that is, retain the ability to recover once stress is alleviated (Blackman et al., 2016). However, we suggest that biofiltration systems should be designed such that vegetation does not experience levels of drought stress which could lead to damage the hydraulic systems of the plant and result in slow recovery of transpiration. Therefore, quantifying drought risk using either the substrate permanent wilting point, or metrics derived from species-specific functions (e.g. S^{ET} or S^w) remain useful indicators. Overall, we suggest that integrating an estimate of drought stress into water balance models will facilitate species selection and allow retention performance to be weighed against drought risk (Szota et al., 2017).

5. Conclusions

We described tree water use strategies to determine their potential to increase ET and therefore retention performance of biofiltration systems. We suggest that species with a high K_c and high

stomatal sensitivity (high S^{ET}) will improve retention performance while minimising the risk of drought stress. The benefits of this approach are more likely to be realised if the volumetric storage capacity of biofiltration systems is increased by increased substrate depth, integration of an internal water storage and use of outlet control structures. We have described the tree water use strategies identified such that they can be integrated into water balance models as part of the design process. We suggest that the strategies identified should now be compared in the field to determine how they will perform in response to realistic inflow regimes, soil moisture and evaporative demand.

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