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Manuscript

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Evapotranspiration of a willow cultivar (Salix miyabeana SX67) 1

grown in a full-scale treatment wetland 2

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Abstract

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Since woody plants like willow are used increasingly in treatment wetlands, there is a growing need to characterize their ecophysiology in these specific growing conditions. For instance, potential evapotranspiration (ET) can be greatly increased in wetlands, due to factors like high water availability as well as oasis and clothesline effects. Few studies report willow ET rates measured in full-scale constructed wetland conditions, and fewer still in a temperate North-American climate. The objective of this study was to measure and model evapotranspiration of a commonly used willow cultivar, Salix mivabeana (SX67), to provide the ET rates and crop coefficient for this species. During two growing seasons, we studied a 48 m² horizontal subsurface flow willow wetland located in eastern Canada, irrigated with pretreated wood preservative leachate. We found a mean monthly evapotranspiration rate of 15 mm/day, for a seasonal cumulative ET value of 2785 mm and a mean crop coefficient of 4.1. Both the evapotranspiration results and leaf area index (LAI) were greater than most results reported for open field willow plantations. Maximal stomatal conductance (\bar{G}_s) was higher than that expected for deciduous trees and even for wetland plants, and mean values correlated well with temperature, solar radiation, relative humidity and day of the year. We demonstrated that an ET model using \bar{G}_s , LAI and water vapor pressure deficit (VPD) as parameters could predict the evapotranspiration rate of our wetland. This simplification of traditional ET models illustrates the absence of evapotranspiration limitations in wetlands. Furthermore, this study also highlights some factors that can enhance ET in treatment wetlands. Our results should both improve the design of treatment wetlands using fast growing willows, and provide a simple ET predictive model based on major evapotranspiration drivers in wetlands.

46 **Keywords**: willow crop coefficient, wetland evapotranspiration, stomatal conductance,

willow leaf parameters, evapotranspiration modelling, zero-discharge wetlands

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1. Introduction

Treatment wetlands, or vegetation filters, are now commonly used for treatment of various types of wastewater (Valipour and Ahn, 2017). "Artificial" wetlands are generally planted with herbaceous plants like *Phragmites*, *Typha*, graminoids or other aquatic and semiaquatic species (Kadlec and Wallace, 2008). More recently, woody species of the Salix genus (willows), generally studied for biomass production, are being tested and used for wastewater treatment purposes. Salix species are mostly hydrophilic and tolerate hypoxic conditions and great water fluctuations well, have a high growth rate and develop a vigorous root system (Kuzovkina et al., 2008), making them good candidates for treatment wetland purposes. Another advantage of using woody plants for water treatment is the added value of biomass production that can be used for bioenergy and biofuel processes (Duggan et al., 2005). Consequently, there is growing interest in willow for use in treatment of landfill leachate, domestic wastewater or other nitrogen rich wastewaters (Białowiec et al., 2003; Dimitriou and Aronsson, 2011; Guidi et al., 2014). Fast growing willows are also known for their great evapotranspiration (ET), which led to the development of a new specific type of treatment wetlands called "zero-discharge wetlands" (ZDWs; Dotro et al., 2017). The design of ZDWs is based mainly on the ET capacity of the plant selected. They operate without liquid effluent, immobilizing contaminants in the wetland substrate and preventing any release of residual contamination in the environment. Depending on the type of water contamination, ZDWs can function as the final step of a

treatment plant or as a secondary treatment. Such wetlands are now well implanted in 69 Scandinavian countries, mainly in Denmark, where the concept was first developed 70 (Gregersen and Brix, 2001; Brix and Arias, 2011), and Ireland (Curneen and Gill, 2014). 71 Conclusive tests have also been performed in Mongolia, under very cold climatic 72 73 conditions (Khurelbataar et al., 2017), and zero-discharge wetlands are currently being 74 tested in other locations. Sound scientific knowledge of the ET rate of the species used is an essential tool to design 75 76 a treatment wetland because of the direct impact it will have on the wetland hydraulics 77 (Kadlec and Wallace, 2008) and its removal performance (Białowiec et al., 2014). It is even 78 more important for zero-discharge wetlands, where ET is the main "treatment" process, 79 ensuring that no liquid waste will flow out of the wetland. While many studies have been published on willow ET, very few concern willows growing in full-scale treatment wetland 80 conditions. However, ET in artificial wetlands can differ greatly from ET measured in a 81 82 plantation, and can significantly surpass potential ET (Dotro et al., 2017). The willow species most studied for ET is Salix viminalis, its hybrids and their numerous 83 cultivars (Frédette et al. 2018). Although widely used in Europe, some long-term studies 84 have pointed out that, in North America, cultivars of S. viminalis are more prone to diseases 85 and insect attacks than other cultivars (Labrecque and Teodorescu, 2005; Nissim et al., 86 2013). Instead, other cultivars from species like Salix eriocephala, S. purpurea, S. nigra 87 and S. miyabeana are frequently used (Smart and Cameron, 2008). In eastern Canada, 88 Nissim et al. (2013) concluded that S. miyabeana and some indigenous species were more 89 90 suited for plantation than S. viminalis. Salix miyabeana has also shown the highest biomass production among cultivars (Labrecque and Teodorescu, 2005; Pitre et al., 2010), good 91

phytoremediation capacity and high stress tolerance (Grenier et al., 2015; Nissim et al., 92 2014). Considering that this species and its cultivars have been proven to be well suited for 93 some regions of North America, there is now interest in using S. miyabeana for treatment 94 wetlands (Lévesque et al., 2017, Grebenshchykova et al., 2017), ET cover (Mirck and 95 96 Volk, 2009) and zero-discharge wetlands (Frédette et al., 2017). However, we found a 97 single study that reported ET rates for this species, based on cultivars grown on a contaminated site for leachate minimization in the north-eastern United States (Mirck and 98 99 Volk, 2009). For all species of willow combined, we found four studies reporting ET rates 100 in treatment wetland conditions, most of them conducted in Europe and none in the 101 Americas. There is thus a clear lack of knowledge regarding the ET capacity of 102 economically important North American willow cultivars, like S. miyabeana, growing in treatment wetlands conditions. 103 104 The first objective of our study was to measure the ET rate and provide a crop coefficient 105 (K_{ET}) for Salix miyabeana (SX67) grown in treatment wetland conditions in a sub-boreal temperate climate. The second objective was to propose a predictive ET model, based on 106 simple meteorological and leaf parameters, which would be coherent with the wetland 107 growing conditions and physiology of fast growing willow species like S. miyabeana. 108 While the first objective would serve as a practical tool for development of a better 109 110 treatment wetland design and add to our knowledge of the ET of North American willow cultivars, the predictive model would enable the transfer of our results to different climatic 111 scenarios and to other willow species that are physiologically similar but have different leaf 112 113 and phenological parameters.

2. Material and methods

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2.1 Study site

The wetland studied is located in an industrial part of the city of Laval, Québec, where mean annual precipitation and temperature are 1000 mm and 6.8 °C, respectively, elevation is 91 m and the growing season is about 170 days. This willow wetland was established in 2012 and serves as a final polishing step connected to a series of other constructed wetlands treating leachate contaminated with utility wood pole preservatives (chromated copper arsenate and pentachlorophenol). The treatment system is operated only during the growing season and when there is no risk of water freezing in the system, generally from May to December. More details about the experimental treatment project are provided in Levesque *et al.* (2017). The willow wetland is a horizontal subsurface flow wetland 8 m wide by 6 m long (Figure 1), lined with a waterproof membrane and filled with a mix of black peat (20%) and sand (80%) with a general porosity of 50%.

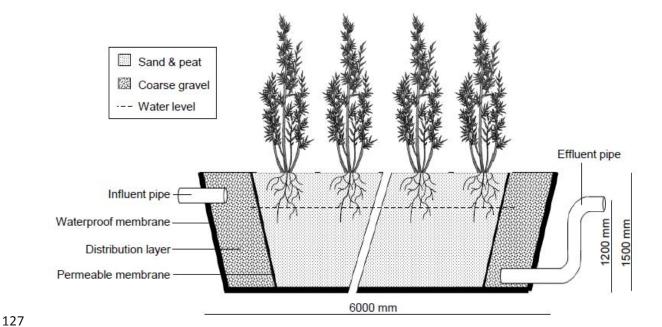


Figure 1. Section view of the horizontal subsurface flow wetland used to measure and model evapotranspiration of *S. miyabeana* in treatment wetland conditions.

Throughout this study, the average hydraulic loading rate of the willow wetland was about 55 L m⁻² d⁻¹ during the operating season, and the affluent contained a low concentration of contaminants (Table 1).

Table 1. Daily volume and general physical and chemical properties of the willow wetland influent, reported as the average value based on the entire growing season, in 2016 and 2017. Absence of values means that measured parameters were below detection limit in all samples.

Parameter	Unit	2016	2017
Daily volume	m ³	3.0	2.3
Hydraulic loading rate	L m ⁻² d ⁻¹	63	48
рН		7.64 ± 0.06	7.73 ± 0.12
DCO	mg/L	40 ± 1	-
PCCD/F	pg TEQ/L	0.32 ± 0.1	1.57 ± 0.46
Chlorinated phenols	μg/L	-	1.4 ±1.4
As	μg/L	82 ± 16	160 ± 82
Cr	μg/L	11 ± 3	12 ± 6
Cu	μg/L	17 ± 7	22 ± 12

The wetland was fertilized in 2014, and again at the beginning of 2017 with a slow-acting fertilizer in (Acer 21-7-14). The shoots were cut back at the end of the 2014 season to maintain a juvenile state and high productivity (Nyland, 2016; Abrahamson *et al.*, 2002). A monitoring station (Campbell Scientific, various sensors) was present on site for basic meteorological data measurement (rainfall, temperature, relative humidity, solar radiation and wind speed).

2.2 Plant material

The wetland was planted with 112 stools of *S. miyabeana* SX67 at a planting density of 2.3 plants/m². *Salix miyabeana* is native to Asia and the cultivar SX67 was developed at the University of Toronto, in Canada (Cameron *et al.*, 2007). It is usually grown from dormant cuttings, and only male clones with no seed production are produced (Cameron *et al.*, 2007). Although it can reproduce vegetatively, it does not propagate laterally (*e.g.* stolon), so the planting density does not change over time. However, the stools produce new stems when they are cut back. They produce 6 stems on average (Tharakan et al., 2005), ranging from 2 to 12 (Fontana et al., 2016). Tharakan et al. (2016) reported a mean leaf area index of 4.9 for this cultivar at the end of a three-year rotation cycle. SX67 present stomata on both abaxial and adaxial sides of leaves (amphistomatic) at the early development stage, and adaxial stomatal density decreases as the leaves mature (Fontana et al., 2017).

2.3 Physiological measurements

To model transpiration of *S. miyabeana*, we measured two main physiological parameters, *i.e.* stomatal conductance and leaf area index.

2.3.1 Stomatal conductance

Instant stomatal conductance (\bar{g}_s), representing the exchange rate of vapor water from leaf to the boundary layer (mmol m⁻² s⁻¹), was sampled on the abaxial side of leaves using a steady state porometer (Decagon, SC-1). In 2016, we sampled \bar{g}_s on 34 days from May 15 to October 11, with measurements in the lower, middle and upper parts of the canopy, both inside and at the border of the wetland, and from 6 AM to 9 PM, for a total of 4003 measurements. Data from 2016 allowed us to optimize sampling for the 2017 campaign, with measurements performed from 10 AM to 2 PM, where mean values of \bar{g}_s were observed, and only in middle and upper part of the canopy, because of the low influence of

the lower part in the general stomatal conductance (\bar{G}_s) of the wetland. In 2017, sampling took place on 43 days from May 11 to October 27, for a total of 3579 measurements. Also, because *S. miyabeana* presents amphistomatic characteristics (Fontana & al., 2017), 150 measurements were made on both adaxial and abaxial sides of the leaves (75 pairs of measurements, taken on four days from May to August 2017) to establish a ratio of transpiration occurring on the upper versus the lower side of the leaf.

2.3.2 Leaf area index

Leaf area index (LAI), which expresses the leaf area covering a given ground area (m² leaf/m² ground), was estimated once a month, in the middle of the month, from May to November and for both growing seasons. We calculated the LAI of the entire wetland based on extrapolation of individual willow leaf area and considering that there could be significant difference between leaf area of willows growing on the border and those growing in the center of the wetland:

$$LAI = (N_{border}IA_{border} + N_{center}IA_{center})/A_{wetland}$$
 (Eq. 1)

Where N is the number of willows growing either on the border or in the center, and their respective mean individual leaf area (IA), and $A_{wetland}$ is the wetland area. IA was estimated for fifteen individual willows, seven growing on the border of the wetland and eight growing in the center, as follows:

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$$IA = A_{leaf}(S_{<1m}N_{leaf} + S_{1-3m}N_{leaf} + S_{>3m}N_{leaf})$$
 (Eq. 2)

 A_{leaf} is the average single leaf area and is measured each month based on 30 to 40 randomly collected leaves and using the software, Mesurim Pro v3.4.4.0. The number of stems (S) were counted on the individuals and divided in 3 height classes (<1m, 1-3m, >3m). Finally,

the average number of leaves (N_{leaf}) present on stems was estimated by direct counting on 5 random stems of each class. Afterwards, we examined the spatial variation of the leaf area by comparing individual area of stools on the edge and stools in the center of the wetland. Because the leaf cover seemed to exceed the actual area of the wetland, we also calculated and adjusted value of LAI based on the projected canopy area (Allen *et al.*, 2011).

2.4 Wetland evapotranspiration calculation

To estimate actual ET of the willows, we used the water balance method, based on the following mathematical equation (Kadlec & Wallace, 2008):

$$ET = \frac{Q_i + IQ_p + Q_r - Q_d - Q_o - \frac{dV}{dt}}{A}$$
 (Eq. 3)

Where ET is the ET rate, Q_l the inflow flowrate, Q_p the precipitation adjusted by a canopy interception factor (I), Q_r the flowrate of runoff entering the wetland, Q_d the underground drainage flowrate, Q_0 the effluent flowrate, $\frac{dV}{dt}$ the variation of the volume of water contained in the wetland and A the wetland area. We considered an interception factor of 25%, determined with an equation provided by Martin and Stephen (2005) and based on leaf area index (see section 2.2.2; I = 3.01LAI + 1.12), meaning that only 75% of the rainfall reaches the wetland substrate, the rest being evaporated directly from the leaf and thus not considered as tree ET $per\ se$. As we will demonstrate below, rapid closure of the wetland canopy makes this high interception factor very suitable. Because of the waterproof membrane and the highly permeable soil surrounding the wetland, it is assumed that Q_r and Q_d are equal to zero. The water volume variation in the wetland is calculated according to the water level and a measured soil porosity of 50%:

$$\frac{dV}{dt} = \theta \cdot A \cdot \Delta L_{(t;t-1)}$$
 (Eq. 4)

Where Θ represents the substrate porosity, A the wetland area and $\Delta L_{(t-1;t)}$ the water level variation for a given period. Water level was measured hourly with two probes (Levelogger Junior Edge, Solinst) placed at two points in the wetland, from May 27 to December 9 in 2016 and from April 21 to November 29 in 2017. Both influent and effluent volume of the willow wetland were monitored with pulse meters (Omega, FTB8000B) throughout the operating season (the system was completely shut down in winter) which represent 214 and 220 days for 2016 and 2017 respectively. Due to a malfunction of the flow meters, 2016 results are overestimated and late season results for both years (October and November 2016 and November 2017) are not presented. Finally, reference ET was calculated according to the modified Penman-Monteith method (Allen & al., 1998), and open water evaporation estimated by pan evaporation.

2.5 Evapotranspiration modelling

In a treatment wetland, there are few limitations on ET. Available energy is greater than direct solar radiation because of both "oasis" and "clothesline" effects (Dotro *et al.*, 2017; Kadlec and Wallace, 2008) that increase ET potential (Allen *et al.*, 1998). Oasis effect provides a vertical energy transfer in the form of sensible heat from the air surrounding the wetland because its moist condition and transpiration make it cooler than the ambient air. The clothesline effect, resulting from the tall wetland plants being surrounded by smaller vegetation, provides a horizontal energy transfer due to wind (Kirkham, 2014). Wind effect is enhanced due to the small size of the wetland and constantly disturbs the boundary layer of plant leaves (Kadlec and Wallace, 2008), meaning that water vapor excreted by the leaves is automatically replaced with fresh air and transpiration potential increases.

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Frequent and high irrigation combined with a saturating water level in the wetland also ensure high water availability and prevent limitation of ET due to water stress. Based on these non-limited conditions, we hypothesized that transpiration of willows in a treatment wetland should be highly correlated to stomatal conductance (i.e. water vapor exchange rate between leaf and air; \bar{G}_s). \bar{G}_s is generally measured in a volume of water per surface of leaf per time unit (e.g. mmol m⁻² s⁻¹), meaning that leaf area capable of transpiring (LAI_{active}) is also required for ET calculation. Because of the relatively constant disturbance of the boundary layer by wind, transpiration rate should also be driven mainly by water vapor pressure deficit (VPD) in the ambient air. Otherwise, the irrigation of the wetland being below the surface, there is no open contact between water and the atmosphere. According Shuttleworth and Wallace's energy partitioning model (1985), the high average LAI of S. miyabeana (> 4 m²; Tharakan et al., 2016) implies that most of the energy available for ET is intercepted by the willows, reducing soil evaporation potential to close to zero. Therefore, in this study, we assume that soil evaporation can be ignored and that willow transpiration can be treated as ET. Daily ET of S. miyabeana grown in a treatment wetland (mm/d) could then be estimated with the following leaf parameter based equation:

$$ET_{SX67} = \bar{G}_{S} \cdot LAI_{active} \cdot VPD$$
 (Eq. 5)

Active leaf area can be calculated throughout the season according to the seasonal leaf development curve and the abaxial/adaxial ratio established by measurements presented in section 2.3, and the vapor pressure deficit can be calculated with daily temperature and relative humidity data. To estimate stomatal conductance, we chose an empirical approach based on environmental parameters known to influence stomata openings (Buckley and

Mott, 2013). We wanted those parameters to be easily accessible, to allow the transpiration rate to be predicted with minimal resources. Through linear regressions, we tested the statistical relation between mean daily stomatal conductance measured on site and the following daily parameters: solar radiation, average and maximal air temperature, average and minimal relative humidity, wind speed and day of the year. Parameters presenting a significant relation with stomatal conductance (p<0.05) were combined to predict canopy general conductance as follows:

$$\bar{G}_{s} = \sum \alpha \bar{g}_{s}^{x}$$
 (Eq. 6)

Where partial stomatal conductance (\bar{g}_s) was calculated according to previously selected parameters (x) having their own relative influence (α) on the general stomatal conductance of the wetland canopy (\bar{G}_s). Finally, crop coefficient was calculated as follows (Kadlec and Wallace, 2008):

$$K_{SX67} = ET_{SX67}/ET_0$$
 (Eq. 7.)

Where K_{SX67} is the crop, or plant, coefficient, ET_{SX67} is the modelled ET rate of the willow stand and ET_0 the reference crop ET.

2.6 Statistical analysis

The relation between meteorological parameters and \bar{G}_s was tested with either linear, quadratic and power regressions. The influence of parameters on a given variable (e.g. influence of leaf face on \bar{G}_s variation) was tested with two-way ANOVAs analysis with a 0.05 significance threshold (α = 0.05). Tukey's post-hoc statistical test was used when necessary to better interpret the results of the analysis of variance (α = 0.05). All statistical analysis were done using R 3.5.1 software.

3. Results

The summer of 2016 was hot and dry, with a mean temperature of $18.0 \,^{\circ}\text{C}$ (± 6.0) and 569 mm of rainfall from May to October. Mean temperature was similar in 2017 (17.9 $\,^{\circ}\text{C}$ ± 4.8), but with less days on which maximum temperature rose above 30 $\,^{\circ}\text{C}$. Also, 2017 saw much higher rainfall, with 819 mm for the same period. A summary of solar radiation, rainfall and daily mean temperature for both growing seasons is shown in Figure 2.

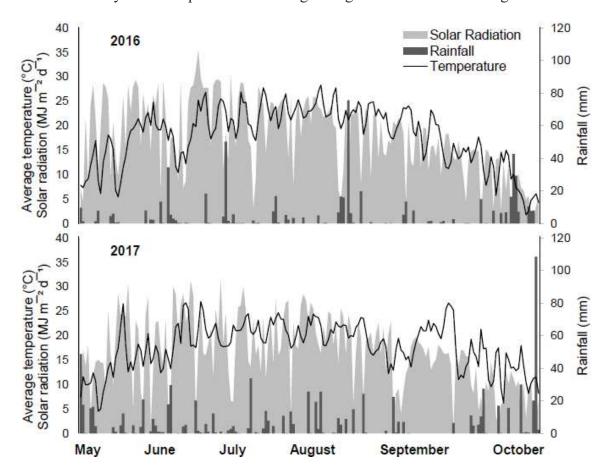


Figure 2. Summary of the meteorological conditions at the experimental site for the 2016 and 2017 growing seasons.

Average reference crop ET was 4.5 mm/d in 2016 and 3.2 mm/d in 2017, for a total of 808 mm and 750 mm respectively, from May to November. Pan evaporation measured in 2017 represented 81% of reference ET. For the willow wetland, we calculated a mean daily ET

rate of 30.9 mm/d and a seasonal total ET of 4536 mm from May 9 to September 30 in 2016, and 16.6 mm/d and a seasonal total of 2906 mm from May 15 to October 31 in 2017.

3.2 Physiological measurements

Stomatal conductance values were generally higher and more variable in the 2016 season, with a mean value of 418 (\pm 124) mmol m⁻² s⁻¹ compared to 309 (\pm 59) mmol m⁻² s⁻¹ in 2017. The adaxial/abaxial stomatal conductance ratio was relatively high (0.33 ± 0.17) and variable in the early season, decreasing to relatively constant and low values (0.14 ± 0.06) for the rest of the summer (Figure 3).

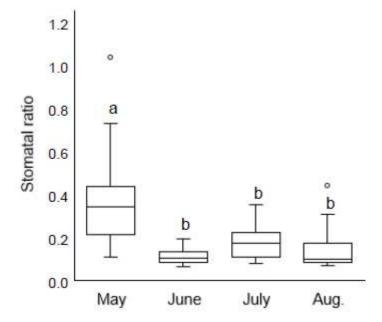


Figure 3. Adaxial/abaxial stomatal conductance ratio of *S. miyabeana* growing in treatment wetland conditions for the 2017 summer season. Different letters represent statistically different values. Thus, overall seasonal transpiration occurring on the upper part (adaxial) of the leaf represents about 20% of the lower side (abaxial) transpiration, and actual stomatal conductance equals approximately 120% of the values measured on the abaxial side of the leaf only. In both the 2016 and 2017 seasons, leaf cover established rapidly, attaining its

highest value in July, with 10.4 and 11.4 m² of leaves per m² of soil respectively. The canopy extended beyond the wetland borders by about 50 cm meter on each side, for a projected canopy area of 63 m² compared to the actual wetland area of 48 m². Peak LAI measured using the projected canopy area was 7.9 in 2016 and 8.7 in 2017. In 2017, the global leaf area was a little higher than in 2016, attained its maximal value earlier and retained active foliage later in the season (Figure 4).

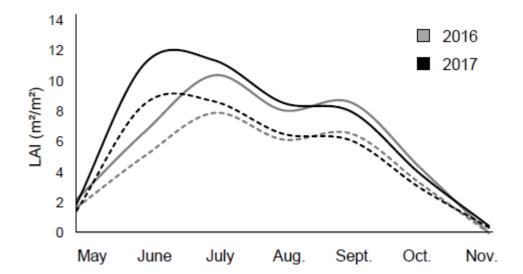
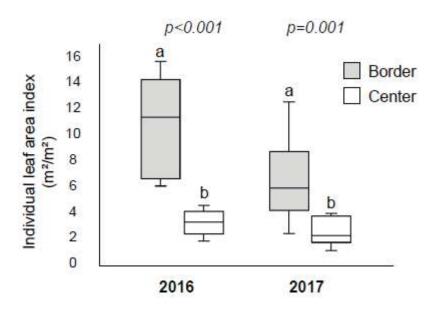


Figure 4. Evolution of the leaf area index of a 48 m² wetland (solid line) planted with *S. miyabeana* throughout 2 successive growing seasons, and the corresponding values adjusted for a 63 m² projected canopy area (dashed line).

Trees on the edge of the wetland had up to three times more leaf area than those in the center (Figure 5).



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Figure 5. Leaf area, measured in the month of July, of 15 individuals of *S. miyabeana* growing either at the border or in the center of a 48 m² constructed wetland. Different letters represent statistically different values.

3.3 Evapotranspiration modelling

We found a significant effect of temperature, solar radiation, relative humidity and day of the year on stomatal conductance (Table 2), but no effect of wind speed.

Table 2. Parameters of the relations found between stomatal conductance of *S*. *miyabeana* and temperature (T), day of year (DOY), solar radiation (Rad) and relative humidity (RH). Parameter importance (α) and predictive equations used for stomatal conductance modelling are presented.

Parameter	Type of relation	p _{value}	R²	α	Equation
Т	Power	<0.001	0.21	0.48	88.4x ^{0.5}
DOY	Quadratic	0.002	0.13	0.30	$-0.02x^2 + 9x - 572$
Rad	Quadratic	0.05	0.05	0.11	$-0.005x^2 + 2x - 177$
RH	Linear	0.03	0.05	0.11	2.9x + 168

For temperature and relative humidity, mean daily values were better predictors than maximum and minimum values respectively. Correlation between \bar{G}_s and each factor

separately was relatively weak (r² from 0.05 to 0.21), but together they explained half of stomatal conductance variation throughout the season (Figure 6.), which can be considered satisfying due to the many other factors driving this parameter but not measured here (Buckley and Mott, 2013).

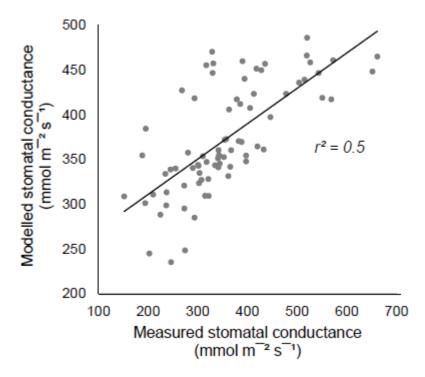


Figure 6. Results of \bar{G}_s modelling, based on temperature, solar radiation, relative humidity and day of year, over \bar{G}_s measured on the field under the same parameters.

The model was good at predicting mean \bar{G}_s , with a predicted mean seasonal value of 428 mmol m⁻² s⁻¹ over 418 mmol m⁻² s⁻¹ measured in 2016, and 329 mmol m⁻² s⁻¹ predicted over 309 mmol m⁻² s⁻¹ measured in 2017. Daily variation was captured more accurately in 2017 than in 2016 (Figure 7).

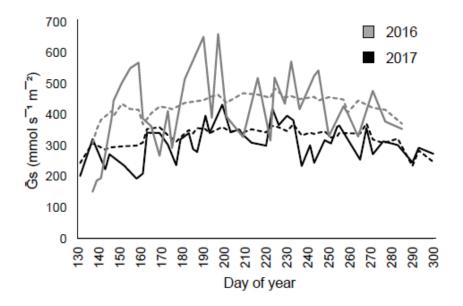


Figure 7. Stomatal conductance (Gs) field measurements (solid line) and modelling results

(dashed line) over the 2016 and 2017 growing seasons.

Using the general stomatal conductance predicted with this model and the previously established leaf area parameters, we calculated the ET rate (Eq. 5) and the corresponding crop coefficient (Eq. 7; Table 3). Willow ET was higher in 2016, as was reference ET, with a mean daily rate of 17.1 mm/d compared to 12.9 mm/d in 2017 (Table 3). Calculated seasonal ET was 3170 mm in 2016 and 2400 mm in 2017. Crop coefficients were constant in both years, with an average of 4.1, and values slightly above 5 times the reference ET for the months of July, August and September (Table 3). Highest ET rates were calculated in August 2016 (44.8 mm/d on August 13) and in July 2017 (34.3 mm/d). Modelled crop coefficients are very close to those calculated with the water balance for most of the 2017 season, but lower than water balance ET in 2016, probably due to the overestimation of actual ET for this season (section 2.4).

Table 3. Mean daily Penman-Monteith reference evapotranspiration (ET₀), estimated active leaf area index of

the 48 m² treatment wetland (LAI), modelled willow evapotranspiration (ET_{SX67}) and crop coefficient (K_{SX67}) presented as monthly and seasonal averages, for the 2016 and 2017 growing seasons.

	2016				2017			
	ET ₀	LAI _{active}	ET _{SX67}	K _(SX67)	ET ₀	LAI _{active}	ET _{SX67}	K _(SX67)
May	5.2 ± 0.9	3.3 ± 1.3	4.3 ± 5.1	0.8 ± 0.7	4.0 ± 2.0	3.4 ± 1.9	3.9 ± 3.5	1.0 ± 0.7
June	5.5 ± 0.9	8.2 ± 1.4	15.5 ± 10.4	2.8 ± 1.0	3.9 ± 1.9	12.1 ± 2.0	16.5 ± 8.7	4.3 ± 1.1
July	5.4 ± 0.6	11.6 ± 0.5	26.8 ± 9.5	5.0 ± 0.9	3.8 ± 1.4	13.3 ± 0.5	19.4 ± 5.6	5.1 ± 1.1
August	5.0 ± 0.5	10.1 ± 0.3	27.4 ± 9.8	5.5 ± 0.9	3.5 ± 1.1	10.7 ± 0.7	17.8 ± 4.3	5.1 ± 0.7
Sept.	3.9 ± 0.6	9.5 ± 0.9	20.0 ± 5.8	5.2 ± 1.2	2.6 ± 1.1	9.1 ± 0.8	13.4 ± 4.6	5.1 ± 1.1
October	1.8 ± 0.5	4.5 ± 1.9	8.7 ± 3.8	4.8 ± 1.3	1.4 ± 0.9	4.8 ± 1.4	6.4 ± 2.3	4.4 ± 0.8
Average	4.5 ± 2.0	7.9 ± 3.2	17.1 ± 12.0	4.0 ± 1.9	3.2 ± 1.8	8.9 ± 3.9	12.9 ± 8.6	4.2 ± 1.6

4. Discussion

The mean monthly ET rate measured for *Salix miyabeana* in treatment wetland conditions ranged from 3.9 to 27.4 mm/d, with a mean seasonal cumulative ET of 2785 mm. Although ET was greater in 2016 than in 2017, crop coefficients were similar for both years, ranging from 0.8 to 5.5 with a mean value of 4.1 times the Penman-Monteith reference ET. These ET results differ from those reported in the very few studies conducted in comparable conditions (Curneen and Gill, 2014; Gregersen and Brix, 2001; Brix and Arias, 2005; Kučerová *et al.*, 2001), although crop coefficients are similar (Table 4). On the other hand, LAI is very high compared to the only study we found for another cultivar of *S. miyabeana* (SX64; Mirck and Volk, 2009; Table 4), grown in open field plantation, with low water input and soil contamination.

Table 4. Evapotranspiration results obtained for fast growth willow cultivars in treatment wetland conditions (A) compared to results obtained for a plantation of Japanese willow (B)

Species (cultivar)	Country	Seasonal Peak		Seasonal	Annual	Ref.
Species (cultival)	Country	ET	K_{ET}	K_{ET}	\mathbf{K}_{ET}	Nei.
S. miyabeana (SX67)	Canada	2785 mm	5.5	4.1	2.5	1
S. viminalis (Bjorn, Tora, Jorr)	Denmark	1113 mm	-	-	2.5	2
S. viminalis	Ireland	669 mm	5.1	3.0	-	3
S. cinereal	Belgium	-	6.7	-	-	4
S. miyabeana (9882-34, 9870-23, SX61, SX64)	USA	515 mm	1.4	1.2	-	5

Note: 1: present article; 2: Gregersen & Brix, 2001; 3: Curneen & Gill, 2014, 4: Kučerová et al., 2001; 5: Mirck & Volk, 2009.

Average seasonal ET rates reported for other fast growing willow cultivars grown in field plantation are also generally much lower than our results (1.4 mm/d, Linderson et al., 2007; 3.0 mm/d, Lindroth et al., 1994; 2.9 mm/d, Personn, 1995; 1.0 mm/d, Mata-Gonzalez; 3.1 mm/d, Budny and Benscoter, 2012). In comparison, similar rates (from 10 to 23 mm/d) were measured for young *S. babylonica* grown in water saturated conditions in the northeastern United States (Pauliukonis *et al.*, 2001). Such high ET rates can be explained by both enhancing factors linked to the treatment wetland itself (*i.e.* oasis and clothesline effect, high water availability, important border effect) and by *S. miyabeana* ecophysiology (*i.e.* high stomatal conductance and leaf area index).

In this study, a simple model based mainly on two leaf parameters was sufficient to model ET. As expected, the model ET results were lower than the water balance results in 2016 (see section 2.4). However, 2017 simulation results closely resembled water balance results. The fact that our simplified ET model yielded conclusive results supports our

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premise that typical ET limiting factors are greatly attenuated in small wetlands. Other studies presenting ET modelling methods for willows often include several limiting factors (Irmak et al., 2013, Iritz et al., 2001), ignore heat advection effect (Přibáň and Ondok, 1986) or focus on soil hydrology (Personn, 1995, Hartwich et al., 2016; Borek et al., 2010) or complex physiological processes (Tallis et al., 2013). Although based on sound scientific assumptions, those models hardly apply in treatment wetland conditions where water level is constant, limitations are attenuated and heat advection effect is very important. The few input parameters required for the operation of the model also represent an opportunity for managers working with treatment wetlands to easily include ET estimation in their planning activities. However, to be used for other taxa, a basic knowledge of the LAI dynamic and general stomatal conductance for the species is needed, and could require additional \bar{g}_s measurement in the field to adjust the model. Regarding ET related characteristics specific to S. miyabeana, we found that mean stomatal conductance (0.4 mol m⁻² s⁻¹) was consistent with published results for other willows (0.4 mol m⁻² s⁻¹, Budny and Benscoter, 2016; 0.2-0.7 mol m⁻² s⁻¹, Hall et al., 1998) or higher (0.2 mol m⁻² s⁻¹, Kučerová et al., 2001). Leaf area index values were higher than those reported in the literature for other willow cultivars, even when using the projected canopy area for the calculation (Figure 8).

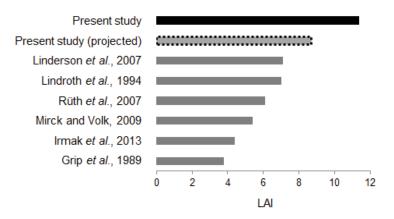


Figure 8. Maximal leaf area index (LAI) reported for willow stands (different cultivars) in various studies including the present results, and the corresponding value adjusted with projected canopy area.

As for stomatal conductance, it is also interesting to note that the highest mean daily value measured ($661 \text{ mmol m}^{-2} \text{ s}^{-1}$) is much higher than the values proposed for deciduous trees and even plants from wet habitats (Jones, 2013). The ratio between the conductance of the upper and lower side of the leaf is consistent with the literature predicting higher adaxial activity or adaxial stomatal density in younger leaves (Fontana *et al.*, 2017). Meteorological factors could only explain about half of the stomatal conductance values and variability. Stomatal aperture is also driven by many biochemical and environmental factors (Buckley and Mott, 2013) that were not studied here. Aging of the willows, or negative effects of contaminant accumulation in the substrate are also factors affecting long term variability of \bar{G}_s in a wetland that should be considered. A sampling campaign (data not shown) conducted in June of 2017 in Denmark on *S. viminalis* clones used for zero-discharge wetlands showed significantly greater stomatal conductance in willows recently coppiced, compared to older individuals growing in the exact same conditions, which supports the aging hypothesis. Those factors should be investigated thoroughly in the future. Leaf area

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of the willow wetland attained its maximal value (complete canopy closure) with two-yearold shoots, peaking in July at around 12 m² of leaves per m² of ground. Planting density and methodological differences could partially explain why LAI of our wetland was very high compared to findings reported in the literature. Furthermore, all results presented in Figure 8 are based on field plantation or natural river bands of much greater size than our wetland and the effect of increased leaf area at the border is negligible. Our finding comparing individual leaf area at the edges versus in the center of the wetland is also interesting because it means we could modulate ET rate directly in the wetland design. Indeed, if ET is directly related to LAI as demonstrated here, adjusting the edge or aspect ratio of the surface area of a wetland could enhance (higher ratio) or limit (lower ratio) ET per ground unit, according to management objectives. Fertilization applied at the beginning of 2017 seemed to have accelerated the establishment of the leaf cover but did not significantly increase maximal LAI. Since the fertilizer used consisted of solid granules applied directly on the soil, with degradation regulated by rainfall and temperature, it is possible that rapid closure of the canopy and high rain interception by willows prevented the fertilizer from appropriately degrading and penetrating the substrate. This hypothesis is supported by the absence of nitrogen in the wetland effluent throughout the season (result not shown). In 2016, the canopy already seemed completely closed by mid-season and it is possible that maximum leaf area index was already attained. Indeed, in 2017, stems grew higher but there was little or no leaf development at the bottom of the stems (as was observed in 2016), probably because canopy closure was achieved and all available light was intercepted in the upper part of the trees. Therefore, we conclude that maximal LAI

was achieved with two-year-old shoots, without a need for fertilization, and that coppicing should be scheduled on a two-year basis.

5. Conclusions

S. miyabeana ET in treatment wetland condition was very high throughout this study. We highlighted several factors related to treatment wetlands that can significantly increase potential ET. Because there are few limitations on ET in wetlands, a model exclusively based on leaf parameters successfully predicted ET values and calculated crop coefficients for the studied willow wetland. Because these results are based on a full-scale wetland, they can be used as design parameters for treatment wetlands using S. miyabeana, and the equation presented for ET calculation can be adjusted for other fast-growing willow species used in similar growing conditions. We also presented a strategy to optimize ET per ground area by changing the aspect ratio of the wetland, and consequently its leaf area index, as well as regularly coppicing the stems. In the future, other parameters possibly affecting ET in treatment wetlands such as tree aging, substrate type and contaminant toxicity could be investigated. This study is a first step towards better ecophysiological characterization of woody plants used in treatment wetlands.

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