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1 **Evapotranspiration of a willow cultivar (*Salix miyabeana* SX67)**

2 **grown in a full-scale treatment wetland**

3

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22 **Declarations of interest:** none

Abstract

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 miyabeana* (SX67), to provide the ET rates and crop coefficient for this species. During two growing seasons, we studied a 48 m^2 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,

47   willow leaf parameters, evapotranspiration modelling, zero-discharge wetlands

48

49   **1. Introduction**

50   Treatment wetlands, or vegetation filters, are now commonly used for treatment of various

51   types of wastewater (Valipour and Ahn, 2017). "Artificial" wetlands are generally planted

52   with herbaceous plants like *Phragmites, Typha*, graminoids or other aquatic and semi-

53   aquatic species (Kadlec and Wallace, 2008). More recently, woody species of the *Salix*

54   genus (willows), generally studied for biomass production, are being tested and used for

55   wastewater treatment purposes. *Salix* species are mostly hydrophilic and tolerate hypoxic

56   conditions and great water fluctuations well, have a high growth rate and develop a

57   vigorous root system (Kuzovkina *et al*., 2008), making them good candidates for treatment

58   wetland purposes. Another advantage of using woody plants for water treatment is the

59   added value of biomass production that can be used for bioenergy and biofuel processes

60   (Duggan *et al.*, 2005). Consequently, there is growing interest in willow for use in

61   treatment of landfill leachate, domestic wastewater or other nitrogen rich wastewaters

62   (Białowiec *et al*., 2003; Dimitriou and Aronsson, 2011; Guidi *et al*., 2014). Fast growing

63   willows are also known for their great evapotranspiration (ET), which led to the

64   development of a new specific type of treatment wetlands called "zero-discharge wetlands"

65   (ZDWs; Dotro *et al*., 2017). The design of ZDWs is based mainly on the ET capacity of the

66   plant selected. They operate without liquid effluent, immobilizing contaminants in the

67   wetland substrate and preventing any release of residual contamination in the environment.

68   Depending on the type of water contamination, ZDWs can function as the final step of a

69 treatment plant or as a secondary treatment. Such wetlands are now well implanted in

70 Scandinavian countries, mainly in Denmark, where the concept was first developed

71 (Gregersen and Brix, 2001; Brix and Arias, 2011), and Ireland (Curneen and Gill, 2014).

72 Conclusive tests have also been performed in Mongolia, under very cold climatic

73 conditions (Khurelbataar *et al*., 2017), and zero-discharge wetlands are currently being

74 tested in other locations.

75 Sound scientific knowledge of the ET rate of the species used is an essential tool to design

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

80 published on willow ET, very few concern willows growing in full-scale treatment wetland

81 conditions. However, ET in artificial wetlands can differ greatly from ET measured in a

82 plantation, and can significantly surpass potential ET (Dotro *et al.,* 2017).

83 The willow species most studied for ET is *Salix viminalis,* its hybrids and their numerous

84 cultivars (Frédette *et al.* 2018). Although widely used in Europe, some long-term studies

85 have pointed out that, in North America, cultivars of *S. viminalis* are more prone to diseases

86 and insect attacks than other cultivars (Labrecque and Teodorescu, 2005; Nissim *et al.*,

87 2013). Instead, other cultivars from species like *Salix eriocephala, S. purpurea*, *S. nigra*

88 and *S. miyabeana* are frequently used (Smart and Cameron, 2008). In eastern Canada,

89 Nissim *et al*. (2013) concluded that *S. miyabeana* and some indigenous species were more

90 suited for plantation than *S. viminalis*. *Salix miyabeana* has also shown the highest biomass

91 production among cultivars (Labrecque and Teodorescu, 2005; Pitre *et al*., 2010), good

92   phytoremediation capacity and high stress tolerance (Grenier *et al.*, 2015; Nissim *et al.*,

93   2014). Considering that this species and its cultivars have been proven to be well suited for

94   some regions of North America, there is now interest in using *S. miyabeana* for treatment

95   wetlands (Lévesque *et al.*, 2017, Grebenshchykova *et al.*, 2017), ET cover (Mirck and

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

98   contaminated site for leachate minimization in the north-eastern United States (Mirck and

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

103 treatment wetlands conditions.

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

106 temperate climate. The second objective was to propose a predictive ET model, based on

107 simple meteorological and leaf parameters, which would be coherent with the wetland

108 growing conditions and physiology of fast growing willow species like *S. miyabeana*.

109 While the first objective would serve as a practical tool for development of a better

110 treatment wetland design and add to our knowledge of the ET of North American willow

111 cultivars, the predictive model would enable the transfer of our results to different climatic

112 scenarios and to other willow species that are physiologically similar but have different leaf

113 and phenological parameters.

114 **2. Material and methods**

115   *2.1 Study site*

116   The wetland studied is located in an industrial part of the city of Laval, Québec, where

117   mean annual precipitation and temperature are 1000 mm and 6.8 °C, respectively, elevation

118   is 91 m and the growing season is about 170 days. This willow wetland was established in

119   2012 and serves as a final polishing step connected to a series of other constructed wetlands

120   treating leachate contaminated with utility wood pole preservatives (chromated copper

121   arsenate and pentachlorophenol). The treatment system is operated only during the growing

122   season and when there is no risk of water freezing in the system, generally from May to

123   December. More details about the experimental treatment project are provided in Levesque

124   *et al.* (2017). The willow wetland is a horizontal subsurface flow wetland 8 m wide by 6 m

125   long (Figure 1), lined with a waterproof membrane and filled with a mix of black peat

126   (20%) and sand (80%) with a general porosity of 50%.
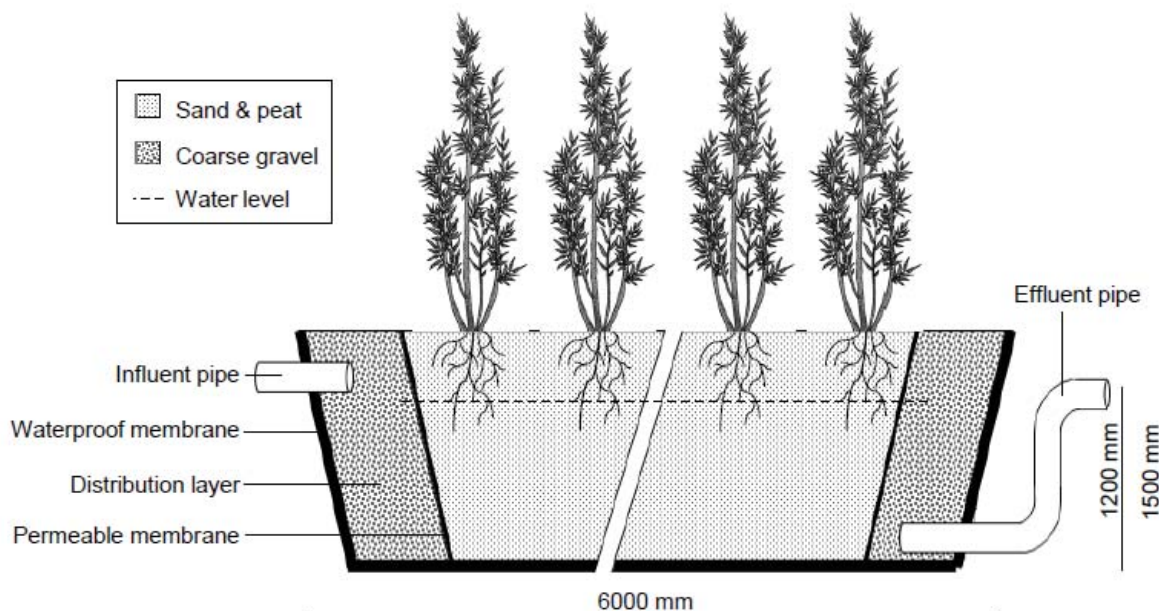
127

128   **Figure 1.** Section view of the horizontal subsurface flow wetland used to measure and model

129   evapotranspiration of *S. miyabeana* in treatment wetland conditions.

130 Throughout this study, the average hydraulic loading rate of the willow wetland was about

131 55 L m^{-2} d^{-1} during the operating season, and the affluent contained a low concentration of

132 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
pH		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

133 The wetland was fertilized in 2014, and again at the beginning of 2017 with a slow-acting

134 fertilizer in (Acer 21-7-14). The shoots were cut back at the end of the 2014 season to

135 maintain a juvenile state and high productivity (Nyland, 2016; Abrahamson *et al.*, 2002). A

136 monitoring station (Campbell Scientific, various sensors) was present on site for basic

137 meteorological data measurement (rainfall, temperature, relative humidity, solar radiation

138 and wind speed).

139 ***2.2 Plant material***

140   The wetland was planted with 112 stools of *S. miyabeana* SX67 at a planting density of 2.3

141   plants/m^2. *Salix miyabeana* is native to Asia and the cultivar SX67 was developed at the

142   University of Toronto, in Canada (Cameron *et al.*, 2007). It is usually grown from dormant

143   cuttings, and only male clones with no seed production are produced (Cameron *et al.*,

144   2007). Although it can reproduce vegetatively, it does not propagate laterally (*e.g.* stolon),

145   so the planting density does not change over time. However, the stools produce new stems

146   when they are cut back. They produce 6 stems on average (Tharakan et al., 2005), ranging

147   from 2 to 12 (Fontana et al., 2016). Tharakan et al. (2016) reported a mean leaf area index

148   of 4.9 for this cultivar at the end of a three-year rotation cycle. SX67 present stomata on

149   both abaxial and adaxial sides of leaves (amphistomatic) at the early development stage,

150   and adaxial stomatal density decreases as the leaves mature (Fontana et al., 2017).

151   ***2.3 Physiological measurements***

152   To model transpiration of *S. miyabeana*, we measured two main physiological parameters,

153   *i.e.* stomatal conductance and leaf area index.

154   *2.3.1 Stomatal conductance*

155   Instant stomatal conductance ($\bar{g}_s$), representing the exchange rate of vapor water from leaf

156   to the boundary layer (mmol m^{-2} s^{-1}), was sampled on the abaxial side of leaves using a

157   steady state porometer (Decagon, SC-1). In 2016, we sampled $\bar{g}_s$ on 34 days from May 15

158   to October 11, with measurements in the lower, middle and upper parts of the canopy, both

159   inside and at the border of the wetland, and from 6 AM to 9 PM, for a total of 4003

160   measurements. Data from 2016 allowed us to optimize sampling for the 2017 campaign,

161   with measurements performed from 10 AM to 2 PM, where mean values of $\bar{g}_s$ were

162   observed, and only in middle and upper part of the canopy, because of the low influence of

163   the lower part in the general stomatal conductance ($\bar{G}_s$) of the wetland. In 2017, sampling

164   took place on 43 days from May 11 to October 27, for a total of 3579 measurements. Also,

165   because *S. miyabeana* presents amphistomatic characteristics (Fontana *& al.*, 2017), 150

166   measurements were made on both adaxial and abaxial sides of the leaves (75 pairs of

167   measurements, taken on four days from May to August 2017) to establish a ratio of

168   transpiration occurring on the upper versus the lower side of the leaf.

169   *2.3.2 Leaf area index*

170   Leaf area index (LAI), which expresses the leaf area covering a given ground area (m^2

171   leaf/m^2 ground), was estimated once a month, in the middle of the month, from May to

172   November and for both growing seasons. We calculated the LAI of the entire wetland

173   based on extrapolation of individual willow leaf area and considering that there could be

174   significant difference between leaf area of willows growing on the border and those

175   growing in the center of the wetland:

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

177   Where N is the number of willows growing either on the border or in the center, and their

178   respective mean individual leaf area (IA), and $A_{wetland}$ is the wetland area. IA was estimated

179   for fifteen individual willows, seven growing on the border of the wetland and eight

180   growing in the center, as follows:

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

182   A_{leaf} is the average single leaf area and is measured each month based on 30 to 40 randomly

183   collected leaves and using the software, Mesurim Pro v3.4.4.0. The number of stems (S)

184   were counted on the individuals and divided in 3 height classes (<1m, 1-3m, >3m). Finally,

185   the average number of leaves (N_{leaf}) present on stems was estimated by direct counting on 5

186   random stems of each class. Afterwards, we examined the spatial variation of the leaf area

187   by comparing individual area of stools on the edge and stools in the center of the wetland.

188   Because the leaf cover seemed to exceed the actual area of the wetland, we also calculated

189   and adjusted value of LAI based on the projected canopy area (Allen *et al.*, 2011).

190   *2.4 Wetland evapotranspiration calculation*

191   To estimate actual ET of the willows, we used the water balance method, based on the

192   following mathematical equation (Kadlec & Wallace, 2008):

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

194   Where ET is the ET rate, Q_i the inflow flowrate, Q_p the precipitation adjusted by a canopy

195   interception factor (I), Q_r the flowrate of runoff entering the wetland, Q_d the underground

196   drainage flowrate, Q_o the effluent flowrate, $\frac{dV}{dt}$ the variation of the volume of water

197   contained in the wetland and A the wetland area. We considered an interception factor of

198   25%, determined with an equation provided by Martin and Stephen (2005) and based on

199   leaf area index (see section 2.2.2; $I = 3.01LAI + 1.12$), meaning that only 75% of the

200   rainfall reaches the wetland substrate, the rest being evaporated directly from the leaf and

201   thus not considered as tree ET *per se*. As we will demonstrate below, rapid closure of the

202   wetland canopy makes this high interception factor very suitable. Because of the

203   waterproof membrane and the highly permeable soil surrounding the wetland, it is assumed

204   that Q_r and Q_d are equal to zero. The water volume variation in the wetland is calculated

205   according to the water level and a measured soil porosity of 50%:

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

207   Where Θ represents the substrate porosity, A the wetland area and $\Delta L_{(t-1;t)}$ the water level

208   variation for a given period. Water level was measured hourly with two probes (Levelogger

209   Junior Edge, Solinst) placed at two points in the wetland, from May 27 to December 9 in

210   2016 and from April 21 to November 29 in 2017. Both influent and effluent volume of the

211   willow wetland were monitored with pulse meters (Omega, FTB8000B) throughout the

212   operating season (the system was completely shut down in winter) which represent 214 and

213   220 days for 2016 and 2017 respectively. Due to a malfunction of the flow meters, 2016

214   results are overestimated and late season results for both years (October and November

215   2016 and November 2017) are not presented. Finally, reference ET was calculated

216   according to the modified Penman-Monteith method (Allen *& al.*, 1998), and open water

217   evaporation estimated by pan evaporation.

218   *2.5 Evapotranspiration modelling*

219   In a treatment wetland, there are few limitations on ET. Available energy is greater than

220   direct solar radiation because of both "oasis" and "clothesline" effects (Dotro *et al.*, 2017;

221   Kadlec and Wallace, 2008) that increase ET potential (Allen *et al.*, 1998). Oasis effect

222   provides a vertical energy transfer in the form of sensible heat from the air surrounding the

223   wetland because its moist condition and transpiration make it cooler than the ambient air.

224   The clothesline effect, resulting from the tall wetland plants being surrounded by smaller

225   vegetation, provides a horizontal energy transfer due to wind (Kirkham, 2014). Wind effect

226   is enhanced due to the small size of the wetland and constantly disturbs the boundary layer

227   of plant leaves (Kadlec and Wallace, 2008), meaning that water vapor excreted by the

228   leaves is automatically replaced with fresh air and transpiration potential increases.

229   Frequent and high irrigation combined with a saturating water level in the wetland also

230   ensure high water availability and prevent limitation of ET due to water stress.

231   Based on these non-limited conditions, we hypothesized that transpiration of willows in a

232   treatment wetland should be highly correlated to stomatal conductance (*i.e.* water vapor

233   exchange rate between leaf and air; $\bar{G}_s$). $\bar{G}_s$ is generally measured in a volume of water per

234   surface of leaf per time unit (*e.g.* mmol m^{-2} s^{-1}), meaning that leaf area capable of

235   transpiring (LAI_{active}) is also required for ET calculation. Because of the relatively constant

236   disturbance of the boundary layer by wind, transpiration rate should also be driven mainly

237   by water vapor pressure deficit (VPD) in the ambient air. Otherwise, the irrigation of the

238   wetland being below the surface, there is no open contact between water and the

239   atmosphere. According Shuttleworth and Wallace's energy partitioning model (1985), the

240   high average LAI of *S. miyabeana* (> 4 m^2; Tharakan *et al.*, 2016) implies that most of the

241   energy available for ET is intercepted by the willows, reducing soil evaporation potential to

242   close to zero. Therefore, in this study, we assume that soil evaporation can be ignored and

243   that willow transpiration can be treated as ET. Daily ET of *S. miyabeana* grown in a

244   treatment wetland (mm/d) could then be estimated with the following leaf parameter based

245   equation:

246    $$ET_{SX67} = \bar{G}_s \cdot LAI_{active} \cdot VPD \qquad \text{(Eq. 5)}$$

247   Active leaf area can be calculated throughout the season according to the seasonal leaf

248   development curve and the abaxial/adaxial ratio established by measurements presented in

249   section 2.3, and the vapor pressure deficit can be calculated with daily temperature and

250   relative humidity data. To estimate stomatal conductance, we chose an empirical approach

251   based on environmental parameters known to influence stomata openings (Buckley and

252 Mott, 2013). We wanted those parameters to be easily accessible, to allow the transpiration

253 rate to be predicted with minimal resources. Through linear regressions, we tested the

254 statistical relation between mean daily stomatal conductance measured on site and the

255 following daily parameters: solar radiation, average and maximal air temperature, average

256 and minimal relative humidity, wind speed and day of the year. Parameters presenting a

257 significant relation with stomatal conductance (p<0.05) were combined to predict canopy

258 general conductance as follows:

259 $$\bar{G}_s = \sum \alpha \bar{g}_s^x \qquad \text{(Eq. 6)}$$

260 Where partial stomatal conductance ($\bar{g}_s$) was calculated according to previously selected

261 parameters (x) having their own relative influence (α) on the general stomatal conductance

262 of the wetland canopy ($\bar{G}_s$). Finally, crop coefficient was calculated as follows (Kadlec and

263 Wallace, 2008):

264 $$K_{SX67} = ET_{SX67}/ET_0 \qquad \text{(Eq. 7.)}$$

265 Where K_{SX67} is the crop, or plant, coefficient, ET_{SX67} is the modelled ET rate of the willow

266 stand and ET_0 the reference crop ET.

267 ### *2.6 Statistical analysis*

268 The relation between meteorological parameters and $\bar{G}_s$ was tested with either linear,

269 quadratic and power regressions. The influence of parameters on a given variable (*e.g.*

270 influence of leaf face on $\bar{G}_s$ variation) was tested with two-way ANOVAs analysis with a

271 0.05 significance threshold ($\alpha = 0.05$). Tukey's post-hoc statistical test was used when

272 necessary to better interpret the results of the analysis of variance ($\alpha = 0.05$). All statistical

273 analysis were done using R 3.5.1 software.

274   **3. Results**

275   The summer of 2016 was hot and dry, with a mean temperature of 18.0 °C (± 6.0) and 569

276   mm of rainfall from May to October. Mean temperature was similar in 2017 (17.9 °C

277   ± 4.8), but with less days on which maximum temperature rose above 30 °C. Also, 2017

278   saw much higher rainfall, with 819 mm for the same period. A summary of solar radiation,

279   rainfall and daily mean temperature for both growing seasons is shown in Figure 2.

280

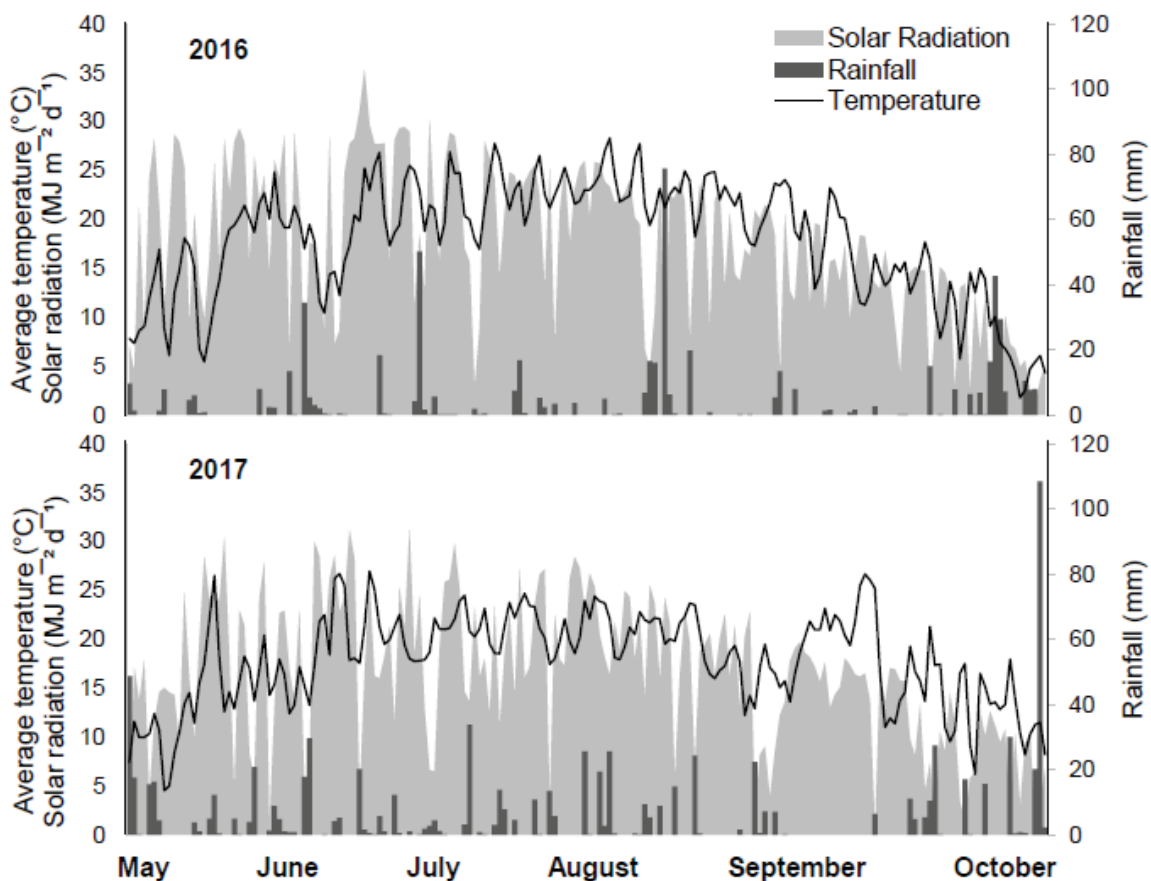281   **Figure 2.** Summary of the meteorological conditions at the experimental site for the 2016 and 2017

282   growing seasons.

283   Average reference crop ET was 4.5 mm/d in 2016 and 3.2 mm/d in 2017, for a total of 808

284   mm and 750 mm respectively, from May to November. Pan evaporation measured in 2017

285   represented 81% of reference ET. For the willow wetland, we calculated a mean daily ET

286   rate of 30.9 mm/d and a seasonal total ET of 4536 mm from May 9 to September 30 in

287   2016, and 16.6 mm/d and a seasonal total of 2906 mm from May 15 to October 31 in 2017.

288   *3.2 Physiological measurements*

289   Stomatal conductance values were generally higher and more variable in the 2016 season,

290   with a mean value of 418 ($\pm$ 124) mmol m^{-2} s^{-1} compared to 309 ($\pm$ 59) mmol m^{-2} s^{-1} in

291   2017. The adaxial/abaxial stomatal conductance ratio was relatively high (0.33 $\pm$ 0.17) and

292   variable in the early season, decreasing to relatively constant and low values (0.14 $\pm$ 0.06)

293   for the rest of the summer (Figure 3).
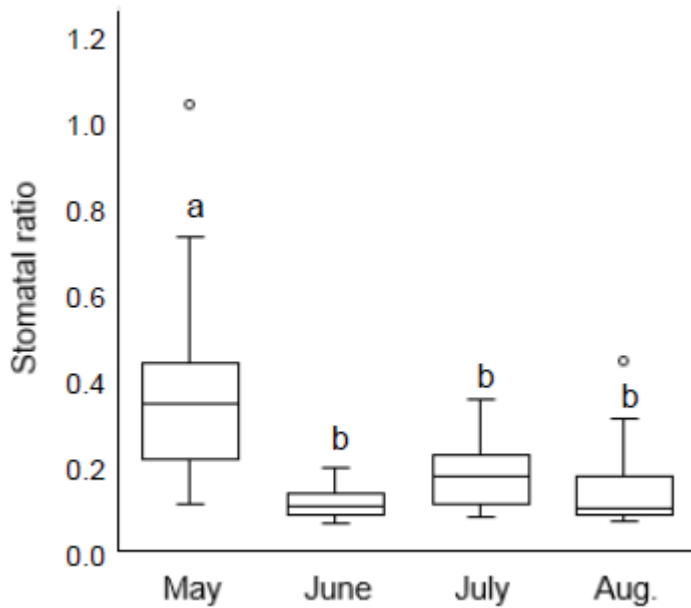

294

295   **Figure 3.** Adaxial/abaxial stomatal conductance ratio of *S. miyabeana* growing in treatment wetland

296   conditions for the 2017 summer season. Different letters represent statistically different values.

297   Thus, overall seasonal transpiration occurring on the upper part (adaxial) of the leaf

298   represents about 20% of the lower side (abaxial) transpiration, and actual stomatal

299   conductance equals approximately 120% of the values measured on the abaxial side of the

300   leaf only. In both the 2016 and 2017 seasons, leaf cover established rapidly, attaining its

301   highest value in July, with 10.4 and 11.4 m^2 of leaves per m^2 of soil respectively. The

302   canopy extended beyond the wetland borders by about 50 cm meter on each side, for a

303   projected canopy area of 63 m^2 compared to the actual wetland area of 48 m^2. Peak LAI

304   measured using the projected canopy area was 7.9 in 2016 and 8.7 in 2017. In 2017, the

305   global leaf area was a little higher than in 2016, attained its maximal value earlier and

306   retained active foliage later in the season (Figure 4).
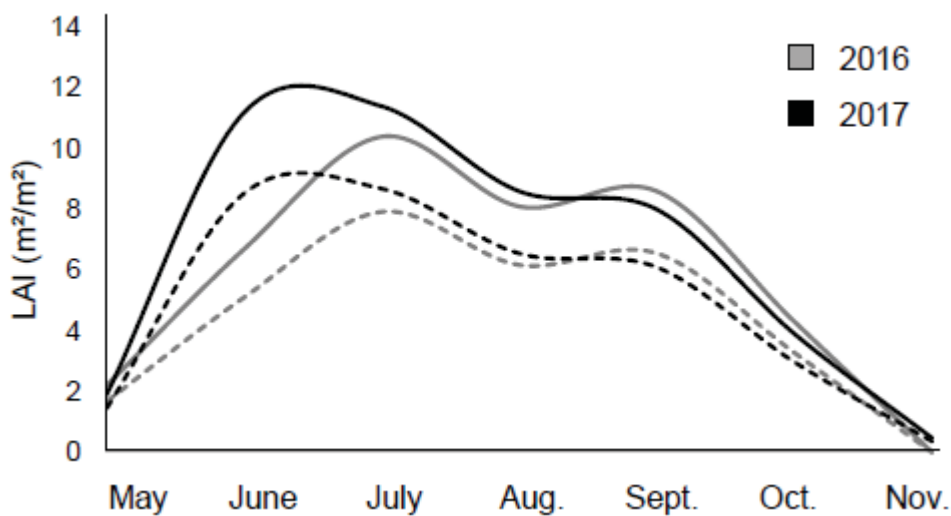

307

308   **Figure 4.** Evolution of the leaf area index of a 48 m² wetland (solid line) planted with *S.*

309   *miyabeana* throughout 2 successive growing seasons, and the corresponding values

310   adjusted for a 63 m² projected canopy area (dashed line).

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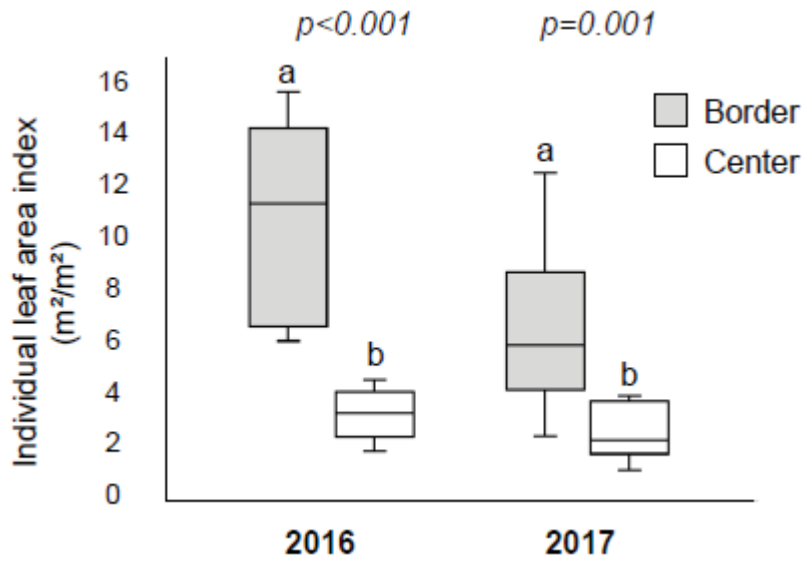
312   center (Figure 5).

313

314   **Figure 5.** Leaf area, measured in the month of July, of 15 individuals of *S. miyabeana* growing

315   either at the border or in the center of a 48 m^2 constructed wetland. Different letters represent

316   statistically different values.

317   *3.3 Evapotranspiration modelling*

318   We found a significant effect of temperature, solar radiation, relative humidity and day of

319   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$_{value}$	R^2	α	Equation
T	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	$2.9x + 168$

320   For temperature and relative humidity, mean daily values were better predictors than

321   maximum and minimum values respectively. Correlation between $\bar{G}_s$ and each factor

322   separately was relatively weak (r^2 from 0.05 to 0.21), but together they explained half of

323   stomatal conductance variation throughout the season (Figure 6.), which can be considered

324   satisfying due to the many other factors driving this parameter but not measured here

325   (Buckley and Mott, 2013).

326

327   **Figure 6.** Results of $\bar{G}_s$ modelling, based on temperature, solar radiation, relative humidity and day

328   of year, over $\bar{G}_s$ measured on the field under the same parameters.

329   The model was good at predicting mean $\bar{G}_s$, with a predicted mean seasonal value of 428

330   mmol m^{-2} s^{-1} over 418 mmol m^{-2} s^{-1} measured in 2016, and 329 mmol m^{-2} s^{-1} predicted over

331   309 mmol m^{-2} s^{-1} measured in 2017. Daily variation was captured more accurately in 2017

332   than in 2016 (Figure 7).

Figure 7. Stomatal conductance (Ḡs) field measurements (solid line) and modelling results (dashed line) over the 2016 and 2017 growing seasons.
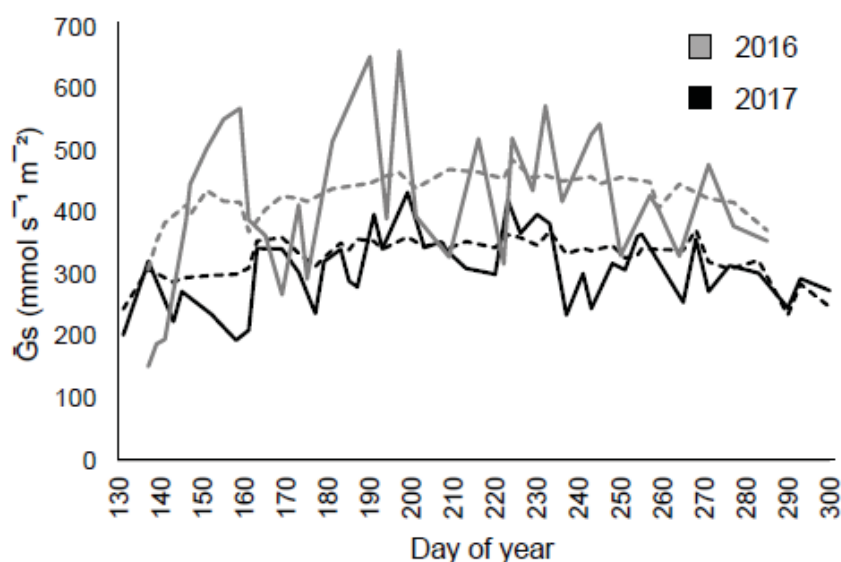
Using the general stomatal conductance predicted with this model and the previously established leaf area parameters, we calculated the ET rate (Eq. 5) and the corresponding crop coefficient (Eq. 7; Table 3). Willow ET was higher in 2016, as was reference ET, with a mean daily rate of 17.1 mm/d compared 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 (ET_0), estimated active leaf area index of

the 48 m^2 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 ET	Peak K_{ET}	Seasonal K_{ET}	Annual K_{ET}	Ref.
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.

358 Average seasonal ET rates reported for other fast growing willow cultivars grown in field

359 plantation are also generally much lower than our results (1.4 mm/d, Linderson et al., 2007;

360 3.0 mm/d, Lindroth et al., 1994; 2.9 mm/d, Personn, 1995; 1.0 mm/d, Mata-Gonzalez; 3.1

361 mm/d, Budny and Benscoter, 2012). In comparison, similar rates (from 10 to 23 mm/d)

362 were measured for young *S. babylonica* grown in water saturated conditions in the north-

363 eastern United States (Pauliukonis *et al.*, 2001). Such high ET rates can be explained by

364 both enhancing factors linked to the treatment wetland itself (*i.e.* oasis and clothesline

365 effect, high water availability, important border effect) and by *S. miyabeana* ecophysiology

366 (*i.e.* high stomatal conductance and leaf area index).

367 In this study, a simple model based mainly on two leaf parameters was sufficient to model

368 ET. As expected, the model ET results were lower than the water balance results in 2016

369 (see section 2.4). However, 2017 simulation results closely resembled water balance

370 results. The fact that our simplified ET model yielded conclusive results supports our

371 premise that typical ET limiting factors are greatly attenuated in small wetlands. Other

372 studies presenting ET modelling methods for willows often include several limiting factors

373 (Irmak et al., 2013, Iritz et al., 2001), ignore heat advection effect (Přibáň and Ondok,

374 1986) or focus on soil hydrology (Personn, 1995, Hartwich *et al*., 2016; Borek et al., 2010)

375 or complex physiological processes (Tallis et al., 2013). Although based on sound scientific

376 assumptions, those models hardly apply in treatment wetland conditions where water level

377 is constant, limitations are attenuated and heat advection effect is very important. The few

378 input parameters required for the operation of the model also represent an opportunity for

379 managers working with treatment wetlands to easily include ET estimation in their planning

380 activities. However, to be used for other taxa, a basic knowledge of the LAI dynamic and

381 general stomatal conductance for the species is needed, and could require additional $\bar{g}_s$

382 measurement in the field to adjust the model.

383 Regarding ET related characteristics specific to *S. miyabeana*, we found that mean stomatal

384 conductance (0.4 mol m^{-2} s^{-1}) was consistent with published results for other willows (0.4

385 mol m^{-2} s^{-1}, Budny and Benscoter, 2016; 0.2-0.7 mol m^{-2} s^{-1}, Hall *et al.*, 1998) or higher

386 (0.2 mol m^{-2} s^{-1}, Kučerová *et al.*, 2001). Leaf area index values were higher than those

387 reported in the literature for other willow cultivars, even when using the projected canopy

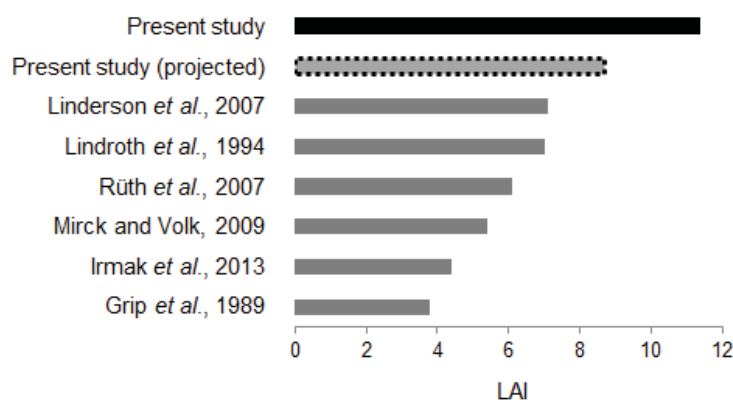388 area for the calculation (Figure 8).

389

390 **Figure 8.** Maximal leaf area index (LAI) reported for willow stands (different cultivars) in various

391 studies including the present results, and the corresponding value adjusted with projected canopy

392 area.

393 As for stomatal conductance, it is also interesting to note that the highest mean daily value

394 measured (661 mmol m^{-2} s^{-1}) is much higher than the values proposed for deciduous trees

395 and even plants from wet habitats (Jones, 2013). The ratio between the conductance of the

396 upper and lower side of the leaf is consistent with the literature predicting higher adaxial

397 activity or adaxial stomatal density in younger leaves (Fontana *et al.*, 2017). Meteorological

398 factors could only explain about half of the stomatal conductance values and variability.

399 Stomatal aperture is also driven by many biochemical and environmental factors (Buckley

400 and Mott, 2013) that were not studied here. Aging of the willows, or negative effects of

401 contaminant accumulation in the substrate are also factors affecting long term variability of

402 $\bar{G}_s$ in a wetland that should be considered. A sampling campaign (data not shown)

403 conducted in June of 2017 in Denmark on *S. viminalis* clones used for zero-discharge

404 wetlands showed significantly greater stomatal conductance in willows recently coppiced,

405 compared to older individuals growing in the exact same conditions, which supports the

406 aging hypothesis. Those factors should be investigated thoroughly in the future. Leaf area

407   of the willow wetland attained its maximal value (complete canopy closure) with two-year-

408   old shoots, peaking in July at around 12 m^2 of leaves per m^2 of ground. Planting density

409   and methodological differences could partially explain why LAI of our wetland was very

410   high compared to findings reported in the literature. Furthermore, all results presented in

411   Figure 8 are based on field plantation or natural river bands of much greater size than our

412   wetland and the effect of increased leaf area at the border is negligible. Our finding

413   comparing individual leaf area at the edges versus in the center of the wetland is also

414   interesting because it means we could modulate ET rate directly in the wetland design.

415   Indeed, if ET is directly related to LAI as demonstrated here, adjusting the edge or aspect

416   ratio of the surface area of a wetland could enhance (higher ratio) or limit (lower ratio) ET

417   per ground unit, according to management objectives. Fertilization applied at the beginning

418   of 2017 seemed to have accelerated the establishment of the leaf cover but did not

419   significantly increase maximal LAI. Since the fertilizer used consisted of solid granules

420   applied directly on the soil, with degradation regulated by rainfall and temperature, it is

421   possible that rapid closure of the canopy and high rain interception by willows prevented

422   the fertilizer from appropriately degrading and penetrating the substrate. This hypothesis is

423   supported by the absence of nitrogen in the wetland effluent throughout the season (result

424   not shown). In 2016, the canopy already seemed completely closed by mid-season and it is

425   possible that maximum leaf area index was already attained. Indeed, in 2017, stems grew

426   higher but there was little or no leaf development at the bottom of the stems (as was

427   observed in 2016), probably because canopy closure was achieved and all available light

428   was intercepted in the upper part of the trees. Therefore, we conclude that maximal LAI

429   was achieved with two-year-old shoots, without a need for fertilization, and that coppicing

430   should be scheduled on a two-year basis.

431   **5. Conclusions**

432   *S. miyabeana* ET in treatment wetland condition was very high throughout this study. We

433   highlighted several factors related to treatment wetlands that can significantly increase

434   potential ET. Because there are few limitations on ET in wetlands, a model exclusively

435   based on leaf parameters successfully predicted ET values and calculated crop coefficients

436   for the studied willow wetland. Because these results are based on a full-scale wetland, they

437   can be used as design parameters for treatment wetlands using *S. miyabeana,* and the

438   equation presented for ET calculation can be adjusted for other fast-growing willow species

439   used in similar growing conditions. We also presented a strategy to optimize ET per ground

440   area by changing the aspect ratio of the wetland, and consequently its leaf area index, as

441   well as regularly coppicing the stems. In the future, other parameters possibly affecting ET

442   in treatment wetlands such as tree aging, substrate type and contaminant toxicity could be

443   investigated. This study is a first step towards better ecophysiological characterization of

444   woody plants used in treatment wetlands.

445   **Acknowledgment**

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