

1 **Aridland constructed treatment wetlands II: Plant mediation of surface hydrology**  
2 **enhances nitrogen removal.**

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12

13 **ABSTRACT**

14  
15 Constructed treatment wetlands have been well established as effective and sustainable  
16 solutions to the problem of urban water treatment and reuse. However, treatment wetlands  
17 located in aridland cities may behave differently relative to their more mesic and humid  
18 counterparts, and this could potentially impact their ability to deliver the ecosystem services that  
19 are expected of them. Specifically, in hot, dry climates large water losses via evaporation and  
20 plant transpiration may comprise a major component of whole-system water budgets. Our  
21 primary goal was to develop a rigorous and informed model of how well these “working  
22 wetlands” function in hot, arid climates by developing and comparing robust water and nutrient  
23 budgets, as our process-based understanding of how mesic constructed wetlands function may  
24 not be readily transferred to arid climates where constructed wetlands are becoming increasingly  
25 widespread. At the Tres Rios constructed treatment wetland in Phoenix AZ USA, we quantified  
26 water losses via plant transpiration and open water evaporation as well as inorganic N loads into  
27 and from the whole wetland system and into the vegetated marsh. We found that water losses  
28 due to transpiration and evaporation were remarkably high when compared to most mesic  
29 constructed wetlands. Total water losses via evaporation and transpiration peaked at 300,000 m<sup>3</sup>  
30 mo<sup>-1</sup> (714 L H<sub>2</sub>O m<sup>-2</sup> mo<sup>-1</sup>) in the hot, dry summer months and averaged more than 70% of the  
31 whole-system water losses over a 27 month time period. At the same time, the vegetated marsh  
32 removed nearly all of the inorganic N that was supplied to it. Large transpirative water losses  
33 moved large volumes of replacement water into the marsh via a “biological tide” that provided  
34 more opportunities for vegetation and soil microbes to process N and other target solutes. This  
35 enhanced the N treatment efficacy of the Tres Rios constructed treatment wetland relative to  
36 humid, mesic systems. To our knowledge, this is the first time that biotically-mediated surface  
37 hydrology has been demonstrated in any wetland.

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40 **Keywords:** Constructed treatment wetlands, water budgets, nutrient budgets, nitrogen removal,  
41 transpiration, urban wetlands

## 42 1. INTRODUCTION

43 The last two centuries have been characterized by a dramatic shift in human  
44 demographics and land use practices, with more people choosing cities and urban areas as their  
45 primary habitat. Specifically, since 1900 the human population living in urban areas has  
46 increased from 10% to over 50% globally, with that proportion projected to grow to 80% by  
47 2050 (Grimm et al. 2008). As urban centers grow in size and number, so do their demands for  
48 resources and services. In particular, the last century has seen the transformation of many cities  
49 into “sanitary cities”, with highly centralized, capitalized, and expensive infrastructure designed  
50 to keep inhabitants healthy (Melosi 2000; per Grove 2009). This development of infrastructure  
51 has imparted considerable inertias on cities that make it more difficult for decision-makers to  
52 envision, let alone implement, novel or transformative new solutions to growing problems  
53 (Childers et al. 2014). Still, there are a number of ways in which urban infrastructure may make  
54 cities more resilient and sustainable by designing to optimize key ecosystem services (Pickett et  
55 al. 2013). One example of using infrastructure to optimize key ecosystem services is the  
56 increasing use of constructed treatment wetlands as part of wastewater treatment plants in place  
57 of expensive and energy-intensive treatment technologies.

58 Constructed treatment wetlands are a relatively low cost and low maintenance solution to  
59 the age-old problem of wastewater treatment and, more recently, water reclamation. They are  
60 especially attractive due to the fact that variable wastewater pathways and often relatively remote  
61 placement of wastewater treatment systems demand simple technologies that require little active  
62 management (Wallace and Knight 2006; Nivala et al. 2013). Most treatment wetlands are  
63 characterized by the presence of macrophytic vegetation, waterlogged or saturated soils, and  
64 municipal effluent that must be treated (Fonder and Headley 2013). While these wetlands share  
65 similar ecosystem service expectations, or “jobs”, of removing nutrients or other solutes (Tanner  
66 et al. 2012), they may be further differentiated with regards to the level of “hard” engineering  
67 components designed into them. These range from totally passive (horizontal flow) systems to  
68 intensely managed systems featuring pulse loading or active aeration (Fonder and Headley 2010).  
69 Design choices for treatment wetlands are often highly dependent on local or regional variables,  
70 including imposed statutory wastewater treatment regulations or site-specific conditions.  
71 Importantly, basic differences, such as climate, may limit general conclusions that might be  
72 drawn from comparative studies of treatment wetland efficacy (Nivala et al. 2013). While

73 treatment wetlands are relatively similar in design and expectations, particular attention must be  
74 paid to the way these systems function in their respective climatic settings.

75 In arid environments, which occupy more than 30% of the earth's land surface, one of the  
76 biggest challenges facing cities is the scarcity of water. As a result, many of these cities are  
77 increasingly turning to the reuse of treated municipal effluent as a means of reclaiming and  
78 recycling water for various urban uses (Greenway 2005). Notably, in the aridland city of Phoenix  
79 AZ USA, virtually all municipal effluent is reused within the city (Metson et al. 2012), and the  
80 only significant export of water from the city is via evaporative losses to the atmosphere. The  
81 challenge facing water reuse is that using reclaimed water in densely populated areas requires  
82 that this water be clean. Increasingly, water service providers are turning to treatment wetlands  
83 that, in addition to cleaning effluent, can also provide auxiliary and serendipitous ecosystem  
84 services such as habitat for birds and other fauna and recreational spaces for urban dwellers.  
85 However, constructed treatment wetlands in hot, arid cities may face unique challenges relative  
86 to similar systems in more mesic and humid settings. In particular, treatment wetlands in hot, dry  
87 climates should lose large amounts of water to evaporation and plant transpiration. How aridland  
88 treatment wetlands perform under these conditions is not well quantified nor well understood,  
89 and our primary goal for this research was to better understand these dynamics. To address this  
90 goal, we quantified water and nutrient budgets for a constructed treatment wetland in Phoenix.

91 In Phoenix, annual precipitation averages 180 mm while potential wetland  
92 evapotranspiration is over 2100 mm (Kadlec 2008). Thus, the objectives driving our research in  
93 this wetland were: 1) to use a nutrient budget approach to determine net inorganic nitrogen (N)  
94 removal by the vegetated marsh and the whole system; 2) to measure transpirational water losses  
95 from the emergent macrophytes; 3) to calculate a dynamic water budget for the system based on  
96 surface water inflows and outflows, precipitation inputs, and estimates for open-water  
97 evaporation and plant transpiration, and; 4) to determine how well this aridland wetland removed  
98 N from municipal wastewater effluent.

99 Historically, phase-change atmospheric losses of water from wetlands have been  
100 presented as a single integrated process: evapotranspiration ( $E_t$ ). Efforts to quantify and  
101 understand  $E_t$  in wetlands have been confounded by large differences in estimates across  
102 wetlands as well as divergences in the many ways that  $E_t$  may be calculated (Goulden et al.  
103 2007). For the purposes of our study, we conceptually and empirically separated plant-mediated

104 transpirational water losses from open-water evaporation. Transpirational water losses have been  
105 reported as both larger and smaller than water losses due to open-water evaporation, but it seems  
106 probable that transpiration dominates water loss in wetlands found in hot, arid climates  
107 (Pauliukonis and Schneider 2001). In fact, this has been documented for a wetland near our study  
108 system (Kadlec 2006). Open-water evaporation should also be high in these systems, and there  
109 are a variety of approaches to this calculation (Kumar et al. 2012). Many approaches to  
110 estimating  $E_t$  in wetlands have calculated it as a residual term in whole-system water and energy  
111 budgets that are based on meteorological data. However, a fundamental limitation with this  
112 approach for our study was that this “subtraction” approach did not articulate spatial  
113 heterogeneity and site-specific nuances that affect  $E_t$ , such as water level, plant community  
114 composition, and plant canopy structure (Lott and Hunt 2001).

115         The objectives we addressed in this study are clearly not new to the constructed treatment  
116 wetland literature. However, researchers have seldom rigorously studied wetlands that are  
117 specifically designed to perform particular ecosystem services but are located in hot, arid  
118 climates. Our research sought to tease apart the key components of the wetland water budget and  
119 combine them with a nutrient budget in order to understand how well the constructed treatment  
120 wetland we studied was performing. Our ultimate goal was to arrive at a more informed model of  
121 aridland wetland ecosystem service performance that can facilitate the best possible adaptive  
122 management of these systems.

123

## 124 **2. METHODS**

### 125 *2.1. Site Description*

126         Our study was conducted at the constructed treatment wetland associated with the 91<sup>st</sup>  
127 Avenue Wastewater Treatment Plant, which is the largest such facility in Phoenix. Construction  
128 of the 3-cell Tres Rios wetland system was completed in 2009-2010 and we began our work  
129 there in Summer 2011. We have focused our work on the largest of three wetland cells, which  
130 was planted first and came online in Summer 2010 (Figure 1). Each wetland cell is bounded by  
131 levee roads, and the cell we studied is 42 ha in size, half of which is fringing vegetated marsh  
132 and half of which is mostly open water with several small upland “islands”. Water depths in the  
133 marsh were consistently about 25 cm while open water depths were 1.5 to 2 m; because of the  
134 way water was managed in the system, these depths did not vary. Our study cell received from

135 95,000 to over 270,000 m<sup>3</sup> d<sup>-1</sup> of effluent, depending on the time of year. The marsh areas were  
136 planted with seven emergent wetland species that are native to Arizona: *Typha latifolia*, *Typha*  
137 *domingensis*, *Schoenoplectus acutus*, *Schoenoplectus americanus*, *Schoenoplectus californicus*,  
138 *Schoenoplectus maritimus*, and *Schoenoplectus tabernaemontani*. *Schoenoplectus maritimus* was  
139 a minor component of the plant community and was out-competed within the first year of our  
140 research (Weller et al. this issue), so we focused our transpiration measurements only on the  
141 other six species.

142 INSERT FIGURE 1 HERE

## 143 2.2. Transpiration Measurements and Evaporation Calculations

144 We utilized a dual-gradient experimental design to study the two major hydrologic  
145 pathways of the wetland system: A whole-system gradient from the inflow to the outflow, and a  
146 within-marsh gradient that included 10 transects from the shore to the open water that were  
147 distributed evenly along the whole system gradient (Figure 1; for details, see Weller et al. this  
148 issue). All of our sampling followed a bimonthly schedule (January, March, May, July,  
149 September, and November), and we present data from July 2011 through September 2013 in this  
150 paper. We measured leaf-specific gas flux along marsh transects that included all species groups  
151 using a LICOR LI-6400 handheld infrared gas analyzer (IRGA). Measurements were made on  
152 individual *T. latifolia*, *T. domingensis*, *S. acutus*, *S. americanus*, *S. californicus*, and *S.*  
153 *tabernaemontani* plants at 50-cm intervals from near the water surface to the top of the canopy.  
154 We collected gas flux data continuously from early morning until as late in the day as was  
155 feasible. For logistical reasons, we sampled one transect at a time, with individual plants chosen  
156 haphazardly until all species present in that transect had been sampled a number of times. Due to  
157 site access restrictions, we were unable to collect nighttime transpiration data and thus assumed  
158 that nighttime transpirative water loss was negligible. Notably, this assumption is one reason that  
159 our transpiration estimates are conservative.

160 Several of the *Schoenoplectus* species have thick stems which required us to modify the  
161 stock LI-6400 IRGA sampling chamber in order to get a gas-tight seal without crushing the plant  
162 stems. To remedy this, we used custom-made foam pads that allowed us to seal the leaf chamber  
163 around these stems while minimizing damage to the plant material. Specifically, we were  
164 interested in IRGA measurements of leaf-specific transpiration rate ( $Tr$ ; mmol H<sub>2</sub>O m<sup>-2</sup> sec<sup>-1</sup>),  
165 photosynthetically active radiation (PAR, μmol photons), ambient air temperature (°C), and

166 relative humidity (%). All of these measurements were taken using the IRGA's default internal  
167 sensors. Daily transpiration data were expressed in units of leaf surface area, and we converted  
168 surface area to dry weight biomass by drying and weighing 8-10 samples of different surface  
169 areas and generating relationships between surface area and dry weight biomass for each species  
170 ( $r^2 > 0.90$  and  $p < 0.01$  in all cases).

171 Half of the 42 ha wetland cell we sampled was open water, as determined by digitizing  
172 and measuring aerial imagery in ArcGIS (ver 10.0, ESRI, Redlands, CA). We calculated open-  
173 water evaporation (E, in  $\text{mm hr}^{-1}$ ) using the Shuttleworth (1993) variation of the Penman (1948)  
174 equation:

$$175 \quad \text{Open Water Evaporation} = ((S * \text{PAR}) + C * 6.43 * (1 + 0.536 * V) * \text{VPD}) / (2.39 * (S * C))$$

176 using hourly data for air temperature (T), relative humidity (RH), irradiance (PAR), barometric  
177 pressure (P), vapor pressure deficit (VPD), slope of the saturation vapor pressure curve (S),  
178 psychrometric constant (C), and wind speed (V) logged at an on-site meteorological station  
179 operated by the City of Phoenix (hereafter referred to as the "meteorological station"). If the  
180 hourly meteorological station data were missing or not reliable for any necessary variable (<5%  
181 of all hourly readings), we did not calculate transpiration or open-water evaporation. Because of  
182 these missing data, we are confident that our evaporation water loss values are underestimates  
183 and are further confident that our transpiration estimates are conservative. Finally, we calculated  
184 internal water loss using a dynamic monthly water budget for the whole system by summing the  
185 whole-system hourly water fluxes via open water evaporation (E) and plant transpiration (Tr) to  
186 monthly totals:

$$187 \quad \text{Internal Water Loss} = E + \text{Tr}$$

188 and compared these data with the monthly whole-system water deficit, which we calculated by  
189 summing daily inflows ( $\text{SW}_i$ ), rainfall (R), and outflows ( $\text{SW}_o$ ) for the month and subtracting the  
190 latter from the former:

$$191 \quad \text{Water Budget Deficit} = \text{SW}_i + R - \text{SW}_o$$

192  $\text{SW}_i$ ,  $\text{SW}_o$ , and R measurements were taken from whole-system data sets provided by the City of  
193 Phoenix (see Section 2.4 for details). After consultation with on-site engineers, we determined  
194 that vertical losses via percolation to groundwater were negligible so this parameter was not  
195 included in our water budget deficit calculations.

196 2.3. *Water Quality Sampling*

197 On the same bimonthly schedule described above, we collected water quality samples  
198 along both gradients. Triplicate water samples were collected by hand in acid-washed Nalgene  
199 bottles at the inflow and outflow points of the study cell, and at the shore and open-water ends of  
200 three marsh transects that represented the inflow-outflow whole system gradient (Figure 1).  
201 Nitrate ( $\text{NO}_3^-$ ), nitrite ( $\text{NO}_2^-$ ), and ammonium ( $\text{NH}_4^+$ ) samples were centrifuged to remove  
202 particulates and analyzed on a Lachat Quick Chem 8000 Flow Injection Analyzer (detection  
203 limit  $0.85 \mu\text{g NO}_3\text{-N/L}$  and  $3.01 \mu\text{g NH}_4\text{-N/L}$ ). For simplicity, we will restrict our water quality  
204 analysis to dissolved inorganic nitrogen ( $\text{NH}_4^+$ ,  $\text{NO}_3^-$ , and  $\text{NO}_2^-$ ). Differences in water quality  
205 data were determined using a paired t-test for whole-system inflow versus outflow data, while an  
206 unequal variance t-test was used for shore versus open water marsh transect data.

207

208 2.4. *Whole System Data*

209 Our research at the Tres Rios constructed treatment wetland is in partnership with the  
210 City of Phoenix Water Services Department, and they provided us with key whole system  
211 datasets for our analyses. We used their daily inflow ( $\text{SW}_i$ ) and outflow ( $\text{SW}_o$ ) data (in mgd,  
212 converted to  $\text{m}^3 \text{d}^{-1}$ ), measured using standard acoustic Doppler flow meters, to calculate total  
213 monthly water flux into and out of our study cell from January 2012 through September 2013.  
214 For the months when we sampled the system, we combined total monthly water flux with  
215 inorganic N concentrations to calculate whole system monthly N loads. We used the previously  
216 discussed hourly meteorological station data to scale our transpiration estimates in time and to  
217 calculate evaporative water losses from the open-water component of our study cell (see Sections  
218 2.2 and 2.5 for details). Because of inconsistencies in the rainfall data collected by the on-site  
219 meteorological station, we summed daily rainfall data from the nearby Goodyear airport to  
220 calculate monthly rainfall contributions (R) to the whole system water budget.

221

222 2.5. *Data-Scaling and Water Budget Calculations*

223 An important step in our whole-system water budget estimates was the scaling of leaf-  
224 specific measurements of transpiration water losses from individual plants to 21 ha of marsh, and  
225 from transpiration measurements made over the course of individual days to a 2+ year time-  
226 series. To scale transpiration water flux across space (Daily  $\text{Tr}_{\text{System}}$ ;  $\text{m}^3 \text{H}_2\text{O d}^{-1}$  whole system<sup>-1</sup>),



227 we combined our IRGA measurements (Instantaneous  $Tr_{IRGA}$ ;  $\text{mmol H}_2\text{O m}^{-2} \text{sec}^{-1}$ ), corrected for  
228 dry-weight plant biomass (Area/Bio;  $\text{cm}^2 \text{gdw}^{-1}$ ), with the whole system live macrophyte  
229 biomass ( $Bio_{System}$ ;  $\text{Mg whole system}^{-1}$ ) calculated from the bimonthly data collected along our  
230 10 marsh transects (see Weller et al. this issue):

$$231 \quad \text{Daily } Tr_{System} = \text{Instantaneous } Tr_{IRGA} * \text{Area/Bio} * Bio_{System} * 74.06$$

232 where 74.06 was a dimensional analysis conversion for  $\text{mmol H}_2\text{O}$  to  $\text{m}^3 \text{H}_2\text{O}$ , seconds to days,  
233 and  $\text{gdw}$  biomass to  $\text{Mg}$  biomass for the 21 ha marsh. To estimate whole system biomass  
234 ( $Bio_{System}$ ) we developed phenometric models that allowed us to non-destructively estimate live  
235 biomass for all plant species by making simple allometric measurements in the field (per Daoust  
236 and Childers 1998; Childers et al. 2006). Every two months, we measured all of the plants in  
237 five  $0.25 \text{ m}^2$  quadrats that were randomly located along each of the 10 marsh transects shown in  
238 Figure 1, for a total of 50  $0.25 \text{ m}^2$  quadrats sampled (for details Weller et al. this issue). We used  
239 simple linear interpolation to extrapolate plant biomass between bimonthly samplings, producing  
240 daily estimates of live macrophyte biomass from July 2011 through September 2013.

241 To scale our plant-specific transpirational water losses in time, we used the  
242 micrometeorological data generated by the IRGA at the time of sampling and the corresponding  
243 hourly data from the on-site meteorological station. Because plant transpiration flux is driven  
244 largely by T, PAR, and RH (Sánchez-Carrillo et al. 2001), we first regressed hourly  
245 meteorological station data for these three variables against the mean of all IRGA measurements  
246 for the same variables for that same hour for each transpiration sampling day. We then generated  
247 multiple regression models that related transpiration flux for each macrophyte species (per  $\text{Mg}$   
248 dry weight of plant biomass) to PAR, T, and RH as measured by the IRGA. These models  
249 combined with the IRGA-meteorological station regressions allowed us to use continuous data  
250 from the meteorological station to interpolate our species-specific transpiration measurements  
251 through time, resulting in whole-system daily transpirative losses (in  $\text{m}^3 \text{H}_2\text{O day}^{-1}$  for the 21 ha  
252 of vegetated marsh) from July 2011 through September 2013. See Table 1 for a summary of all  
253 parameters, applications, methods, and sampling frequencies used to for these calculations.

254 INSERT TABLE 1 HERE

255

### 256 3. RESULTS AND DISCUSSION

#### 257 3.1. Transpiration

258 The phenometric biomass models were the same for *T. latifolia* and *T. domingensis*, and  
259 for *S. acutus* and *S. tabernaemontani* (see Weller et al. this issue for details). For this reason we  
260 combined our leaf-specific transpiration measurements for both species of each genus in both  
261 cases. Transpiration rates for *S. californicus* and *S. americanus* were treated separately and  
262 discretely. On a typical hot, dry day in July, these rates were as high as 10, 14, 13, and 9 mmol  
263 H<sub>2</sub>O m<sup>-2</sup> leaf area s<sup>-1</sup> for *Typha* spp., *S. acutus* + *S. tabernaemontani*, *S. californicus*, and *S.*  
264 *americanus*, respectively. In January, when air temperatures were low and plant biomass was at  
265 its annual minimum, the highest transpiration rates were only 2, 4, 4, and 6 mmol H<sub>2</sub>O m<sup>-2</sup> leaf  
266 area s<sup>-1</sup> for *Typha* spp., *S. acutus* + *S. tabernaemontani*, *S. californicus*, and *S. americanus*,  
267 respectively. These rates equated to July 2011 averages of 30 ± 0.8, 16 ± 0.4, 0.6 ± 0.3, and 9.6 ±  
268 0.02 mm day<sup>-1</sup> and January 2014 averages of 1.7 ± 0.2, 1 ± 0.03, 0.4 ± 0.03, and 0.4 ± 0.03 mm  
269 day<sup>-1</sup> for the entire standing stock of *Typha* spp., *S. acutus* + *S. tabernaemontani*, *S. californicus*,  
270 and *S. americanus*, respectively. By comparison, Pedescoll et al. (2013) found that a *T.*  
271 *angustifolia*-dominated constructed wetland in a similarly arid climate transpired a maximum of  
272 23 mm day<sup>-1</sup>, which is similar to the maximum rates from our treatment wetland. In contrast,  
273 individual *T. domingensis* plants in the humid, mesic Florida Everglades transpired up to 11  
274 mmol m<sup>-2</sup> sec<sup>-1</sup> (Koch and Rawlik 1993)—well below the maximum plant-specific rates we  
275 measured for this species. Reported transpiration rates for *Typha* spp. are even lower in cooler  
276 climates: 5.8 ± 0.9 mm day<sup>-1</sup> in marshes along Oneida Lake NY USA (Pauliukonis and  
277 Schneider 2001) and 6.5 mm day<sup>-1</sup> in marshes in Ohio, USA (Martin et al. 2003). These findings  
278 illustrate the dramatic differences in transpiration water loss between hot, arid and cooler, mesic  
279 wetlands.

280 We scaled these leaf-specific IRGA gas flux measurements in time to generate estimates  
281 of daily water loss by plant transpiration for the entire 21 ha vegetated marsh using hourly data  
282 from the on-site meteorological station. Regression relationships between meteorological station  
283 data and measurements within the canopy allowed us to extrapolate transpiration measurements  
284 between our bimonthly samplings. Multivariate regressions relating transpiration rate to T, RH,  
285 and PAR found that only T and PAR were significant predictors of transpiration by all four plant  
286 species groups, with relatively little interspecies variation in the strength of the relationships ( $r^2 =$

287 0.57 to 0.67; all p-values <0.01). We scaled leaf-specific transpiration measurements in space  
288 using the live macrophyte biomass estimates for each species group from Weller et al. (this  
289 issue) after converting transpiration rates from  $\text{mmol H}_2\text{O m}^{-2}$  leaf area  $\text{s}^{-1}$  to  $\text{ml H}_2\text{O gdw}$   
290  $\text{biomass}^{-1} \text{ day}^{-1}$  and ultimately to  $\text{m}^3 \text{H}_2\text{O day}^{-1}$  for the entire 21 ha marsh. We found  
291 considerable variation in transpirative water losses among the four species groups, with *Typha*  
292 spp. having the largest average transpiration rate ( $2614 \pm 58 \text{ m}^3 \text{H}_2\text{O day}^{-1}$  or  $12.45 \pm 0.28 \text{ L H}_2\text{O}$   
293  $\text{m}^{-2} \text{ day}^{-1}$ ) and *S. californicus* the lowest average rate ( $221 \pm 5 \text{ m}^3 \text{H}_2\text{O day}^{-1}$  or  $1.05 \pm 0.02 \text{ L}$   
294  $\text{H}_2\text{O m}^{-2} \text{ day}^{-1}$ ). Most, but not all, of this variation was driven by differences in the amount of  
295 live biomass (Weller et al. this issue). In fact, although *Typha* spp. has been steadily taking over  
296 the macrophyte community in our study wetland (Weller et al. this issue), *S. acutus* and *S.*  
297 *tabernaemontani* continued to make a contribution to overall daily transpirative water losses that  
298 was disproportionate to their contributions to total biomass (Figure 2). This disproportionate  
299 contribution to daily transpirative water loss was likely due to the fact that these large-stemmed  
300 bulrushes transpire considerably more water per unit biomass than do cattails.

301 INSERT FIGURE 2 HERE

302 As expected, all species groups showed strong seasonality, with the highest  
303 transpirational water losses in the hot, dry summer months and the lowest losses in the winter  
304 months (Figures 2 and 3). While close relationships between transpiration and seasonal variation  
305 in climate and biomass have been well documented in the literature (e.g. Moro et al. 2004;  
306 Pedescoll et al. 2013), the rates of water transpired from our Tres Rios system were considerably  
307 higher than even those from other arid wetlands. For example, Drexler et al. (2008) found that an  
308 arid wetland in California, USA transpired on average  $6 \text{ mm day}^{-1}$ , and Goulden et al. (2007)  
309 reported peak annual transpiration of less than  $5 \text{ mm day}^{-1}$  for another marsh in California. A  
310 study conducted by Bialowiec et al. (2014) in a cooler, more mesic wetland in Poland reported a  
311 maximum system-wide transpiration rate of  $4.6 \text{ mm d}^{-1}$ , and Abtew (1996) found that *Typha*-  
312 dominated marsh systems in the warm, humid Florida Everglades had maximum transpirational  
313 losses of only  $3.6 \text{ mm day}^{-1}$ . Overall, Tres Rios had transpirative water losses that were as much  
314 as an order of magnitude greater than has been reported for other wastewater treatment wetlands.

315 Whole-system macrophyte biomass exhibited regular seasonality, with peak biomass in  
316 July of approximately 400 to 600 Mg for the 21 ha marsh (Figure 3). As expected, whole-system  
317 transpiration losses closely mirrored plant biomass (Figure 3). The highest total monthly

318 transpiration losses of approximately 200,000 to 300,000 m<sup>3</sup> for the 21 ha marsh were in June  
319 and July, when ambient air temperatures regularly exceeded 45°C and relative humidity was  
320 typically below 10%, and often below 5%. As we noted above, during this study, the plant  
321 community was tending towards a bi-specific stand of *T. domingensis* and *T. latifolia*, but the  
322 contribution of several *Schoenoplectus* species to transpiration flux remained strong (Figure 2).

323 INSERT FIGURE 3 HERE

### 324 3.2. Whole System Water Budget

325 Daily inflow and outflow data for our wetland study cell were used to calculate whole-  
326 system monthly water deficits. The inflow sensor was not fully operational until January 2012,  
327 and therefore whole-system water deficit was not calculated until January 2012 (Figure 4). The  
328 whole-system water budget was dominated by transpirational water losses and open water  
329 evaporation, the latter of which represented a lower fraction of our total water budget—on the  
330 order of a third or less—during the hotter months when plant biomass was higher and the plants  
331 were most productive (Figure 4). Notably, precipitation was only a small fraction of the water  
332 budget. During our study, Tres Rios received 105 mm (105 L H<sub>2</sub>O m<sup>-2</sup> yr<sup>-1</sup> or 44,100 m<sup>3</sup> yr<sup>-1</sup>),  
333 122 mm (122 L H<sub>2</sub>O m<sup>-2</sup> yr<sup>-1</sup> or 51,240 m<sup>3</sup> yr<sup>-1</sup>), and 167 mm (167 L H<sub>2</sub>O m<sup>-2</sup> yr<sup>-1</sup> or 70,140 m<sup>3</sup>  
334 yr<sup>-1</sup>) of rainfall annually in 2011, 2012, and 2013 respectively. These inputs were very small  
335 when compared with transpiration and evaporation losses of 2.5 – 3.0 million m<sup>3</sup> y<sup>-1</sup>. In 2012 and  
336 2013, precipitation comprised on average 0.8% (with a maximum of 4.1% in January 2013) of  
337 the whole system monthly water deficit, while from 2011 to 2013 precipitation comprised on  
338 average only 1.9% (with a maximum of 16.2% in December 2012) of the total water losses via  
339 transpiration and open water evaporation.

340 INSERT FIGURE 4 HERE

341 In many months our transpiration + evaporation-based water budget was remarkably  
342 consistent with the inflow-outflow deficits, particularly in 2013 (Figure 4). We found that  
343 transpiration + evaporation represented an average of 70% of the whole-system water deficit  
344 throughout the study period (67% if March-May 2012 are included; see Figure 4 legend for  
345 explanation). This consistency lends confidence in our scaled-up transpiration measurements and  
346 calculations of open water evaporation. Notably, these contributions of evaporation and  
347 transpiration are much higher than those reported for constructed wetlands in mesic, humid  
348 climates. For example, Meuleman et al. (2003) reported that transpiration and evaporation

349 comprised only 13% of the water budget for a constructed wetland in the Netherlands, where the  
350 climate is considerably cooler, more humid, and wetter than in Phoenix. Similarly, Kadlec et al.  
351 (2010) reported that transpiration and evaporation represented only 3% of total water outputs for  
352 a treatment wetland in Columbia, Missouri, USA and Favero et al. (2007) reported that these two  
353 water losses comprised only about 10% of the water budget for a constructed wetland near  
354 Venice, Italy.

355

### 356 *3.3. Water Quality and Nitrogen Budget*

357 We focused our analysis of nutrient sequestration on nitrogen (N), specifically dissolved  
358 inorganic nitrogen ( $\text{NO}_3^-$  and  $\text{NH}_4^+$ ), because this was the focus of the City's Clean Water Act  
359 permit for the treatment wetland. At the whole-system scale, inflow loads of both  $\text{NO}_3^-$  and  $\text{NH}_4^+$   
360 were nearly always higher than outflow loads (Figure 6a and 6b), with inflow loads averaging  
361  $532 \pm 84$  (SE)  $\text{kg d}^{-1}$  and  $169 \pm 19$  (SE)  $\text{kg N d}^{-1}$  and outflow loads averaging  $417 \pm 67$  (SE)  $\text{kg N}$   
362  $\text{d}^{-1}$  and  $87 \pm 17$  (SE)  $\text{kg d}^{-1}$  for  $\text{NO}_3^-$  and  $\text{NH}_4^+$ , respectively. The monthly whole-system N  
363 uptake rates averaged  $3.51 \pm 0.98$  (SE) ( $16.71 \pm 4.66 \text{ g N m}^{-2} \text{ mo}^{-1}$ ) and  $2.48 \pm 0.61$  (SE) ( $11.81$   
364  $\pm 2.90 \text{ g N m}^{-2} \text{ mo}^{-1}$ )  $\text{Mg N mo}^{-1}$  for  $\text{NO}_3^-$  and  $\text{NH}_4^+$ , respectively, from July 2011 through  
365 September 2013. On average, 22% of nitrate inputs and 48% of ammonium inputs were taken up  
366 between the whole-system inflow and outflow in spite of the short water residence time of the  
367 system and the likelihood that much of this water did not come into physical contact with the  
368 vegetated marsh that makes up 50% of the system by area.

369

INSERT FIGURE 5 HERE

370 Within the vegetated marsh, we were able to compare N concentrations along the three  
371 marsh transects but did not have water flux data to calculate actual nutrient removal rates. Still,  
372 we consistently found much lower concentrations of dissolved inorganic N in the near-shore  
373 water overlying the marsh, compared with concentrations at the marsh-open water interface. This  
374 pattern was most dramatic for  $\text{NO}_3^-$ , with near-shore concentrations that were often at or below  
375 the detection limit (Figure 6a). Marsh removal of  $\text{NH}_4^+$  was also consistent through time and  
376 across seasons, but was less dramatic than with  $\text{NO}_3^-$  (Figure 6b). Weller et al. (this issue)  
377 estimated N uptake rates by the plants themselves based on productivity and tissue stoichiometry  
378 and found that 19% of the whole-system N removal could be directly accounted for by plant

379 uptake. Even without actual nutrient flux estimates, it seems clear that the vegetated marsh was  
380 active and efficient in N removal, regardless of time of year.

381 INSERT FIGURE 6 HERE

### 382 3.4. A Plant-Mediated “Biological Tide”

383 The N concentration data from along our marsh transects (Figure 6) suggested that  
384 whole-system efficiency might easily be improved if more N-rich water could directly interact  
385 with the vegetated marsh. One possible mechanism for this is the lateral movement of water into  
386 the marsh to replace losses via plant transpiration, particularly in the summer when the plants are  
387 most productive, soil temperatures are highest, and transpiration is maximal. This lateral  
388 movement of water into the vegetated marsh is what we are calling a plant-mediated “biological  
389 tide.” We refer to it as a “tide” because it is an active horizontal advection from open water areas  
390 into the vegetated marsh that is analogous to astronomical tides seen in coastal wetlands. To our  
391 knowledge, this is the first time that such control of surface hydrology by plants has been  
392 documented in a wetland.

393 We investigated this “biological tide” by calculating the volume of water overlying the  
394 marsh that was lost daily to transpiration. Water depth measurements at the 50 transect points  
395 where aboveground plant biomass was measured showed that water depth was consistent across  
396 space and time, with a mean of 24 cm. We subtracted the portion of this water volume that was  
397 occupied by plant stems by scaling up stem culm diameter measurements, and used this  
398 corrected volume to estimate transpirational losses. During the summer months, as much as 15 –  
399 20% (approximately 8,000 to 11,000 m<sup>3</sup> H<sub>2</sub>O day<sup>-1</sup>) of the total volume of water overlying the  
400 marsh was transpired daily and must have been replaced by a lateral flow from the open-water  
401 areas to maintain the observed constant water depths (Figure 7). These values are conservative  
402 estimates because we were not able to account for the volume of water occupied by extensive  
403 thatched dead vegetation (i.e., wrack) on the marsh surface, thus suggesting that summertime  
404 water residence times in the marsh were likely less than 4-5 days.

405 INSERT FIGURE 7 HERE

406 These transpirative losses from the vegetated marsh, and lateral flow to replace them,  
407 were thus substantial. We argue that this plant-driven control of wetland hydrology, or  
408 “biological tide”, may be an important mechanism in arid systems to move additional nutrients  
409 into the marsh for uptake and processing (Figure 8). This “biological tide” is pulling water and

410 nutrients both laterally into the marsh and vertically down into the soils, creating more  
411 opportunities for plants to take up N and for soil microbes to process N and other bioactive  
412 solutes as Martin et al. (2003) and others have suggested. In fact, we argue that this “biological  
413 tide” phenomenon makes the Tres Rios treatment wetland more efficient at nutrient removal, and  
414 thus more effective at providing desired ecosystem services, than its counterparts in more mesic,  
415 humid climates. This phenomenon may actually be at work in many other treatment wetlands  
416 located in hot, dry climates. However, the high volume of water loss that drives the “biological  
417 tide” means that there is a trade-off between the provision of water-cleansing ecosystem services  
418 and the volume of water with improved water quality that can be exported from these systems for  
419 local reuse.

420 INSERT FIGURE 8 HERE

### 421 3.5. *Summary*

422 We found that water losses due to transpiration and evaporation were remarkably high  
423 compared with constructed wetlands in more humid, mesic climates. Total water losses via both  
424 processes peaked at over 300,000 m<sup>3</sup> month<sup>-1</sup> in the hot, dry summer months and averaged more  
425 than 70% of the whole-system water losses from June 2011 through September 2013. We found  
426 that large transpirative water losses actually appeared to enhance N removal efficacy relative to  
427 humid, mesic systems by drawing large volumes of replacement water and solutes into the marsh  
428 via a “biological tide”. This plant-mediated “tide” appears to be replacing more than 20% of the  
429 total volume of water overlying the marsh during hot, dry summer months. Transpiration-driven  
430 movement of shallow subsurface water has been documented in a number of wetlands, including  
431 into tree islands in the Okavango Delta in Botswana (Bauer-Gottwein et al. 2007, Ramberg and  
432 Wolski 2008) and in the Florida Everglades (Bazante et al. 2006, Troxler-Gann and Childers  
433 2006, Sullivan et al. 2012). But, to our knowledge, this is the first time that biotically-mediated  
434 surface hydrology has been demonstrated in any wetland.

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## FIGURE LEGENDS

Figure 1: The Tres Rios constructed treatment wetland cell used in this study. Blue arrows show the effluent inflow and outflow points and white lines are the approximate locations of the 10 marsh transects (each 50 – 60 m long). Water quality samples were collected at the inflow and outflow points and at 1) the northernmost transect closest to the inflow, 2) the southernmost transect nearest to the outflow, and the 3) eastern transect in between them. Total cell area = 42 ha; 21 ha of vegetated marsh and 21 ha of open water.

Figure 2: Total daily water loss via plant transpiration from the 21 ha of vegetated marsh. TYPHA=*Typha domingensis* and *T.latifolia*; SAC/STAB=*Schoenoplectus acutus* and *S. tabernaemontani*; SAM=*S. americanus*, and; SCAL=*S. californicus*.

Figure 3: Total live aboveground macrophyte biomass and total monthly transpiration water losses from the 21 ha marsh. Note that MT is equivalent to Mg.

Figure 4: Monthly total water losses due to open water evaporation and plant transpiration (stacked bars) compared with the inflow – outflow whole system water deficit. Note that the inflow sensor was not working properly prior to January 2012 or in March through May 2012.

Figure 5: Whole system nitrate (A) and ammonium (B) flux into the study cell (circles) and out of the study cell (squares).

Figure 6: Vegetated marsh concentrations of nitrate (A) and ammonium (B) flux near the shoreline (circles) and at the marsh-open water interface (squares) of the three water quality marsh transects.

Figure 7: Estimates of the volume of water overlying the 21 ha of vegetated marsh that was transpired daily by the plants and had to be replaced by the “biological tide”.

616 Figure 8: Conceptual schematic of our hypothesized “biological tide” in which high rates of plant  
617 transpiration, particularly during the hot, dry summer months, drive a lateral movement  
618 of water and solutes into the marsh and vertically down into marsh soils from the open-  
619 water areas. This biotically-mediated water flow may be replacing the entire volume of  
620 water overlying the marsh every 4 – 5 days at peak transpiration rates.  
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# FIGURES

623 Figure 1:

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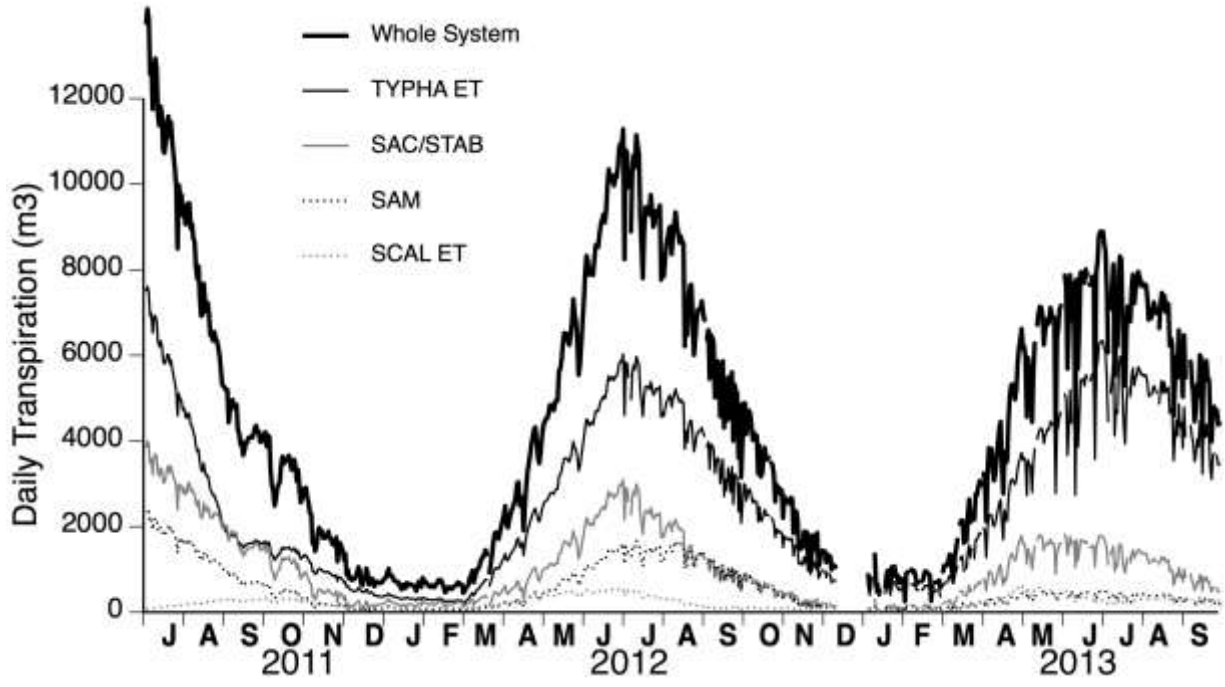
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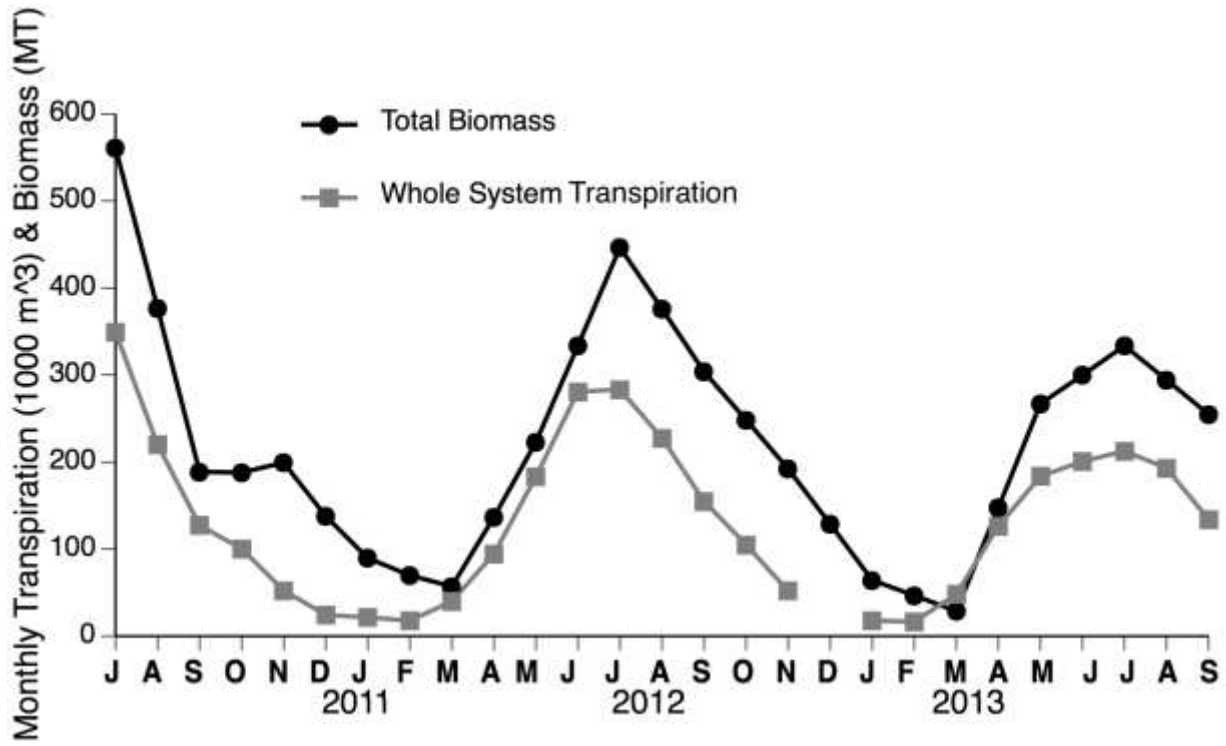
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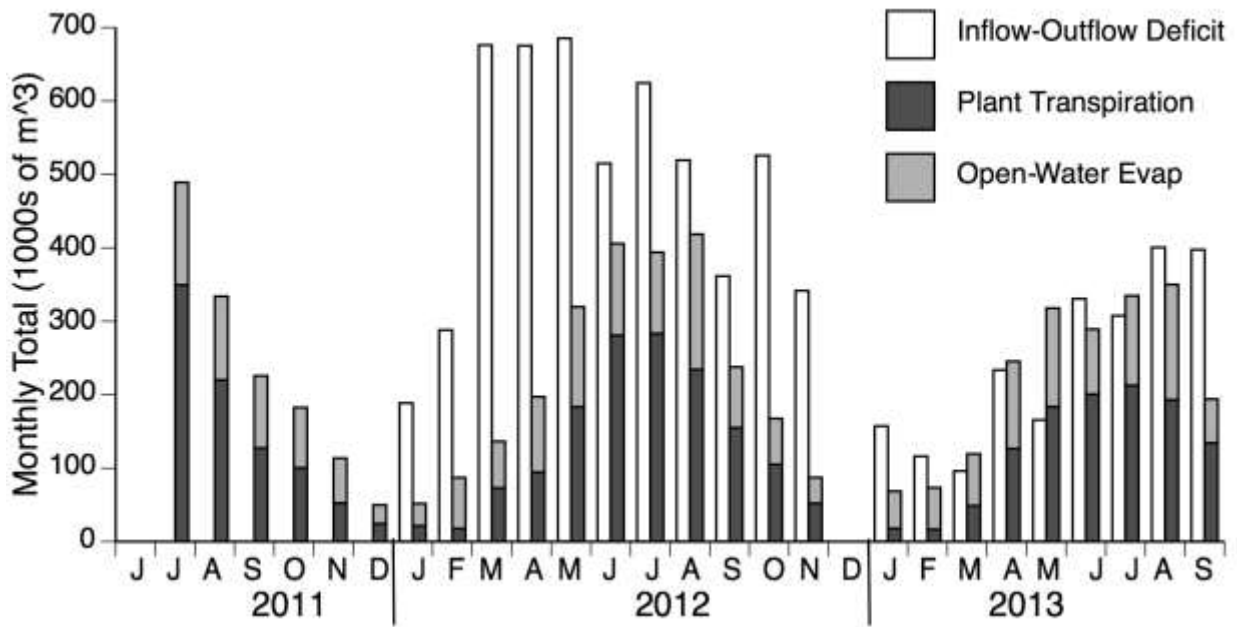


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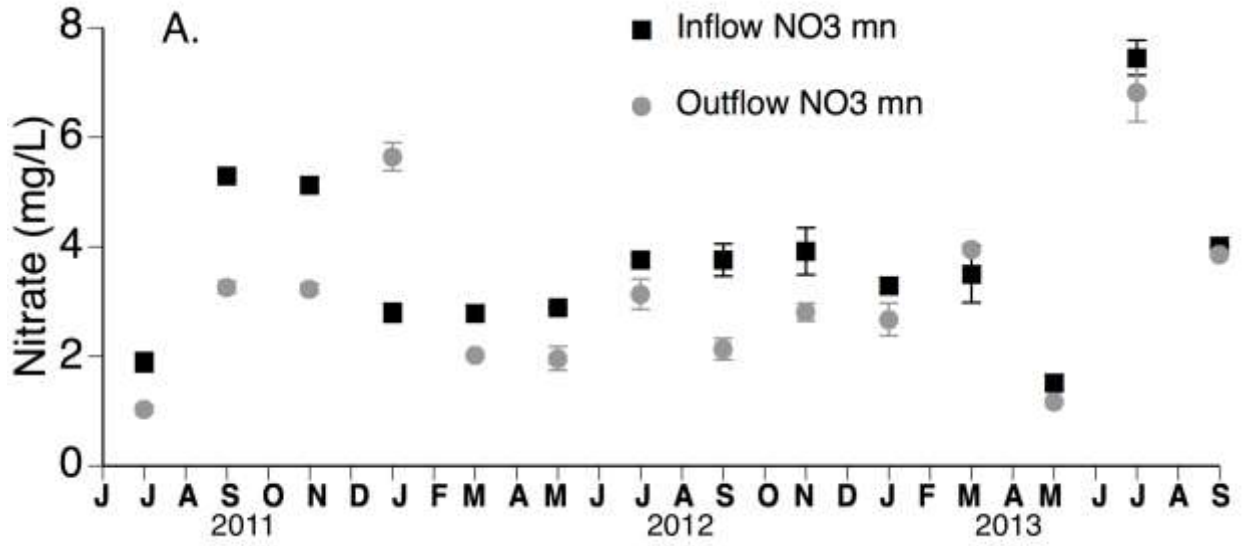
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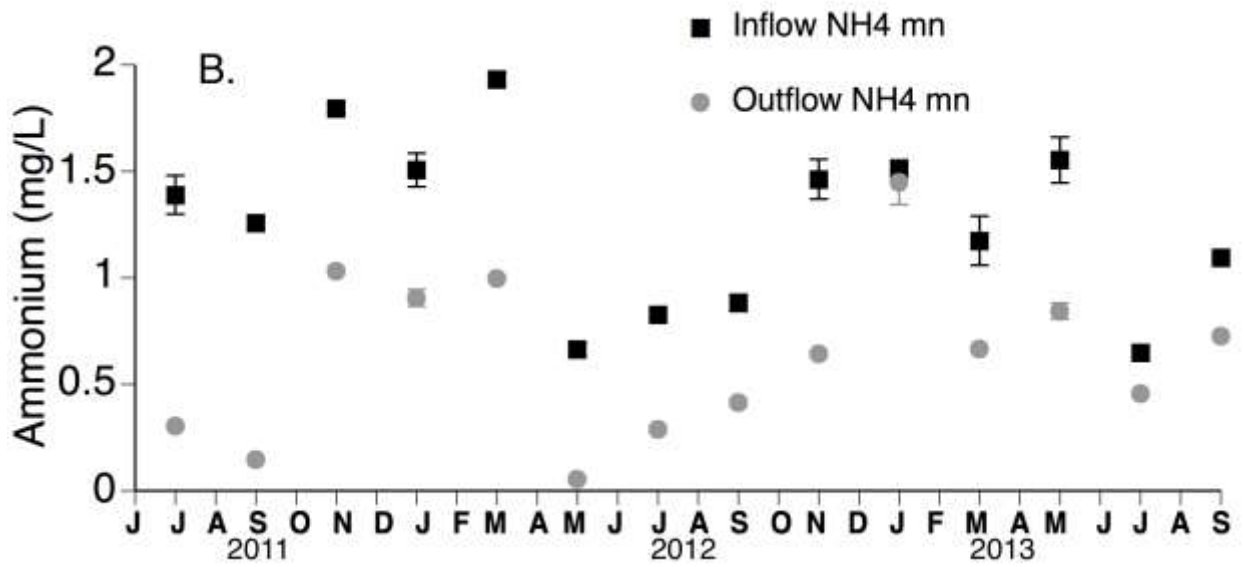


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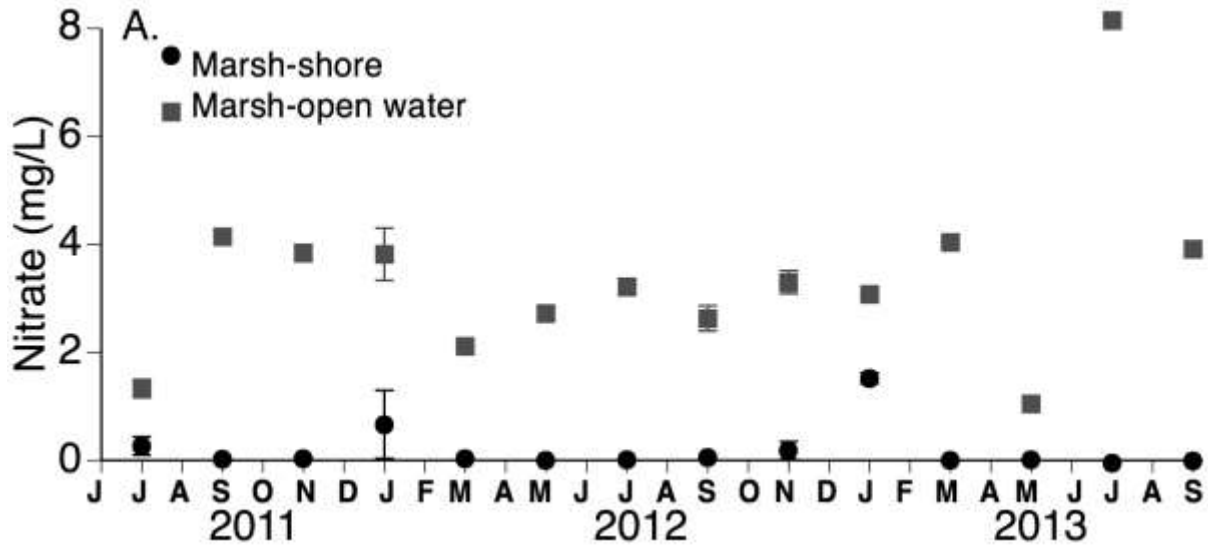


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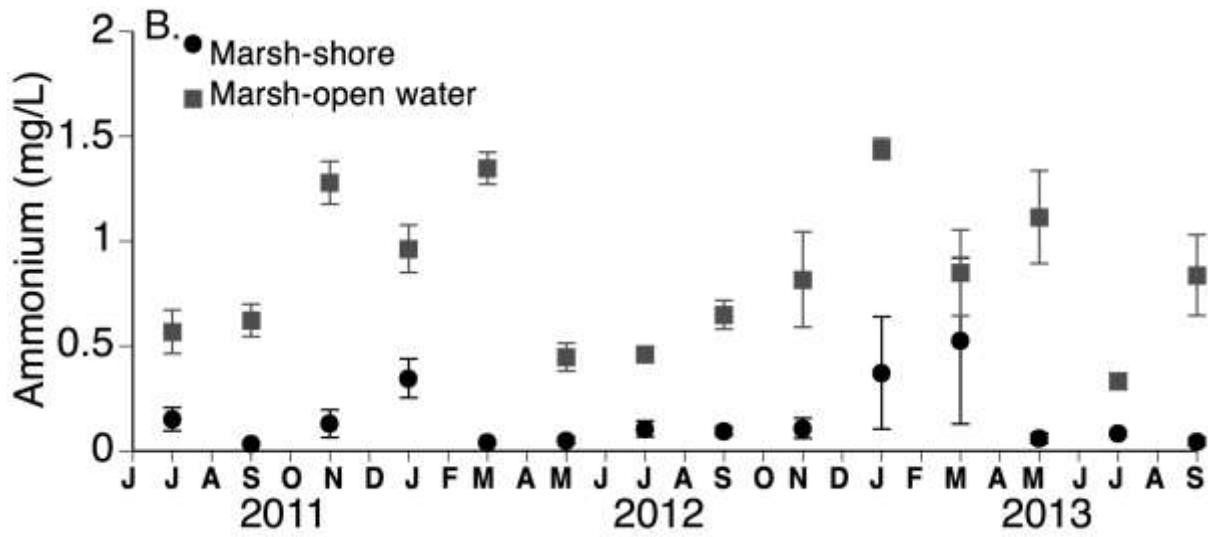


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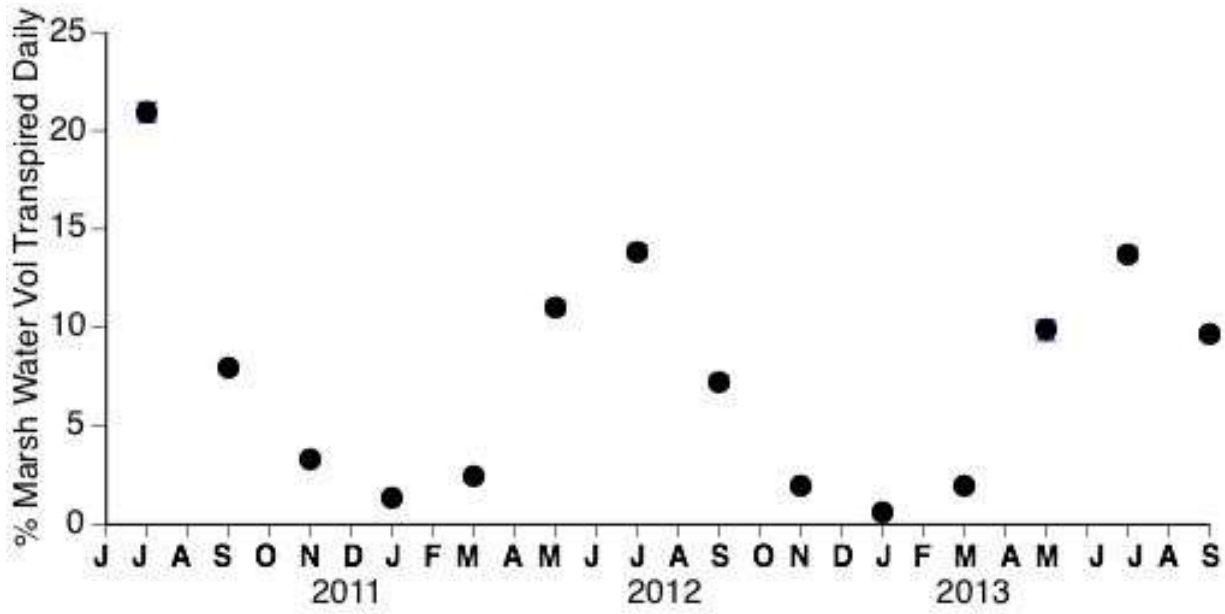


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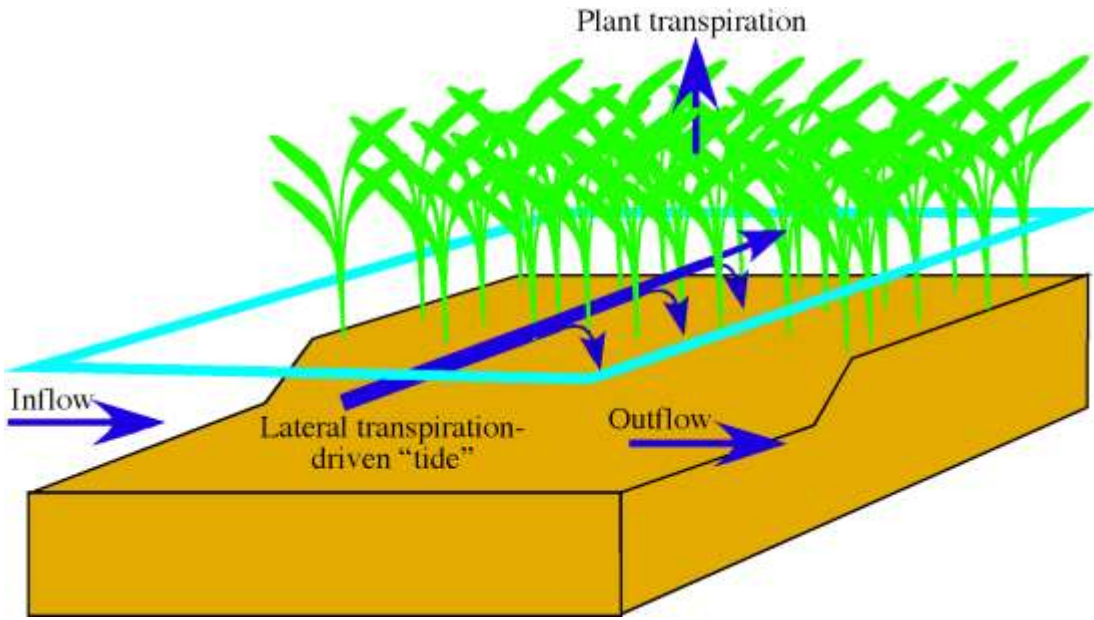
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## TABLE LEGENDS

672 Table 1: Summary of the parameters, applications, methods, and sampling frequencies for all  
673 variables used to estimate whole-system plant transpiration, open water evaporation, and  
674 water budgets.

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## TABLES

Table 1:

<b>Parameter and Application</b>	<b>Methods</b>	<b>Data Frequency</b>
Transpiration, leaf area, leaf-level RH, leaf-level PAR, leaf-level T; used to estimate whole system plant transpiration	Infrared Gas Analyzer (IRGA)	Bi-monthly
Ambient RH, ambient PAR, ambient T, wind speed, vapor pressure deficit, R; used to calculate open water evaporation and to scale plant transpiration in time	On-site meteorological station (City of Phoenix)	Hourly
Surface water inflow, surface water outflow; used to calculate water budget deficit	Acoustic Doppler flow gauges (City of Phoenix)	Daily
Aboveground plant biomass for 4 species groups; used to scale plant transpiration in space	Non-destructive phenometric technique (Weller et al. this issue)	Bi-monthly

681