WATER USE, EFFICIENCY, AND STOMATAL SENSITIVITY IN EASTERN COTTONWOOD AND HYBRID POPLAR VARIETALS ON CONTRASTING SITES IN THE SOUTHEASTERN UNITED STATES

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EXTENDED ABSTRACT

The Southeastern United States has widescale potential to achieve high productivity from elite eastern cottonwood (*Populus deltoides* W. Bartram ex Marshall) and hybrid poplar varietals to produce renewable bioenergy and bioproducts (U.S. Department of Energy 2016). In order to determine their impact to groundwater supplies and streamflow and maintain growth during drought periods, varietals that use water efficiently and/or tolerate water stress conditions are needed to make planting recommendations across a variety of sites (Petzold and others 2011, Zalesny and others 2016). Poplars have been shown to exhibit a range of water use strategies with some exhibiting a conservative (isohydric) strategy in which they restrict water use and, in turn, productivity under water stress conditions and others exhibiting a more risky (anisohydric) strategy in which they continue to function under increasing water stress (Attia and others 2015, Navarro and others 2020). In addition, inoculation with nitrogen-fixing endophytic bacteria may improve water stress tolerance (Rho and others 2018) by increasing root biomass and reducing damage from reactive oxygen species (Khan and others 2016, Rogers and others 2012). This research sought to address the following objectives: (1) Determine water use strategies for six *Populus* varietals (three eastern cottonwood and three hybrid poplars) planted on the Upper Gulf Coastal Plain (Coastal Plain) and the Lower Mississippi Alluvial Valley (LMAV) and determine if strategies differ depending on planting site, (2) Determine if inoculation with diazotrophic endophyte bacteria impacts growth and water use strategies in these tested varietals and if site conditions impact the endophyte effect, and (3) Determine two-year aboveground biomass production for all varietals at both sites and identify physiological parameters that are most correlated with growth.

Tested varietals consisted of three eastern cottonwood varietals (*P. deltoides* × *P. deltoides*, “D×D”) including 110412, S7C8, ST66, and three hybrid poplar varietals, two of which were *P. deltoides* × *P. maximowiczii* (A. Henry) crosses (“D×M”; 6329 and 8019) and one *P. trichocarpa* (Torr. & Gray ex Hook.) × *P. deltoides* cross (“T×D”; 5077) supplied by GreenWood Resources Inc. (Portland, OR, USA). Individuals were planted as cuttings in 15-tree blocks at a 6 by 9 feet spacing in 2018 on a Gulf Coastal Plain site in northeastern Mississippi and a site in the Mississippi Delta portion of the LMAV. The LMAV site was

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Citation for proceedings: Willis, John L.; Self, Andrew B.; Siegert, Courtney M., eds. 2022. Proceedings of the 21st Biennial Southern Silvicultural Research Conference. Gen. Tech. Rep. SRS-268. Asheville, NC: U.S. Department of Agriculture Forest Service, Southern Research Station. 262 p. https://doi.org/10.2737/SRS-GTR-268.

more poorly drained and had a history of row crop production compared with the Coastal Plain site. Half of the blocks were inoculated with diazotrophic endophyte bacteria from Intrinsyx Bio (Moffett Federal Airfield, CA, USA). In the second growing season, Granier heat dissipation sapflow sensors (Granier 1987) were installed into two or three trees per varietal and endophyte treatment. Half-hourly sapflow rates (J_S) were combined with environmental data, tree-level sapwood and leaf areas (see below) to calculate canopy stomatal conductance (canopy G_S) as follows:

$$Canopy\ G_S = \frac{K_G \times \frac{J_S \times A_S}{A_L}}{VPD} \tag{1}$$

where

K_G is a conductance coefficient, A_S is tree sapwood area, A_L is tree leaf area, and VPD is atmospheric vapor pressure deficit.

Canopy G_S was plotted vs. the natural log of VPD at both low and average to high soil moisture conditions (θ) to determine reference stomatal conductance (G_{Sref} at VPD = 1 kPa), stomatal sensitivity (m; slope of the GS vs. VPD relationship) and scaled stomatal sensitivity (m/G_{Sref}) (Oren and others 1999). At the end of the second growing season but before leaf fall, trees were harvested to estimate sapwood area, leaf area and total dry woody biomass. Biomass at the end of the first growing season was estimated from tree heights and diameters. Whole-tree water use efficiency (WUE) was calculated as the ratio of dry biomass accumulated during the second growing season and water used during that time period for each individual.

We found that, at the Coastal Plain site, varietal 8019 (D×M) used more water during the early portion of the growing season with water use declining after June while other varietals, including S7C8 (D×D), ST66 (D×D), and 6329 (D×M) increased water use during the growing season using the most water in August or used similar amounts of water for most months of the growing season (fig. 1A). At the LMAV site, S7C8 (D×D) and 5077 (T×D) used the most water in June and July, while other varietals either maintained or increased water use throughout the growing season (fig. 1B). Biomass was significantly higher across varietals at the Coastal Plain site compared with the LMAV site. At the Coastal Plain site, biomass was greatest in 8019 (D×M) and S7C8 (D×D) and greatest for all eastern cottonwoods at the LMAV site (fig. 1C, D). We found that whole-tree WUE was similar across varietals at 5.2 g biomass per kg water used and that water use scaled positively with tree size. In terms of the relationship between canopy G_S and lnVPD, we found that water use strategies in terms of scaled stomatal sensitivity converged across varietals under stressful soil water conditions (low soil moisture at the Coastal Plain site, high soil moisture at the LMAV site), but that varietal 8019 (D×M) and 110412 (D×D) tended to exhibit the highest plasticity in stomatal sensitivity under different soil moisture conditions. We found that inoculation with endophytes caused significant impacts at the more nitrogen limited, LMAV site leading to increased biomass production, whole-tree WUE, and scaled G_S sensitivity under low soil moistures. Across sites and varietals, tree leaf area and whole-tree WUE were positively correlated with woody biomass production while plasticity in scaled G_S sensitivity was important with lower sensitivity under high soil moisture as well as higher sensitivity under low soil moisture being positively correlated with biomass. Overall, these findings can be used to model hydrologic impacts of large-scale *Populus* biofuel production as well as to recommend varietals with efficient water use strategies. More information and results from this study can be found in Renninger and others (2021).

Table 1— Analysis of variance results presenting significant effects of site (S), varietal (V) and endophyte inoculation (E), as well as their interaction (×) on productivity, water use, and canopy stomatal conductance (G_S) parameters under low and average to high soil moisture (θ)

Parameter	Site	Varietal	Endophyte	S×V	S×E	V×E	S×V×E
Biomass	**	**	–	–	*	–	–
Specific gravity	*	***	**	–	*	–	–
Leaf area	–	***	–	–	*	–	–
Seasonal water use	**	***	*	*	*	–	–
Whole-tree WUE	–	–	–	–	–	–	–
G_{Sref}, low θ	–	***	**	*	–	–	*
G_S sensitivity, low θ	–	***	–	**	–	–	–
Scaled G_S sensitivity, low θ	–	*	***	**	***	**	–
G_{Sref}, high θ	–	**	–	**	–	–	–
G_S sensitivity, high θ	–	*	–	**	–	–	–
Scaled G_S sensitivity, high θ	–	**	*	**	–	–	–

*= p<0.05; ** = p<0.01; *** = p<0.001; WUE = water use efficiency; G_{Sref} = reference canopy conductance; – = not significant.

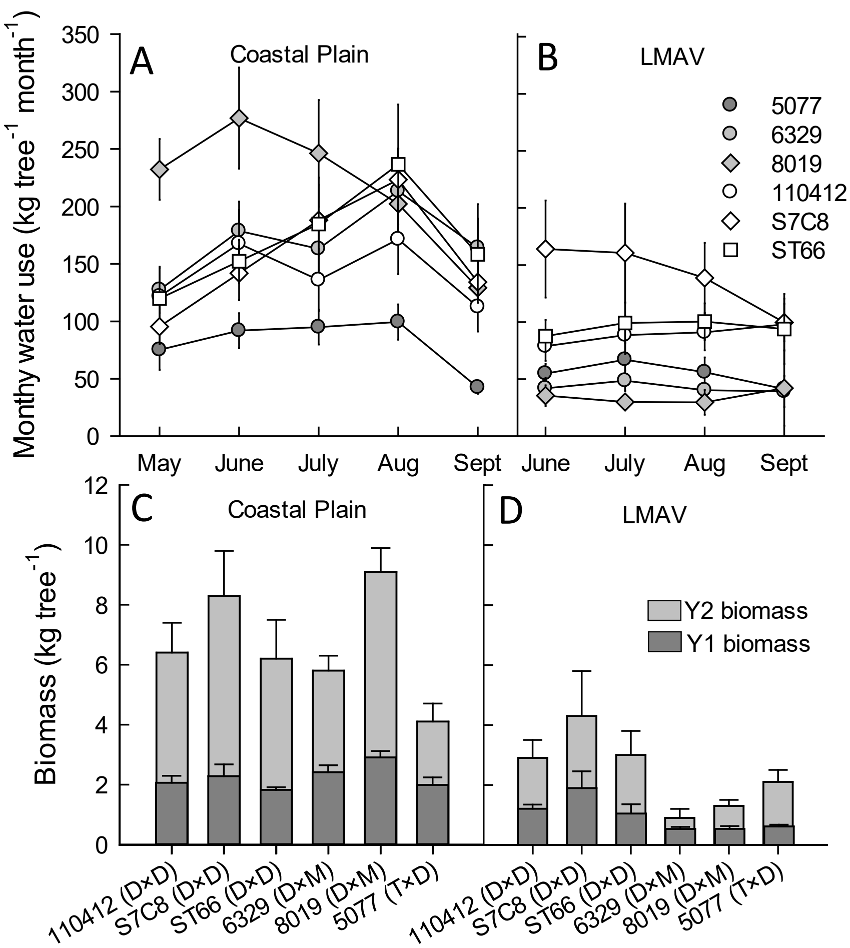

Figure 1—Total monthly water use (kg water tree^{-1} month^{-1}) in the second growing season at (A) the Coastal Plain site and (B) the LMAV site for eastern cottonwoods including varietals 110412 (white circles), S7C8 (white diamonds) and ST66 (white squares) and hybrid poplars in the *P. deltoides* × *P. maximowiczii* taxa including 6329 (light gray circles) and 8019 (light gray diamonds) and the *P. trichocarpa* × *P. deltoides* taxa including varietal 5077 (dark gray circles) and biomass (kg tree^{-1}) for the first growing season (Y1; dark gray) and second growing season (Y2: light gray) at the (C) Coastal Plain, and (D) LMAV sites.

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