

Flooding Treatment Effects on Sap Flow of Deciduous Trees in the Chesapeake Bay

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ABSTRACT

Sea level rise and changes in precipitation regimes due to global warming have impacted coastal forest ecophysiology over past decades, leading to widespread tree mortality and ‘ghost forests’ – areas where large swaths of forest are converted to marsh habitat. However, it is difficult to predict rapid changes observed in coastal areas, as most models independently incorporate terrestrial and oceanic habitats. Sap flow, the upwards movement of water through trees, is a useful indicator of water use and vitality. Using data from a large-scale manipulative field experiment, we examined the rates of sap flow in three deciduous tree species in the Chesapeake Bay region- red maple, tulip poplar, and American beech- subjected to annual floods of fresh or brackish water. These species are expected to exhibit variation in flood and salinity tolerance. Sap flow was measured using a thermal dissipation heat probe system, and ANOVA and t-test analyses were used to determine effects of flood treatments, species, and abiotic drivers on sap flux (sap flow over an area). Preliminary results show that the sap flux of tulip poplars is disproportionately and negatively affected by saltwater treatment. These results aid us in accurately predicting the impacts of climate change on coastal ecosystems.

Introduction

Global warming has led to coastal sea level rise and altered precipitation regimes over the past several decades. The stress of these impacts has negative implications for the overall health of coastal upland forests (Hopple, 2023), resulting in rising rates of ‘ghost forest’ formation; i.e., large-scale woody plant mortality over the course of a transition from upland to wetland forest (McDowell, 2022) (Figure 1). However, understanding and predicting such state changes remains a significant challenge, and little is known about what potential indicators might be used to assess ecosystem risk and resilience.

Following intense flooding events, trees may exhibit a variety of unseen responses despite appearing healthy. For instance, the saturation of soil with fresh or saltwater leads to hypoxic soil conditions, resulting in root mortality and reduced water uptake (McDowell, 2022). The complex, cascading

interactions between abiotic drivers and these physiological changes can lead to hydraulic failure and carbon starvation, and eventually mortality. Thus, a thorough understanding of the impacts of such events is necessary to assess exactly how and to what extent trees are affected by freshwater and saltwater flooding.

One crucial process in trees is sap flow, the upwards movement of water through a tree’s xylem, or water-conducting tissue (Bovard, 2005). As the xylem is the primary conduit of water use from the soil to the canopy, where photosynthesis occurs, it is a useful indicator of tree vitality. Soil salinity increases as a result of saltwater inundation, which will increase soil water potential relative to that of the canopy and thus weaken the water potential gradient, and the rate of sap flow, of the tree. Unusually low sap flow velocity is therefore an early sign of stress in trees in the process of conversion to ghost forest state (Teobaldelli, 2004).

However, there is limited available data on the effects of extreme events on coastal

areas, as these changes are often rapid and unpredictable. Additionally, few studies incorporate interactions in coastal regions as opposed to solely terrestrial or oceanic areas (Hopple, 2023). To better understand the impacts of sea level rise and increased inundation frequency on coastal forests, the Terrestrial Ecological Manipulation to Probe the Effects of Storm Treatments (TEMPEST) experiment simulates extreme, disruptive, hydrologic events in a deciduous coastal forest in Maryland, U.S.A. Three tree species- *Liriodendron tulipifera* (tulip poplar), *Acer rubrum* (red maple) and *Fagus grandifolia* (American beech)- are inundated annually with fresh or brackish water, and an extensive sensor network monitors a variety of tree and soil responses throughout the day.

Methods

TEMPEST Experimental Design

The TEMPEST study site consists of three 50 m x 40 m experimental plots—freshwa-

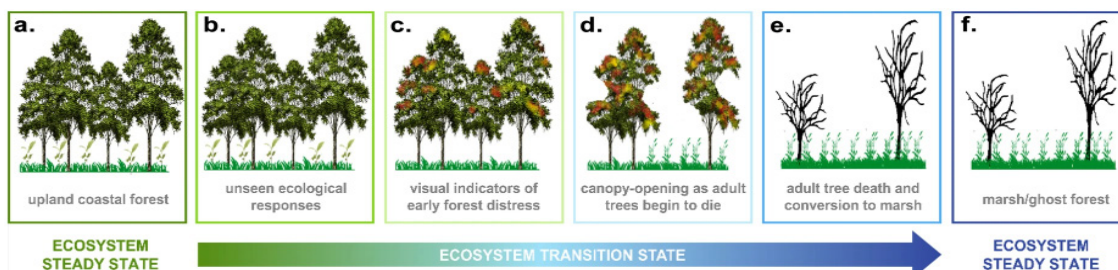


Figure 1: Stages in ghost forest formation, from healthy upland coastal forest to marshland (Hopple, 2023).

ter, estuarine, and control—in a mid-successional temperate deciduous forest on the western shore of the Chesapeake Bay, Maryland, U.S.A (38.876°N, 76.553°W). Each TEMPEST treatment uniformly applies 300 m³ over a period of 5 hours of freshwater or saltwater in a single day and approximates a 15 cm rainfall event, comparable to a 10-year storm or hurricane for the region.

Sap Flow Measurements

Sap flow velocity was measured using a thermal heat probe dissipation system containing two 2.5 cm long probes with a 1 cm effective sensing length (Plant Sensors, 1994; Granier, 1985). Both probes were inserted perpendicular to the tree, one approximately 1.3 m above the ground on the north-facing side of the tree, and the second approximately 10 cm below the first (Hopple et al., 2023). The upper probe, downstream of xylem sap flow, was heated at a constant power of 0.2 W while the unheated lower probe acted as a reference for temperature difference measurements due to heat dissipation (Lu, 2004). The probe systems were covered with reflective insulation to ensure isolation of the system. Approximately 18-20 trees (at least 6 of each focal species) in each treatment plot were fitted with sap flow sensors, although the exact number of active sensors varied due to technical difficulties or tree death. Raw data was outputted as a difference in voltage between the two probes, expressed every 15 minutes; this can be converted to a difference in temperature (°C) following Granier, 1985: $F = 118.99 \cdot 10^{-6} (T_{max} - T) 1.231$ (Equation 1)

In which T = the difference in temperature between the heated and reference probe, T_{max}= the maximum difference in temperature (i.e. no sap flow) in a 24-hour period, and F = sap flow velocity in m/s. The constants $\alpha = 118.99 \cdot 10^{-6}$ and $\beta = 1.231$ are derived

Year	TEMPEST Event Date	Pre-Flood Dates	Post-Flood Dates
2021	N/A	June 6 - June 12	June 13 - June 19
2022	June 22	June 15 - June 21	June 23 - June 29
2023	June 6, June 7	May 30 - June 5	June 8 - June 14

Table 1: TEMPEST flood event dates and the “pre-flood” and “post-flood” periods used in this analysis. Each consecutive year of TEMPEST supplies an additional day of flooding (i.e. TEMPEST 1 in 2022 had one day of flooding, TEMPEST 2 in 2023 had two days of flooding, etc.). Note that there was no flooding event in 2021, so two weeks were chosen from the middle of June 2021 to serve as a control for June sap flux.



Figure 2: TEMPEST experimental design; (a) aerial view of the three TEMPEST plots (b) forest layout and experimental infrastructure (Hopple, 2023)

from the original Granier probe calibration and depend on the magnitude of constant power (0.2 W) supplied to the probe.

It was necessary to account for the differences in area of sapwood, or actively conducting xylem, between species and individual trees. Thus, sap flow velocity measurements were scaled via the following allometric equations (Brantley, 2016):

$$A_s = a \cdot D^b \text{ (Equation 2)}$$

For which A_s = estimated sapwood area (m²), D = diameter of tree at breast height (m), and a and b are species-specific coefficients calculated from Brantley et al. (Table 2). Diameter at breast height for all trees has been measured annually in cm using dendrometer bands at 1.4 m above ground.

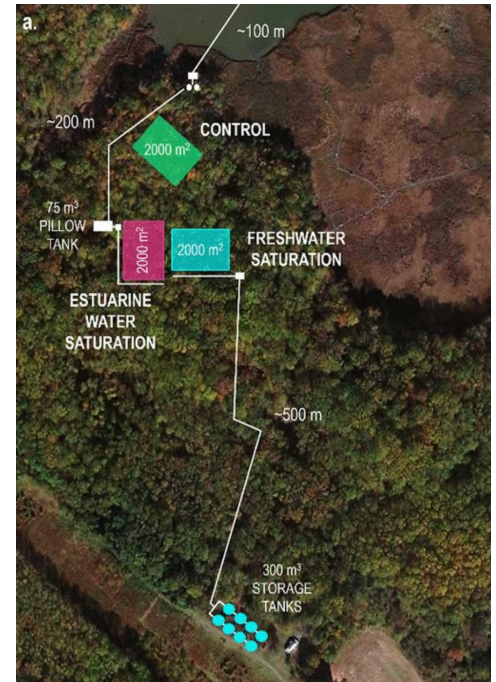
Combining both our point measurement of sap flow velocity and our estimation of sapwood area, sap flux density (the rate of sap flow over the entire volume of sapwood) can be obtained via the following equation (Lu, 2000):

$$F_d = A_s \cdot F \text{ (Equation 3)}$$

In which F_d = sap flux density (m³/s).

Computational Methods

The sap flow data used for this analysis



was recorded at 15-minute intervals from January 1, 2021 to May 2, 2024. Soil volumetric water content was measured over the same time frame at 15 cm via 46 TERS-12 moisture sensors distributed evenly across each treatment plot. Photosynthetically active radiation data was obtained for April 19, 2022 to May 2, 2024 from the Global Change Research Wetland (GCRew) synoptic site, which overlaps with the TEMPEST freshwater experimental plot.

After raw sap flow data was converted to sap flux density, values were filtered to identify the times of day at which peak sap flux was occurring. This was determined to be from 11 am to 12 pm, and all subsequent analysis was conducted using measurements from this time frame. Sap flux density data was further filtered for outlier values (< 2e-06 m³/s) for all subsequent analysis. Furthermore, examinations of abiotic drivers were conducted for the weeks preceding and following TEMPEST flood events (Table 2). Linear regression was performed to create the graphs of sap flux density plotted against soil volumetric water content and photosynthetically active radiation as well as calculated

Species	a (SE)	b (SE)	R ²
ACRU	0.5973 (0.0414)	2.0743 (0.0375)	0.999
LITU	0.8086 (0.2110)	1.8331 (0.1332)	0.991
FAGR	0.8198 (0.0702)	1.8635 (0.0474)	0.987

Table 2: Species-specific coefficients for allometric equations as calculated by Brantley, 2016. ACRU = *Acer Rubrum* (Red maple), LITU = *Liriodendron tulipifera* (tulip poplar), FAGR = *Fagus grandifolia* (American Beech). Note that Brantley et al. did not calculate coefficients for FAGR specifically, so estimates for all trees with diffuse-porous xylem were used instead (Phillips, 1996).

their R-squared and p-values to determine the strength and sign of their linear relationships (Whickham, 2024). Lastly, an Analysis of Variance (ANOVA) test for difference in means was conducted to determine if there were statistically significant variability in the sap flux density of different treatments and species, as well as a statistically significant association between treatment and species (to confirm and account for the fact that each species will respond differently to a given treatment effect).

Results

Sap flux density displays a clear seasonal signal, with peaks during the growing season from approximately April through September as the tree actively engages in photosynthesis and water transport. Additionally, clear differences are seen in the total sap flux density of different species, with tulip poplar generally having the highest relative sap flux density (average in control plot during growing season for 2021-2023 is $8.15 \cdot 10^{-7}$ m³/s), followed by American beech ($4.91 \cdot 10^{-7}$ m³/s), then red maple ($8.38 \cdot 10^{-8}$ m³/s). Treatment effects in comparison to the control plot display less stark variation, with comparable sap flux density values in the freshwater plot and slightly lower values in the saltwater plot (Figure 3).

The linear correlation between midday sap flux and photosynthetically active radiation (PAR) as a potential environmental driver during the weeks immediately preceding and following TEMPEST flood events was also examined. A linear regression model examining the effects of PAR displayed a statistically significant relationship

with sap flux density for all species (p values $\ll 0.05$). However, the model does not display a strong linear fit to the data; red maple has the strongest positive fit with an R² value of 0.15.

Discussion

These results show that flooding has noticeable negative effects on tree sap flow across species. These effects differ across treatment plots, with saltwater providing the greatest repression of sap flow. When examining differences between species, tulip

poplar consistently has the highest sap flux density and red maple consistently has the lowest sap flux density, with American beech averaging values in between the two. This is likely due to differences in sapwood area; tulip poplar is the largest by diameter and sapwood area of the three species (Hopple et al., 2023) and thus has a greater area over which sap flow can occur.

Freshwater flooding does not seem to have a negative effect on sap flux density and seems to in fact increase the overall rate of sap flow (Figure 1). This is likely due to the time frame of these data; were sap flux isolated for the weeks during or following TEMPEST flood events, sap flow might have displayed an overall decline. Furthermore, depending on the physiology of the tree and predisposition to flooding effects such as xylem embolism and crown loss, flooding effects may take several years to outwardly manifest (Johnson, 2011). Analysis over the next several TEMPEST flood events would be expected to reveal a decrease in sap flux density, though less extreme than that of saltwater, emphasizing the importance of such multi-year, long term experiments for understanding the slow effects of global climate change (Norby, 2010). Species-specific physiological differences between tulip poplar, red maple, and American beech may account for their varied responses to saltwater treatment.



Figure 3: Sap flux density from the TEMPEST experiment, averaged by plot and species every 15 minutes from January 2021 to May 2024 during peak sap flow hours (11 am - 12 pm local time). The vertical blue rectangles in 2021 represent the time frames of Hurricane Elsa and Hurricane Ida, respectively, two coastal storms that impacted the TEMPEST experimental site and could account for the lower sap flux values during those months.

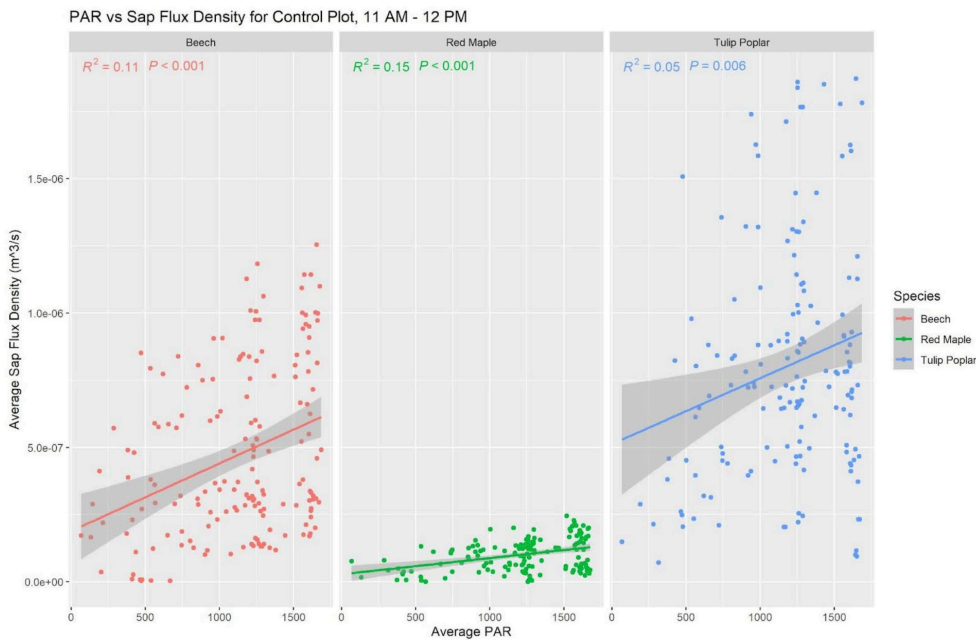


Figure 4: Sap flux density and photosynthetically active radiation isolated for the weeks before and after TEMPEST flood events from 2022-2023, including “pre-flood” and “post-flood” weeks in 2021. Values are averaged in 15-minute increments for peak sap flow hours (11 am - 12 pm). In an ANOVA test (Table 2), there were statistically significant differences in sap flux density within species, within treatments, and between different species in different treatments. The p values for both species and treatment effects were highly significant ($P < 0.001$). These results corroborate what can be visually observed in the box plot of sap flux density (Figure 5).

	df	Sum of Squares	Mean Squares	F value	P Value
Species	2	1.414e-10	7.068e-11	963.32	< 2e-16
Treatment	2	1.139e-11	5.690e-12	77.62	< 2e-16
Species : Treatment	4	6.360e-12	1.590e-12	21.68	< 2e-16
Residuals	2080	1.526e-10	7.000e-14		

Table 3: ANOVA test results for examining statistically significant differences in sap flux density between treatments and species for the weeks of pre and post TEMPEST flood events. Statistically significant differences between the interactions of these variables were also tested.

Tulip poplar is primarily an upland species and has shallow root depth and diameter in comparison to red maple and American beech (Kattge, 2020), which may lead to a greater reduction in water uptake in saline conditions. This could lead to more rapid xylem embolism, a primary driver of hydraulic failure (McDowell, 2022).

The TEMPEST experimental site is in a well-watered ecosystem not prone to drought (Hoppole et al., 2023), so soil volumetric water content is not a strong limiting factor for tree vitality. However, PAR is expected to have relatively strong controls on sap flow (Bovard, 2005), and may be limited

instead by the time frame of the observed data and a lack of controls for confounding variables. Linear regression was conducted solely for midday values, without filtering for days of low sunlight or unfavorable weather conditions. Were PAR to be measured over a larger, more variable timescale (i.e. more low and high values rather than centralized values), a stronger positive linear relationship would be expected. Future analysis will likely involve an assessment of these potential confounding variables, and a continuation of examining the abiotic factors and treatment effects influencing sap flow over the next several years of the TEMPEST experiment.

Data Availability

All analyses were conducted using R version 4.4.0 (R Core Team 2024). The complete analytical code is publicly available at <https://github.com/radha-vs/TEMPEST-SULI-2024>. All TEMPEST data, metadata, and further project information is publicly available at <https://github.com/COMPASS-DOE/TEMPEST>.

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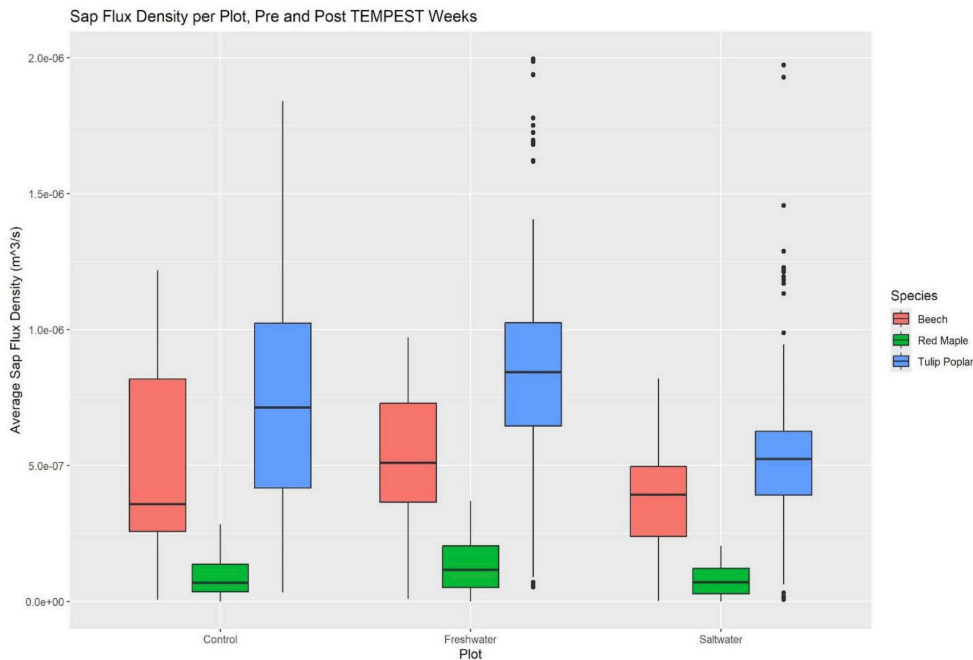


Figure 5: Boxplot of all sap flux density data from weeks immediately preceding and following TEMPEST flood events from 2022-2023. Sap flux density is averaged for midday (11 am - 12 pm) values and plotted against treatment plots. Species identity is represented by color.

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