

Ecosystem Services from New York Green Roofs: Differences in Water-Use Between Plant Species

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Abstract

Poor wastewater management has negative effects on local ecosystems, and human health around the world (Holeton et al. 2010). Green roofs can reduce storm-water runoff by converting impervious grey surfaces into hydrologically-active green surfaces. Most previous research on the plants grown on green roofs has focused on which species can survive the unique and often-hostile green roof environment, but the differential ability of plant species to contribute to storm water management may have major impacts on the benefits provided by roofs. First, I measured stomatal conductance and leaf temperature over the course of a day to capture how these respond to changing light and thermal conditions. These diurnal courses were measured on six days across three green roofs and in a greenhouse experiment. Second, I compared the water use of a suite of green roof plants grown in a greenhouse at different watering frequencies, and weighed on subsequent days to estimate water-loss from evapotranspiration. Species showed significant differences in stomatal conductance over the course of the day, suggesting species-specific differences in water-use. Leaf temperature was loosely associated with stomatal conductance, but was complicated by the effects of environmental variables. Like stomatal conductance in the diurnal course, soil moisture drawdown experiment also revealed significant differences in water-loss, at the whole plant scale. Drawdown data also revealed species-differences in water-use in response to drought-stress. These results should be used to inform future green infrastructure planting to improve urban wastewater management.

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Introduction

In 2014, 54% of the global population lived in urban centers (World Health Organization, 2014). As urban areas continue to grow, it will become increasingly important to consider the effects of urban infrastructure on local ecosystems, human health, and the global environment. Green infrastructure projects are generally designed to address the specific needs of an increasingly urban environment (Tzoulas et al. 2007). Green infrastructure can refer to traditional urban green spaces, such as parks and street trees, as well as include newer projects, such as green streets (green areas installed into unused space on city-streets), right-of-way bioswales (expanded tree pits at property line boundaries designed to both reduce and direct storm water runoff) and green roofs. Ecosystem services provided by green infrastructure include: improvements in wastewater management, mitigation of urban heat island effect, removal of air and water pollutants, and others (Orberndorfer 2007, Pataki et al. 2011). As policy-makers around the world begin to consider the utility of green infrastructure, a comprehensive understanding of the services provided by green infrastructure will be vital to effective design and implementation of green infrastructure projects (Agence France-Presse via Aljazeera America 2015, CNN 2008).

In New York City, the primary motivation for green infrastructure is wastewater management (NYC DEP, 2015). New York uses a combined sewer system: storm water runoff and sewage waste are collected in the same underground drainage network, and directed to a wastewater treatment facility before being discharged into a local body of water. During storm surges, the combined drainage system is not sufficient to hold the increased volume of water, and a combination of storm water runoff and sewage waste is

discharged into local bodies of water without treatment (NYC DEP, 2015). Once the wastewater has contaminated local watersheds, it can have negative impacts on local ecosystems and human health (Vymazal 2005, Holeton et al. 2011). Since planted surfaces can absorb and store more water than the impervious surfaces that cover most urban areas, green infrastructure can mitigate the effects of combined sewer overflow by reducing storm water runoff (Bliss et al. 2007).

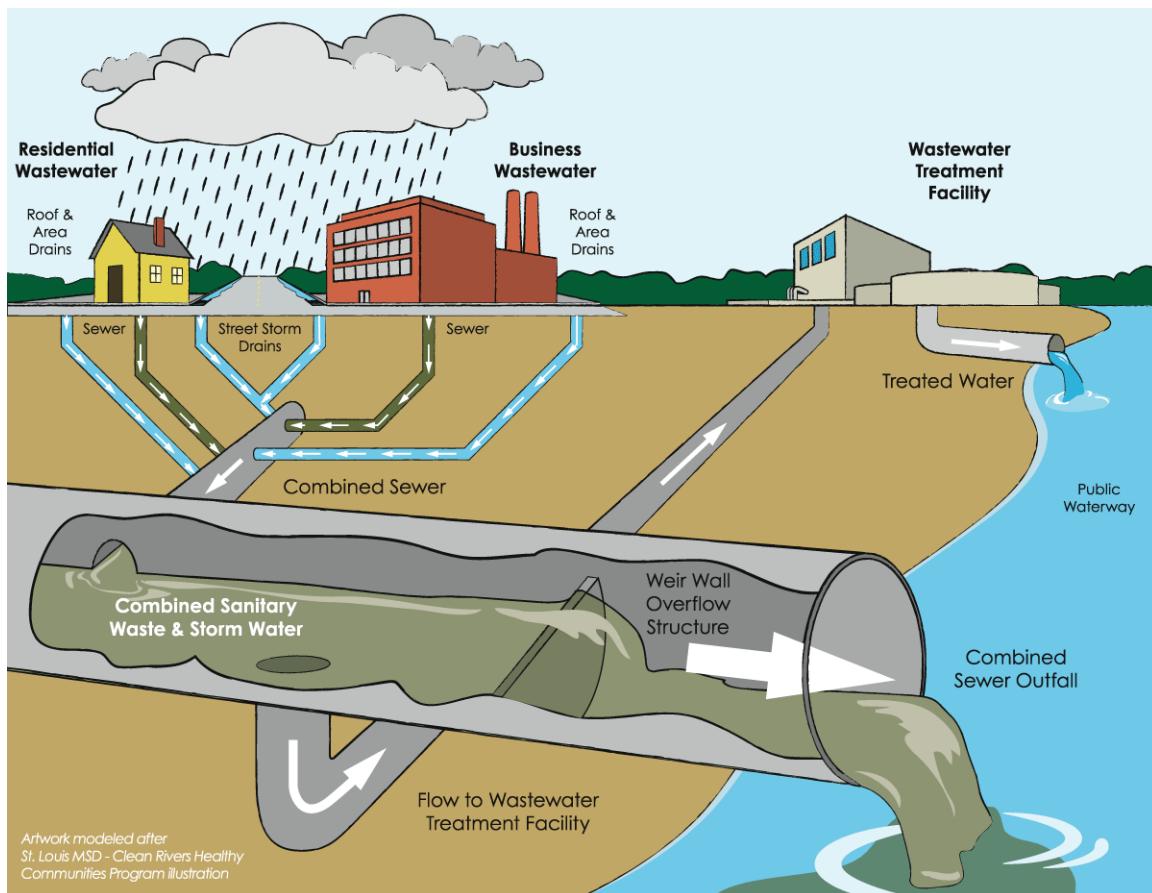


Fig. 1 Illustration showing dry weather and wet weather conditions in cities with combined sewer systems (Graphic courtesy of St. Louis MSD via Capital Region Water)

Green roofs reduce storm water runoff through moisture retention by growing media (soil designed for green roofs) and subsequent evapotranspiration by plants (Poe et al. 2015). Soil often has clay and organic particles that can become heavy when saturated with water, so growth media with low organic matter content and high porous mineral

content have been developed in order to absorb moisture and grow plants without crushing the roof (Bianchini and Hewage 2012). During a precipitation event, moisture is absorbed by the growth media, and held within the soil matrix, until it reaches field capacity, or the point at which the moisture exceeds the amount that can be held by the substrate under gravity (Stovin et al. 2012). When growth media reaches field capacity, water flows through the green roof media, into a drainage layer, where it is temporarily stored before becoming runoff (Stovin et al. 2012). Following a precipitation event, water tied up in the soil is evapotranspired by plants (Beretta et al. 2015).

The rate of evapotranspiration during dry periods influences the green roof's moisture-retention-capacity, and therefore the ability of the roof to mitigate runoff during the next precipitation event (Beretta et al. 2015). Evapotranspiration is the process by which plants take up water from the roots, to be returned to the atmosphere through the leaves. Pressure differences between the air outside the leaf and the water inside pull water from the roots to the leaf, such that the water is effectively evaporated through the stomata (Pereira et al. 1999). This process decreases soil moisture, such that by the time the following precipitation event occurs, the initial level of moisture held in the growth substrate is further from field capacity than it would be without evapotranspiration by plants (Beretta et al. 2015). Thus, the ability of plants to decrease soil moisture through transpiration determines the ability of the growth substrate to reduce runoff during precipitation events.

The green roof environment does not lend itself to supporting the plants that are most likely to help reduce storm water runoff. Urban rooftop environments are characterized by lack of shade, elevated temperatures, high wind speeds, and increased

drought conditions (Oberndorfer et al. 2007). Weight constraints on rooftops also limit the range possible green roof growing media types, as well as depth of media. In order to survive, species grown on green roofs must have relatively shallow rooting depths and be tolerant of high levels of UV radiation, elevated temperatures, drought, and green roof growth media (Orbendorfer et al. 2007, Bianchini and Hewage 2012). These traits are generally found in species that transpire less water, and therefore are likely to contribute less to storm water runoff mitigation (Nagase and Dunnett, 2012).

Most of the species planted on green roofs are selected for stress-tolerance, rather than ecosystem services (Van Mechelen et al. 2013). Chosen species are often drought tolerant, sometimes including species with photosynthetic pathways (C4, CAM, facultative CAM) specifically adapted to drought conditions (Orberndorfer, 2007). Although drought tolerance is an ideal trait for plant survival on the roofs, it can be counterproductive in terms of ecosystem services. Drought-tolerant plants generally conserve water through reduced transpiration, which leads to reduced moisture uptake by plant roots, and reduced contribution to storm water runoff mitigation (Schroeder et al. 2001). In order to provide storm water runoff management services, green roof plant species should transpire enough to regenerate water storage while still being stress-tolerant enough to survive.

Many studies have demonstrated the effects of plant biomass on moisture retention, but have not necessarily considered the effects of inter-species variation (Poe et al. 2015, Beretta et al. 2015, VanWoert et al. 2005). When species-effects have been examined, one popular approach is direct measurement of soil moisture and water-loss from different plant species, in a controlled environment (Nagase and Dunnett 2012,

Wolf and Lundholm 2008). Another approach involves measuring plants themselves. Direct measurement of plants has been attempted on green roofs (McIvor and Lundholm, 2011), but these studies do not necessarily provide comprehensive representation of species-differences in contribution to ecosystem services, unless they take into account changes in plant activity over the course of the day. Since diurnal (i.e. over the course of the day, as opposed to night) changes in ambient light and temperature have been found to determine plant activity (Tuzet et al. 2003, Whitehead 1997), spot measurements of plant activity in different species on green roofs do not provide a full picture of plant physiological effects on green roof ecosystem services. In light of diurnal changes in plant activity, a diurnal course experiments, where dynamic plant traits are measured at regular intervals throughout the day, was used by Gillner et al. 2015 to detect species-differences in water-use in studies on urban street-trees. For plants in the green roof environment, a diurnal course experiment is expected to reflect species-differences in water-use more comprehensively than spot measurements.

In this study, evapotranspiration was evaluated at the leaf-level using thermal imagery and stomatal conductance in a diurnal course. Stomatal conductance is a measurement of gas flux at the leaf's stomates, including both carbon flux in and water flux out (Farquhar and Sharkey, 1982). Since it measures water-flux out, stomatal conductance is reflective of the transpiration occurring through the leaf at a specific time (Damour et al. 2010). Thermal imagery estimates transpiration by measuring leaf temperature, and stands to increase efficiency of field measurements of transpiration, because taking thermal images is quicker than taking stomatal conductance measurements. Since transpiration is evaporative and therefore a cooling process, we

expect leaves transpiring more water to be cooler (Jones, 1999). Stomatal conductance is an established indicator of transpiration (Damour et al. 2010), but methods for measuring transpiration with thermal imagery is a less conventionalized. Here, thermal imaging methods of measuring transpiration are tested and evaluated against stomatal conductance: a negative relationship between leaf temperature and stomatal conductance is expected.

Water-use was also examined in a soil-moisture drawdown trial, where water-loss at the whole-plant scale was measured in plants of nine different species, at two different levels of drought-stress. Similar to studies measuring the effects of plant species on runoff and water-retention in growth media (Nagase and Dunnett 2012, Wolf and Lundholm 2008), this experiment investigated species differences in water-use, as well as drought-effects on water-use.

I examined traits indicative of water-use in plant species grown on green roofs in New York City. I used leaf-level and whole-plant scale measurements to detect species-differences in water-use, and investigate the methods by which species-differences may be identified, including thermal imagery and stomatal conductance in a diurnal course. I predicted measureable species-differences in water-use, which could then be used to determine optimal species composition with respect to storm water runoff mitigation.

Methods

Site Descriptions

Jackie Robinson Recreation Center Roof

The Jackie Robinson Recreation Center green roof has 12 planting boxes, each with a dimension of 4 m by 2 m (McGuire et al. 2013). Each box is divided into two plots, representing two ecosystem types: “Hempstead Plains” and “Rocky Summit,” both plant communities that support species tolerant of high UV radiation, high winds, limited water storage, and drought conditions (McGuire et al. 2013). The Hempstead Plains is a threatened prairie community native to Long Island, dominated by species in the Poaceae and Asteraceae families (Slater, 1987). The Rocky Summit is a grassland community occurring on the tops of mountains, ridges, and outcrops in Lower New England and the Hudson Highlands of New York State (Reschke, 1990).

Diana Center Green Roof

The Sybil Levy Golden '38 Ecological Learning Center, located on the 6th floor of the Diana Center in Barnard College, includes the Diana Center Green Roof. The vegetation here is also split into areas organized by “Hempstead Plains” and “Rocky Summit” ecosystem types, as on the Jackie Robinson Recreation Center roof.

Ranaqua Green Roof

Located in the southeastern corner of Bronx Park, Ranaqua is a three-story brick building and the headquarters of the Bronx Department of Parks and Recreation of the City of New York. The roof is split into four quadrants, each of which has an associated cistern for collecting and measuring runoff. Quadrants 1, 2, and 4 were all planted with the same species, but differ in growing media. Quadrant 3 is not vegetated. Recent data has shown that growing media did not significantly affect runoff, but that quadrants with

plant cover had significantly less runoff than the quadrant with no plant cover (Newman and Kativar, 2014; unpublished data).

Diurnal Course

For each diurnal course, thermal images and stomatal conductance data were taken on 6 individuals per species, every two hours. Diurnal courses took place twice per roof site and once in the Schermerhorn greenhouse. At each roof site, species were selected based on frequency across roofs and abundance within roofs (Table 1). Each diurnal course was scheduled to begin one hour before dawn and end in the mid-afternoon, but due to administrative constraints, early-morning measurements were not possible on all roofs (Table 1). At the greenhouse site, the same plants that were used for the soil-moisture drawdown experiment were used in the diurnal course (described under “Soil Moisture Drawdown,”). Greenhouse plants were measured 5:00-17:00 on September 21. For the duration of each diurnal course, A HOBO U30 Remote Monitoring System was used to record ambient temperature and Photosynthetically Active Radiation (PAR).

Table 1 Diurnal courses took place twice at each of three roof sites, but were not identical in duration. Species were selected based on frequency and abundance, but selection was not identical across roofs, or across diurnal courses.

	Diana Center		Ranaqua		Jackie Robinson	
	7/8/15	7/31/15	7/16/15	8/13/15	7/24/15	8/12/15
	5:00-16:00		9:00-15:00		9:00-15:00	
<i>Baptisia tinctoria</i>	1	1	0	0	0	0
<i>Digitaria ischaemum</i>	0	0	0	0	0	1
<i>Erigeron annuus</i>	0	0	0	0	1	0
<i>Lespedeza capitata</i>	0	0	0	0	1	1
<i>Monarda fistulosa</i>	0	0	1	1	0	0
<i>Oenothera biennis</i>	1	1	0	0	0	0
<i>Oxalis stricta</i>	0	0	1	0	0	0
<i>Panicum virgatum</i>	1	1	1	1	1	1
<i>Pycnanthemum tenuifolium</i>	0	1	1	1	1	1
<i>Schizachyrium scoparium</i>	1	1	1	1	1	1
<i>Setaria faberi</i>	0	0	0	0	1	1
<i>Solidago juncea</i>	0	0	1	1	0	0
<i>Solidago nemoralis</i>	0	0	1	1	0	0
<i>Solidago odora</i>	1	1	0	0	1	1
<i>Sorghastrum nutans</i>	0	0	0	0	0	1
<i>Symphotrichum laeve</i>	1	1	1	1	1	1
<i>Symphotrichum novae-angliae</i>	1	1	1	1	0	0

Stomatal conductance was measured using two Model SC-1 handheld Steady State Diffusion Porometers, by Decagon. To measure stomatal conductance, the leaf is clipped inside a sensor head, where gaseous water-flux from the underside of the leaf is measured by putting it in a series with two known humidity levels (Decagon Devices, 2011). For one of the porometers, the known humidity levels are determined by ambient temperature. For the other, known humidity levels are determined using a desiccant chamber—a compartment filled with Drierite, to dry out the surrounding air to a measured humidity level. All stomatal conductance is measured in reference to a known calibration, determined by taking initial porometer measurements on a wet piece of filter paper (Decagon Devices, 2011).

Thermal images were taken using a T60SC FLIR Thermal Camera. Reference thermal images were taken every 6 test-images, to control for the effect of ambient temperature and light (PAR) on leaves. For references images, filter paper was used to simulate heat conductance of a non-transpiring leaf. To find leaf temperature, images were converted to *.csv files using FLIR ExaminIR software, and then analyzed as Text Images in ImageJ Image Processing and Analysis software. In ImageJ, the free-hand tool was used to outline the relevant leaf, and the histogram tool was used to find mean temperature within the outlined area. The same procedure was used on filter paper for reference image temperatures.

Soil Moisture Drawdown

Plants of 9 different species were grown from plugs at Greenbelt Native Plant Center (3808 Victoria Blvd, Staten Island, NY 10314). The 9 species were: *Apocynum cannabinum*, *Andropogon gerardii*, *Carex pensylvanicum*, *Danthonia spicata*, *Ionactis linariifolia*, *Pycnanthemum virginiana*, *Solidago nemoralis*, *Sorghastrum nutans*, and *Schizachyrium scoparium*. Once grown to maturity, half the individuals of each species were watered once per week (“drought-stressed”), and the other half were watered twice per week (“well-watered”). After one month of living under differing watering schedules, the plants were transported to the greenhouse on the top floor of Schermerhorn Hall in Columbia College, where all plants underwent a soil moisture drawdown trial: plants were watered to saturation and subsequently weighed regularly to determine water-loss. Pots were weighed on Days 1, 2, 5, 6, 7, 9, 11, 12, 13, 14, 16, and 20. Differences

in water-loss were detected by comparing plant pot weights proportional to their weight on Day 1 of the drawdown experiment.

Biomass data was taken on the greenhouse plants. At the end of the drawdown, all plants were uprooted and dried in a Fisher Scientific Isotemp drying oven set to 50°C, to be weighed for dry aboveground biomass. Biomass data was used to determine how much of the water-loss from the drawdown was due to specific plant physiology, and how much was due to plant size.

Statistical Analysis

All statistical analysis was done using R 3.2.2 GUI 1.66 Mavericks build (6996).

Generalized Additive Modeling (GAM) was used to build best-fit curves for diurnal stomatal conductance of each species on each site (Fig 2, S1-6). GAM was used over Generalized Linear Modeling (GLM) because we do not expect diurnal stomatal conductance to follow a linear trend, and we do not expect variation in conductance to follow the statistical distributions necessary for GLM. GAM forms a smoothing curve to fit the points, rather than fitting the points to a linear or transformed curve. Statistically significant difference of intercepts is measured by assuming each species curve can be explained with one smoothing curve, finding the intercept of each species curve using this smoother, and finding t-values for each intercept. Statistically significant difference of overall trend is measured by assuming each species' stomatal conductance needs to be plotted with a separate smoothing curve, and comparing each smoothing curve to the aggregate smoothing curve of each of the other species. It should be noted that p-values for GAM are approximate, and must be considered carefully: p-values less than 0.001

indicate statistically significant difference (between intercepts or trends), and p-values greater than 0.1 indicate lack of statistically significant difference, but p-values on the order of 0.05 can be considered neither statistically significant nor non-significant. (Zuur, 2009)

Leaf temperature data was normalized by subtracting the mean temperature of the corresponding reference image (filter paper) from the mean temperature of each test image. Normalized leaf temperature was then plotted against leaf conductance, at times of day where plant transpiration rates were expected to be highest (11am-12pm).

In the drawdown experiment, proportional weight was calculated for each plant pot on each day of measurement, by dividing weight of the plant pot on Day n by weight of the same plant pot on Day 1. Median and Inter-quartile Range (IQR) of each treatment were calculated for each day, and represented visually, using box and whisker plots. Species and treatment effects were analyzed using two-way factorial ANOVA of proportional plant weight on Day 20.

Median and IQR of final aboveground biomass are reported within each species/watering-treatment, and representing using box and whisker plots.

Results

Diurnal Course

GAM revealed significant differences in the intercepts and trends of diurnal stomatal conductance for some species on the Diana Center diurnal course on July 8, 2015 (Fig. 2). For this diurnal course, ambient temperature was 24.5°C at 5:00, reached a peak of 33.5°C at 14:30, and dropped to 24°C by 16:20 (Fig. 2). PAR followed a similarly diurnal trend, starting at 1.2 µE, reaching a peak of 1858.7 µE at 14:00, and dropping again to 1.2 µE by 16:20 (Fig. 2). GAM intercepts were found to be significantly distinct in *Sympyotrichum laeve* ($p=5.36\times 10^{-6}***$), *Sympyotrichum novae-angliae* ($p=0.00018***$), and *Schizachyrium scoparium* ($p=0.000659***$). GAM intercept of *Oenothera biennis* was not significantly distinct from aggregate GAM ($p=0.720007$). GAM analysis was inconclusive for intercepts on *Panicum virgatum*, *Solidago nemoralis*, and *Baptisia tinctoria*. GAM curves were found to be significantly distinct only in *Sympyotrichum novae-angliae* ($F_{8.3,5.5}=7.269$, $p=6.82\times 10^{-7}***$) and *Schizachyrium scoparium* ($F_{4.3,8.0}=7.579$, $p=3.49\times 10^{-9}***$). GAM analysis on all other stomatal conductance curves was inconclusive. Though GAM was used to make visualizations of stomatal conductance trends on all roofs (S1-S6), statistical analyses of GAM were only evaluated for the July 8 diurnal course on the Diana Center.

Range of stomatal conductance varied across study sites. On the Diana Center, stomatal conductance ranged from 26.8 mmol m⁻² s⁻¹ to 1830.0 mmol m⁻² s⁻¹ on July 8, and from 8.0 mmol m⁻² s⁻¹ to 2064.0 mmol m⁻² s⁻¹ on July 31. On the Ranaqua roof, conductance ranged from 2.3 mmol m⁻² s⁻¹ to 1167.5 mmol m⁻² s⁻¹ on July 16, and from 38.6 mmol m⁻² s⁻¹ to 1273.6 mmol m⁻² s⁻¹ on August 13. On the Jackie Robinson roof,

conductance ranged from $8.3 \text{ mmol m}^{-2} \text{ s}^{-1}$ to $612.5 \text{ mmol m}^{-2} \text{ s}^{-1}$ on July 24, and from $30.9 \text{ mmol m}^{-2} \text{ s}^{-1}$ to $773.4 \text{ mmol m}^{-2} \text{ s}^{-1}$ on August 12. In the Schermerhorn greenhouse, conductance ranged from $8.4 \text{ mmol m}^{-2} \text{ s}^{-1}$ to $417.6 \text{ mmol m}^{-2} \text{ s}^{-1}$ on September 21.

Normalized leaf temperature from leaves measured 11am-12pm July 8, on the Diana Center is plotted against stomatal conductance (Fig. 3). The aggregate trend, including all species, appears to be negative (Fig. 3), but correlation was not significantly different from zero ($R^2=0.00484$, $p=0.666$). Correlation was only significantly negative in *Symphyotrichum laeve* ($R^2=0.6558$, $p=0.00816^{**}$).

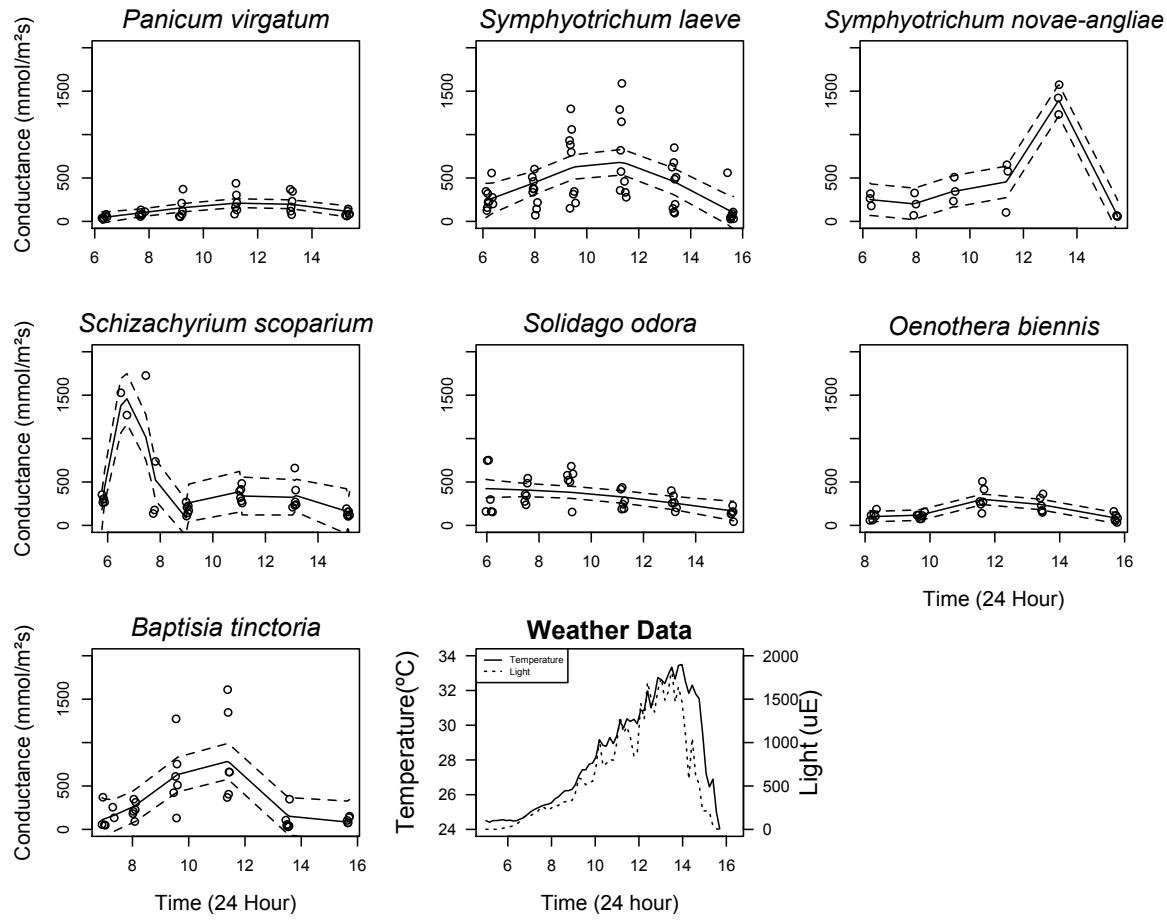


Figure 2 Diana Center 7/8/15. Measurements began at 5am and ended at 4pm. First 7 panels show GAM curves for each of 7 species, with dashed lines indicating standard error of the curves. The last panel shows ambient temperature and Photosynthetically Active Radiation (PAR) over the same 5am-4pm timescale. 6 individuals per species were tracked for stomatal conductance over the course of the day, except for *Sympphyotrichum laeve* (n=9) and *Sympphyotrichum novae-angliae* (n=3).

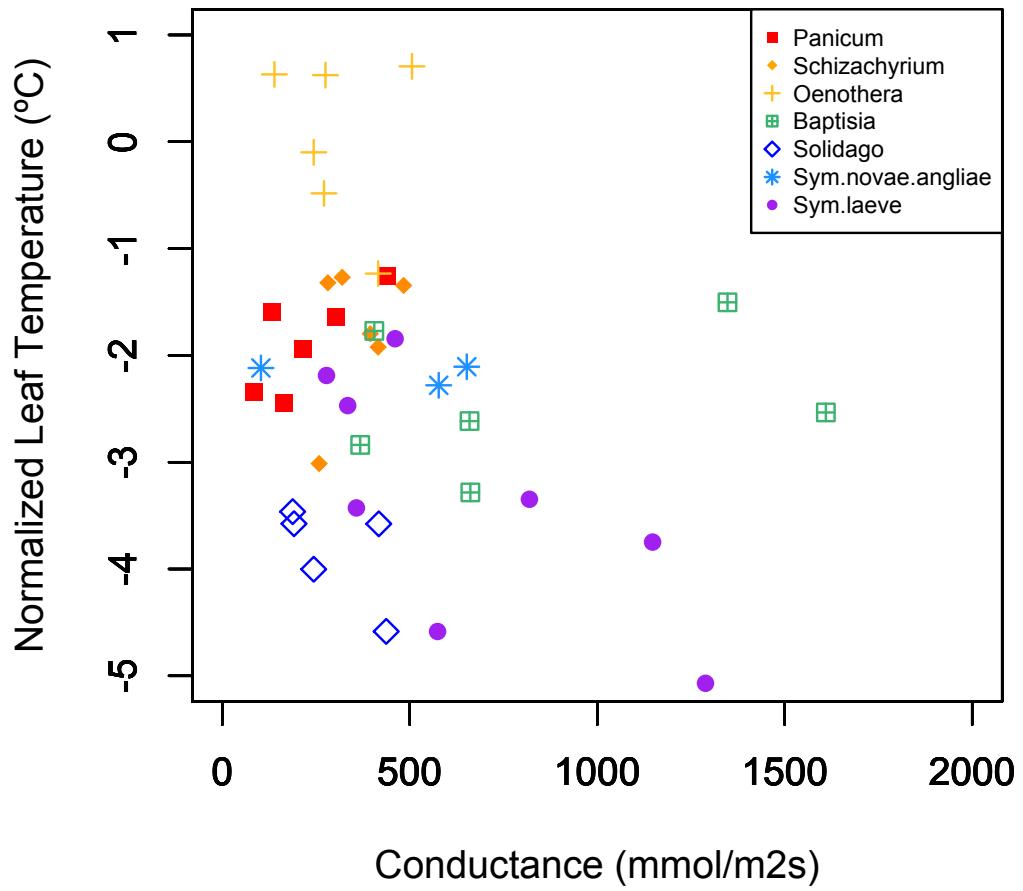


Figure 3 Normalized Leaf Temperature ($^{\circ}\text{C}$) vs. Conductance ($\text{mmol m}^{-2}\text{s}^{-1}$) for all species on Diana Center 7/8/15 11am-12pm. Leaf Temperature normalized by subtracting temperature measured in reference images from temperature measured in corresponding leaf images.

Soil Moisture Drawdown

Two-way factorial ANOVA revealed significant effects of species ($F_{8,168}=4.201$, $p=2.38\times 10^{-11}***$) and watering-treatment ($F_{1,168}=77.441$, $p=1.60\times 10^{-11}***$) on final proportional plant pot weight. Interaction between species and watering-treatment was also statistically significant ($F_{7,168}=4.201$, $p=0.000271***$). In *Pycnanthemum virginiana* and *Solidago nemoralis*, IQR of proportional weight for drought and well-watered plants did not overlap past Day 1, and in *Sorghastrum nutans* and *Andropogon gerardii*, IQR of proportional weight for drought and well-watered plants overlapped minimally (Fig. 4). For all other species where plants were split into two watering treatments, overlap in IQR between drought and well-watered plants was evident (Fig. 4). Where differences in proportional plant weight between watering treatments was apparent, drought-stressed plants had greater proportional plant weight than well-watered plants (Fig. 4).

Species where proportional plant pot weight was different between watering treatments were not necessarily the same species for which dry aboveground biomass was different between watering treatments. IQR of aboveground biomass was non-overlapping between watering treatments in *Apocynum cannabinum* and *Solidago nemoralis* (Fig. 5). In both *Apocynum* and *Solidago*, median aboveground biomass was lower for drought-stressed plants than well-watered plants (Fig. 5). For all other species where plants were split into two watering treatments, IQR was overlapping between treatments (Fig. 5).

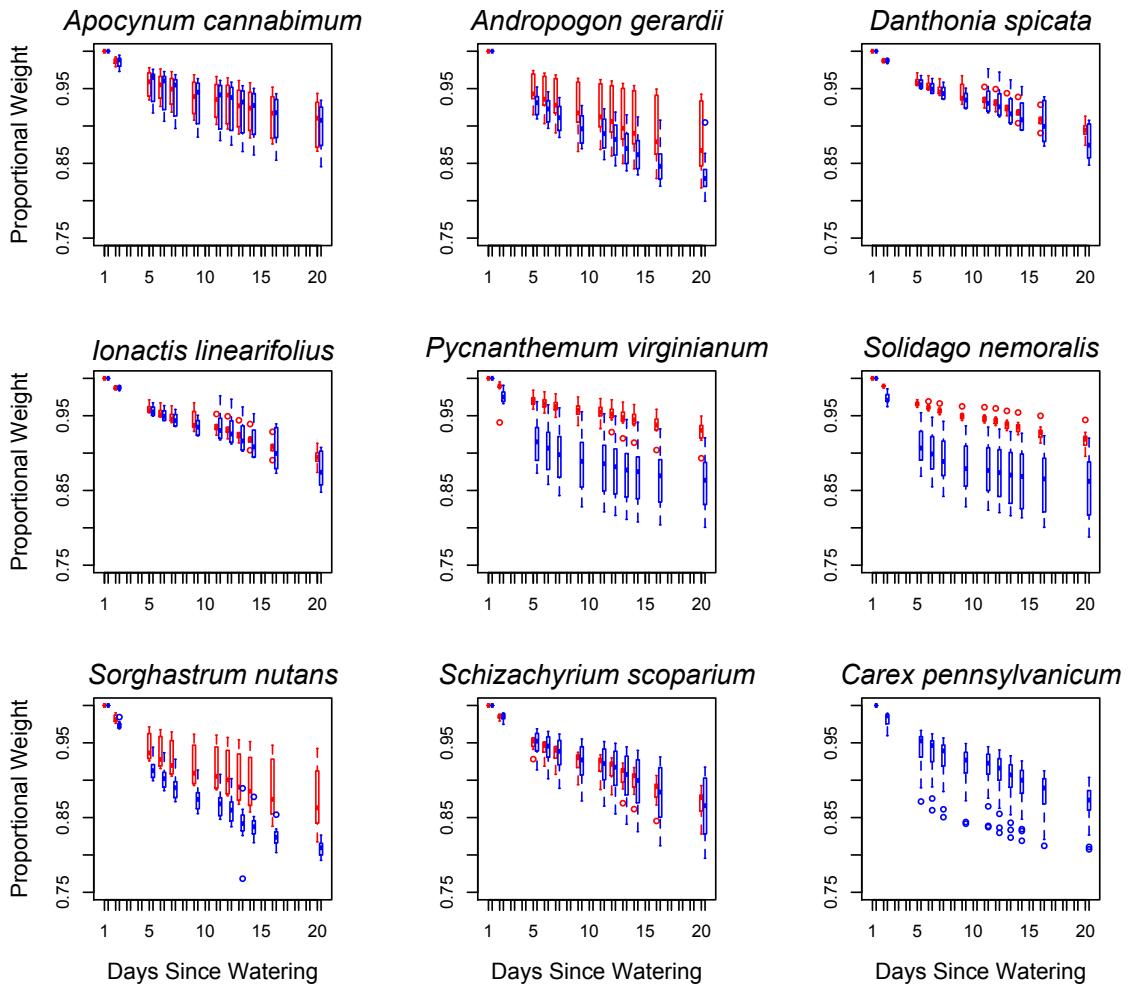


Figure 4 Soil-moisture drawdown trial: weight of plants proportional to weight on Day 1, over 20-day drought period. Red box plots represent plants grown in drought-stressed conditions, and blue box plots represent plants grown in well-watered conditions. Tops and bottoms of boxes indicate third and first quartiles; horizontal lines inside boxes indicate median proportional weight, whiskers indicate extreme (max/min) values, and open circles indicate outliers.

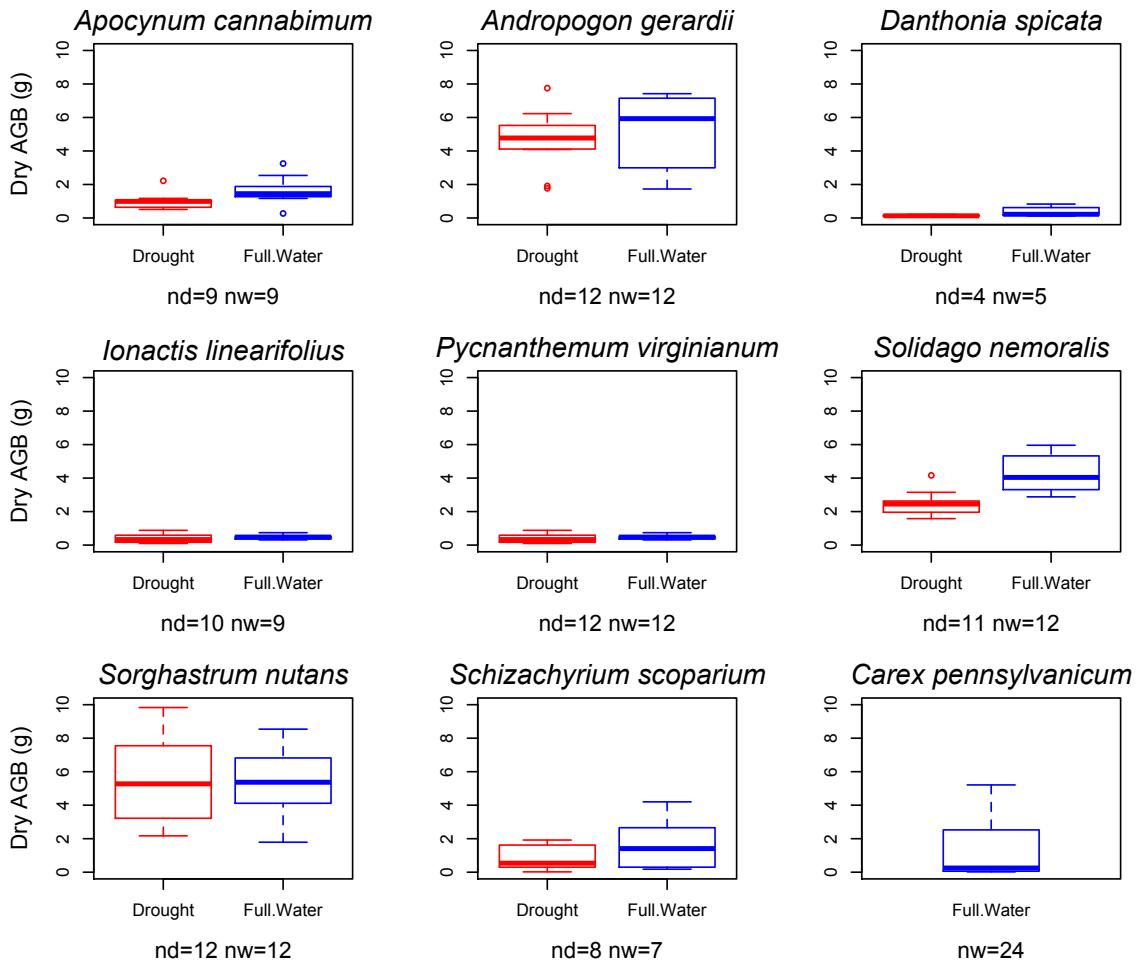


Figure 5 Box and whisker plots of dried aboveground biomass for each treatment in the greenhouse experiment. Tops and bottoms of boxes indicate third and first quartiles; horizontal lines inside boxes indicate median proportional weight, whiskers indicate extreme (max/min) values, and open circles indicate outliers. Nd=number of drought-stressed plants, Nw=number of full-watered plants.

Discussion

Species-differences in water-use were found at both the leaf-level and the whole-plant scale; this result is important because water-use differences between species make plant species increase moisture retention in green roof growth substrate at different rates, thereby contributing differently to urban storm water management (Beretta et al. 2015). Over the course of a day, significant species-differences were found in overall rates of stomatal conductance, as well as diurnal trends. Significantly distinct GAM intercepts in *Symphyotrichum laeve* ($p=5.36\times 10^{-6}***$), *Symphyotrichum novae-angliae* ($p=0.00018***$), and *Schizachyrium scoparium* ($p=0.000659***$) suggest that in each of these species, the overall rate of stomatal conductance is either significantly higher or lower than average stomatal conductance across all seven species. Based on visual inspection of the GAM curves (Fig. 2), *Symphyotrichum laeve*, *Symphyotrichum novae-angliae*, and *Schizachyrium scoparium* all exhibit higher stomatal conductance than average, suggesting that they transpire the most water, and contribute the most effectively to green roof moisture retention and storm water runoff mitigation.

Though the relationship between normalized leaf temperature and stomatal conductance was visually consistent with the predicted trend, leaf temperature was not a reliable indicator of transpiration, as analyzed here. Consistent with the idea that transpiration cools the leaf (Jones, 1999), normalized leaf temperature appears to decrease with increased conductance (Fig. 3). However, there was no significant correlation between normalized leaf temperature and conductance ($R^2=0.00484$, $p=0.666$). It is possible that significant effects of transpiration on leaf temperature are obscured or complicated by the effects of ambient conditions on leaf temperature.

Although the reference image is meant to account for environmental effects (Jones et al., 2002), there are a number of reasons why the filter paper reference image may not be an accurate in normalizing for the effects of ambient light and temperature. The white filter paper has a higher albedo than the green leaves, and may therefore be cooler than a non-transpiring leaf would be at the same light and temperature (Doulos et al. 2004). Artifacts of the reference's albedo may be apparent in normalized leaf temperature, causing normalized leaf temperature to be artificially high, particularly if the reference image was taken in the sun. Differences in microclimate may also have an affect—if the reference image was not taken in exactly the same location as the test image, ambient effects on the two images may not be comparable, and may complicate normalization of test image data using reference images (Doulos et al. 2004). Some of these complications could be rectified in the future by placing references next to tested leaves, and taking thermal images with both leaf and reference in the same frame.

It should be noted that normalized leaf temperature was more correlated for some species than others—unlike all other tested species, in *Symphyotrichum laeve*, normalized leaf temperature was significantly negatively correlated to stomatal conductance ($R^2=0.6558$, $p=0.00816^{**}$). Since *Symphyotrichum laeve* showed some of the highest stomatal conductance and the lowest normalized leaf temperature, this result could imply that at high levels of transpiration, the effect of transpiration can overcome artifacts of environmental variables due to issues with reference temperatures. On the other hand, one reference image was taken per 6 leaf images of plants in the same species, so differences it is also possible that species differences in level of correlation between leaf temperature and conductance was due to differences in the reference temperature.

Along with species-differences in overall conductance, significant species-differences in diurnal stomatal conductance trend curves were found in *Symphyotrichum novae-angliae* ($F_{8,3,5,5}=7.269$, $p=6.82\times 10^{-7}***$) and *Schizachyrium scoparium* ($F_{4,3,8,0}=7.579$, $p=3.49\times 10^{-9}***$), suggesting that spot measurements do not provide a comprehensive understanding of water-use in different species. Visual inspection of the GAM curves of each species reveals that in *Schizachyrium*, stomatal conductance is highest earlier in the day than most other species, and in *Symphyotrichum novae-angliae*, conductance is highest later in the day than most other species (Fig. 2). Though most plants have been found to follow similar diurnal patterns in response to environmental variables, usually characterized by high stomatal activity in the morning, followed by a midday depression (Roessler and Monson 1985, Tuzet et al. 2003), species-differences in diurnal behavior are well-documented in plant ecophysiology literature (Jia-Ding et al. 2004, de Mattos et al. 1997). Despite evidence for variability in diurnal plant activity between species, the diurnal course approach is rarely applied to green infrastructure. Significant differences between diurnal stomatal conductance curves in *Schizachyrium scoparium* and *Symphyotrichum novae-angliae* suggest that, dynamic plant traits affecting green roof ecosystem services must be measured within the context of a full day, in order to be fully representative of species difference. These results put previous work using spot measurements (McIvor and Lundholm, 2011), into question.

The range in stomatal conductance was greatest during the Diana Center diurnal courses ([26.8, 1830.0], [8.0, 2064]), followed by diurnal courses on Ranaqua ([2.3, 1167.5], [38.6, 1273.6]), and on Jackie Robinson ([8.3, 612.5], [30.9, 773.4]), with the lowest range in stomatal conductance on the September 21 diurnal course in the

Schermerhorn greenhouse ([8.4, 417.6]) (All intervals reported in $\text{mmol m}^{-2}\text{s}^{-1}$). Some of the differences in range of stomatal conductance measurements may be attributable to differences in the timeframe of measurements during each diurnal course. Since Ranaqua and Jackie Robinson diurnal courses began later in the day than the Diana Center diurnal courses, any plants with high stomatal conductance early in the morning would not have been recorded as having high conductance at Ranaqua or Jackie Robinson. Other factors that likely come into play are senescence and drought-stress. The lowest range of stomatal conductance was in the diurnal course at Schermerhorn greenhouse, which took place on September 21. By this time, plants were already beginning to show signs of senescence, and therefore may not have been conducting as much water (Zhang et al. 1998). In addition, plants in the greenhouse as well as on the Jackie Robinson roof were relatively drought-stressed—unlike Ranaqua and the Diana Center, plants on the Jackie Robinson green roof are not irrigated, and plants in the greenhouse diurnal course were being purposefully drought-stressed. Drought-stress can decrease the level of conductance, as plants may acclimate to low water-availability by closing their stomates in order to conserve water (Duan et al. 2015). This phenomenon was explored in detail in the soil moisture drawdown experiment.

In the soil moisture drawdown experiment, water-loss was greater in some species than others, and different species responded differently to drought, with respect to water-loss. Two-way factorial ANOVA on final proportional weight of plant pots revealed significant differences in water-loss by species ($F_{8,168}=4.201$, $p=2.38\times 10^{-11}***$) and watering-treatment ($F_{1,168}=77.441$, $p=1.60\times 10^{-11}***$), with significant interaction between the effects of species and watering-treatment ($F_{7,168}=4.201$, $p=0.000271***$). Not only

do these results indicate that species vary in water-use, they also suggest that different species respond differently to drought, with respect to water-use. This phenomenon is apparent in non-parametric analysis of the drawdown experiment: final IQR of drought-stressed vs. fully-watered *Andropogon gerardii*, *Pycnanthemum virginianum*, *Solidago nemoralis*, and *Sorghastrum nutans* is non-overlapping, whereas final IQR between watering-treatments in all other species split by watering-treatment is overlapping (Fig. 4). In fact, throughout the drawdown trial, overlap in IQR between watering treatments for *Andropogon*, *Pycnanthemum*, *Solidago*, and *Sorghastrum* is minimal, with fully-watered plants losing more water than drought-stressed plants (Fig. 4).

For *Andropogon*, *Pycnanthemum*, and *Sorghastrum*, differences in water-loss between watering-treatments are not explained by differences in available leaf area for respiration, suggesting that these species are acclimating to drought-conditions, rather than taking up less water as a result of declines in growth or health. In *Solidago nemoralis*, the difference in water-loss by watering treatment corresponds to a difference in aboveground biomass, demonstrated by non-overlapping IQR between higher aboveground biomass of drought-stressed plants and lower aboveground biomass of well-watered plants (Fig. 5). In this case, decreased water-loss in drought-stressed *Solidago* plants is likely a function of size of plants, rather than physiological acclimation to drought. In *Andropogon*, *Pycnanthemum* and *Sorghastrum*, on the other hand, aboveground biomass is similar across watering-treatments, as demonstrated by overlapping IQR, so differences in water-loss between watering-treatments are not explained by size of plants. Water-use response to drought-stress is therefore attributable to physiological drought-responses particular to these species. These results suggest that

Andropogon, *Pycnanthemum*, and *Sorghastrum* are isohydric, meaning that they close their stomates in response to drought, in order to conserve water (Duan et al. 2015). Isohydric species behave in contrast to anisohydric species, which maintain stomatal conductance in drought in order to continue taking in CO₂ through the stomates (Duan et al. 2015). Anisohydric species, here *Apocynum cannabinum*, *Danthonia spicata*, *Ionactis linariifolia*, *Solidago nemoralis*, and *Schizachyrium scoparium*, will take up more water under drought-stress than isohydric species, and therefore are more likely to contribute positively to storm water runoff mitigation on non-irrigated roofs (such as Jackie Robinson) than isohydric species.

Conclusions

When measured at the leaf-level, plant species grown on green roofs can vary significantly both in overall rates, and in diurnal trends of water-flux. Species-differences in transpiration are most conspicuous through measurements of stomatal conductance, but despite difficulty in normalizing for environmental variables, thermal imagery is promising in its potential to detect significant variation in transpiration between species—with alterations to analysis and methodology, could be a quicker, more convenient tool for measuring transpiration on green roofs than stomatal conductance. Measurements of water-loss at the whole-plant scale support the hypothesis that water-use varies significantly between plants grown on green roofs, and also reveal which species reduce water-use in response to drought (*Andropogon gerardii*, *Pycnanthemum virginiana*, and *Sorghastrum nutans*), which may be applied to planting choices on particularly drought-stressed roofs.

Recommendations

Of the plants that are able to survive on green roofs, those that use the most water should be planted most frequently, because they are the most effective at restoring moisture-capacity in green roof growing substrate, thereby reducing storm water runoff most effectively. In determining which plants are the highest water-users, diurnal behavior must be taken into account in order to obtain an accurate understanding of which species are using the most water. Further study of thermal imagery as it relates to measurement of plant transpiration is warranted to improve efficiency of water-use assessment in the field. Anisohydric species (those that maintain stomatal conductance under drought-stress, including *Apocynum cannabinum*, *Danthonia spicata*, *Ionactis linariifolia*, *Solidago nemoralis*, and *Schizachyrium scoparium*) should be used on non-irrigated green roofs that are particularly drought-stressed environments.

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Appendix

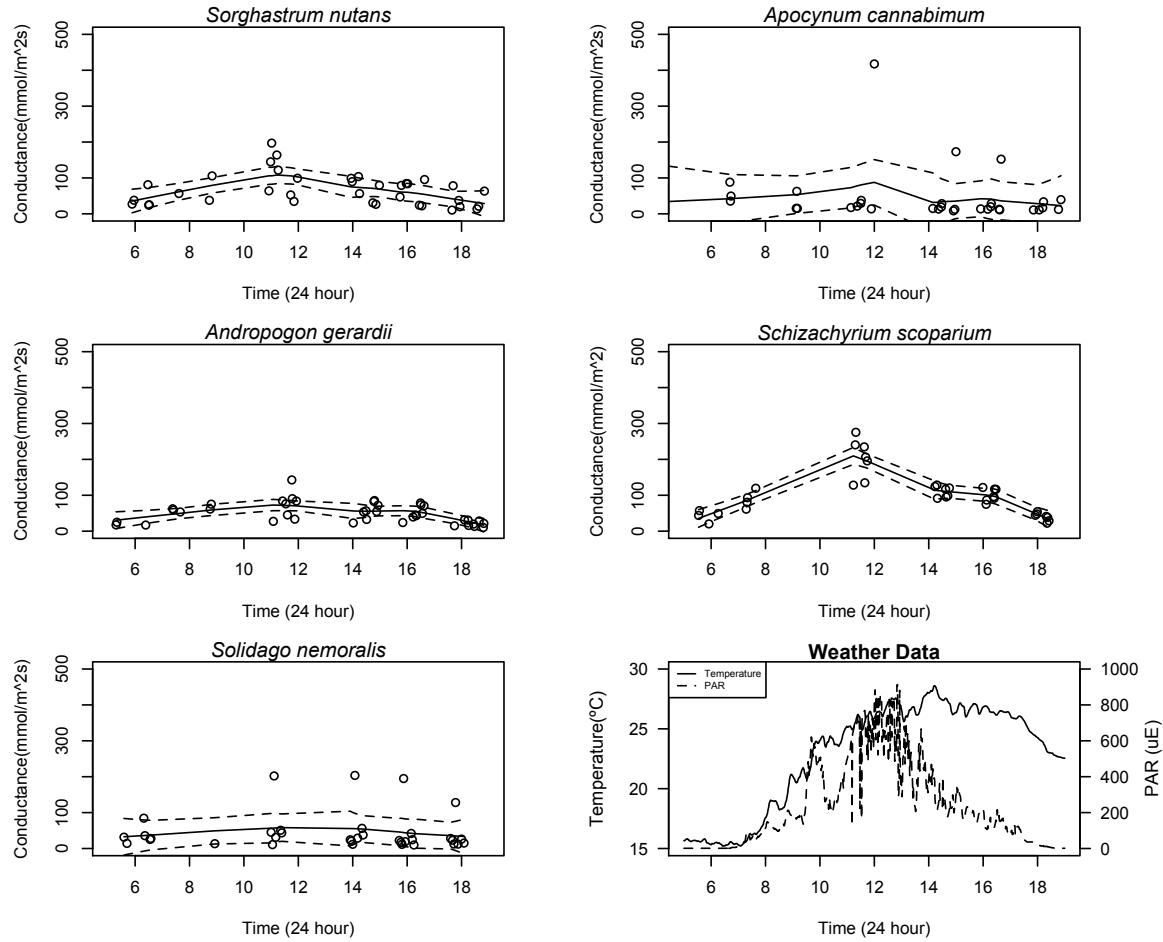


Figure S1 Schermerhorn greenhouse 9/21/15. Measurements began at 5am and ended at 7pm. First 5 panels show GAM curves for each of 5 species, with dashed lines indicating standard error of the curves. LOESS smoother set at 0.5. The last panel shows ambient temperature and Photosynthetically Active Radiation (PAR) over the same 5am-7pm timescale.

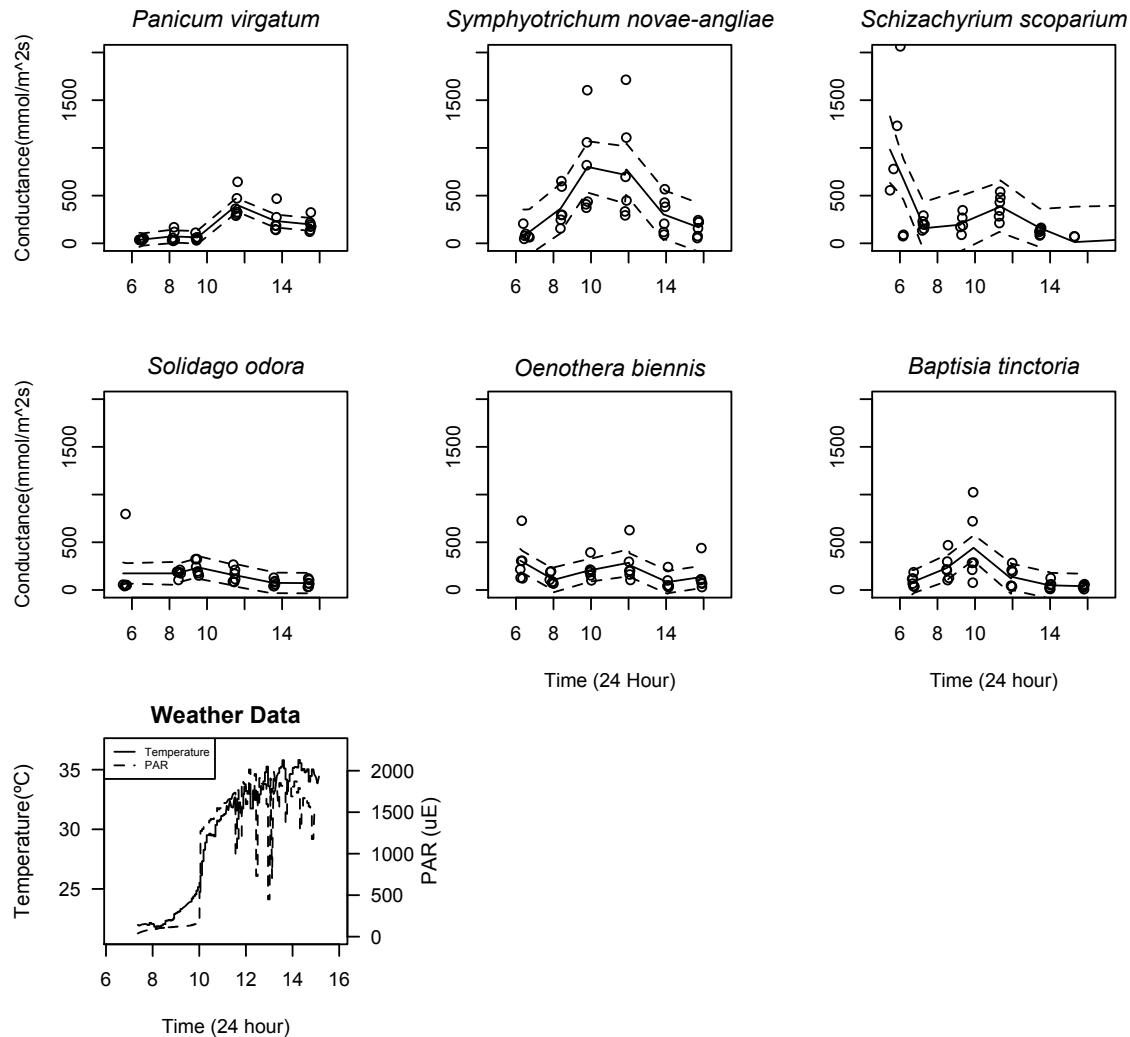


Figure S2 Diana Center 7/31/15. Measurements began at 5am and ended at 4pm. First 7 panels show GAM curves for each of 7 species, with dashed lines indicating standard error of the curves. LOESS smoother set at 0.5. The last panel shows ambient temperature and Photosynthetically Active Radiation (PAR) over the same 5am-4pm timescale.

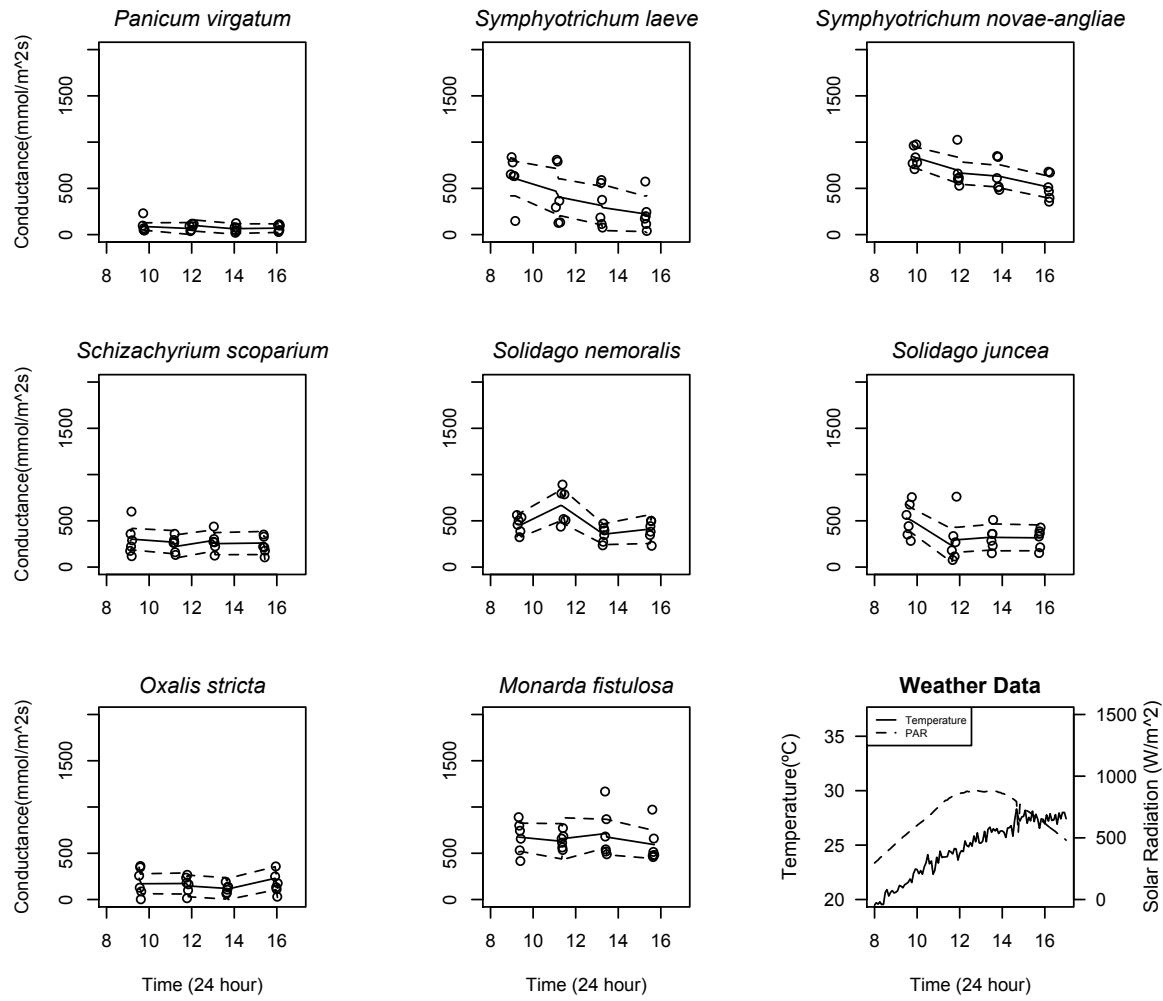


Figure S3 Ranaqua 7/16/15. Measurements began at 9am and ended at 4pm. First 8 panels show GAM curves for each of 8 species, with dashed lines indicating standard error of the curves. LOESS smoother set at 0.5. The last panel shows ambient temperature and solar radiation (W/m^2) over the same 9am-4pm timescale.

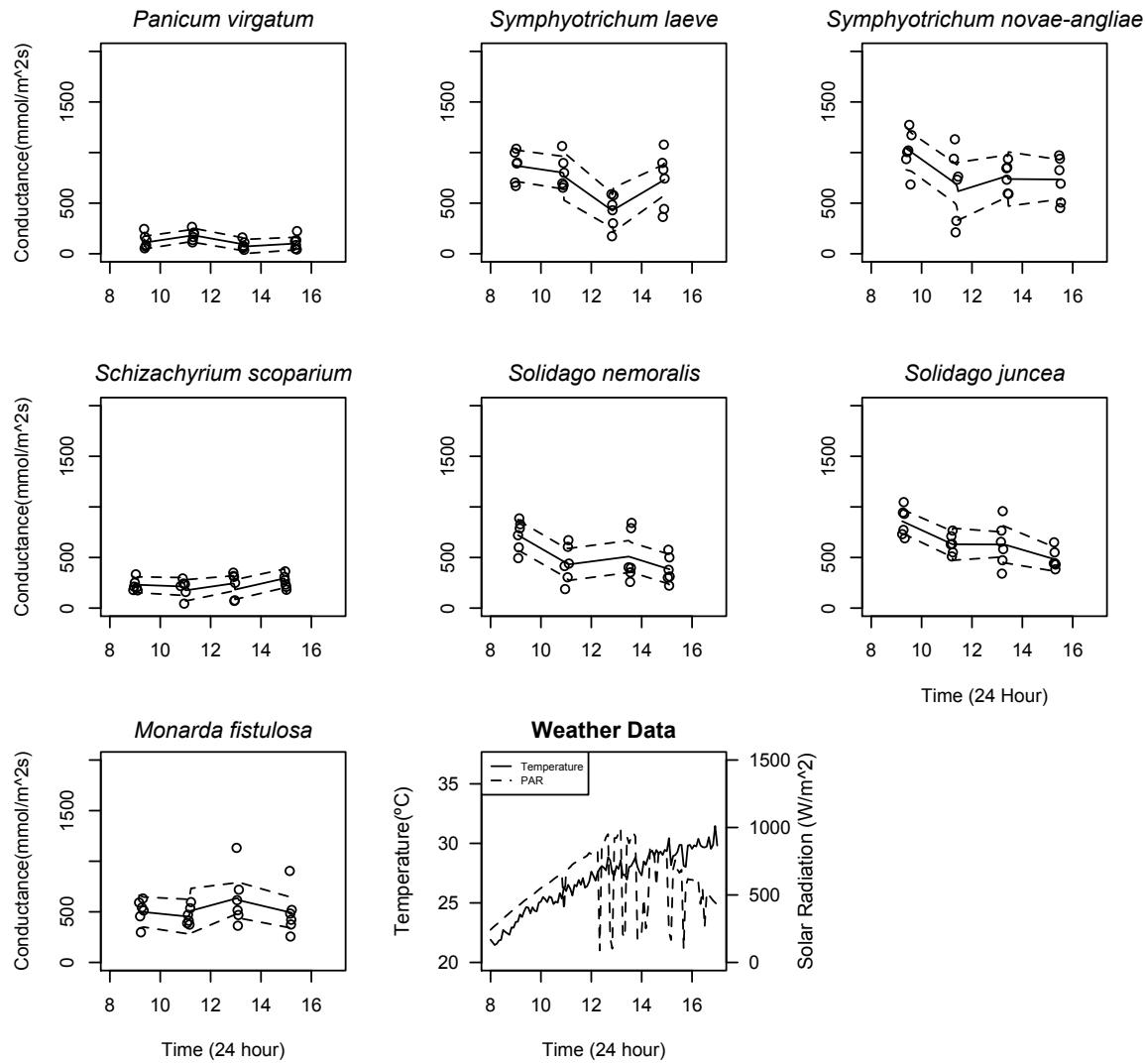


Figure S4 Ranaqua 8/13/15. Measurements began at 9am and ended at 4pm. First 7 panels show GAM curves for each of 7 species, with dashed lines indicating standard error of the curves. LOESS smoother set at 0.5. Weather data is not yet available for this date.

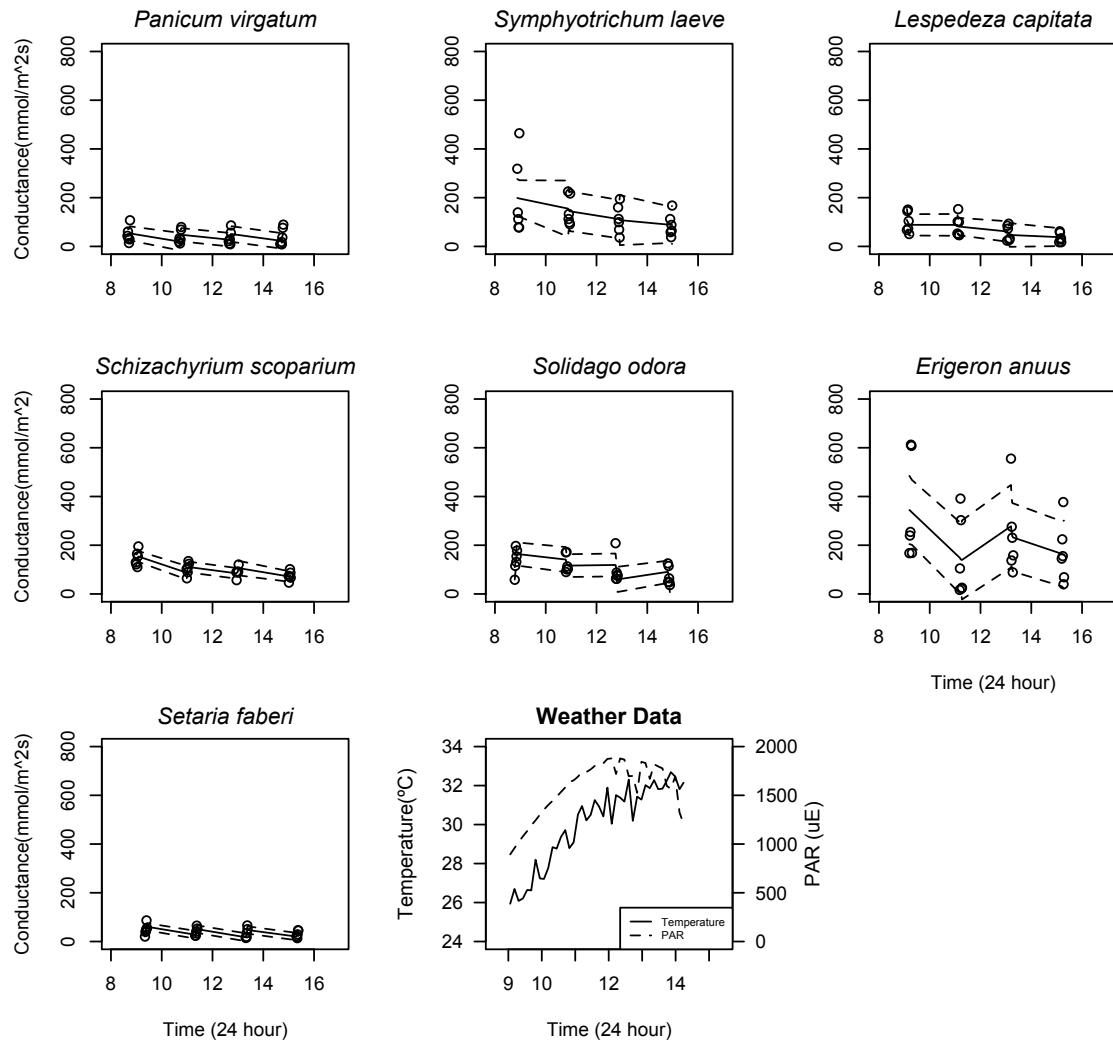


Figure S5 Jackie Robinson 7/24/15. Measurements began at 9am and ended at 3pm. First 7 panels show GAM curves for each of 7 species, with dashed lines indicating standard error of the curves. LOESS smoother set at 0.7. The last panel shows ambient temperature and Photosynthetically Active Radiation (PAR) over the same 9am-3pm timescale.

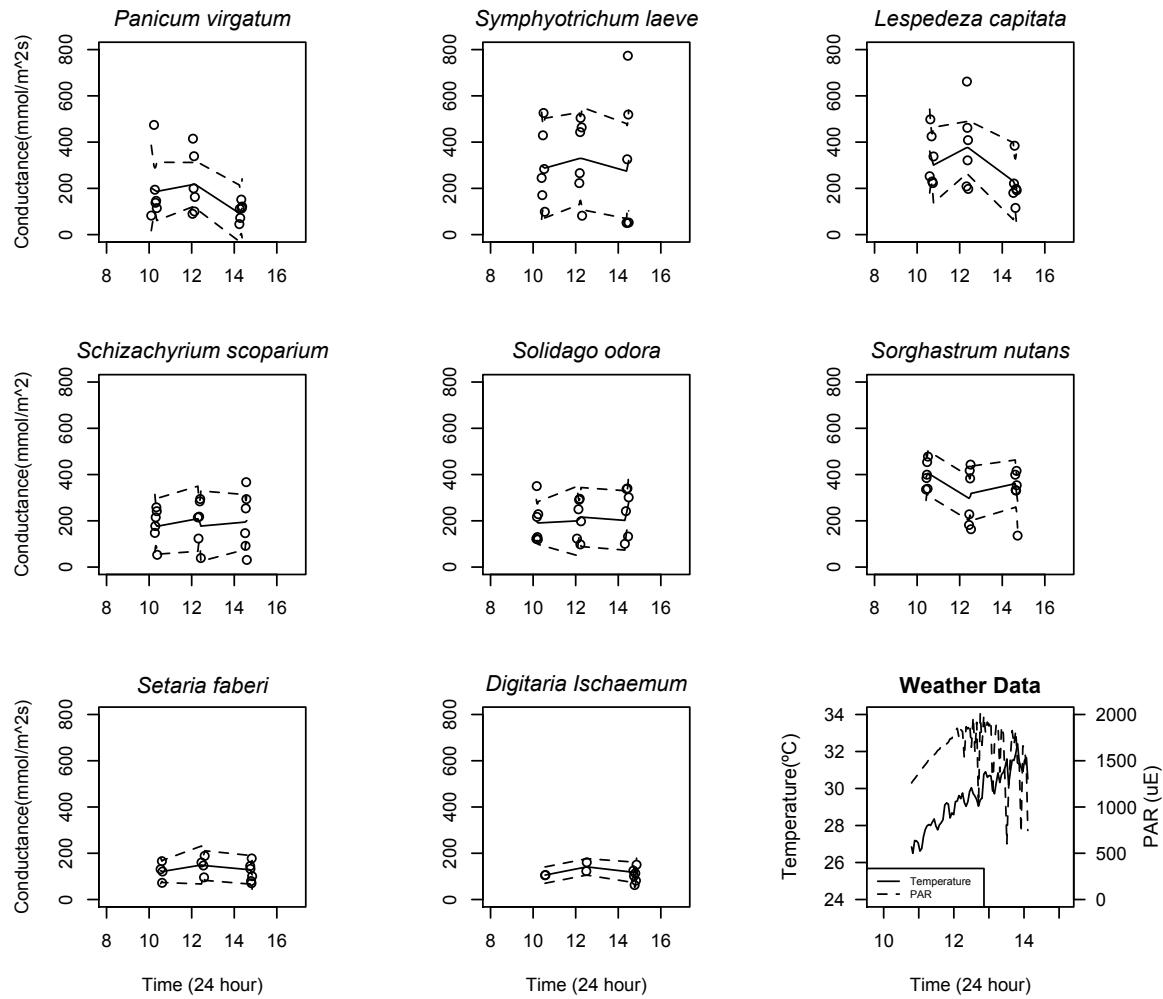


Figure S6 Jackie Robinson 8/12/15. Measurements began at 10am and ended at 3pm. First 7 panels show GAM curves for each of 7 species, with dashed lines indicating standard error of the curves. LOESS smoother set at 0.7. The last panel shows ambient temperature and Photosynthetically Active Radiation (PAR) over the same 10am-3pm timescale. For *Setaria faberi*, n=4 for first two time-steps and n=6 for the last time-step. For *Digitaria Ischaemum*, n=2 for the first two time-steps and n=6 for the last time-step.

Table S1 Mean final weight of plant pots proportional to weight of plant pots on Day 1, for drought-stressed and full-watered plants of each species.

	Drought	Full-Water
<i>Apocynum</i>	0.9468	0.9406
<i>Andropogon</i>	0.9353	0.9099
<i>Carex</i>	n/a	0.8674
<i>Danthonia</i>	0.9483	0.9448
<i>Ionactis</i>	0.9263	0.9294
<i>Pycnanthemum</i>	0.9628	0.8962
<i>Solidago</i>	0.9568	0.8914
<i>Sorghastrum</i>	0.9296	0.8904
<i>Schizachyrium</i>	0.9352	0.9345

Table S2 Mean Root: Shoot Ratios for 9 species in greenhouse experiments. First column shows mean root: shoot for drought-treatment plants only, second column shows mean root: shoot ratios for fully-watered plants only, and the last column shows mean root: shoot for all plants together per species.

	Drought	n	Full-Water	n	Mean Root: Shoot	n
<i>Andropogon</i>	0.738495974	2	1.089246726	2	0.91387135	4
<i>Apocynum</i>	3.341323935	3	3.341323935	3	3.220276705	6
<i>Carex</i>	n/a	0	0.629382257	4	0.629382257	4
<i>Danthonia</i>	0.272893773	2	0.641389086	2	0.457141429	4
<i>Ionactis</i>	0.71969697	3	0.943655914	3	0.831676442	6
<i>Pycnanthemum</i>	3.116444444	3	4.625383217	3	3.87091383	6
<i>Schizachyrium</i>	0.335712304	5	0.580250955	2	0.40558049	7
<i>Solidago</i>	0.231508768	4	0.703481881	2	0.388833139	6
<i>Sorghastrum</i>	0.957759007	2	3.062862128	3	2.22082088	5

Table S3 Percent mortality for each species within full-water watering treatment, drought-stressed treatment, and overall.

	Full-Water	Drought	Both treatments
<i>Andropogon</i>	0.17	0.00	0.08
<i>Apocynum</i>	0.00	0.25	0.11
<i>Danthonia</i>	0.17	0.33	0.25
<i>Ionactis</i>	0.10	0.00	0.05
<i>Pycnanthemum</i>	0.33	0.00	0.17
<i>Solidago</i>	0.25	0.75	0.50
<i>Sorghastrum</i>	0.08	0.00	0.04
<i>Schizachyrium</i>	0.25	0.00	0.13