SWORD FERN ECOPHYSIOLOGY REPORT 2018 – SEWARD PARK, WA

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Overview:

Seward Park has been experiencing a decline in Western sword fern (*Polystichum munitum*) populations since 2013. Efforts to investigate the cause of the decline have not been conclusive to date, yet the severely impacted areas of the park are now in need of follow-up restoration. To determine whether sword fern recovery is possible at sites that have experienced complete die-off, we examined the survival and growth of planted sword fern seedlings at "Ground Zero" (the epicenter of the sword fern decline at the park) and two more nearby locations. Additionally, to understand the physiological status of stands of sword fern throughout the park, we set up a network of monitoring plots to survey existing sword fern plants in various stages of decline. All measurements were standardized across the three study areas to establish a baseline data set for referenced over time. We treated "plot types" as separate measurement area and therefore experimental findings are primarily reported on a plot type basis (Figure 1). This is the first series of studies conducted at Seward Park that examined the mechanisms and dynamics behind sword fern decline and recovery using a physiological approach.

Methods:

Restoration potential of affected sites

In March of 2018, sword fern plants previously grown into ½ gallon containers were planted into plots in the area known as "Ground Zero" at Seward Park (Figure 1, "restoration transect" plots). Seedlings were planted in three blocks (47-57 seedlings block^{-1}). Planting blocks were positioned across "Ground Zero" and direct surrounding area downslope to account for differences in slope position, soil moisture, and overstory conditions (i.e. differences in canopy architecture from trees dominating the site). Each block consisted of 5 transects of 9-15 ferns with 2-m spacing between ferns within each transect and 3-m spacing between transects. Each transect was arranged from North to South. Ferns were hand planted into pre-selected locations and remained un-irrigated and otherwise unaided for the duration of the study. Following planting, each fern tagged with an alphanumeric code to enable tracking during subsequent measurement periods. Measurements were taken on a sub-sample of the full planted population, which included every other transect, for a total of 3 transects per block. Within those transects, either every other or all seedling were measured.

Figure 1. Overview of the locations and plot types (yellow squares – "Eco-phys" plots established in 2018, black dots – monitoring plots established in 2015, and red squares – restoration "planting transect" plots planted near the original "die-off" location ["Ground Zero"]) included in the ecophysiological assessment of sword fern decline at Seward Park, Seattle, WA.

Monitoring established sword fern population within Seward Park

A series of sword fern monitoring plots were established in and around the “Ground Zero” area at Seward Park in 2015 (Figure 1, “2015 plots”). The plots were designed to capture existing populations of sword fern during a decline. The plots are ca. 3 m^2 and consist of a dynamic population of sword fern (3 to 17 individuals $plot^{-1}$). Due to the severity of the decline in and around “Ground Zero”, additional plots were established throughout the full length of the park in the spring of 2018 (“Eco-phys” plots), in an effort to capture the spatial spread of the decline and assess plants in different stages of decline severity (Figure 1). Eleven plots were established using a randomized approach, each containing 3-9 ferns per plot.

Ecophysiological measurements

All three plot types were subject to monthly measurements May-October, 2018 and again in April 2019. Measurements included: chlorophyll fluorescence (F_v/F_m), chlorophyll content (SPAD), soil moisture (%), and the extent of crown decline (foliar browning and frond number). Plant physiological status (i.e. stress response) was estimated through measurements of chlorophyll fluorescence and relative chlorophyll content. Fluorescence was measured with a portable fluorometer (OS30P+, Opti-Sciences, Inc., Hudson, NH, USA). First, foliage was dark adapted for at least 20 minutes then a weak modulating light was used to calculate maximum fluorescence (F_o), followed by a pulse of an intense saturating light to calculate maximum fluorescence (F_m), variable fluorescence (F_v=F_m-F_o), and the F_v/F_m ratio (the potential quantum efficiency of photosystem II [PSII]), an indicator of plant photosynthetic performance and photo-inhibition during plant stress (Maxwell and Johnson, 2000). Relative chlorophyll content was measured in SPAD units using a Konica Minolta SPAD 502 meter (Konica Minolta, Ramsey, NJ, USA). SPAD measurements indicate relative chlorophyll content of leaf tissue by measuring the transmittance of the tissues in the red and infrared regions (Bussotti and Pollastrini, 2015). For both the chlorophyll fluorescence and content measurements three points per plant were surveyed.

In tandem with physiological measurements, the number of live fronds per fern and the percentage of foliar browning (of the live crowns) were estimated. Finally, we assessed soil moisture status using a moisture probe, and measured at three locations around the base of each living fern (inserted 10-15 cm from the base of each fern).

Results:

Restoration potential of affected sites

While seedling survival one year following outplanting was high (75-87%), moisture played a critical role in plant physiological and morphological responses. For example, during the driest portion of the first growing season (August 2018), the values of F_v/F_m were well below optimal (0.79-0.84, Maxwell and Johnson, 2000) and indicate that plants were experiencing substantial stress (Figure 2). This was particularly evident for ferns planted at the original "Ground Zero" planting block (GZ 1), which had the lowest F_v/F_m values among seedlings. The chlorophyll content (SPAD) values also declined with the onset of drought and did not recover fully by the following spring. However, the lack of chlorophyll content recovery was likely due to a high proportion of new fronds, which tends to provide lower SPAD values.

The seasonal decrease in soil moisture differed slightly between the three planting blocks, with the upper (GZ 1) and middle (GZ 2) planting blocks exhibiting the greatest decrease in soil moisture during August 2018 (Figure 2). Unsurprisingly, in the absence of supplemental irrigation, the number of fronds per fern (Figure 3) and the percentage of foliar browning (Figure 4) increased at the onset of drought. Changes in frond number and foliar browning were inversely correlated with soil moisture. Overall, frond number decrease by 62% and foliar browning increased by 31% during the first year of growth among ferns established at restoration plots at Ground Zero (Table 1).

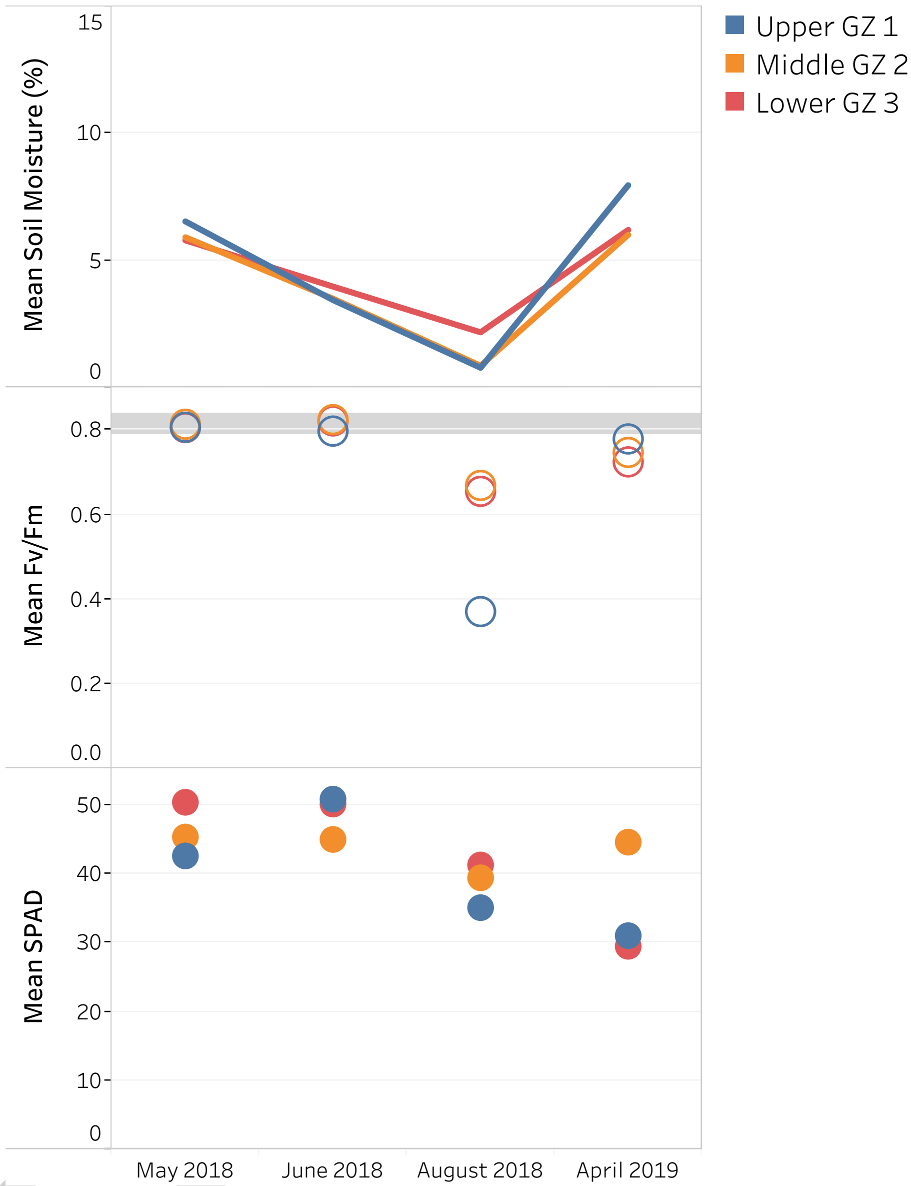


Figure 2. Soil moisture (%), mean chlorophyll fluorescence (F_v/F_m), and chlorophyll content (SPAD) measurements of one-year old western sword fern (*Polystichum munitum*) seedlings planted at "Ground Zero", Seward Park, Seattle, WA. Colors indicate planting blocks, which varied across slope location, moisture, and canopy cover conditions; where blue indicates the "Upper GZ 1" planting block, which is the original location of the decline and yellow "Middle GZ 2" and red "Lower GZ 3" planting blocks located downslope. Measurements are shown for the periods that correspond with early seedling establishment (May and June, 2018), mid-season (August, 2018), and first spring (April, 2019) following outplanting. Gray bar in the F_v/F_m figure represents the approximate optimal range of values, below which plants are considered to be experiencing stress (Maxwell et al. 2000).

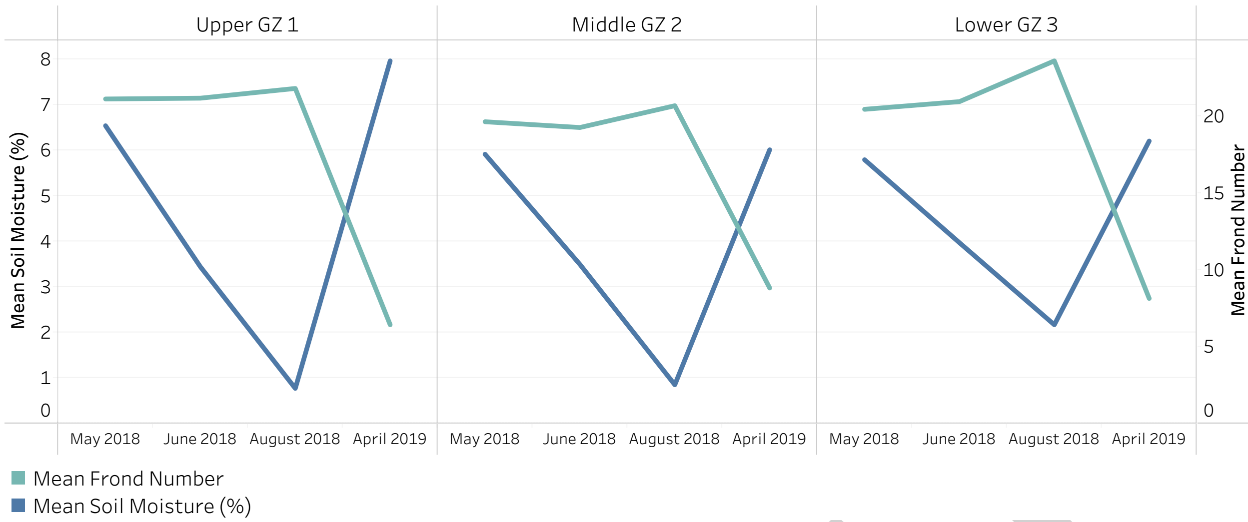


Figure 3. Mean soil moisture percentage (blue) plotted against mean frond count (teal) for western sword fern (*Polystichum munitum*) seedlings planted at "Ground Zero", Seward Park, Seattle, WA. Measurements are shown for the periods that correspond with early seedling establishment (May and June, 2018), mid-season (August, 2018), and first spring (April, 2019) following outplanting. Figure is segmented by planting block, including: "Upper GZ 1" – corresponding to the original decline location, "Middle GZ 2" and "Lower GZ 3" indicate the downslope sites.

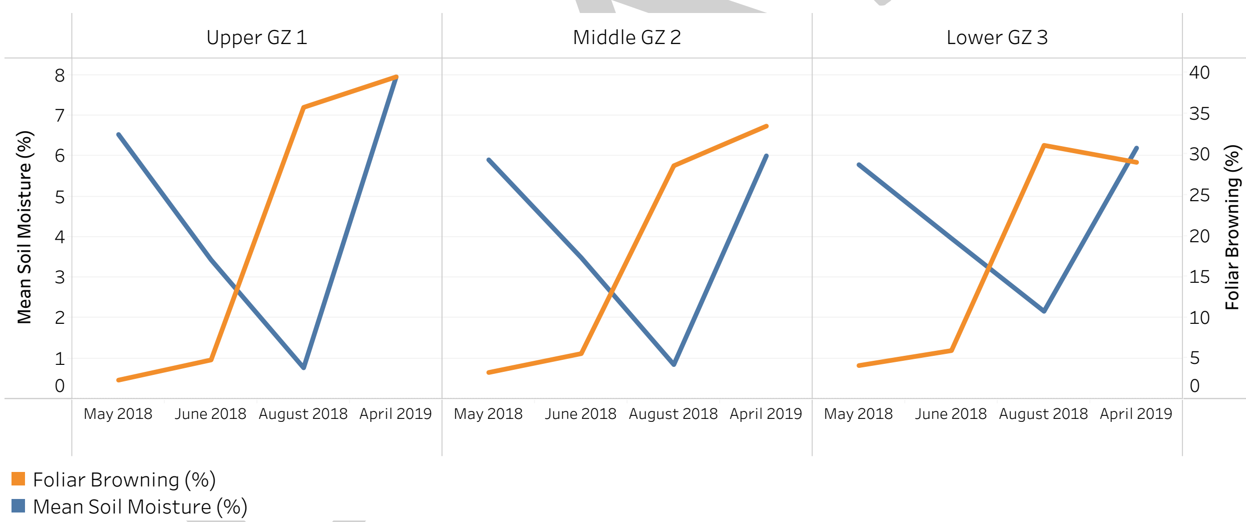


Figure 4. Mean soil moisture percentage (blue) plotted against percent foliar browning (orange) for western sword fern (*Polystichum munitum*) seedlings planted at "Ground Zero", Seward Park, Seattle, WA. Measurements are shown for the periods that correspond with early seedling establishment (May and June, 2018), mid-season (August, 2018), and first spring (April, 2019) following outplanting. Figure is segmented by planting block, including: "Upper GZ 1" – corresponding to the original decline location, "Middle GZ 2" and "Lower GZ 3" indicate the downslope blocks.

Monitoring the status of established sword fern population within Seward Park

Late season measurements showed a high degree of plant stress, near complete loss of sword fern canopy, and ferns appeared unlikely to rebound; however, following April 2019 measurements, 90% of the impacted populations showed some degree of recovery (defined here as the presence of new fronds).

2018 "Eco-phys" Plots

When averaged across all plots, mean foliar browning percentage decreased slightly during June, but returned back to baseline levels in April 2019. Ferns had a lower number of live fronds in August, which corresponded with the driest portion of the year. However, these ferns were able to regenerate new fronds in the following spring (Figure 5, Table 1). This may indicate that 15-20% foliar browning of the live fern crown is a manageable amount of seasonal canopy loss from which adult ferns can recover, but follow up measurements are needed to account for potential differences between years. The mean F_v/F_m values were within the normal range, indicating that ferns were not stressed at the beginning of the growing season. However, as moisture declined F_v/F_m values decreased below the optimal threshold, suggesting that the photosynthetic capacity of plants was compromised. Interestingly, while soil moisture levels returned back to the original spring levels by April 2019, ferns had not exhibited a parallel recovery in F_v/F_m. It is unclear whether this decreasing trend in F_v/F_m values will continue throughout the growing season, and warrants continued investigation. Chlorophyll florescence values (SPAD) also decreased during August 2018, but rebounded in the following spring, to near original levels.

"2015 Plots"

The ferns in the monitoring plots established in 2015, located in the vicinity of "Ground Zero", have been showing signs of significant decline over the course of the past three years. In general, the percent foliar browning and decrease in frond numbers among ferns in these plots corresponded directly with soil moisture, reaching the highest foliar browning and decrease in frond number at the driest measurement period, in August 2019 (Figure 6). These plants appeared to be under continuous stress from May to April, with F_v/F_m values below the optimal range in May 2018. As with the 2018 plots, these values declined with decreases in soil moisture but did not rebound once moisture returned back to its original levels in April 2019. Chlorophyll fluorescence (SPAD) values remained fairly static from March 2018 to April 2019.

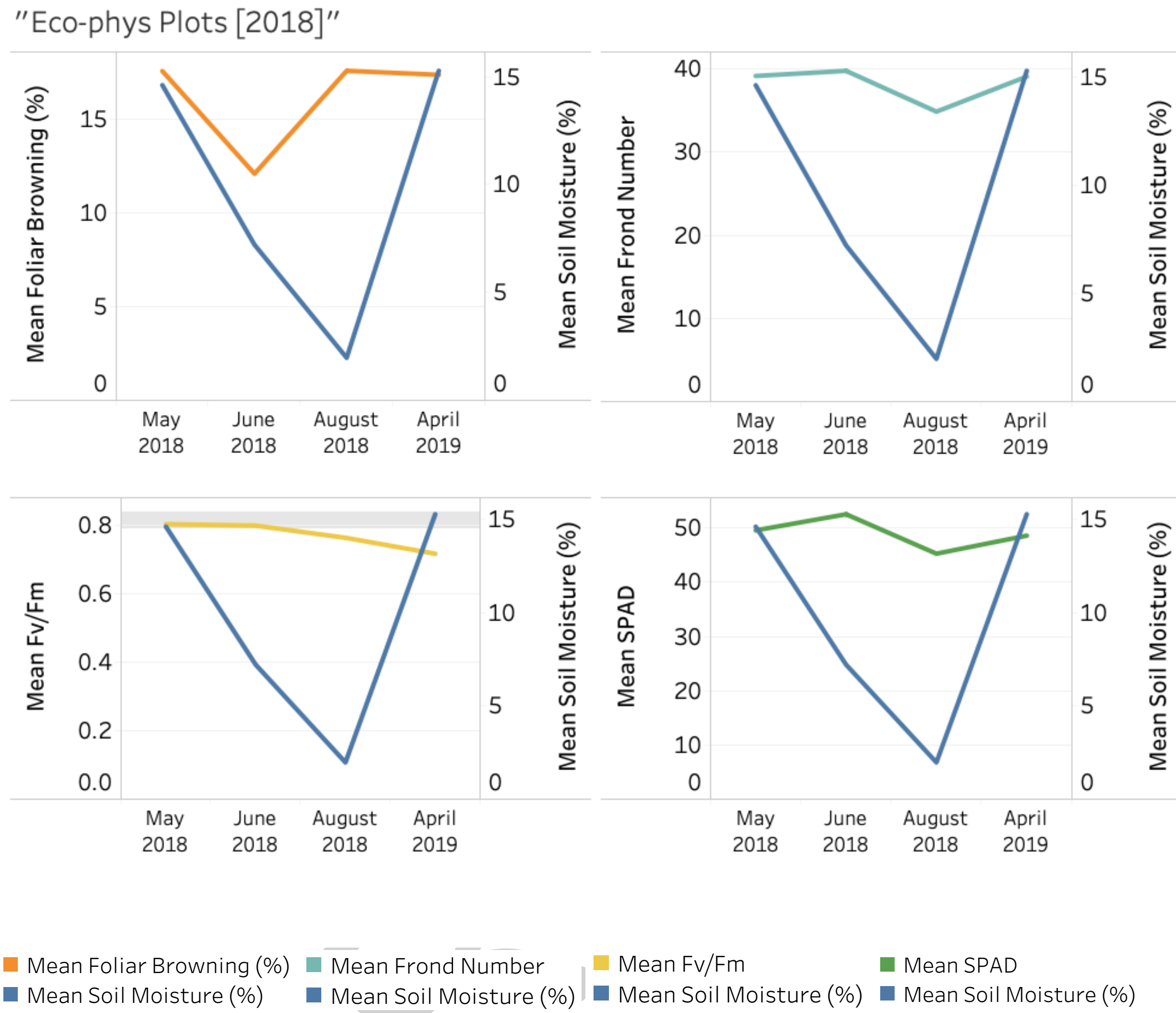


Figure 5. Changes in mean foliar browning (%; orange), mean frond number (teal), mean chlorophyll fluorescence (F_v/F_m; yellow) and mean chlorophyll content (SPAD; green) plotted against soil moisture (%; blue) measured on western sword fern plants in the "2018 Eco-phys plots" located throughout Seward Park, Seattle WA.

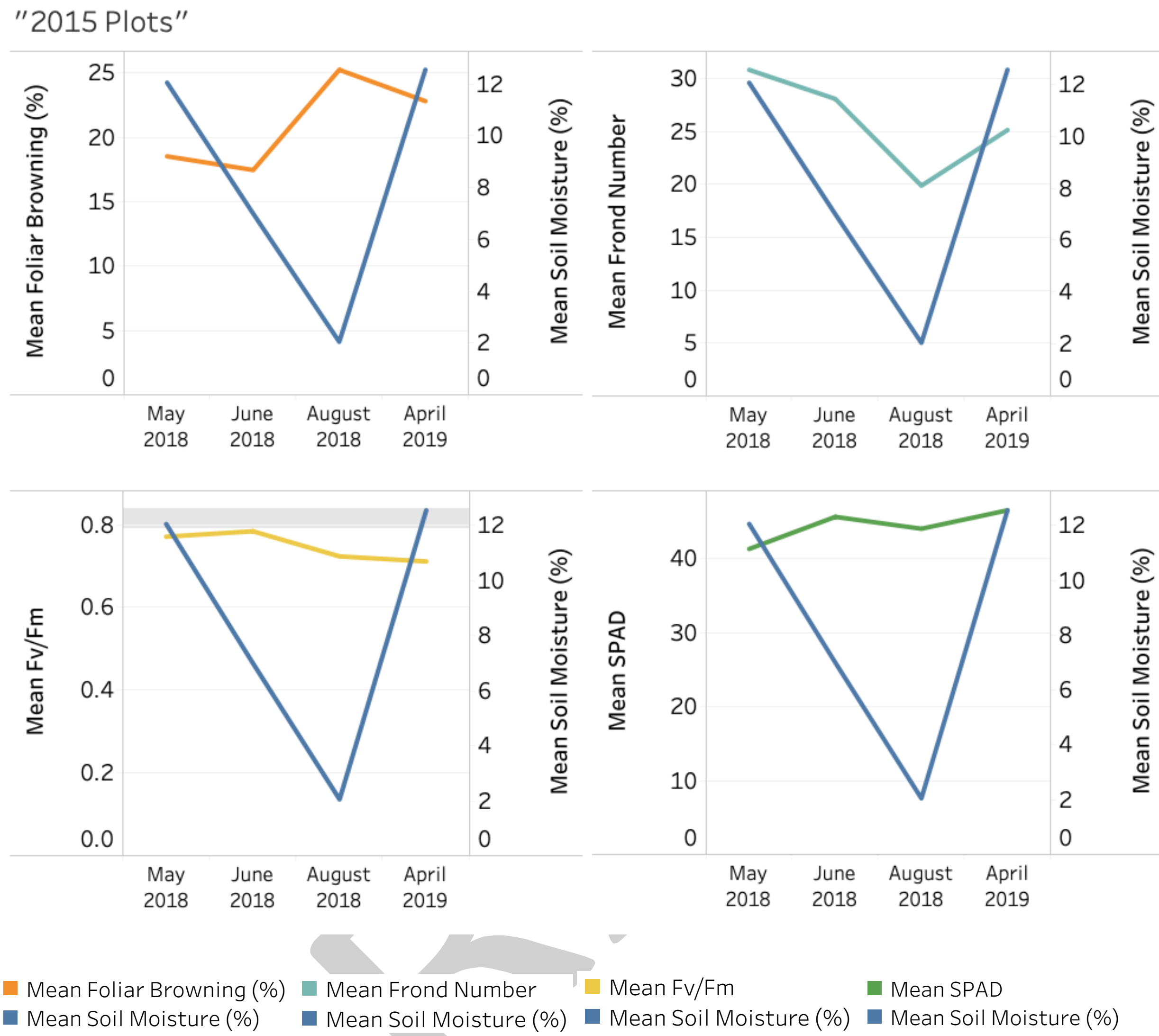


Figure 6. Changes in mean foliar browning (%; orange), mean frond number (teal), mean chlorophyll fluorescence (F_v/F_m; yellow) and mean chlorophyll content (SPAD; green) plotted against soil moisture (%; blue) measured on western sword fern plants in the "2015 monitoring plots" near the original decline location "Ground Zero" at Seward Park, Seattle WA. Ferns in these plots have been experiencing symptoms of decline over the course of the past three years.

Discussion:

First-year seedling survival ranged between 75 and 87%, which is above the regional estimates of average seedling survival (70-83%) for restoration plantings in Washington state (Warnick and Zaniewski, 2008). Transplant shock is a significant cause of plant stress (Paknejad et al., 2007) and can at least partially explain the negative response exhibited by seedlings outplanted on the “Ground Zero” site. Though these findings provide hope for the potential recovery, continued monitoring is needed to establish whether seedlings will persist past the first growing season.

While the different plot types in this study were assessed separately, they provide a basis for informal comparison across different sword fern populations within Seward Park (Figure 7, Table 1). Not surprisingly, seedlings planted in and surrounding the “Ground Zero” area experienced the most intense foliar decline (i.e. 31% increase in foliar browning and 62% decrease in frond number) as well as decreases in F_v/F_m and chlorophyll content (SPAD). Ferns in the “2015 plots”, which have been in decline over the course of the past three years, also showed a 4% increase in foliar browning and an 18% decrease in frond number compared to the previous spring. The healthier populations of sword fern in the “2018 Eco-phys” plots which were located further away from “Ground Zero” were the least impacted and showed no overall net change in foliar symptoms, despite significant impacts exhibited during the dry portion of the year (Table 1).

Although plots in the vicinity of “Ground Zero” tend to show the most persistent signs of physiological stress, these locations also have lower soil moisture content throughout the year (Figure 8). Thus, drought clearly plays a substantial role in plant stress and recovery. However, it remains unclear whether additional stresses are disproportionately exacerbating drought severity and/or interfering with the capacity of fern seedlings at to recover from prolonged periods of drought stress at “Ground Zero” compared to other areas of the park. To address this question, future efforts should examine the differences in canopy structure (i.e. light intensity and soil surface temperature) and soil parameters (i.e. type, moisture holding capacity) across Seward Park.

Evidence out of coastal California suggests that sword fern plants are remarkably resilient to drought and are able to recover fully from drought stress upon a substantial soil rehydration event (Baer et al., 2016; Pittermann et al., 2011). However, Baer et al. 2016 also highlight that it is unclear if several years of continued drought without full plant recovery can completely exhaust the belowground resources, leading to plant mortality akin to the “die-off” phenomenon observed at Seward Park. The inability of the sword fern plants in the “2015 plots” to recover following season drought, suggests that moisture levels either stay below those needed for rehydration, or that plant water relations may be compromised. A number of insects and vascular pathogens have been shown to inhibit plant capacity to recover from plant embolism and cavitation and exacerbate the impacts of seasonal drought (Castello et al., 1995; Kloepper et al., 2013; McElrone et al., 2003; Yadeta and Thomma, 2013). Thus, to fully understand the driving forces of decline, future work should focus more specifically on understanding the differences between water relations in “healthy” and “declining” ferns across a gradient of experimentally controlled soil moisture conditions.

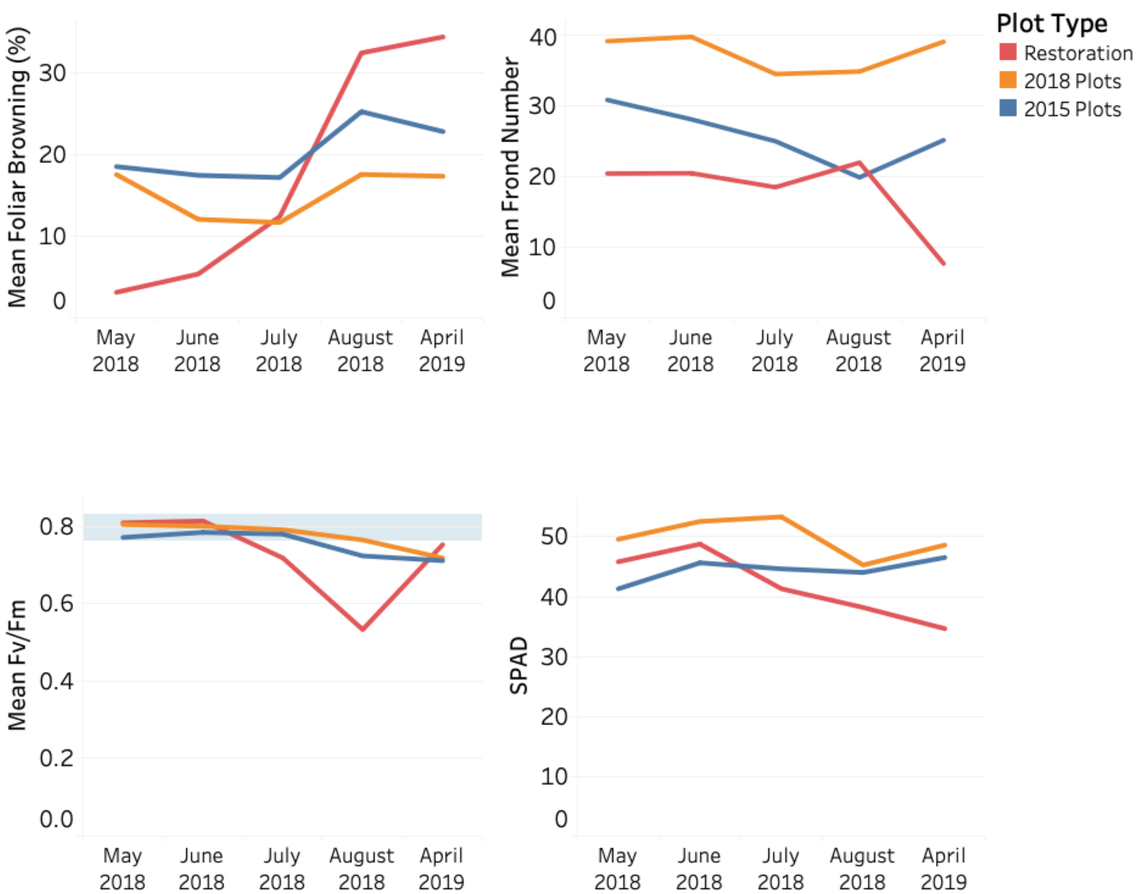


Figure 7. Summary of four ecophysiological measurements across five measurement points and three plot types (red – "restoration" plots, yellow – "2018 Eco-phys" plots, blue – "2015 plots) included in the ecophysiological assessment of western sword fern decline at Seward Park, Seattle, WA. Measurements included: mean foliar browning (%), mean frond number, mean chlorophyll fluorescence (F_v/F_m), and mean chlorophyll content (SPAD).

Lastly, we observed substantial variation in all measurement parameters between plots of the same type (Appendix Figures 1-8) indicating plot- and plant-level differences across sword fern populations within Seward Park. Taken together, the continued ecophysiological monitoring of sword fern populations throughout the 2019 growing season is critical to gaining a more comprehensive understanding of the factors driving sword fern responses to drought across space and time.

Table 1. Changes in mean foliar browning (%) and frond number across the three plot types located throughout Seward Park, Seattle, WA. Green upward arrows represent percent increase, while red downward arrows represent percent decrease.

Date	Mean Foliar Browning (%)	Change in Foliar Browning	Frond Number	Change in Frond Number
		"Restoration" Plots		
May-18	3.2	↑31%	20	↓62%
Jun-18	5.4		21	
Jul-18	12.4		19	
Aug-18	32.4		22	
Apr-19	34.4		8	
		2018 "Eco-phys" Plots		
May-18	17.6	0%	39	0%
Jun-18	12.1		40	
Jul-18	11.7		35	
Aug-18	17.6		35	
Apr-19	17.4		39	
		2015 Plots		
May-18	18.5	↑4%	31	↓18%
Jun-18	17.5		28	
Jul-18	17.2		25	
Aug-18	25.3		20	
Apr-19	22.8		25	

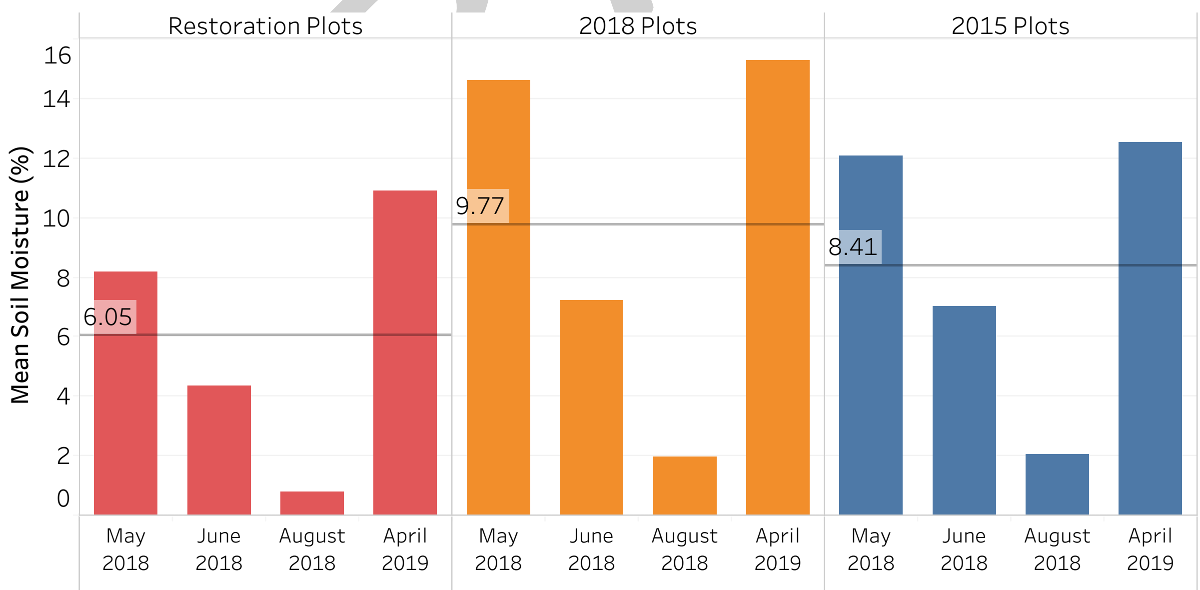


Figure 8. Mean soil moisture content (%) across the three plot types located throughout Seward Park, Seattle, WA. Red bars represent the "restoration" transect plots, yellow bars represent the 2018 "Eco-phys" plots located throughout Seward Park, and the blue bars denote the "2015 plots" located near "Ground Zero". The gray bar in each panel indicates the mean moisture percentage across plots between May 2018 and April 2019.

Appendix

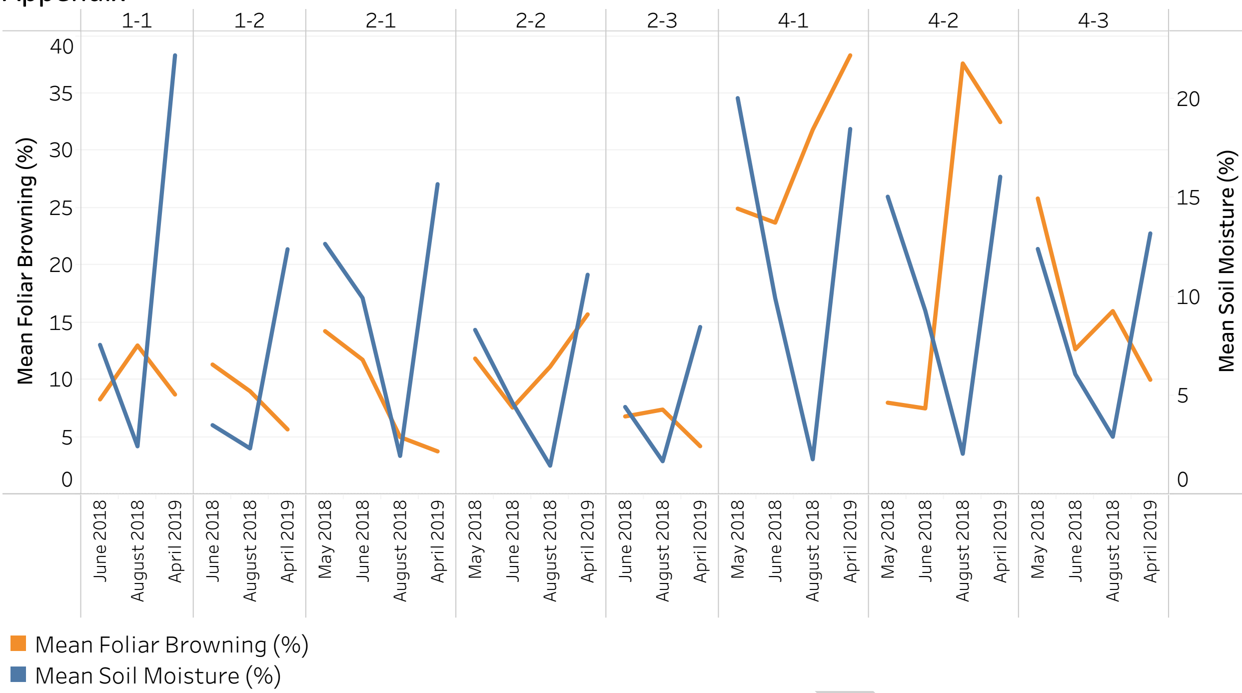


Figure 1. Mean foliar browning (%; orange) plotted against mean soil moisture (%; blue) across time, measured from sword fern plants in the "2018 Eco-phys" plots located throughout Seward Park, Seattle, WA. Each panel in the figure represents a mean value for all ferns within individual plots. Some plots do not have data for May and June and these measurement periods are thus excluded from some panels.

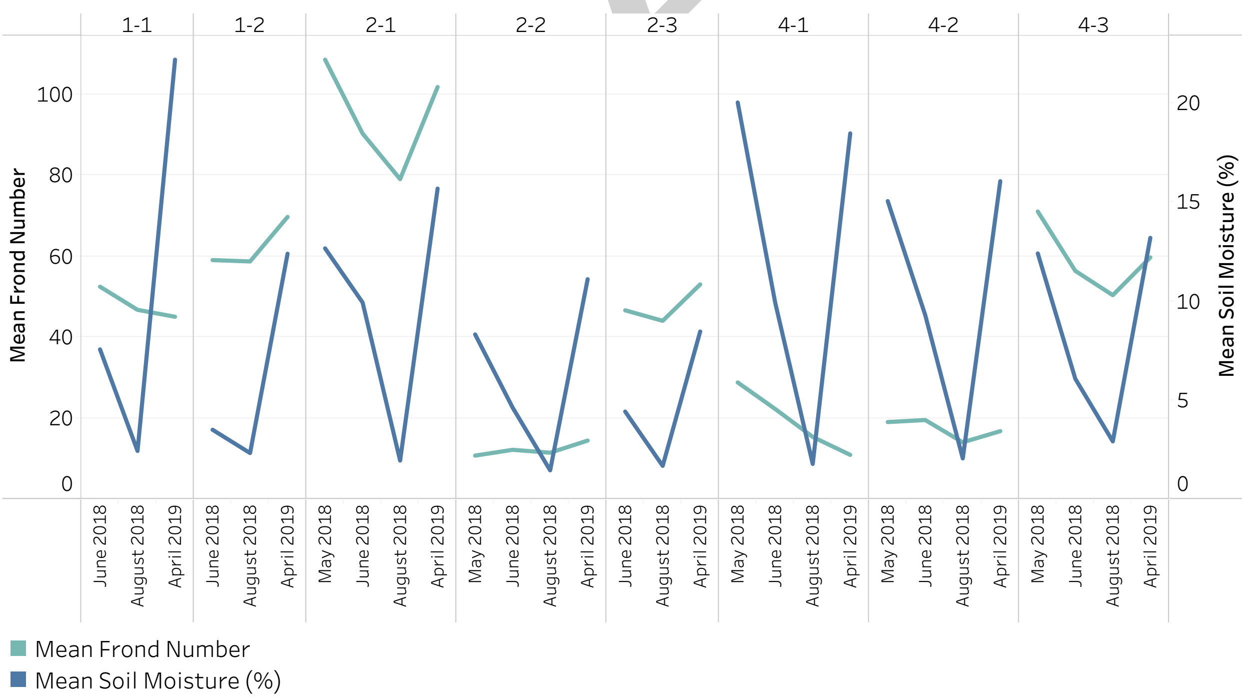


Figure 2. Mean frond number (teal) plotted against mean soil moisture (%; blue) across time, measured from sword fern plants in the "2018 Eco-phys" plots located throughout Seward Park, Seattle, WA. Each panel in the figure represents a mean value for all ferns within individual plots. Some plots do not have data for May and June and these measurement periods are thus excluded from some panels.

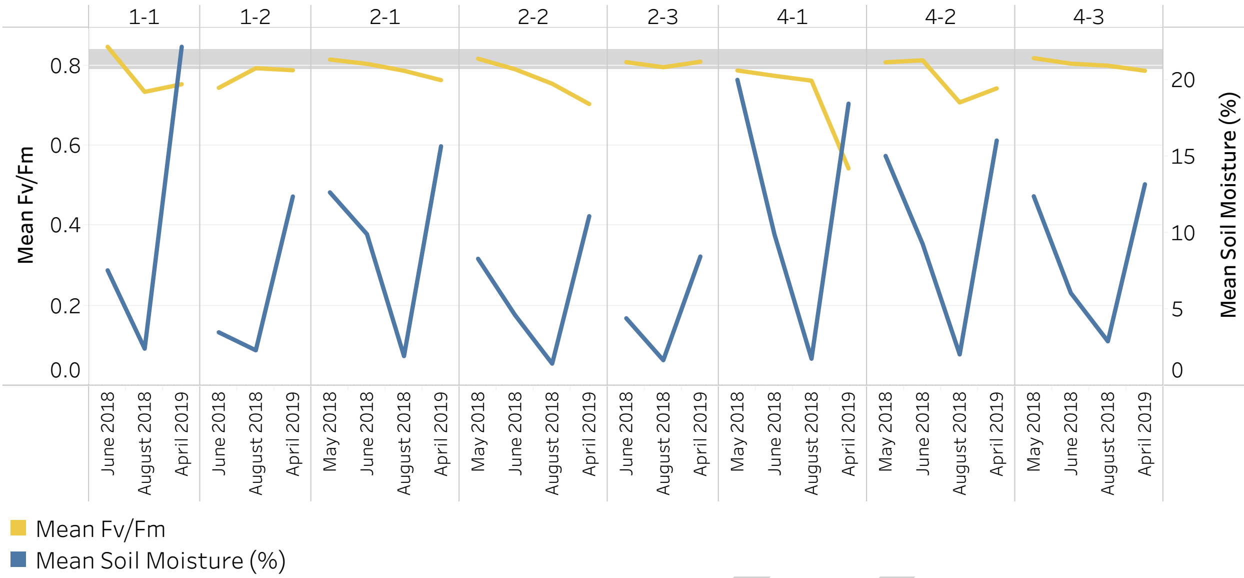


Figure 3. Mean chlorophyll fluorescence (F_v/F_m; yellow) plotted against mean soil moisture (%; blue) across time, measured from sword fern plants in the "2018 Eco-phys" plots located throughout Seward Park, Seattle, WA. Each panel in the figure represents a mean value for all ferns within individual plots. Some plots do not have data for May and June and these measurement periods are thus excluded from some panels.

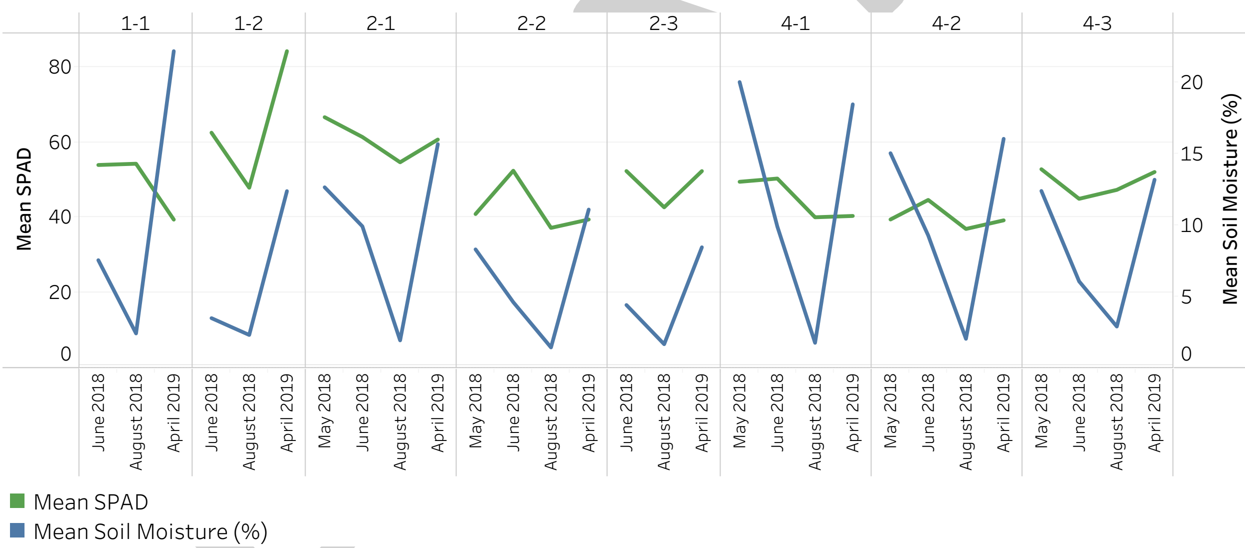


Figure 4. Mean chlorophyll content (SPAD; green) plotted against mean soil moisture (%; blue) across time, measured from sword fern plants in the "2018 Eco-phys" plots located throughout Seward Park, Seattle, WA. Each panel in the figure represents a mean value for all ferns within individual plots. Some plots do not have data for May and June and these measurement periods are thus excluded from some panels.

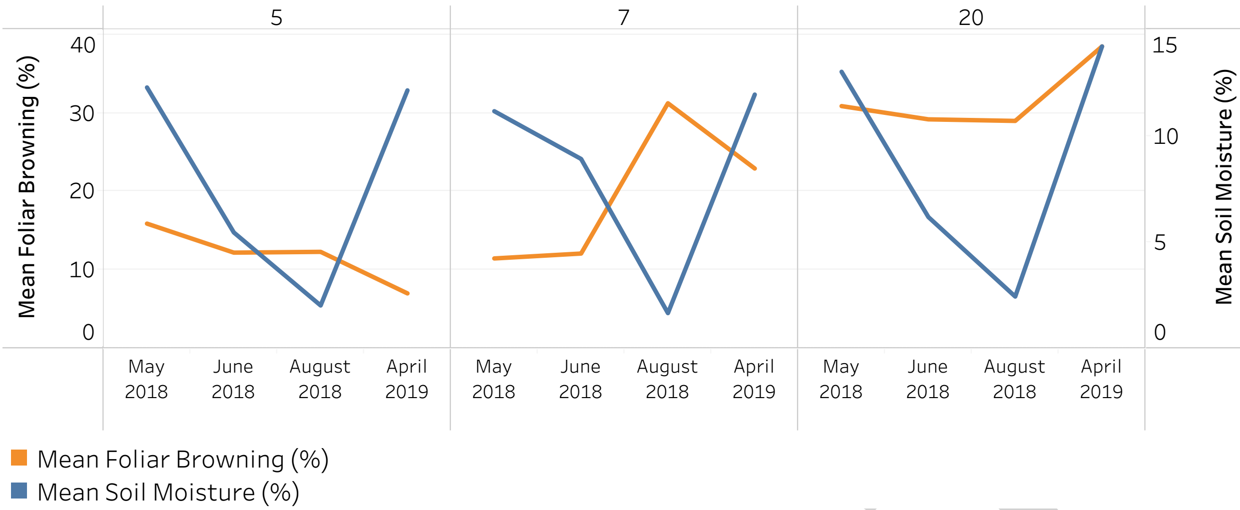
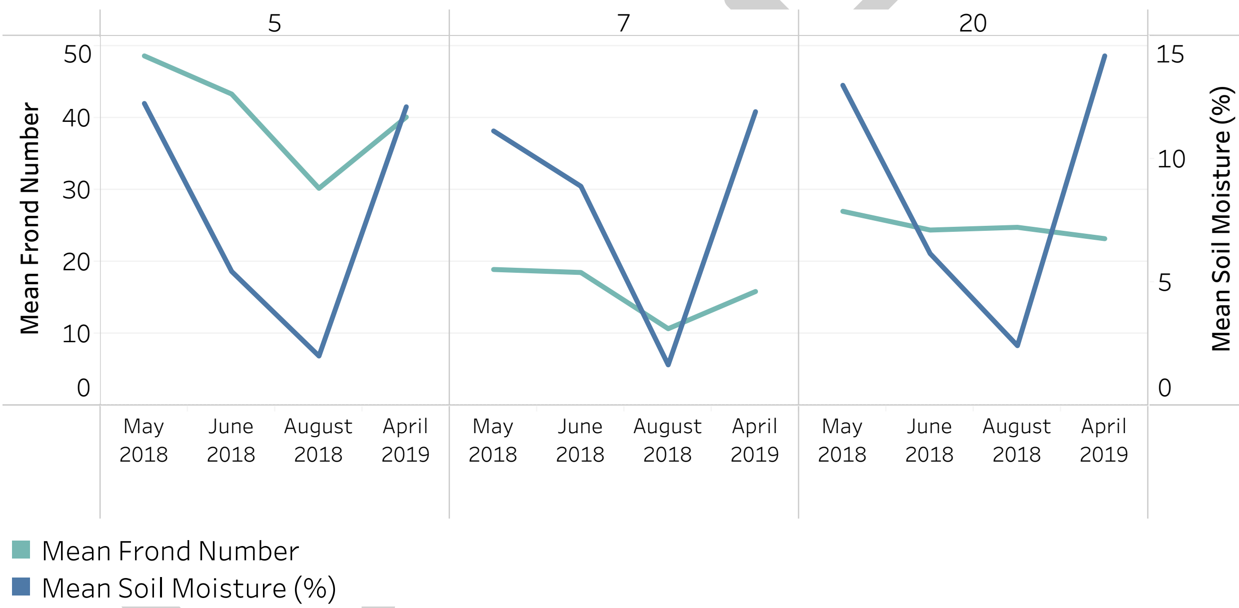


Figure 5. Mean foliar browning (%; orange) plotted against mean soil moisture (%; blue) across time, measured from sword fern plants in the “2015 plots” located near the original die-off location near “Ground Zero” at Seward Park, Seattle, WA. Each panel in the figure represents a mean value for all ferns within individual plots.



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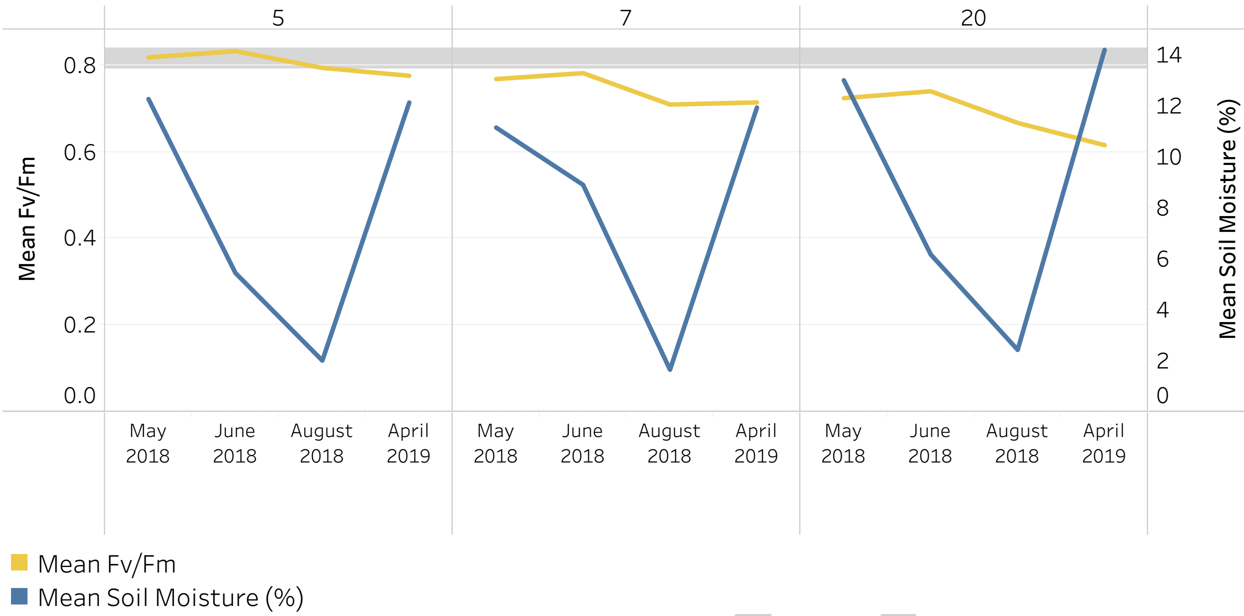

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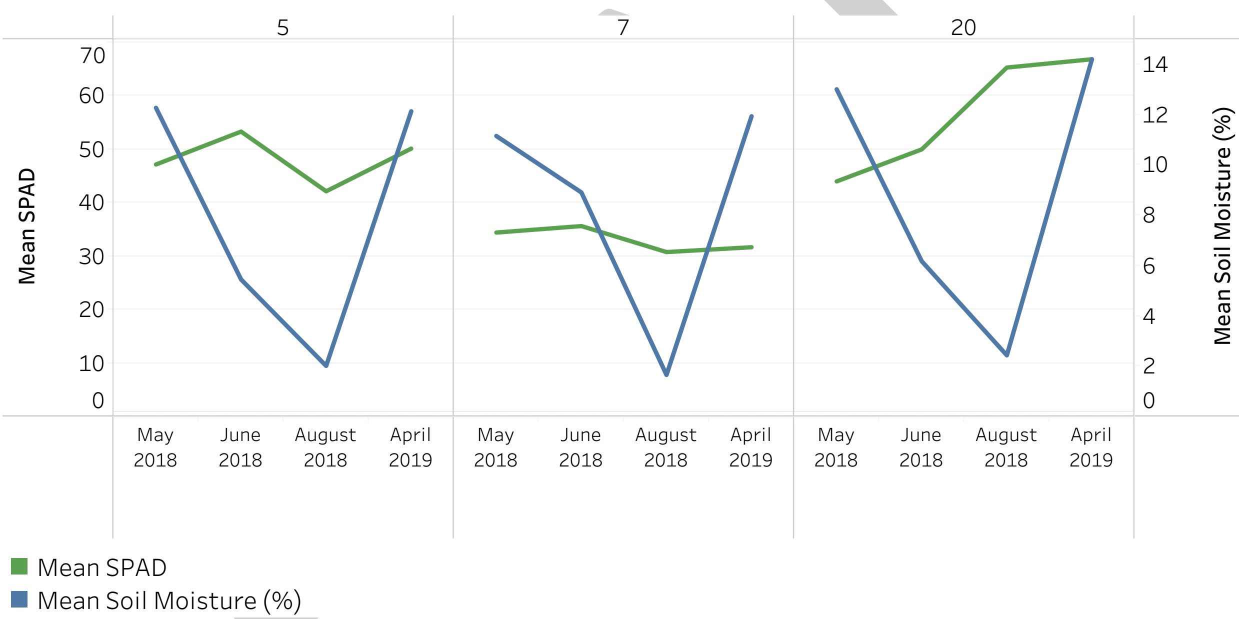

Figure 8. Mean chlorophyll content (SPAD; green) plotted against mean soil moisture (%; blue) across time, measured from sword fern plants in the "2015 plots" located near the original die-off location near "Ground Zero" at Seward Park, Seattle, WA. Each panel in the figure represents a mean value for all ferns within individual plots.

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