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Research Article

Drought stress affects interactions between potato plants, psyllid vectors, and a bacterial pathogen

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Abstract

Transmission of insect-borne pathogens is mediated by interactions between insects and plants across variable environments. Water stress, for example, affects the physiology, defense, chemistry, and nutritional balance of plants in ways that alter their tolerance to herbivores and pathogens. However, few studies have explored interactions between water stress and insect-borne pathogens as well as the molecular mechanisms mediating these interactions. Here, we address these knowledge gaps by assessing effects of plant water stress on the transmission of a bacterial pathogen, *Candidatus* Liberibacter solanacearum (CLs), by the vector *Bactericera cockerelli* Šulc (potato psyllid). We hypothesized that plant water stress would promote pathogen transmission by inducing plant gene transcripts and phytohormones involved in defense. Our results showed water stress was associated with decreased CLs titer with two psyllid haplotypes. Our analysis of plant gene transcripts suggested water stress affected phytohormone pathways in ways that altered plant tolerance to the CLs pathogen. Our study shows that abiotic stressors like drought may mediate the spread of plant pathogens by altering plant signaling pathways in ways that affect pathogen transmission.

Keywords: *Bactericera cockerelli*, *Candidatus* Liberibacter solanacearum, environmental context, species interactions, vector-borne pathogen, water stress

Introduction

Interactions between abiotic and biotic stressors affect the function of natural and managed ecosystems (Tillman et al. 2011, Atkinson and Unwin 2012). Water stress, e.g. can alter the spread of vector-borne plant pathogens by affecting vector fecundity and behavior as well as pathogen transmissibility (Davis et al. 2015, Martini and Stelinski 2017, Nalam et al. 2020). Interactions among abiotic and biotic stressors such as herbivores and pathogens may often be plant-mediated and driven by alterations of plant signaling pathways (Hara et al. 2012, Erb and Reymond 2019). However, few studies have assessed interactions between abiotic and biotic stressors while also conducting an examination of the molecular mechanisms underlying these interactions.

Water stress often increases plant attractiveness to insects and can mediate plant–insect–pathogen interactions by inducing changes in plants (Huberty and Denno 2004). For example, the concentration of sugars and amino acids, and defensive compounds like glucosinolates, often increase in plants subjected to water stress (Khan et al. 2010, Mewis et al. 2012). Plants respond to water stress by closing their stomata, a process largely controlled by the hormone abscisic acid (ABA; Hara et al. 2012), and induction of ABA can affect other hormones involved in herbivore and pathogen defense such as jasmonic acid (JA) and salicylic acid (SA; Szczepaniec and Finke 2019). As plants have limited energic capacity to express defenses, there is strong reason to predict that water stress could alter herbivore and pathogen tolerance through plant-mediated mechanisms.

Assessing how plant–vector–pathogen interactions are shaped by environmental context is key to protecting crops from disease, particularly as drought events become common (Zhao and Dai 2022). One area where this may be the case is the Columbia River Basin region in the US States of Washington and Oregon. Potatoes in this region are host to several hemipteran pests that transmit pathogens, such as potato psyllid (*Bactericera cockerelli* Šulc), the vector of *Candidatus* Liberibacter solanacearum ('CLs'), a Gram-negative α-proteobacterium that causes the disease zebra chip (Hansen et al. 2008, Butler and Trumble 2012). Aboveground symptoms of zebra chip can resemble those of other potato diseases like psyllid yellows and purple top and include leaf rolling, chlorosis and discoloring, aerial tubers, and plant death; and distinctive symptoms in the tuber like vascular browning, necrosis in the internal tissue, and streaking in the medullary ray tissue (Munyaneza 2012). Costs for potato psyllid control in Washington, Oregon, and Idaho are nearly $10 million annually, making potato crops unprofitable for many growers over the past decade (Greenway and Rondon 2018). While there are four identified *B. cockerelli* subpopulations (Northwestern, Western, Central, and Southwestern), the Western and Northwestern haplotypes are most common in this region (Swisher et al. 2015). These insects can survive outside of crop fields in shrub–steppe habitats with noncrop hosts, and thus are exposed to environmental stressors within and outside crop fields (Thinakaran et al. 2017). Under the Köppen–Geiger climate classification, continuous water stress during the summer is common for plants in this region (Beck et al. 2018) and overlaps with the potato growing season, although effects on *B. cockerelli* and CLs are largely unknown.

Here, we assessed the effects of continuous water stress on transmission of the CLs pathogen, and measured multiple traits

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of vectors and host plants that may mediate these effects. Prior research with *B. cockerelli* has shown that water stress may decrease the amount of feeding on potato plants but may increase overall psyllid survival (Huot and Tamborindeguy 2017, Nalam et al. 2020). We wanted to test for potential changes in plant defense and nutrition that may be driving the changes in behavior seen in previous studies. To accomplish this, we conducted experiments to test effects of water stress on CLs transmission and induction of gene transcripts associated with defensive-gene pathways and sugar biosynthesis. We hypothesized that water stress would limit the ability of plants to induce genetic pathways associated with pathogen defense. We, thus predicted there would be higher CLs titer in water stressed plants.

Methods

Study system

The *B. cockerelli* colonies used in this experiment were started using a colony descended from specimens collected from a potato field in Zillah, WA in 2013, and thereafter reared in a greenhouse on potato. The initial colony contained individuals of both Western and Northwestern haplotypes, which were isolated upon collection and then reared separately. To ensure each colony was not contaminated, haplotypes were confirmed with high resolution melting analysis at establishment and every 3 months thereafter (Swisher et al. 2012). After the initial Western and Northwestern colonies were established, we split these colonies in two and reared half the insects on plants infected with CLs and half on plants without CLs; these colonies contained infectious or uninfectious psyllids, respectively. Before experiments began, we confirmed insect infection status using PCR and CLs specific primers and took a sample of 10 psyllids from each colony to test CLs presence or absence in colony insects and found 100% infection in the CLs+ colonies and no infection in the CLs− colonies.

Effects of water stress on pathogen transmission

We first assessed effects of water stress on transmission of CLs by infectious psyllids of the Western and Northwestern haplotypes. To determine a continuous drought stress treatment that would visibly stress the plant without killing it within the duration of the experiment, we grew four potato plants for 8 weeks in a greenhouse. We then removed the aboveground biomass, took a measure of wet soil weight, and then took a dry soil weight measurement after baking soil at 100°C for 48 h. The average ratio between wet and dry soil weight was 4:1. In our experiment control plants thus received the amount of water lost from fully watered soil after drying (320 ml), and the water stress treatment was set at 25% of that amount of water (80 ml).

The experiment was initiated by planting Russet Burbank seed potatoes in 10 cm pots in a greenhouse with a 16:8 h photoperiod (light:dark) (21–24°C light; 16–18°C dark). Plants were watered three times a week for 2 weeks until there were 96 viable plants over 5 cm tall, with the average height of 15 cm. After this period, each plant was randomly assigned to a water treatment, with 48 plants receiving 100% water and 48 receiving 25% water thereafter. Plants received treatments for 7 d before psyllids were added. In total, four adult psyllids (two of each sex) from the infectious CLs colony were added to each plant in the CLs+ treatments, while the control plants received four noninfectious psyllids, to determine a baseline level of phytohormone-associated gene expression. We used only three replicates in the control compared to seven or eight replicates per treatments as we wanted less variation for our baseline measurements. This experimental setup was replicated four times so that we could collect tissue through destructive sampling at four separate psyllid exposure times (3 h, 1d, 2d, and 5d) without inducing a damage response in the plant. In the NW experiment, there were seven CLs+ replicates and three CLs− replicates at each time point for each stress treatment, while in the W experiment, there were eight CLs+ and three CLs− replicates. To sample, we removed three leaves total from the plant, one from the top, middle, and bottom of the plants. The experiment was a factorial 2 × 2 × 4 (two stress treatments, two infection treatments, and four time points). Replication varied between the NW and the W experiments because the *B. cockerelli* colony populations fluctuated heavily from week to week, making it difficult to determine how many psyllids would be available when the plants were ready. Leaves were wrapped in foil, placed in liquid N2 to flash-freeze, then chilled with dry ice and stored at −80°C. This experiment was in March 2019 using Northwestern haplotype psyllids and in May 2020 using Western haplotypes.

Analysis of pathogen titer and transcripts related to phytohormone signaling

For measuring CLs titer, plant leaves were wrapped in aluminum foil, frozen in liquid N_2, and snap chilled in dry ice before storing in −80°C. Samples were ground into powder in liquid N_2 using mortar and pestles. Homogenized tissue (100 mg) was used for RNA extraction using Promega SV kits (Promega, Madison, WI) and cDNA from 1 μg of total RNA using Bio-Rad iScript cDNA synthesis kits. CLs primers (Table S1, Supporting Information) were used in qRT-PCR reactions (10 μl) containing 3 μl of ddH_2O, 5 μl of iTaq Univer SYBR Green Supermix, 1 μl of diluted primer mix [forward and reverse (concentration 10 μM)], and 1 μl of diluted (1:25) cDNA template. The qRT-PCR program included an initial denaturation for 3 min at 95°C followed by 40 cycles of denaturation at 95°C for 15 s, annealing for 30 s at 60°C, and extension for 30 s at 72°C. For melting curve analysis, a dissociation step cycle was added (55°C for 10 s, and then 0.5°C for 10 s until 95°C). The relative transcript abundance of CLs (per 100 mg biomass) at different times were calculated using the delta-delta Ct method, ($2^{-\Delta\Delta Ct}$) with *Solanum tuberosum–Elongation factor1 α* (*StEF1α*) as a housekeeping gene, which gives us a baseline to compare transcript abundance of our genes of interest (Kozera and Rapacz 2013, Otulak-Kozieł et al. 2020).

To assess if water stress affected phytohormones in host plants, we measured relative transcript abundance of three defense-associated genes, *Pathogenesis-related Protein 1* (*StPRIb*) (downstream of SA signaling; Makarova et al. 2018), *Protease Inhibitor II* (*StPIN2*) (downstream of JA signaling; Herde et al. 1999), and *9-cis-epoxycarotenoid dioxygenase 1* (*NCED1*) (upstream ABA biosynthetic gene, catalyzing oxidative cleavage of 9-cis-epoxycarotenoids neoxanthin and violaxanthin to xanthoxin, a key step in the ABA biosynthetic pathway; Changan et al. 2018, Liu et al. 2020) over multiple time points. To test the effects of water stress on nutritional quality, we measured relative transcript abundance of a sugar biosynthetic gene, *Sucrose Synthase 4* (*SuSy4*), which encode a cytoplasmic enzyme, glycosyl transferase responsible for sugar metabolism mainly in the plant sink organs (Van Harsselaar et al. 2017, Stein and Granot 2020). We hypothesized that water stress may reduce induction of sugar biosynthesis. After treatments were complete, aboveground plant tissue was harvested and prepped for RNA extraction as described earlier. After processing, primers specific to each gene transcript (Table S1, Supporting Information) were used in qRT-PCR reactions (10 μl) fol-

lowing procedures described earlier for measuring relative transcript accumulation of CLs. Relative transcript abundances of *PR1b*, *PIN2*, *NECD1*, and *SuSy4* were then calculated using the delta-delta Ct method, ($2^{-\Delta\Delta Ct}$) with *StEF1α* as a housekeeping gene (Livak and Schmittgen 2001, Kozera and Rapacz 2013). CLs infection in the infected plants were confirmed through PCR using CLs specific primers. The 2019 plants were tested for *PR1b*, *PIN2*, and *NECD1* and the 2020 plants were tested for *PR1b*, *PIN2*, *NECD1*, and *SuSy4*.

Data analysis

Data from each experiment were analyzed using general linear models. To analyze factors affecting CLs transmission, the model had water, treatment, and time as explanatory variables, and CLs delta CT (ΔCT) values as the response, with each psyllid haplotype analyzed separately. To analyze the factors affecting phytohormone-associated gene expression, water and pathogen treatments were the explanatory variables. For each gene of interest, a separate model was used, with the ΔCT value of each gene transcript as the response. The CLs results were subset to 1, 2, and 5 d, because there was no transmission at 3 h in the Western haplotype experiment. Phytohormone-associated gene expression increased over time, but there was no change in the relationship between treatments at any timepoint (Figures S1–S3, Supporting Information), so data was subset to 5 d and reanalyzed without time as a factor.

We also used least square means for each analysis, which were calculated using the emmeans package in R statistical software (Lenth et al. 2019, R Core Team 2022). The fold change in gene titer was analyzed with ANOVA to examine the overall effect of treatment on delta CT values. Parameter estimates and subsequent calculations for delta-delta CT ($2^{-\Delta\Delta Ct}$) are plotted on the log10 scale. Delta-delta CT values are a measure of relative expression and compare the estimated marginal mean of each treatment to the mean of our control (100 CLs−). We use this method to graphically compare gene expression because it is a more precise measure than the ΔCt used in general linear models.

Results

Effects of water stress on CLs titer

We found that water stress is associated with CLs titer and that water stressed plants had lower titer than full water control plants, but only in one of the experiments. We also see relative expression increase over time in both experiments. In the Northwest experiment, there was no significant difference in mean CLs titer between stressed and control plants ($\chi 2 = 0.31$, $P = .57$), and changes in titer over time were similar across treatments ($\chi 2 = 0.81$, $P = .67$). The relative expression at each time point also has high standard error and no significant differences (Fig. 1A, Table 1). In contrast, the Western experiment had water stressed plants with significantly lower CLs titer compared to control plants ($\chi 2 = 13.6$, $P < .001$) and these differences in relative titer increased over time ($\chi 2 = 22.7$, $P < .001$), which can also be seen in the relative expression (Fig. 1B, Table 1). Expression is not only higher overall in the nonstressed treatment, it also increases more over time. The stressed plants go from an EFC of 0.320 at 1 d to 1.938 at 5 d, while the unstressed plants go from 1.0 to 6.056 (Table 1).

Effects of water stress and CLs on plant relative gene transcript expression

When we examine the expression of genes related to defensive hormone synthesis, we do not find any consistent correlations across both sets of experiments that suggest the difference in CLs titer is caused by a plant defensive response. In the NW experiment, the ABA-associated gene transcript *NCED1* is significantly affected by pathogen presence ($\chi^2 = 14.0837$, $P = .002$), but not water stress. Within CLs+ treatments, stressed plants have an EFC of 5.53 compared to the nonstressed, with an EFC of 2.09 (Table 1; water stress × pathogen interaction: $\chi^2 = 4.4427$, $P = .035$; Fig. 2A). In the Western haplotype experiment, water stress affected *NCED1* ($\chi^2 = 20.1$, $P < .001$), but CLs did not ($\chi^2 = 1.13$, $P = .29$), and there was no significant interaction between water stress and pathogen infection ($\chi^2 = 0.073$, $P = .39$; Fig. 2B). Here, we see that all treatments had lower expression relative to the control, with stressed treatments decreasing more than the 100 CLs+ (Table 1, Fig. 2B).

The relative transcript abundance of the SA-associated gene transcript (*PR1b*) was significantly lower when CLs was present in the Northwest haplotype experiment, with fold change more than half of the control (Table 1; $\chi^2 = 11.29$, $P = .007$). However, this gene transcript was not significantly affected by water stress ($\chi^2 = 1.92$, $P = .17$) or the interaction between water stress and CLs ($\chi^2 = 0.040$, $P = .84$; Fig. 3A). While the same pattern of relative expression for *PR1b* was present in Western haplotype experiment, the effects of water stress ($\chi^2 = 0.24$, $P = .62$), CLs infection ($\chi^2 = 2.22$, $P = .13$) and the interaction between water stress and pathogen infection ($\chi^2 = 0.17$, $P = .68$) were not significant, likely due to the higher associated standard error (Fig. 3B, Table 1).

The relative transcript abundance of the JA-associated gene transcript (*PIN2*) was not significantly affected by CLs in the Northwest haplotype experiment ($\chi^2 = 3.47$, $P = .062$), and there was no effect of water stress ($\chi^2 = 0.017$, $P = .90$) nor an interaction between water stress and CLs ($\chi^2 = 0.53$, $P = .47$; Fig. 4A). We see that all treatments had higher EFC than the control, but that the largest fold change is in the 100 CLs+ treatment. In the Western haplotype experiment, there is a significant interaction between CLs and water stress ($\chi^2 = 6.97$, $P = .0083$; Fig. 4B). The 25% CLs+ treatment is the only one where fold change is higher than control, but water stress ($\chi^2 = 0.26$, $P = .61$) or infection ($\chi^2 = 0.99$, $P = .32$) have no effect by themselves (Fig. 4B).

Discussion

We expected water stress would heighten induction of ABA and weaken plant defense by suppressing induction of SA, an outcome that has been observed in tomatoes (Thaler and Bostock 2004) and *Arabidopsis* plants (Asselbergh et al. 2008). While water stress affected CLs transmission, it was not in the direction we hypothesized. We found that water stress was correlated with lower relative CLs titer in experiments with both psyllid haplotypes (Fig. 1), but did not correlate with any consistent pattern of defensive gene induction (Figs 2–4). This suggests continuous water stress may have affected pathogen transmission by altering psyllid behavior or plant physiology. However, given the research on how pulsed water stress affects plants and hemipteran feeding behavior (Mody et al. 2009), it may be that intermittent or less severe water stress would have a different effect. In a study of continuous- and pulsed-drought stress on brassica-feeding aphids, the highest growth rate and fecundity occurred on medium stressed plants (50% of control water treatment), and pulsed-stressed plants had higher foliar nitrogen concentration without a significant change in glucosinolate concentration (Tariq et al. 2012).

We expected that the continuous water stress would induce the ABA-biosynthetic gene transcript (*NCED1*), which can mediate induction of the JA and SA pathways as well (Szczepaniec and Finke 2019, Kapoor et al. 2021). Research on how drought and in-

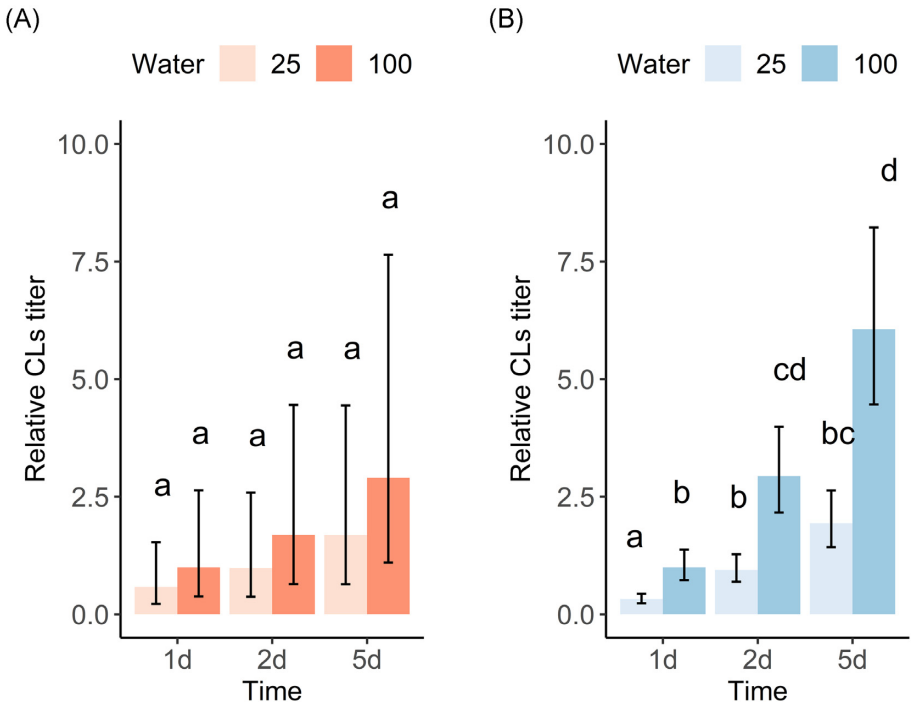

Figure 1. Relative CLs titer (per 100 mg plant biomass) over time (1, 2, and 5 d) in experiments with the (A) Northwestern and (B) Western *B. cockerelli* haplotypes. Error bars are 1 SE, and letters indicate a significant difference between treatments within a time point ($\alpha = 0.05$). Cls titer increases over time in both experiments, with titer significantly higher in the 100% water treatments.

Table 1. A list of mean estimated fold change (EFC) values for each treatment and gene of interest, with the upper and lower standard error (SE) and Tukey letter. These results correspond to the bar plots of each gene of interest.

Haplotype	Gene	CLs	Water	EFC	Upper SE	Lower SE	.group
Northwestern	NCED1	CLs−	25	0.394291	0.672118	0.231307	a
Northwestern	NCED1	CLs−	100	1	1.704624	0.58664	ab
Northwestern	NCED1	CLs+	25	5.534773	7.847589	3.903582	c
Northwestern	NCED1	CLs+	100	2.099115	2.976273	1.480471	bc
Northwestern	PIN2	CLs−	25	1.639588	2.739949	0.981131	a
Northwestern	PIN2	CLs−	100	1	1.67112	0.598401	a
Northwestern	PIN2	CLs+	25	2.682487	3.754314	1.916659	a
Northwestern	PIN2	CLs+	100	3.080352	4.311151	2.200937	a
Northwestern	PR1b	CLs−	25	1.728955	2.755369	1.084894	a
Northwestern	PR1b	CLs−	100	1	1.593662	0.627486	ab
Northwestern	PR1b	CLs+	25	0.457201	0.586533	0.356387	bc
Northwestern	PR1b	CLs+	100	0.307159	0.394047	0.23943	c
Western	NCED1	CLs−	25	0.394291	0.488541	0.318224	a
Western	NCED1	CLs−	100	1	1.239036	0.807079	b
Western	NCED1	CLs+	25	0.3799	0.433183	0.333171	a
Western	NCED1	CLs+	100	0.711374	0.811147	0.623872	b
Western	PIN2	CLs−	25	0.316074	0.476639	0.209598	a
Western	PIN2	CLs−	100	1	1.507999	0.66313	ab
Western	PIN2	CLs+	25	1.089563	1.401201	0.847236	b
Western	PIN2	CLs+	100	0.570876	0.734158	0.443909	ab
Western	PR1b	CLs−	25	1.728955	3.32928	0.897877	a
Western	PR1b	CLs−	100	1	1.925603	0.519318	a
Western	PR1b	CLs+	25	0.642486	0.891553	0.462999	a
Western	PR1b	CLs+	100	0.570886	0.792196	0.411402	a

sect feeding affect ABA found that drought upregulated ABA signaling, which decreased SA-dependent defense (Guo et al 2016). Antagonistic interactions between the ABA pathway and *PR* genes have been found in grapes (Hatmi et al 2018) and rice (Xu et al 2013). When we look at the overall expression across infection treatments, we do see higher *NCED1* and lower *PR1b* in the CLs+ treatment, suggesting the antagonistic interaction we hypothesized, but without an associated increase in pathogen titer. This suggests that decreased expression of the SA-pathway alone is not enough to affect pathogen susceptibility. The ERF branch of the JA pathway is also activated by infection (Aerts et al 2021), but while we do see a higher mean EFC of the JA-associated *PIN2*

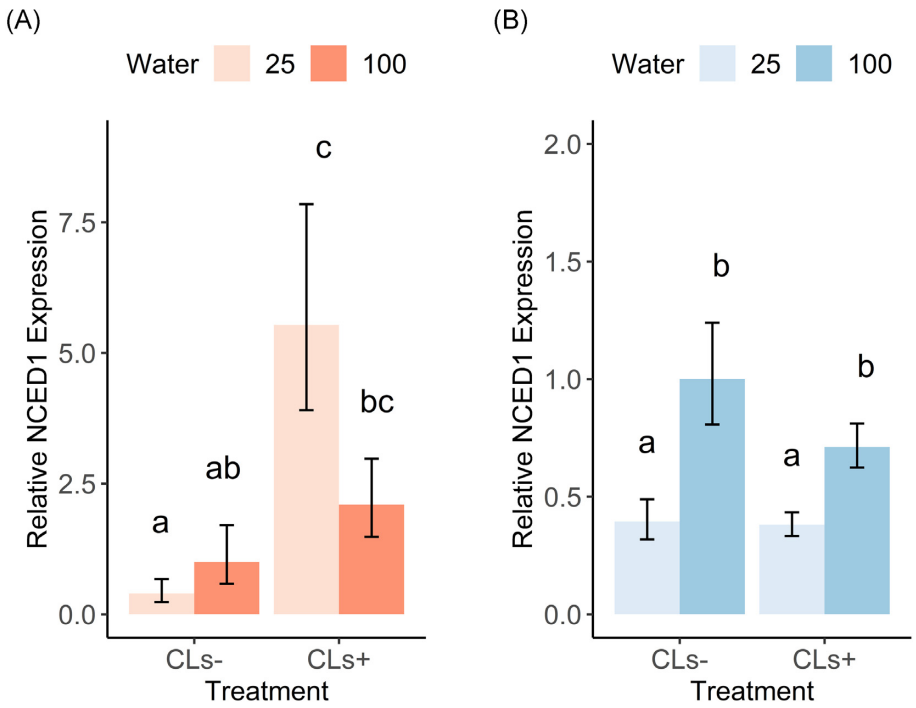

Figure 2. Relative gene transcript expression of the *NCED1* gene (ABA) after 5 d in experiments with the **(A)** Northwestern and **(B)** Western *B. cockerelli* haplotypes. Error bars are 1 SE, and letters indicate a significant difference between treatments within a time point ($\alpha = 0.05$). In the NW experiment, expression is significantly affected by infection but not water stress, while the W experiment is affected by water stress but not infection.

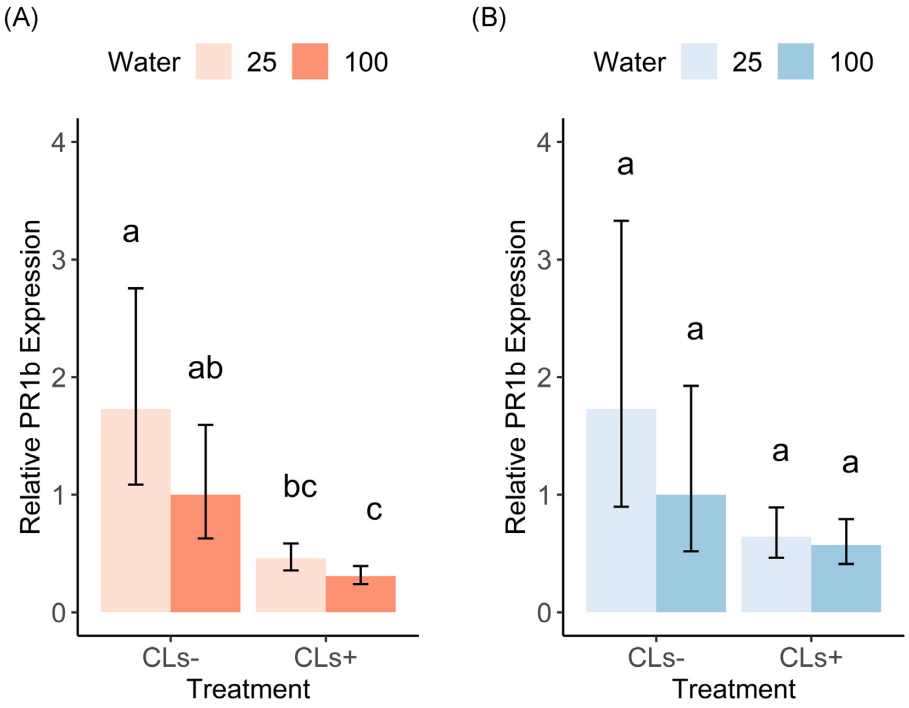

Figure 3. Relative gene transcript expression of the *PR1b* (SA) gene after 5 d in experiments with the **(A)** Northwestern and **(B)** Western *B. cockerelli* haplotypes. Error bars are 1 SE, and letters indicate a significant difference between treatments within a time point ($\alpha = 0.05$). In the NW experiment, expression is significantly affected by infection but not water stress, and there are no significant interactions in the W experiment.

gene, there are no consistent patterns. Furthermore, water stress only affected ABA in Western haplotype experiments with relatively weak gene expression differences across treatments (Fig. 2). CLs infection affected plant gene expression only in the Northwest haplotype experiment, while water stress did not, and interactions between these factors were minimal. These results make any inference about the effects of these stressors on plant signaling pathways difficult and did not provide the clear relationship between water stress treatment and induction of ABA that we expected.

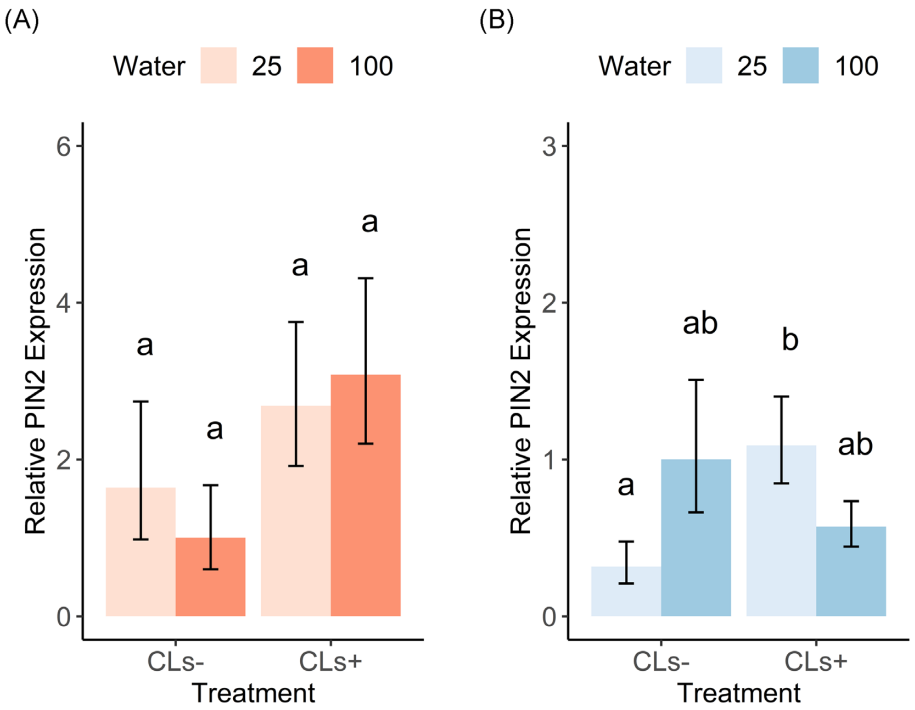

Figure 4. Relative gene transcript expression of the *PIN2* (JA) gene after 5 d in experiments with the **(A)** Northwestern and **(B)** Western *B. cockerelli* haplotypes. Error bars are 1 SE, and letters indicate a significant difference between treatments within a time point ($\alpha = 0.05$). In the NW experiment, there are no significant interactions, while in the W experiment there only the interaction between water stress and infection is significant.

While studies on *B. cockerelli* and water stress have found that it can decrease feeding behavior (Huot and Tamborindeguy 2017, Nalam et al. 2020) we still predicted that water stress and CLs would generate differences in induction of plant defenses. In the Northwest haplotype experiments, CLs infection affected each gene transcript of interest, where water stress only affected *PR1b*. In the Western haplotype experiment, water stress only affected induction of *NCED1*, but CLs had weak effects. While there appeared to be some support in our data of negative crosstalk between the SA and JA/ABA pathways in the Northwest haplotype experiment, the same was not observed in the Western haplotype experiment (Figs 3 and 4). This reflects the complexity of plant chemical defense. In a study that also combined water and pathogen stress, JA and SA biosynthesis were found to be enhanced by the combined stressors and negatively regulated ABA, but these effects were not observed when only a single stressor was applied (Gupta et al. 2020).

Water stress can also influence pathogen transmission by influencing vector life history and behavior. *B. cockerelli* had increased survival on water-stressed mature tomato plants (Huot and Tamborindeguy 2017), which may be because of the potential for increased nutrient availability in water-stressed plants (Khan et al. 2010, Nachappa et al. 2016). Soybean aphids have reduced transmission of *soybean mosaic virus* on drought-stressed plants because of shorter feeding bouts (Nachappa et al. 2016). Similarly, CLs-infected *B. cockerelli* tend to feed for longer on unstressed plants compared to stressed plants, and water stress decreases the frequency of feeding (Nalam et al. 2020). We found that CLs+ treated plants had lower *PR1b* and higher *PIN2* titer (Figs 3 and 4), suggesting that they may have been under herbivory stress for longer due to increased feeding activity. This suggests vector behavior may mediate infection more than changes in plant chemistry. Vector choice may also be affected by the physical effects of water stress on plants, as water stress may alter feeding behavior by decreasing the sugar to water ratio in the phloem (Khan et al. 2010), and stressed plants can become more sugar dense without increased sugar biosynthesis. Thus, while water stress may not affect the survival of *B. cockerelli* directly, it may affect psyllid feeding behaviors and indirectly decrease pathogen transmission.

Our results add to the literature suggesting water stress alters tolerance to pathogens in plants (Nachappa et al. 2016, Del Cid et al. 2018, Nalam et al. 2020). However, further research may be needed to untangle the interactions between defensive pathways. When there is simultaneous defense against an insect predator and a pathogen, there are complex interactions occurring that go beyond the defensive pathways studied here. Our observed results may also vary based on the way a plant experiences herbivory by a pathogen-carrying insect. A paper on pea plant responses to herbivory by both a vector and nonvector herbivore found that the plant exhibited different defense responses depending on the order of attackers. When the plant was first infected by the vector insect, then fed by the nonvector, herbivory inhibited the antipathogen defense signals. When the order was reversed, pathogen infection enhanced antiherbivory defense signaling (Basu et al. 2021).

Research suggests that water stress can mediate pathogen transmission through altering feeding behavior. The act of phloem-feeding requires a piercing–sucking insect to puncture the cell wall and ingest the phloem, and the thickness or viscosity of those plant elements depends on the amount of water available. In the soybean aphid system, water stress increases phloem sap quality but is associated with decreased feeding time, and thus decreased virus transmission (Nachappa et al. 2016). A similar result was also observed in the potato psyllid system, with CLs-infected psyllids spending longer in the salivation and ingestion stages of feeding on well-watered plants, while infection status did not affect feeding on drought-stressed plants (Nalam et al. 2020). Under no-choice conditions, pathogen transmission in-

creased with increasing water stress in grapevines, but when vectors were given a choice between uninfected, well-watered plants and stressed-infected plants, there was a decrease in transmission with increasing water stress (Del Cid et al. 2018).

While water stress is not always associated with decreased pathogen transmission in crop systems, there are systems where a possibility of harnessing it for management exists. Records of drought indices all show increasing aridity since 1950, and global drought frequency is expected to greatly increase even under the RC4.5 low-to-moderate emissions scenario (Zhao and Dai 2022). As these new climate norms are forcing producers to adjust approaches to integrated pest management at every level, we may be able to build systems that manipulate water stress to control insect feeding and pathogen transmission. However, the other effects of a changing climate will need to be balanced as well. A warming climate may decrease the development time for insects (Bale et al. 2002) and increase risk of pathogen transmission, and that same climate may alter plant attractiveness to vectors. Further research on physiological effects of water stress on vector feeding and survival may aid in elucidating the mechanisms that protect water-stressed plants from herbivory and vector-borne pathogens. More broadly, if water stress affects pathogen transmission in an agroecosystem, irrigation strategies may be developed to find the minimum water needed to maintain yields while reducing risk from disease.

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Supplementary data

Supplementary data are available at FEMSEC online.

Conflict of interest statement. None declared.

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