Sodium Chloride Application Decreases Photosynthetic Activity in *Vitis* spp. Regardless of Exposed Rootstock Selection

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Abstract

As our climate changes and temperatures rise, water scarcity and high evapotranspiration has led to a greater buildup of sodium chloride in agricultural soils. Grapevines, like most perennial crops, are glycophytic and lack sufficient responses to high soil salinity in their immediate rooting zone. Salt toxicity can lead to detrimental effects on growth, yield, and overall vine health. There is a need to understand how NaCl affects grapevine photosynthesis and address the potential for different forms of salt tolerance in grapevines. Measurements of transpiration, stomatal conductance, and net assimilation rates were conducted across two trials on grapevine rootstocks exposed to NaCl concentrations of 0, 25, 50, 75, and 100mM. Because several rootstock selections varied between experiments, five selections were shared across both trials for continuity. The preliminary trial tested the photosynthetic response of eight rootstocks when irrigated with NaCl solution at concentrations of 0, 25, 75, or 100mM NaCl over 21 days. Differences between selections were only significant at concentrations of 75mM or greater. A second trial was conducted with nine rootstock selections all grafted to Cabernet Sauvignon and similarly exposed to 50mM NaCl. Three measurement time points showed a delineation in photosynthetic response early in the latter trial. However, the observed differences were no longer significant following 21 days of exposure. These trials quantify the response of different *Vitis spp.* rootstocks under varying NaCl concentrations. More importantly, they explore the potential for translating salt tolerant qualities of rootstocks to grafted scions in field conditions.

Introduction

With a rapidly increasing global population, higher water prices, and the desire to maximize the utility of scarce resources, there has been a move to use reclaimed water for irrigation in many vineyards worldwide (Laurenson et al., 2012). Among the concerns is the accumulation of chlorine and sodium in reclaimed water. Unlike many perennial crops, grapevines are relatively tolerant of high sodium levels in the rhizosphere. However, chlorine in the root-zone can be excessively damaging and result in early leaf senescence, poor fruit set, lower transpiration rates, decreased carbon assimilation rates, and high levels of NaCl in ripe berries (Moya et al., 2003; Munns and Tester 2008). These responses may directly contribute to poor photosynthetic capacity, lowered yields, less vigorous shoot growth, and the harvest of unsellable fruit of poor quality. Chloride accumulation in vineyard soils can be an issue of great concern and is likely to become more prevalent as agriculture changes to accommodate a growing global population.

The accepted threshold for what is considered a saline soil in vineyards is vague. These studies have recorded changes in growth rate, total biomass accumulation, photosynthetic response, and more parameters in response to varying applied NaCl concentrations.

Using nine rootstock-genotypes previously screened at 75mM NaCl for chloride tolerance (data not shown), this study sought to compare the responses of tolerant, moderately-tolerant, and susceptible individuals to four concentrations of NaCl: 0, 25, 75, and 100mM to elucidate a general threshold where an accession considered 'salt-tolerant' becomes most beneficial with regard to its aversion to chloride toxicity. Weekly measures of key physiological processes, such as stomatal conductance (g_s), net carbon assimilation (A_{net}), and transpiration rates (E), were recorded as a way to track the performance of each genotype to exposure of variable sodium chloride concentrations ([NaCl]).

Methods

Vineyard Site and Plant Materials: Nine accessions were chosen for this study and meant to represent three categories of Cl^- accumulators in photosynthetic tissues. The highly Cl^- tolerant individuals were 99 Ruggeri, 110 Richter, and *V. acerifolia* 9018; moderately tolerant individuals were 140 Ruggeri, Dog Ridge, and Ramsey; susceptible individuals were 101-14 Mgt, *V. riparia* 'Gloire', and 44-53M. For consistency in experimental units, herbaceous cuttings of each accession were taken from a single mother vine found within the University of California Davis, Department of Viticulture and Enology collection at Hopkins Vineyard in Davis, California. A singular propagule type was chosen to remain consistent with previous findings of variability by propagule type (Fort et al., 2013)

Chloride Tolerance Screening: Samples were taken from 50-day-old seedlings grown from vegetative, or hardwood, cuttings of mature plants with no known diseases; irrigated with 0, 25, 75, or 100mM NaCl water for 21 days. Samples were washed in DI water and dried at 80ºC for 72 hours, ground to a fine powder, and chloride extracted using 25mL DI water for 0.25g dry mass. Controls were treated identically except were not given NaCl solution whatsoever.

[Cl^-] Determination and Analysis: An acid buffer solution and gelatin reagent were used to quantify leaf chloride concentrations via silver-ion titration. A chloridometer (Model 926, Nelson-Jameson Inc., Marshfield, WI, USA), was used for quantification of Cl^-; each sample measured a minimum of three times. Data was compiled using R statistical software, using packages "agricolae" and "stats" (R core Team, 2018; Felipe de Mendiburu, 2019).

Results

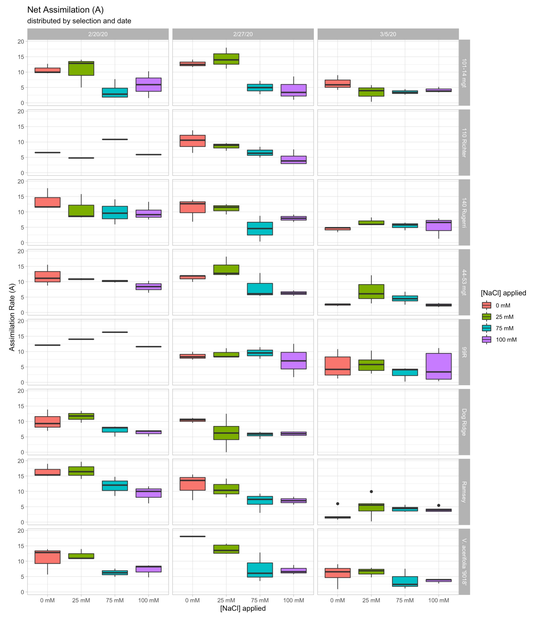

Figure 1. Net carbon assimilation rate (A_{net}) of all tested genotypes; applied NaCl separated by color; columns separated by date of measurement.

	Selection							
Selection	E		A_{net}		CO_2		g_s	
101-14 mgt	0.0018 ± 0.0006	a	7.39 ± 1.39	a	203.78 ± 24.54	ab	0.1 ± 0.04	a
110 Richter	0.0015 ± 0.0005	a	7.2 ± 1.22	a	195.49 ± 26.04	ab	0.08 ± 0.03	a
140 Rugerri	0.0017 ± 0.0003	a	8.61 ± 1.1	a	202.96 ± 12.91	ab	0.09 ± 0.02	a
44-53 mgt	0.0015 ± 0.0004	a	8.03 ± 1.19	a	201.13 ± 17.26	ab	0.09 ± 0.02	a
99 Richter	0.0012 ± 0.0003	a	7.13 ± 1.16	a	178.64 ± 14.91	ab	0.07 ± 0.02	a
Dog Ridge	0.0017 ± 0.0004	a	7.72 ± 1.04	a	216.18 ± 22.25	a	0.09 ± 0.02	a
V. acerifolia '9018'	0.002 ± 0.0006	a	8.48 ± 1.25	a	214.47 ± 19.99	a	0.12 ± 0.04	a
Ramsey	0.0013 ± 0.0004	a	7.94 ± 1.27	a	171.75 ± 14.46	ab	0.08 ± 0.03	a
p value	0.302		0.775		0.018**		0.409	

Table 2.6. Photosynthesis based on LiCOR measurements by selection tested

Table 1: Transpiration (E), net carbon assimilation (A_{net}), intracellular CO2, and stomatal conductance (g_s) by genotype.

	Applied [NaCl]							
[NaCl]	E $mol * m^{-2} * s^{-1}$		A_{net} $mol * m^{-2} * s^{-1}$		CO_2 $mol * mol^{-1}$		g_s $mol * m^{-2} * s^{-1}$	
0 mM	0.002 ± 0.0004	a	9.10 ± 0.9	a	220.45 ± 14.8	a	0.13 ± 0.03	a
25 mM	0.002 ± 0.0003	a	9.45 ± 0.9	a	216.71 ± 12.4	a	0.12 ± 0.02	a
75 mM	0.001 ± 0.0001	b	6.41 ± 0.7	b	177.54 ± 11.2	a	0.06 ± 0.01	b
100 mM	0.0009 ± 0.0001	b	6.35 ± 0.6	b	169.37 ± 11.3	a	0.05 ± 0.01	b
p value		< 0.001*		< 0.001*		0.573		< 0.001*

Table 2: Transpiration (E), net carbon assimilation (A_{net}), intracellular CO2, and stomatal conductance (g_s) by applied [NaCl]

Conclusion

This trial has shown that in grapevines, a threshold of 2 dS * m^{-1}, or approximately 25 mMol NaCl, is too low to be considered a threat to photosynthetic activity or vine productivity. Disregarding accession, our results show that applications of 25 mMol NaCl are not significantly different from 0 mMol applied NaCl in g_s, E, A_{net}, or internal CO2. However, at applied amounts of 75 mMol NaCl or greater significant decreases in the measured photosynthetic parameters are observed.

Our results suggestthat NaCl concentrations of 75 mMol, or approximately 7.5 dS * m^{-1}, have the potential to reduce photosynthetic activity in rootstocks by as much as half that of the corresponding, non salt-stressed individual. Higher NaCl concentrations reduce transpiration rates, stomatal conductance, and net carbon assimilation in all varieties we tested here. However, it is unknown if this response is transmissible to a grafted scion, which would be assuming the responsibilities of photosynthesis in production settings.