Scientia Horticulturae 237 (2018) 81–88

Contents lists available at ScienceDirect

Scientia Horticulturae

journal homepage: www.elsevier.com/locate/scihorti

Physiological response of selected avocados (*Persea americana*) subjected to NaCl and $NaHCO_3$ stress

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ARTICLE INFO

Keywords:
Alkali stress
Chlorophyll fluorescence
Photosynthesis
Pigments
Salt stress
Tocopherols

ABSTRACT

Alkali stress on crops has gained importance around the world in recent years. Avocado (*Persea americana* Mill.) is considered a salt-sensitive species, but the effect of alkaline water on avocados has not been sufficiently studied. Biomass, gas exchange, chlorophyll-*a* fluorescence, photosynthetic pigments and tocopherols were evaluated in response to alkali salt ($NaHCO_3$) and neutral salt (NaCl) stresses on avocado. Three irrigation treatments were applied: Control (no added salt), $1\,g\,l^{-1}$ NaCl and $1.5\,g\,l^{-1}$ $NaHCO_3$ for a period of 5 months. Six clonally propagated avocado rootstocks were employed, four local selections of West Indian and two Mexican race rootstocks. Biomass reduction was observed in both salt treatments, especially in the roots under the alkali stress. A reversible photoinhibition, a lower photochemical efficiency of PSII and a greater oxidative stress were detected in the alkali stressed avocados. Although biomass was reduced under salt and alkali stress, alkali stress harmed the photosynthetic apparatus to a greater extent than salt stress. Further research is necessary, the problem of alkali stress is increasing, especially in places with alkali waters where salt sensitive species such as avocado is cultivated.

1. Introduction

The total area in the world affected by alkaline (sodic) soils is 434 million ha, and 397 million ha are affected by saline soils (FAO/UNESCO, 1980; Jin et al., 2008). Alkaline soils are mainly due to the accumulation of $NaHCO_3$ and Na_2CO_3, while saline soils are caused by NaCl accumulation (Shi and Sheng, 2005; Yang et al., 2009). In most cultivated plants, yields start declining even at relatively low salinity in irrigation water, ie., electrical conductivity (EC_w) $> 0.8\,dS\,m^{-1}$ (Ayers and Westcot, 1994) or soil $EC_{se} > 1\,dS\,m^{-1}$ in saturated soil extracts (Chinnusamy et al., 2005). Alkali stress has gained importance as a threat to crop production in the recent years (Gong et al., 2013; Shi and Sheng, 2005; Yang et al., 2009). Researchers in countries with arid and semi-arid areas have extensively studied the response of higher plants to salinity stress, but alkalinity stress has not been so thoroughly studied (Javid et al., 2012). Alkali waters (with dissolved $NaHCO_3$) is used for irrigation in the Canary islands islands, and it is usually extracted from horizontal water mines "galleries" (Soler and Lozano, 1988). On the other hand, salty water is a concern on coastal southern Spain where tropical crops are cultivated (Durán-Zuazo et al., 2004), and also in the Canary islands. Coastal wells in the region have been subjected to overexploitation causing salt water intrusion (Rodríguez Rodríguez et al., 1991), worsening the situation among irrigated agriculture (González Soto et al., 1991).

Avocado (*Persea americana* Mill.) is considered a salt-sensitive species, especially the Mexican and Guatemalan races (var. *drymifolia* and *guatemalensis* respectively). Many authors consider the West Indian race (var. *americana*), the most tolerant to salinity (Ben-Ya'acov and Michelson, 1995; Mickelbart and Arpaia, 2002; Schaffer et al., 2012b). Canary islands is the second most important region in Spain for avocado production (MAGRAMA, 2014; Sayadi et al., 2005). Given the high variability of West Indian avocado (*Persea americana* Mill.) genotypes in the Canary islands, that were brought to the islands in the XIX century mostly from Cuba (Galan Sauco, 1990; Rodríguez García, 1979), a research searching for West Indian rootstock's tolerance to *Phytophthora cinnamomi* (Gallo Llobet et al., 1999) was initiated in 1984. From this research, four local rootstocks were selected due to its' tolerance to *P. cinnamomi*, nevertheless its' salinity tolerance has not been evaluated.

High salt concentration may inhibit growth as the water relationship within the plant is altered (Bar et al., 1996; Oster et al., 1985). A

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https://doi.org/10.1016/j.scienta.2018.04.010
Received 25 December 2017; Received in revised form 3 April 2018; Accepted 4 April 2018
0304-4238/ © 2018 Published by Elsevier B.V.

Table 1
Substrate analysis.

pH	EC dS m^{-1}	Ca^{2+} meq l^{-1}	Mg^{2+} meq l^{-1}	Na^{+} meq l^{-1}	K^{+} meq l^{-1}	CO_3^{2-}meq l^{-1}	HCO_3^{-}meq l^{-1}	Cl^{-}meq l^{-1}	SO_4^{2-}meq l^{-1}
6.54	0.73	2.11	1.89	3.61	0.29	0.00	1.65	1.64	3.39

NO^{3-} ppm	XCa^{2+} meq 100 g^{-1}	XMg^{2+} meq 100 g^{-1}	XNa^{+} meq 100 g^{-1}	XK^{+} meq 100 g^{-1}	CEC meq 100 g^{-1}	P ppm
14.21	12.51	5.64	1.15	1.53	25.20	31.04

Fe ppm	Mn ppm	Zn ppm	Cu ppm	O.M. %
24.24	14.50	1.65	0.93	3.01

Table 2
Water analysis of the irrigation treatments.

Treatment	pH	EC dS m^{-1}	Ca^{2+}meq l^{-1}	Mg^{2+}meq l^{-1}	Na^{+}meq l^{-1}	K^{+}meq l^{-1}	CO_3^{2-}meq l^{-1}	HCO_3^{-}meq l^{-1}	Cl^{-}meq l^{-1}
Control	8.92	0.79	0.68	2.28	5.90	0.33	1.28	4.87	1.85
1.5 g l^{-1} $NaHCO_3$	8.50	2.11	0.74	2.10	25.4	0.40	2.07	20.48	1.50
1 g l^{-1} NaCl	8.88	2.63	0.74	2.20	25.2	0.39	1.53	4.75	19.20

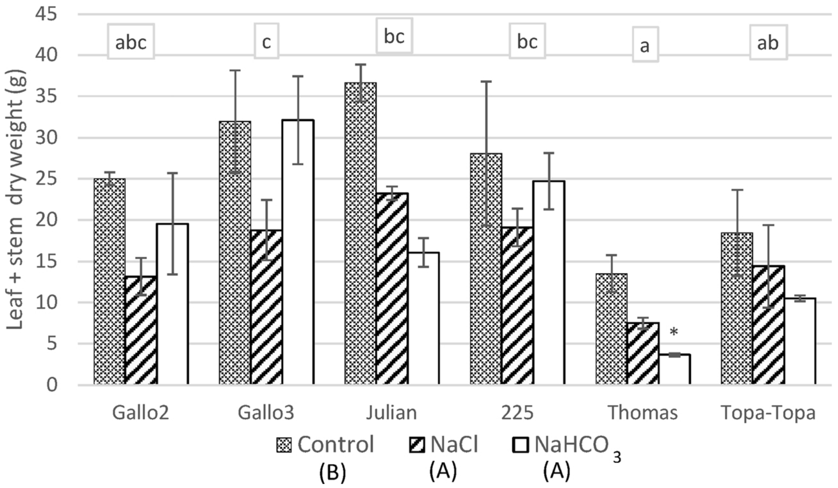

Fig. 1. Leaf + stem dry weight. Each bar represents the mean (± 1 std. error) of 9 replicates (for rootstocks) and 18 replicates (for treatments). Bars with different letters compare rootstocks, and capital letters compare treatments which are significantly different $P < 0.05$ according to a two-way ANOVA, using Tukey's HSD test.
* 50% of 'Thomas' under $NaHCO_3$ treatment died.

decrease in stomatal aperture is the most dramatic and readily measurable whole plant response to salinity. Stomatal responses are undoubtedly induced by the osmotic effect of the salt outside the roots (Munns and Termaat, 1986). Salinity affects stomatal conductance immediately (Shabala and Munns et al., 2012). At high salinity, salts can concentrate in leaves to excessive levels, and it is more likely that the damage is caused by Na^+ rather than Cl^- (Munns and Tester, 2008). A toxic effect of Na^+ could affect directly on the photosystems or on pH homeostasis due to H-coupled Na^+ efflux mechanisms (Shabala and Munns et al., 2012). The rate of cell expansion in growing tissues and the degree of stomatal aperture in leaves are reduced due to the osmotic effect of salts. A reduction of photosynthesis is caused by a reduction in stomatal conductance of CO_2, which together with the slower formation of photosynthetic leaf area, reduces the flow of assimilates to the meristematic and growing tissues of the plant *i.e.* leaves and roots, although leaves are often more affected than roots, due to the ability to efflux excessive salts and control uptake in order to avoid toxicity (Munns and Sharp, 1993). The cellular and metabolic processes involved are in common with drought-affected plants (Munns, 2002).

Pigment composition changes do not occur until high salinity levels (Chaves et al., 2012). In other tree crops such as olive trees (*Olea europaea* L.), chlorophyll concentration have been observed to descend with increasing salinity levels (Mousavi et al., 2008). Only a few investigations on alkaline stress to plant photosynthesis have been reported in which lower pigment content and net photosynthetic rate (Pn) were measured compared to salt-treated plants (Chen et al., 2011; Li et al., 2010). In order to understand better the efficiency of light harvesting of avocado, a functional evaluation is required to evaluate the dynamics of non-photochemical quenching of Chl fluorescence (NPQ) (Takayama et al., 2013). To our knowledge, there is no report comparing effects of salt and alkali stress on physiological response of avocado. In the present study, our objective was to compare physiological responses of avocado to alkali salt and neutral salt stresses, by studying biomass, photosynthesis, fluorescence, photosynthetic pigments and tocopherols. The expected results could provide a basis for understanding the response to alkali salt stress which has not been yet described on avocado from a physiological point of view, and compare it with NaCl salt stress.

2. Materials and methods

2.1. Study site and plant material

The experiment was conducted in a greenhouse at the Canarian Institute of Agricultural Research (I.C.I.A.) in Tenerife, Canary Islands, Spain (28° 30.795′ N 16° 23.253′ W). The greenhouse had climate control mechanisms by which mean daily high and low temperatures were held to 22.3 °C ± 3.1 and 17.3 °C ± 2.5, respectively, during the course of the experiment.

Although 'Duke 7′ is commonly used on salinity studies (Castro

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