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Influence of salinity and boron on germination, seedling growth and transplanting mortality of guayule: A combined growth chamber and greenhouse study

Hui Zhu[a,b], Gary Bañuelos[b,*]

[a] Key Laboratory of Wetland Ecology and Environment, Northeast Institute of Geography and Agroecology, Chinese Academy of Sciences, Changchun 130012, PR China
[b] USDA, Agricultural Research Service, San Joaquin Valley Agricultural Science Center, 9611 South Riverbend Avenue, Parlier, CA 93648-9757, USA

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ABSTRACT

Guayule (*Parthenium argentatum* A. Gray), a drought tolerant plant, originating from southwestern United States and northern Mexico, is considered to be a promising rubber-producing plant for arid and semiarid areas. To evaluate the potential of guayule as an alternative crop for saline boron-laden soils in the West side of central California, four experiments were conducted to evaluate germination, seedling growth and transplanting mortality of six guayule lines (AZ-1, AZ-2, AZ-3, AZ-4, AZ-5, and AZ-6) at different levels of salinity (electrical conductivity (EC) of 5, 10, 15, and 20 dS/m) and boron (B) levels (5, 10, 20, 30, and 40 mg/L) under both growth chamber and greenhouse conditions. Overall, increased salinity (especially at levels of EC > 10 dS/m) inhibited both germination and seedling growth of all guayule lines. In contrast, B positively influenced (to varied degrees) germination and growth of specific guayule lines (AZ-1, AZ-2, and AZ-4) and had no significant negative influence on the other three lines. The combination of 5 mg B/L and salinity (EC) of 5 dS/m significantly ($P < 0.05$) increased both germination percentage and speed of germination of AZ-4 and AZ-6, and the seedling vigor index (SVI) and length of AZ-1 and AZ-6, SVI of AZ-2, and length of AZ-5, respectively. There was no statistical difference on germination percentages for most guayule lines (except for AZ-3) grown in saline B-laden (EC of 10 dS/m, 10 mg B/L) and control (EC of 0.4 dS/m, 0.12 mg B/L) soils under greenhouse conditions. AZ-4 showed even a greater germination percentage in saline B-laden soil than in control soil, while AZ-6 showed very small difference between both types of soils. AZ-1 and AZ-6 showed greater germination percentage and speed of germination than the other lines, although greater mortality percentage was observed when AZ-1 and AZ-6 seedlings were transplanted into saline B-laden soils. Our results from both growth chamber and greenhouse experiments indicate that AZ-4 and AZ-6 can successfully germinate in typical saline B-laden soils present in the westside of central California.

1. Introduction

Guayule (*Parthenium argentatum* A. Gray), a native plant in the southwestern United States and northern Mexico, was exploited as a potential source of natural rubber for commercial production (Ray et al., 2005). Although latex content in guayule is dependent on different lines and cultivars, area of cultivation, soil type/quality, harvest time, and amounts of irrigation water, an average rubber yield of 300–2000 kg ha^{-1} $year^{-1}$ has been reported for field production in a review paper by Rasutis et al. (2015). Besides its promising latex rubber production, guayule is also a source of biomass for biofuel and non-toxic adhesives (USDA, 2009). Due to guayule's large commercial value and its drought tolerance, guayule has been developed and considered as an alternative crop for arid and semiarid areas of the southwestern United States, north central Mexico, and other regions with the similar arid climates throughout the world (McMahan et al., 2015; Thompson et al., 1988).

The field establishment of guayule is commonly achieved by transplanting seedlings initially started in greenhouse-grown cell packs (Sanchez et al., 2014; Bucks et al., 1986; Foster and Coffelt, 2005). Guayule seedlings are, however, reported to be more sensitive to environmental stress than mature plants (Miyamoto et al., 1982; Sanchez et al., 2014), therefore, the transplanting mortal-

* Corresponding author.
E-mail address: gary.banuelos@ars.usda.gov (G. Bañuelos).

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ity and early growth rate of guayule needs to be tested before transplanting the plant into a new agricultural environment with any potential environmental stress. Compared with seedling transplanting, direct seeding is considered to be less expensive and more efficient. The feasibility of direct seeding, however, needs to be thoroughly evaluated under different environmental conditions, e.g., excessive salinity, temperature, trace elements, etc., and the optimal sowing conditions need to be identified for an ideal germination rate. In this regard, Sanchez et al. (2014) and Miyamoto et al. (1982) have reported on the influence of temperature, salinity, and water stress on the germination of different guayule lines direct seeded, although the effect of salinity and trace elements, and their combined effect is still unclear on guayule germination. In addition, information is also not available on these environmental effects on various new lines that have been bred in recent years.

In arid regions of central California, high soil salinity in conjunction with severe drought is a serious threat to irrigated agriculture (Centofanti and Bañuelos, 2015), especially in one of the greatest producing agricultural regions in westside San Joaquin Valley (SJV). Soils in this area are mainly derived from Cretaceous shale rocks that contain naturally high levels of salts and other trace elements like boron (B) and selenium (Se). Due to the salinization and reduction of available water resources, growers in the westside of the SJV are facing great challenges to even grow crops. Therefore, identifying alternative crops that are both salt, B and drought tolerant is imperative for sustaining some economic viability for growers in this region of California.

Salinity generally inhibits the germination and early growth of many plant species (Zhang et al., 2015), while B is an essential element for plant growth and is reported to have both negative and positive influence on seed germination. For example, Bañuelos et al. (1999) found that B at 20–40 mg/L inhibited the germination of corn (*Zea mays* L.), carrots (*Daucus carota*), tomato (*Lycopersicum esculentum*), and alfalfa (*Medicago sativa* L.). In contrast, Memon et al. (2013) reported that B promoted germination of broccoli (*Brassica oleracea*) when seeds were soaked in 0.01% B solution. Acar et al. (2010) reported a negative effect of B on pistachio (*Pistacia vera L.*) germination when seeds were soaked in boric acid (H_3BO_3) solutions at levels ranging from 10 to 50 mg/L, while a positive effect on germination was observed at H_3BO_3 concentrations between 75 and 100 mg/L. Similarly, Mirshekari (2012) reported that seedling vigor and yield of dill (*Anethum graveolens*) were enhanced when they combined 1% B with 1.5% iron (Fe), while B alone restricted the germination vigor. Besides boron's separate effect on plant germination, B was also reported to have the potential to mitigate salt toxic symptoms occurring during seed germination and early growth of some plants, e.g., pea (*Pisum sativum*) (Bonilla et al., 2004). Consequently, we hypothesized the following for guayule: 1) B might reduce salinity toxicity symptoms during seed germination; 2) guayule could be grown by direct seeding or by transplanting seedlings in saline B-laden soils of westside of central California.

The objectives of this study were to (1) evaluate both individually and combined influence of salinity and B on the germination percentage and speed of germination of six guayule lines (AZ-1, AZ-2, AZ-3, AZ-4, AZ-5, and AZ-6) under growth chamber conditions; 2) test both the germination percentage and speed of germination of the above guayule lines in saline and B-laden soil collected from westside of central California under greenhouse conditions; and 3) evaluate the transplanting mortality and early growth of guayule lines in the above saline and B-laden soil. This study provided information for determining the feasibility of direct seeding and/or transplanting guayule in saline B-laden soil from the westside of central California.

2. Materials and methods

2.1. Materials

Four experiments were carried out at USDA, San Joaquin Valley (SJV) Agricultural Research Service Center in Parlier, CA under growth chamber and greenhouse growing conditions in 2015. Seeds of six guayule lines; AZ-1, AZ-2, AZ-3, AZ-4, AZ-5, and AZ-6, were collected from National Arid Land Plant Genetic Resource Unit (NALPGRU) in Parlier, CA, US. The seeds were initially harvested in 2013 from 10 year old guayule trees at NALPGRU in Parlier, CA, which were previously identified to possess important traits related to greater biomass and rubber production (Stelljes and Senft, 1997). In addition, we manually selected only filled seeds for the experiment; plumped seeds were shown to have highest quality (Jorge et al., 2007; Sanchez et al., 2014). Two types of soil (saline and non-saline) were collected at Red Rock Ranch in westside of California (Five Points, CA), and at the SJV Agricultural Research Service Center in Parlier, the eastside of California, respectively. Soils were collected from 0 to 25 cm, cleaned of rocks and other debris, homogenized thoroughly, and subsampled for chemical analysis prior to planting. The saline soil, consisted of an Oxalis silty clay loam, was tested to have a mean electrical conductivity (EC) of 10.2 dS/m, pH of 8.02, and water extractable B, Se and sodium (Na) concentrations of 10.5 mg/L, 0.2 mg/L, and 1469 mg/L, respectively. The non-saline soil (designated as control), consisted of a Hanford sandy loam, was tested to have a mean EC of 0.4 dS/m, pH of 7.8, and water extractable B, Se and Na concentrations of 0.12 mg/L, 0.002 mg/L, and 96.7 mg/L, respectively. Additional chemical characteristics of both soil types are described in greater detail in Centofanti and Bañuelos (2015); Bañuelos (2002), and Bañuelos and Lin (2010) respectively. All chemical reagents used in this study were purchased from Fisher Scientific, Pittsburgh, PA.

2.2. Methods

2.2.1. Guayule germination in petri dishes with different levels of salinity and B (Experiment 1)

In this experiment, the treatments consisted of four salinity levels (EC) of 5, 10, 15, and 20 dS/m and five B concentrations of 5, 10, 20, 30, and 40 mg/L, and one control (deionized water). There were three replicates for each individual treatment. Salinity solutions were prepared with sodium sulfate (Na_2SO_4), one of the dominating salts in westside soils of central California (Centofanti and Bañuelos, 2015; Bañuelos, 2009), and distilled water. Boron solutions were prepared with H_3BO_3 and DI water. Waterman filter paper was placed on the bottom of the plastic Petri dishes (diameter of 14.5 cm) which were bleached, rinsed and dried in advance. The filter paper was wetted with 10 mL of the designated treatment solution, and 50 seeds from each respective guayule line were evenly distributed in each Petri dish. Petri dishes with seeds and treatment solution were sealed with parafilm and then placed in an environment-controlled growth chamber with an alternating 9 h and 15 h temperature of 25/18 °C within each 24 h period. There was a minimum and maximum light intensity of 200 and 450 μE (photosynthetic photon flux) m^{-2} s^{-1}, respectively, during daylight hours. Radical emergence through the seed coat was counted for each Petri dish and data were recorded every 24 h. The germination percentage and speed of germination were calculated, and the lengths of seedlings were measured after 7 days of germination.

2.2.2. Guayule germination in petri dishes with salinity and B mixed solution (Experiment 2)

Due to the observed low germination percentages and weakness of guayule seedlings under higher salinity treatments, as well as the observed promotive effect of B on guayule germination (See