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Effect of the scarification methods on the germination and growth of seeds of the Fabaceae family members in presence of silver nanoparticles

Efecto de los métodos de escarificación sobre la germinación y crecimiento de semillas pertenecientes a la familia Fabaceae en presencia de nanopartículas de plata

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Abstract. The effect of scarification methods and silver nanoparticles on plantlets growth were tested on seeds of plants belonging to the Fabaceae family (*Prosopis laevigata*, *Acacia farnesiana* and *Erythrina americana*), which are predominant species in semi-arid ecosystems in Mexico. The scarification methods consisted in using coarse sand paper and two different concentrations of sulphuric acid (H_2SO_4 98% and H_2SO_4 50%); immersion of seeds in distilled water was used as a control. The percentage of germination was calculated and the Kotowski´s coefficient was determined. After scarification, the seeds were immersed in silver nanoparticles solutions at different concentrations i.e., 100 mg/L, 500 mg/L and 1000 mg/L. Thereafter, seeds were incubated in Petri dishes. Root and shoot lengths and dry biomass were measured at 7, 14 and 30 days. The mechanical scarification showed the maximum level of germination in the three tested plants. A negative effect was observed on the root/shoot of the plantlets exposed to silver nanoparticles solutions at different concentrations.

Keywords: Semiarid Ecosystem; Fabaceae; Silver Nanoparticles, Scarification, Germination.

Resumen. Se probó el efecto de los métodos de escarificación y la presencia de nanopartículas de plata sobre la germinación de semillas y crecimiento de plántulas pertenecientes a la familia Fabaceae (*Prosopis laevigata*, *Acacia farnesiana* y *Erythrina americana*), las cuales son especies predominantes en ecosistemas semiáridos de México. Los métodos de escarificación consistieron en tratamientos mecánico hecho con una lija granular y tratamiento químico, consistente en inmersión en soluciones de ácido sulfúrico a dos diferentes concentraciones p.e., H_2SO_4 98% and H_2SO_4 50%. La inmersión de semillas en agua destilada se usó como control. Se calculó el porcentaje de germinación y se determinó el coeficiente de Kotowski. Luego de la escarificación, las semillas se sumergieron en soluciones de nanopartículas de plata a diferentes concentraciones (100 mg/L, 500 mg/L y 1000 mg/L). Luego, las semillas se incubaron en cajas de Petri. La longitud de raíces y tallos y la biomasa seca se midieron a los 7, 14 y 30 días. La escarificación mecánica tuvo el máximo nivel de germinación en las tres plantas analizadas. Se observó un efecto negativo en la longitud de raíz/tallo de las plántulas expuestas a las soluciones de nanopartículas de plata para las tres concentraciones.

Palabras clave: Ecosistema Semiárido; Fabaceae; Nanopartículas de Plata; Escarificación; Germinación.

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INTRODUCTION

The industry of nanotechnology has had a fast development around the world, impacting on the economy, society and environment. The interest in the potential benefits of nanomaterials, and a greater production of these materials have naturally led to an increased concern about the (1) potential toxic effects resulting from their usage or (2) unintentional release into environmental natural resources such as soil, air and water (Service, 2004; Moore, 2006; Nel et al., 2013).

According to USEPA, (2005) the engineered nanomaterials could be classified in four types (1) carbon-based materials, usually including fullerene, single walled carbon nanotube (SWCNT) and multi-walled carbon nanotube (MWCNT); (2) Metal-based materials (3) dendrimers; and (4) composites, which combine nanoparticles with other nanoparticles or with larger, bulk-type materials.

Depending on the type, nanoparticles may be released to the atmosphere in the form of aerosols, as well as to the soil and surface water. For example, nanoparticles released to the atmosphere may be deposited in the soil, and affect not only the soil microbial communities but also the plants (Bystrzejewska-Piotrowska et al., 2009). Some studies have reported toxic effects of nanoparticles on the germination and/or root growth of some plant species (Yang et al., 2006; Lin et al., 2007). Zhu et al. (2008) reported some mechanisms involved in the presence of nanoparticles in plants (i.e., absorption, translocation, and accumulation).

The information about the effect of nanoparticles on plants in specific ecosystems, such as semi arid and arid ecosystems, still remains entirely unknown; these ecosystems play an important role in biogeochemical cycles such as carbon, nitrogen and phosphorus but the nanoparticles effects are not clearly understood, as well as their participation on ecosystem maintenance and its importance on environmental system functioning (Vitousek et al., 2002; Houlton et al., 2008; Menge et al., 2014).

The predominance of some species is clear in the Mexican semi arid ecosystems. For instance, a high abundance of tree members of the Fabaceae family [i.e., mesquite (*P. laevigata*), huizache (*A. angustissima*) and colorin (*E. americana*)] play a key role on N fixation, rainwater capture and fertility of soils (Bouillet et al., 2008; Eldridge et al., 2011).

The semiarid ecosystems represent around 40% of the total land area, and contribute strongly to the carbon dioxide sink in the world (Poulter et al., 2014). These ecosystems could be receptors of nanomaterials and nanoparticles, and depending on the nanoparticle nature, some characteristics might be modified (i.e., mobility to entry into the environment, and its interaction with the soil microorganisms and plants: Musee, 2011). De la Rosa et al. (2011) evaluated the effect of ZnO nanoparticles (NP) on the germination rate of three desert plants [i.e., *Prosopis juliflora-volutina* (velvet mesquite), *Parkinsonia florida* (blue palo verde) and *Salsola tragus* (tumbleweed)] and observed significant effects of ZnO NP on the root size of the plantlets but no effects on the germination rate.

This study evaluated the effect of the scarification method and the silver nanoparticles (AgNP) concentration on the germination rate, and root and shoot elongation in three typical plants of the Mexican semi-arid ecosystem [mesquite (*P. laevigata*), huizache (*A. angustissima*) and colorin (*E. americana*)]. The obtained results allowed us to understand the effects of nanoparticles after seeds had been scarified on the germination and subsequent growth of seedlings.

MATERIALS AND METHODS

Collection of plant material. Seeds of *E. americana, P. laevigata and A. farnesiana* were collected in the Valle del Mesquital (19° 55´ N 99° 32´ W). The average altitude was 2400 m a.s.l. The climate is a semi-hot and semi-dry with a mean annual temperature of 16 °C, and an average annual precipitation of 650 mm, mainly from May to September (htttp://www.inegi.gob.mx).

Pods were collected from April through May, and the manually removed seeds were stored at 4 °C in plastic bags in the dark. The broken and insect-damaged seeds were discarded.

Scarification of seeds. All seeds of *E. americana*, *P. laevigata and A. agustissima* were immersed in distilled water during 6 hours. After this pre-treatment, the following treatments were applied to fifty seeds per replicate for each treatment: abrasion of seeds mechanically scarified with coarse sand paper until part of the outer layer of the endocarp was removed (ABR treatment); immersion in distilled water (v/v) (WAT treatment); immersion in H_2SO_4 at 50% (v/v), and immersion in H_2SO_4 at 98% (v/v) (Ac50 and Ac98 treatments, respectively). The seeds were immersed for 60 minutes.

Preparation of silver nanoparticles solution. Silver nanoparticles (AgNP) provided by ID-Nano® (<100 nm, surface area 80 – 120 m^2/g, colloidal suspension) were prepared by adding colloidal solid powder to a beaker containing distilled water. Mixtures were stirred for 5 min. Adequate dilution was performed to obtain the desired AgNP concentration (100 mg/L, 500 mg/L and 1000 mg/L). Suspensions were prepared the same day of the experimental set-up.

Germination of seeds. After scarification, the seeds were immersed in the AgNPs solutions at the described concentrations (i.e., 100 mg AgNPs/L, 500 mg AgNPs/L and 1000 mg AgNPs/L of distilled water) for 6 hours at 20 °C.

The seeds were placed in Petri-dishes on filter paper (Whatman® 1:11 μm) moistened with the AgNPs solution, according to the treatment conditions. Distilled water was used as control. The Petri dishes were sealed and incubated at 12-h photoperiod and environmental temperature. Occasional observations were done in order to follow the germination of the seeds. The percentage of germinated seeds was calculated after 7, 14 and 30 days.

Growth plantlets measurement. The percentage of germination was calculated using equation 1; Relative Germination (RG) was determined using equation 2, Growth Reduction (GR) for roots and shoots was calculated using equation 3; and the Kotowski´s coefficient was calculated using the equation reported by Makhlouf et al. (2015).

$$\%G = [(\#\text{Germinated seeds})/(\text{Total \# Seeds})] \times 100 \tag{1}$$

$$RG = [(\%\text{Germination in treatment})/(\%\text{Germination in control})] \times 100 \tag{2}$$

$$GR = \%\text{Growth of roots or shoots in treatments} - \%\text{Growth of roots or shoots in controls} \tag{3}$$

Statistical analysis. All germination tests and growth experiments were carried out in triplicate. The results were analyzed using ANOVA, followed by the Tukey´s HSD test, using the statistics software SPSS 16.0 (Statistical Package for the social Sciences, Chicago, IL.).

RESULTS

Plant species description. The Fabaceae is the third largest family of angiosperms worldwide, with approximately 650 genera and more than 18000 species (Gao et al., 2011). This family is one of the most important groups among Mexican plants in abundance and richness terms; the description of the selected plant species is given in Table 1.

Effect of scarification method and nanoparticle concentration on seed germination. The application of the abrasion method resulted in 28%, 77% and 8% germination for *P. laevigata, A. farnesiana* and *E. americana* after 30 days, respectively. The lowest values were obtained for the WAT treatment (i.e., 18%, 21% and 4% for *P. laevigata, A. farnesiana* y *E. Americana*, respectively, in the same period of time).

The observed percentages of germination for *E. americana* after 7, 14 and 30 days were lower to those in *P. laevigata, A. farnesiana* for all treatments through the sampling days.

The highest ($P<0.05$) percentages of germination and Kotowski´s coefficient were obtained in the ABR method for the three plant species (Table 2).

Effect of nanoparticles concentration on root and shoot growth. Figure 1 shows the root length and GR of roots (RGR) for huizache, colorin and mesquite germinated in the AgNPs solutions. The size of roots of colorin (Fig. 1 a) in the WAT treatment was between 4.5 ± .12 cm and 5.1 ± 0.2 cm. The highest value was observed in the control of the ABR treatment (5.3 ± 0.08 cm).

The lowest values of root elongation were detected in all treatments for all treated seeds at 1000 mg AgNP/L. The lowest RGR value was found in *E. americana* for the Ac98 treatment at 1000 mg AgNP/L (31 ± 5%). It was the lowest value for all the treatments and plant species evaluated.

In huizache (Fig.1 b), root elongation was reduced when seeds were immersed in solution at 1000 mg AgNP/L for treatments WAT, Ac50 and Ac98; the root growth reduction values were 29 ± 9 %, 24 ± 4% and 22 ± 2%, respectively. The root length was not significantly affected by immersion in 100 mg AgNP/L compared to the control treatment, but in Ac50 root growth increased. The highest root length was observed in the control of the WAT method treatment, where seeds were immersed in both distilled water without AgNPs (5.6 ± 0.04 cm) and at 100 mg AgNP /L (5.6 ± 0.1 cm).

The mesquite roots (Fig.1 c) had the lowest root elongation (2.3 ± 0.1 cm in ABR and Ac50 treatments at 1000 mg AgNP/L) compared to huizache and colorin plants. The highest value was observed in the control seeds immersed in sulphuric acid at 98% in absence of silver nanoparticles (3.4 ± 0.1 cm). No significant differences ($P<0.05$) were observed among the lowest values of root elongation, which were detected for all the scarification method treatments at 1000 mg AgNP /L. The lowest value for RGR in mesquite plantlets was observed in both ABR and Ac50 treatments at 1000 mg

Table 1. Characteristics of the study species.
Tabla 1. Características de las especies en estudio.

Plant species	Common name	Family	N fixer	Number of cotyledons	
Prosopis laevigata	Mesquite	Fabaceae	Yes	Dicotyledonous	Perennial herbaceous/tree
Acacia angustissima	Huizache	Fabaceae	Yes	Dicotyledonous	Perennial herbaceous/tree
Erithryna americana	Colorin	Fabaceae	Yes	Dicotyledonous	Perennial Herbaceous/tree

Table 2. Effect of the scarification method and nanoparticle concentration on seed germination, the Kotowski's coefficient and dry biomass of the study species.

Tabla 2. Efecto del método de escarificación y la concentración de nanopartículas sobre la germinación de semillas, el coeficiente de Kotowski y la biomasa seca de las especies en estudio.

Treatment	**Concentration (mg Ag NP/L)**	**Percentage of germination**			**Kotowski´s coefficient after 30 days**	**Dry biomass (g)**
		7 days	**14 days**	**30 days**		
Prosopis laevigata						
Control	0	0 ± 0	7 ± 2	18 ± 3	4.2*	0.60 ± 0.03 a
	100	0 ± 0	6 ± 3	17 ± 4	3.8	0.35 ± 0.02 b
	500	0 ± 0	7 ± 4	16 ± 3	4.0	1.81 ± 0.01 c
	1000	0 ± 0	5 ± 3	12 ± 4	4.0	1.40 ± 0.01 c
Abrasion	0	11 ± 4	18 ± 4	28 ± 4	5.1*	0.40 ± 0.01 a
	100	10 ± 4	15 ± 3	20 ± 3	5.0	0.42 ± 0.02 a
	500	11 ± 5	14 ± 4	20 ± 4	5.0	0.23 ± 0.03 b
	1000	7 ± 3	10 ± 5	19 ± 5	4.7	0.20 ± 0.02 b
H_2SO_4 50%	0	2 ± 2	15 ± 4	19 ± 4	4.8*	0.54 ± 0.02 a
	100	2 ± 2	9 ± 3	13 ± 4	4.5	0.42 ± 0.02 b
	500	0 ± 0	7 ± 3	15 ± 5	4.0	0.40 ± 0.02 b
	1000	1 ± 1	6 ± 4	15 ± 3	4.0	0.39 ± 0.01 b
H_2SO_4 98%	0	4 ± 2	12 ± 3	21 ± 6	4.7*	0.44 ± 0.01 a
	100	2 ± 1	9 ± 3	19 ± 2	4.2	0.35 ± 0.03 b
	500	0 ± 0	5 ± 2	17 ± 3	3.8	0.40 ± 0.03 a
	1000	0 ± 0	5 ± 3	15 ± 2	3.8	0.23 ± 0.02 c
Acacia agustissima						
Control	0	2 ± 3	12 ± 4	21 ± 4	4.5*	0.60 ± 0.09 a
	100	0 ± 0	5 ± 2	15 ± 2	3.8	0.62 ± 0.05 a
	500	0 ± 0	5 ± 3	15 ± 1	3.8	0.55 ± 0.05 a
	500	0 ± 0	2 ± 1	12 ± 2	3.6	0.49 ± 0.03 a
Abrasion	0	46 ± 8	69 ± 10	77 ± 10	5.6*	0.72 ± 0.02 a
	100	35 ± 3	52 ± 7	65 ± 4	4.8	0.74 ± 0.03 a
	500	32 ± 3	50 ± 5	60 ± 5	5.2	0.64 ± 0.05 ab
	1000	28 ± 4	45 ± 9	60 ± 9	5.0	0.60 ± 0.06 ab
H_2SO_4 50%	0	16 ± 5	52 ± 8	62 ± 8	3.7	0.53 ± 0.04 a
	100	8 ± 3	45 ± 7	52 ± 5	4.8*	0.58 ± 0.05 a
	500	7 ± 4	40 ± 5	50 ± 7	4.5	0.52 ± 0.04 a
	1000	5 ± 5	40 ± 9	45 ± 5	4.5	0.49 ± 0.04 a
H_2SO_4 98%	0	26 ± 7	60 ± 7	69 ± 8	5.2*	0.54 ± 0.05 a
	100	20 ± 5	56 ± 6	58 ± 5	4.8	0.52 ± 0.04 a
	500	20 ± 4	50 ± 9	52 ± 5	4.6	0.50 ± 0.06 a
	1000	15 ± 8	46 ± 6	52 ± 7	4.6	0.50 ± 0.05 a
Erythrina americana						
Control	0	0 ± 0	1 ± 1	4 ± 1	4.0*	1.03 ± 0.02 a
	100	0 ± 0	0 ± 0	2 ± 1	3.3	0.94 ± 0.08 a
	500	0 ± 0	0 ± 0	2 ± 2	3.3	0.90 ± 0.04 a
	1000	0 ± 0	0 ± 0	0 ± 0	0	0.82 ± 0.03 b

Abrasion	0	3 ± 2	6 ± 2	8 ± 6	6.0*	1.12 ± 0.02 a
	100	4 ± 2	4 ± 3	4 ± 5	5.8	0.95 ± 0.05 b
	500	3 ± 3	4 ± 2	3 ± 3	5.2	0.90 ± 0.06 b
	1000	1 ± 1	1 ± 1	4 ± 2	4.2	0.85 ± 0.06 b
H_2SO_4 50%	0	0 ± 0	4 ± 2	8 ± 8	4.3	0.90 ± 0.01 a
	100	0 ± 0	2 ± 4	2 ± 1	4.5*	0.90 ± 0.04 a
	500	0 ± 0	2 ± 2	1 ± 1	4.1	0.83 ± 0.06 b
	1000	0 ± 0	1 ± 1	1 ± 0	4.5	0.80 ± 0.05 b
H_2SO_4 98%	0	2 ± 2	3 ± 2	13 ± 4	4.3*	0.76 ± 0.01 a
	100	2 ± 1	2 ± 2	10 ± 3	4.0	0.70 ± 0.04 b
	500	1 ± 1	0 ± 0	9 ± 3	3.6	0.68 ± 0.04 b
	1000	1 ± 1	1 ± 1	9 ± 4	3.7	0.60

Data are average of three triplicates ± SD.
* Represents highest Kotowski´s value per treatment.
Percentage of germination was calculated by using the equation 1. Same letters do not show statistically significant differences among treatments by the Tukey test (P<0.001).
Los datos son el promedio de tres réplicas ± SD.
* Representa los valores máximos para el coeficiente de Kotowski por tratamiento.
El porcentaje de germinación fue calculado empleando la ecuación 1. Letras iguales no muestran diferencias estadísticamente significativas entre tratamientos aplicando la prueba de Tukey (P<0,001).

AgNP/L with a reduction of 25 ± 3% with respect to their controls.

The length of shoots and GR of shoots (SGR) are shown in Figure 2. Colorin (Fig. 2 a) shoots showed, on average, the minimum negative effect on SGR values. The seeds scarified by using sulphuric acid at 98% of concentration reduced the shoot length in a 25 ± 6%. No significant difference was observed for the SGR values under the other three scarification methods. The length of shoots of plantlets after immersion in 1000 mg AgNP/L solutions did not show a significant difference among the applied treatments (P>0.05).

In huizache (Fig.2 b), it was not possible to detect a significant difference between the SGR of the WAT and the ABR method Similarly, the lowest value for the shoot length was not different statistically among the scarification treatments at 1000 mg AgNP/L (4.6 ± .72 cm, 5.1 ± 0.6 cm, 4.8± 0.1 cm, 4.8 ± 0.54 cm). A positive effect on shoot length was observed in seeds immersed in AgNP solution at 100 mg/L for WAT, ABR and Ac50 treatments, but it was not the case for the Ac98 treatment.

When the concentration of the nanoparticles solution was increased from 100 to 1000 mg AgNP/L, the elongation of the shoots and the rate of growth in mesquite was decreased (Fig.2 c). There were no observed statistical differences between the shoot length in the controls of treatments and those immersed into solutions at 100 mg AgNP/L. The lowest values of shoot elongation were detected in the control, abrasion and sulphuric acid at 98% scarification methods (-62 ± 5.6%, -56 ± 3.2%).

DISCUSSION

Scarification method on germination rate. The effect of the scarification method on the germination was measured using the Kotowski´s coefficient; the mechanical abrasion increased the percentage of germination compared to the WAT, and chemical treatments. The percentage of germination was higher for this method for mesquite and huizache; colorin seeds had a similar value for the H_2SO_4 at 98%.

Myint et al. (2010) reported that mechanical scarification of oil palm seeds resulted in higher germination rates. Alderete-Chávez (2010) proved chemical and physical methods (i.e., sulphuric acid and heat). Immersion in sulphuric acid responded positively in all cases (i.e., 7 min, 15 min and 30 min of immersion for *Lupinus lepthophyllus*). All treatments yielded a positive effect compared to the control. It is also known that concentrated H_2SO_4 is known to be a consistent and uniform method for high levels of germination in different plant seeds (Tian et al., 2010; Ebrahimi et al., 2012).

Pipinis et al. (2011) demonstrated that the germination of *Cercis siliquastrum* (Fabaceae*)* increased when the immersion time in acid increased. In our experiment, the immersion time was not variable. However, an increment of the Kotowski´s coefficient of mesquite and huizache was observed when the sulphuric acid concentration increased. In our case, the control treatment obtained the lowest values compared to the other scarification methods.

In general terms, the *A. angustissima* seeds obtained the maximum values for the germination rates on all treatments.

Fig. 1. Root length (cm) and Root Growth Reduction (RGR) (%) for scarified seeds of (a) colorin (*E. americana*), (b) huizache (*A. farnesiana*) and (c) mesquite (*P. laevigata*) after 30 days of incubation and immersed in AgNPs solutions at different concentrations (100 mg AgNp/L, 500 mg AgNp/L and 1000 mg AgNp/L). Bars are minimum significant difference. Different letters represent significant differences (P<0.05) according to the Tukey´s test.

Fig. 1. Longitud de raíz (cm) y Reducción de Crecimiento de Raíz (RCR) (%) para las semillas escarificadas de (a) colorin (*E. americana*), (b) huizache (*A. farnesiana*) y (c) mesquite (*P. laevigata*) después de 30 días de incubación e inmersas en soluciones de AgNPs a diferentes concentraciones (100 mg AgNp/L, 500 mg AgNp/L and 1000 mg AgNp/L). Las barras representan las diferencias mínimas significativas. Diferentes letras representan diferencias significativas (P<0,05) de acuerdo con la prueba de Tukey.

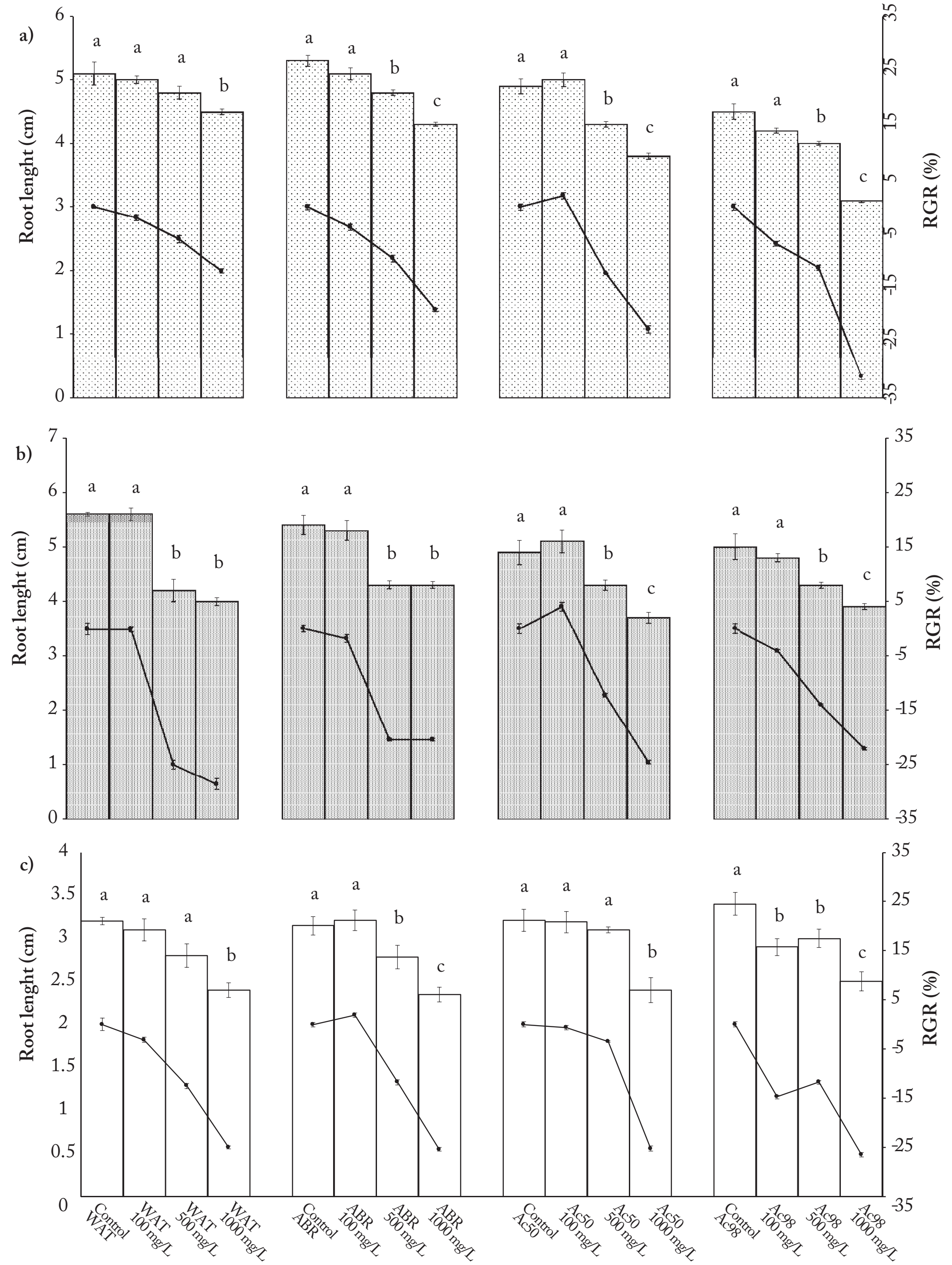
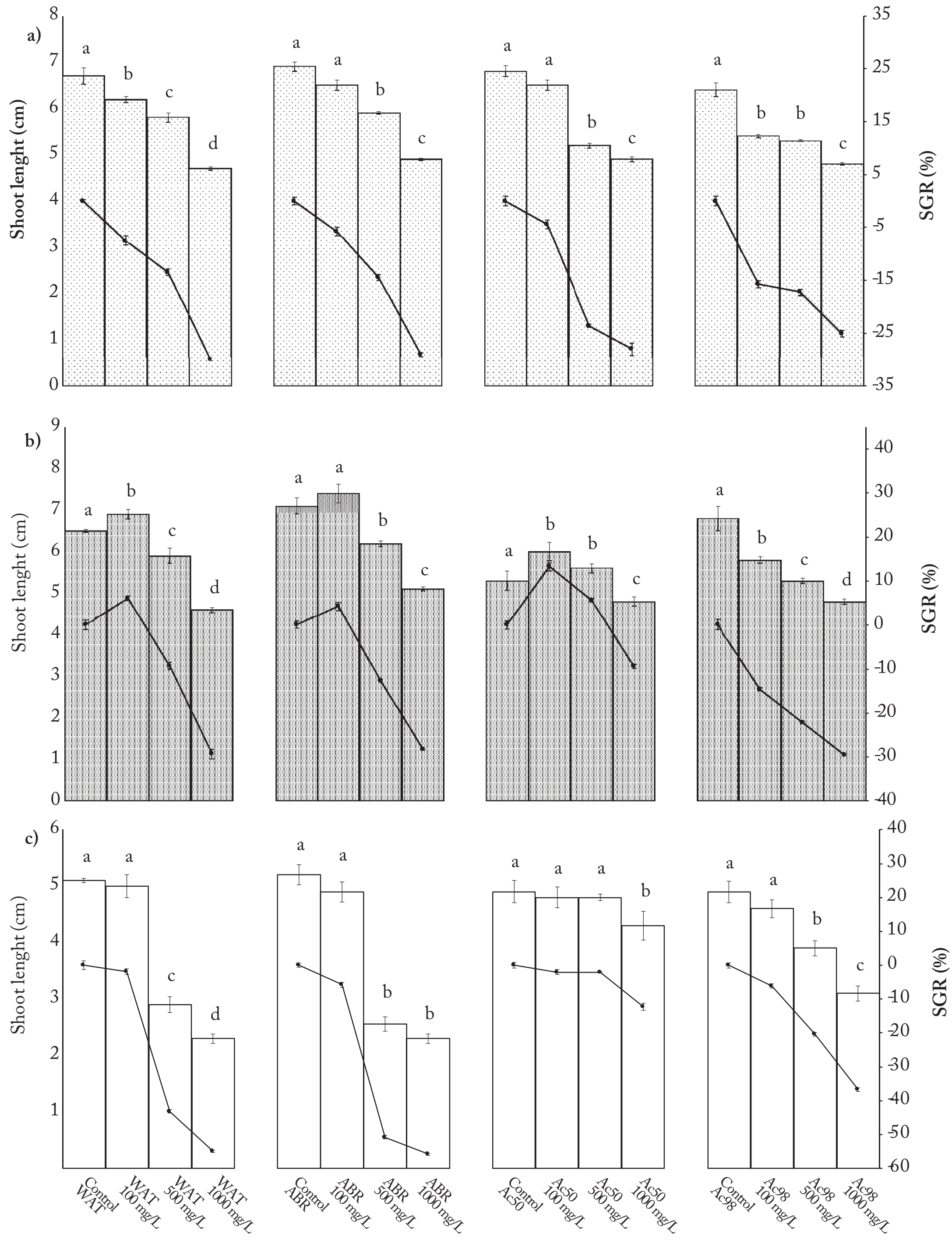


Fig. 2. Shoot length (cm) and Shoot Growth Reduction (SGR) (%) for scarified seeds of (a) colorin (*E. americana*), (b) huizache (*A. farnesiana*) and (c) mesquite (*P. laevigata*) after 30 days of incubation and immersed in AgNPs solutions at different concentrations (100 mg AgNp/L, 500 mg AgNp/L and 1000 mg AgNp/L). Bars are minimum significant difference. Different letters represent significant differences (P<0.05) according to the Tukey´s test.

Fig. 2. Longitud de tallo (cm) y reducción de crecimiento de tallo (SGR) (%) para semillas escarificadas de (a) colorin (*E. americana*), (b) huizache (*A. farnesiana*) y (c) mesquite (*P. laevigata*) después de 30 días de incubación e inmersas en soluciones de AgNPs a diferentes concentraciones (100 mg AgNp/L, 500 mg AgNp/L and 1000 mg AgNp/L). Las barras representan las diferencias mínimas significativas. Diferentes letras representan diferencias significativas (P<0,05) de acuerdo con la prueba de Tukey.



Effect of the nanoparticles on biomass and root and shoot growth. It has been reported that the presence of nanoparticles affects the root and shoot elongation in higher plants species (Lin et al., 2007). The physical and chemical properties of nanoparticles are an important factor for facilitating the accessibility to organisms; Lee et al. (2008) studied the effect of no miscible water nanoparticles on *Phaseolus vulgaris* and *Triticum aestivum.* They observed a growth inhibition of seedlings exposed to different concentration of Cu nanoparticles. In addition, they also observed bioaccumulation of nanoparticles when the concentration was increased. El-Temsah and Joner (2012) evaluated the bioavailabity of silver and zero-valent iron nanoparticles (ZVI) in water and two contrasting soils. Their results suggested that ZVI at low concentrations (i.e., 0 to 5000 mg/L) did not affect seed germination and plant growth. On other hand, the silver nanoparticles inhibited seed germination at low concentrations (i.e., 0 to 100 mg/L) without impeding complete germination.

De la Rosa et al. (2011) reported reduction of the mesquite root growth as an indicator of nanoparticles toxicity. However, the mesquite resulted more tolerant to nanoparticles compared to plants belonging to the genus *Salsola* and *Parkinsonia*, species which predominate in semiarid ecosystems. Similarly, *Prosopis laevigata* seeds showed the same tolerance values as that of mesquite roots. In our experiment, the root and shoot elongation of the three plant species were inhibited, and it was related to the concentration of silver nanoparticles. At higher concentration of NPs, the inhibition increased.

Colorin roots growth was inhibited on an average than the mesquite and huizache plantlets. There is no scientific evidence available about the effect of scarification methods and nanoparticles presence on the germination and growth of *E. americana* seeds. This article is very likely the first scientific document evaluating the parameters above explained.

The shoot length for the tested plants was affected by the silver nanoparticles, independently of the scarification method. Dimpka et al. (2012) tested the effect of metallic nanoparticles (Cu and Zn) on the shoot growth of wheat (*Triticum aestivum*) grown in sand, observing a negative effect for the Cu nanoparticles. The negative effects of the silver nanoparticles on elongation of shoots have been widely described (Cheng et al., 2011; Lee et al., 2012). Pandey et al. (2014) found a positive effect of the silver nanoparticles on the root/shoot length of *Brassica juncea* L.; it was correlated with the chlorophyll and protein concentration. In this experiment the RGR, SGR and shoot length for all tested plants were negatively affected. Mesquite was the plant species with GR of root and shoot most affected with -62±2% in the control scarification treatment at 1000 mg AgNP/L. The effect of the silver nanoparticles was negative in all plants regardless of the concentration or scarification method.

CONCLUSION

It was possible to determine the effect of the scarification method and the silver nanoparticles immersion of seeds of three typical plants of semi arid ecosystems i.e., *Prosopis laevigata*, *Acasia angustissima* and *Erythrina americana.* We found that the mechanical method resulted in higher values for the Kotowski´s coefficient and germination rate for all plants. *Acasia angustissima* seeds showed the highest level of germination compared to colorin and mesquite. The RGR values were negative for all the treatments and seeds. Huizache plantlets showed a better response against the toxicity of the nanoparticles expressed as a minor reduction of the root/shoot length ratio.

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