

## EFFECT OF SCARIFICATION AND TEMPERATURE ON SEED GERMINATION OF TWO MEXICAN SPECIES OF *MIMOSA* (LEGUMINOSAE-MIMOSOIDEAE)

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**Abstract:** It has been considered that *Mimosa aculeaticarpa* var. *aculeaticarpa* and *M. luisana* have the potential to restore temperate and semi-arid environments, respectively. Mature seeds were collected from eight mother plants per taxon to determine the effects of scarification and temperature on seed germination of both taxa, and to define the optimum temperature for germination. To obtain a representative sample of each taxon, seeds with the same weight and size were mixed. Five replicates of 20 seeds each (100 seeds per taxon/temperature, plus the control) were established. The seeds were mechanically scarified and incubated under controlled conditions: 5 to 45 °C with 5 °C intervals, and a 12h light/12h darkness photoperiod. Results showed that scarification and temperature were significant factors in seed germination. In both taxa, germination was epigeal and phanerocotylar. The highest seed germination percentages were obtained when seeds were scarified. 88 % of seeds of *M. aculeaticarpa* var. *aculeaticarpa* germinated at 25 °C. Hundred percent of seeds of *M. luisana* germinated at 30 °C. The optimum temperature for germination of seeds of both taxa was established at 25 °C. *Mimosa* taxa differ in the germination rate: *M. luisana* germinated at 6.6 seeds/day and *M. aculeaticarpa* var. *aculeaticarpa* at 3.4 seeds/day. Overall results suggest that, for both taxa, an increase or decrease in temperature will not inhibit seed germination, supporting the recommendation to use these plants for reforestation, as part of environmental restoration projects in temperate and semi-arid Mexican regions.

**Key words:** arid tropical scrub, conservation, restoration, temperate forest.

**Resumen:** *Mimosa aculeaticarpa* var. *aculeaticarpa* y *M. luisana* son taxa con potencial para la restauración ambiental. Se evaluaron los efectos de la escarificación y la temperatura en la germinación de semillas de ambos taxa y se definió la temperatura óptima de germinación, se colectaron semillas maduras de ocho individuos por especie. Las semillas (igual peso y tamaño) se mezclaron para obtener una muestra compuesta por taxon. Se establecieron cinco repeticiones de 20 semillas por taxon (100 semillas por taxon, por temperatura, más el control). Las semillas fueron escarificadas mecánicamente, realizando un corte en el extremo opuesto al eje del embrión e incubadas en un gradiente de 5 a 45 °C, con intervalos de 5 °C y un fotoperíodo de 12h luz/12h oscuridad. Los resultados indican que la escarificación y la temperatura son factores significativos para la germinación de las semillas. En ambos taxa, la germinación fue epígea y fanerocotilar. Los mayores porcentajes de germinación se obtuvieron cuando las semillas fueron escarificadas. *Mimosa aculeaticarpa* var. *aculeaticarpa* logró el 88 % de germinación a 25 °C; mientras que *M. luisana* alcanzó 100 % a 30 °C; sin embargo, para ambos taxa la temperatura óptima de germinación fue de 25 °C. La tasa de germinación difiere, en *M. luisana* se registraron 6.6 semillas/día; mientras que en *M. aculeaticarpa* var. *aculeaticarpa* sólo 3.4 semillas/día. Los resultados sugieren que ante un incremento o decremento en la temperatura, la germinación de las dos especies estudiadas no será inhibida; por lo tanto, se apoya la recomendación del uso de ambas especies en reforestación como parte de proyectos de restauración ambiental en regiones templadas y semiáridas de México.

**Palabras clave:** bosque templado, conservación, matorral xerófilo; restauración.

In Mexico, species of *Mimosa* L. (Leguminosae-Mimosoideae) have attracted attention due to their multiple uses, their ability to survive in harsh environments, to resprout after having been cut or burnt, because the genus has many

ecological, economic, and cultural values (Camargo-Ricalde *et al.*, 2001). *Mimosa* comprises ca. 530 species in the world, and it is one of the three largest genera of Mimosoideae, along with *Acacia* and *Inga* (Simon *et al.*, 2011).

After Brazil, Mexico is the second diversification center of *Mimosa*, being the most diverse genus of the subfamily with ca. 110 species, 60 % endemic to the country, many of them occurring as dominant or co-dominant elements in arid ecosystems, widely distributed in tropical vegetation, and few species growing in temperate forests (Grether *et al.*, 1996; Camargo-Ricalde *et al.*, 2002). Within arid and semi-arid environments, some *Mimosa* species are resource-island formers, enriching the soil with organic matter and nutrients (Camargo-Ricalde *et al.*, 2002, 2010a), and as reservoirs of arbuscular mycorrhizal fungal spores (Camargo-Ricalde and Dhillion, 2003), and nitrogen-fixing bacteria (Camargo-Ricalde *et al.*, 2010a, b). In addition, the nurse effect is very clear between *Mimosa luisana* Brandegees and *Neobuxbaumia tetetzo* (F.A. C. Weber ex K. Schum) Backeberg, a columnar cactus endemic to Mexico (Valiente-Banuet *et al.*, 1991; Valiente-Banuet and Ezcurra, 1991). Hence, some *Mimosa* species are considered potentially useful for ecological restoration projects (Dhillion *et al.*, 2004; Moreno-Calles and Casas, 2010; Pavón *et al.*, 2011).

As a first step, it is known that seeds of some *Mimosa* species are able to germinate in a wide range of temperatures (De Souza-Araújo and De Castro-Andrade, 1983; Camargo-Ricalde *et al.*, 2004; Pavón *et al.*, 2011), and that scarification increases the percentage of germination (Ferreira *et al.*, 1992; Camargo-Ricalde and Grether, 1998; Camargo-Ricalde *et al.*, 2004; González-Castañeda *et al.*, 2004; Silveira and Fernandes, 2006; Leal and Biondi, 2007; Chahuan and Johnson, 2008; Pavón *et al.*, 2011; Gehan-Jayasuriya *et al.*, 2013). However, it is not yet possible to generalize about how seeds will respond to both seed scarification and temperature fluctuation because only 5 % of the species of *Mimosa* have been studied around the world. For the Mexican *Mimosa*, seed germination has merely been studied in 13 species occurring in arid ecosystems (*i.e.* Camargo-Ricalde *et al.*, 2004, Pavón *et al.*, 2011) and only one from temperate forests (Martínez-Pérez *et al.*, 2006).

Hence, and under the assumption that species are adapted to the environments where they have evolved, arid and semi-arid ecosystem species will be able to germinate at higher and more extreme temperatures than temperate ecosystem species. Therefore, the aims of this study were: (i) To determine the effects of scarification and temperature on seed germination percentage, germination rate and saturation rate; and (ii) To define the optimum temperature for seed germination, of two *Mimosa* taxa occurring in two contrasting ecosystems: *Mimosa aculeaticarpa* var. *aculeaticarpa* Ortega (temperate forest, endemic to Mexico) and *M. luisana* (arid tropical scrub, narrow endemic to Mexico).

## Materials and Methods

*Experimental design.* Mature fruits of *M. aculeaticarpa* var. *aculeaticarpa* and *M. luisana* were collected during the

maximum productivity season of each taxon (November and October, respectively), from eight different mother plants with similar height and foliage cover. Fruit harvesting was random, spanning the entire area of the canopy per plant.

*Mimosa aculeaticarpa* var. *aculeaticarpa* was collected from a temperate forest, in the Municipality of Tiripetío, State of Michoacán, 19° 58.218' N, 101° 31.664' W, at 1,950 m a.s.l., and *M. luisana* from an arid tropical scrub, in the Valley of Tehuacán, State of Puebla, 18° 15' 23.7" N, 97° 09' 03.3" W, at 2,232 m a.s.l. Voucher specimens were deposited at the Herbario Metropolitano (UAMIZ), Universidad Autónoma Metropolitana-Iztapalapa.

*Seed characterization and treatments prior to sowing.* After collection, seeds were removed from the fruits. Approximately, 10,000 seeds, per species, were obtained and stored in plastic containers, at room temperature (20 °C), in Mexico City. For two weeks, bruchids (Insecta, Coleoptera) emerging from the seeds were collected and fixed in FAA for identification.

Since seed size and weight can influence the rate of germination (Matilla, 2004; Skogen *et al.*, 2010), 3,600 healthy mature seeds per taxon, were characterized. The evaluated variables were: length, width and thickness, as well as seed weight. Seed size was measured with a digital vernier caliper (Absolute Digimatic, CD-6"CS), and seed weight was determined individually using an analytical balance with 10<sup>-4</sup> g resolution (Adventurer, Ohaus, Melrose, USA). Healthy seeds, of both taxa, with a similar range of size and weight, were mixed to obtain a combined sample. It is important to mention that for assuring the adequate development of the whole assay, the range of seed size and weight was chosen in terms of the minimum number of seeds needed for it.

*Germination assays under laboratory conditions.* Legume seeds are known to have physical dormancy (Baskin, 2003; Van Assche *et al.*, 2003; Venier *et al.*, 2012; Gehan-Jayasuriya *et al.*, 2013), as *Mimosa* species (Martínez-Pérez *et al.*, 2006; Gehan-Jayasuriya *et al.*, 2013). Prior to any assay, all *Mimosa* seeds were washed with commercial detergent (3 g/100 ml water/3 min) (Camargo-Ricalde and Grether, 1998). Seeds were mechanically scarified: the seed coat was cut at the side opposite to the embryo's axis in order to avoid damage. Germination assays took place in an incubator (Convion T 38/Lb/AP), using trays (30 × 15 × 20 cm), with agar (3 %) as substrate. A temperature gradient with nine intervals of 5 °C each (from 5 to 45 °C), and a photoperiod of 12h light/12h darkness was set.

For every thermal treatment, five replicates of 20 scarified seeds each (100 seeds per taxon, 1,800 seeds in total), as well as five replicates of 20 unscarified seeds each as control (100 seeds per taxon, 1,800 seeds in total) were established. Seeds were considered as germinated when the radicle reached 1 mm in length (Bewley and Black, 1994). All the assays continued for 30 days.

To analyze the effect of temperature fluctuations in relation to the seed germination behavior of plants, diverse authors (*i.e.* Piedrahita-Cardona 1997, 1998; Enríquez-Peña *et al.*, 2004; Weng and Hsu, 2006; Pavón *et al.*, 2011) consider the parameters listed below, which will be used, as well, for the *Mimosa* taxa studied here:

Total Percentage of Germination (PG).- it is defined as the percentage of the total germination at the end of the assay.

Half Germination Rate ( $G_{50}$ ).- it is the number of days, after sowing, needed to reach 50 % of germination, and it was estimated by interpolating the day before and the day after in which 50 % of the total seed germination was obtained.

Germination Rate (GR).- it is defined as the correlation between the number of germinated seeds and the germination time:  $GR = \Sigma(n_i)/t$ , where  $n_i$  = number of germinated seeds in one day (i), t = time from sowing until the last seed germinates.

Saturation Rate (SR).- it is defined as the time for germination in relation to the germination percentage:  $SR = \Sigma(n_i t_i)/N$ , where  $n_i$  = number of germinated seeds in one day (i);  $t_i$  = number of days after sowing, N = total number of seeds sown.

Optimum Temperature (OT).- it was calculated as the average of all temperatures, weighted with the total seed germination percentage, at each temperature (Olf *et al.*, 1994).

*Statistical analyses.* Seed weight was compared between taxa using a *t*-student test ( $P < 0.05$ ). Homogeneity of variance was examined for all temperature treatments (Levene's test,  $P < 0.05$ ). When homogeneity of variance was found, a one-way ANOVA was used to analyze the intraspecific variation within the germination percentages and between control and scarified seeds, and a two-way ANOVA to examine the interspecific variation within the germination percentages, where the taxa and the temperatures were the variables (ANOVA,  $P < 0.05$ ). A multiple comparison test was employed (Tukey's HSD,  $P < 0.05$ ) to form groups. In *Mimosa aculeaticarpa* var. *aculeaticarpa*, the homogeneity of variance of SR was not found; therefore, the non-parametric Kruskal-Wallis test was used ( $P < 0.05$ ), and medians were compared by the Mann-Whitney test ( $P < 0.05$ , Sokal and Rohlf, 1995). All analyses were performed with the NCSS Software (Hintze, 2001).

## Results

*Seed size, weight, and production.* The two studied *Mimosa* taxa produce numerous seeds of variable weight and size. When comparing the weight of seeds between both taxa and within each taxon, no significant differences were showed ( $t = 0.7442$ ,  $P > 0.05$ ). In this sense, no significant differences were found, when comparing the size between and within both taxa; though, *M. aculeaticarpa* var. *aculeaticarpa* seeds were the biggest (Table 1).

Not all collected seeds were healthy; many of them were infested by bruchids (Insecta, Coleoptera). The mean percentage of infestation was 11.9 % for *M. aculeaticarpa* var. *aculeaticarpa*, and 48.7 % for *M. luisana* (Table 1). In the case of *M. aculeaticarpa* var. *aculeaticarpa*, two species of seed predator bruchids were identified, *Acanthoscelides chirichahuae* (Fall) and *A. mexicanus* (Sharp). *Mimosa luisana* seeds were infested by three species, two belonging to genus *Acanthoscelides* (*Acanthoscelides mexicanus* and *A. chirichahuae*) and one to genus *Stator* [*Stator pruininus* (Horn)].

*Scarification effect on the total percentage of seed germination (PG).* The two *Mimosa* taxa showed both epigeal and phanerocotylar germination, which started the first day after sowing. In both taxa, germination was stimulated by scarification (Figures 1-3; Table 2). The PG of unscarified seeds was low and slow. Scarified seeds registered a significantly higher PG ( $F = 143.88$ ,  $P < 0.001$ ; Table 2). Scarified seeds of *M. aculeaticarpa* var. *aculeaticarpa* germinated 57 times faster than those of *M. luisana*. In the case of *Mimosa luisana*, unscarified seeds germinated five times faster than those of *M. aculeaticarpa* var. *aculeaticarpa* (Figures 1-3). Although these data were not statistically significant, seeds of *Mimosa luisana* tended to germinate in greater numbers than those of *M. aculeaticarpa* var. *aculeaticarpa* (Table 2).

*Temperature effect on total seed germination percentage (PG).* The two-way ANOVA analysis showed a significant effect of the two factors, taxon and temperature, on the PG (Tables 3, 4). The lowest germination percentage of both taxa was recorded at 5 °C (Figures 1-3), differing significantly from the other temperature ranges ( $F = 33.03$  and  $F = 18.63$ ; both with  $P < 0.005$ , respectively). The highest PG (88 %) was recorded at 25 °C in *M. aculeaticarpa* var. *aculeaticarpa* ( $F = 33.03$ ;  $P < 0.005$ ); although, 88 % of

**Table 1.** *Mimosa* taxa mature seed size, weight, production, and percentage of infested and non-infested seeds by bruchids (Insecta, Coleoptera).

Variables / Taxon		<i>M. aculeaticarpa</i> var. <i>aculeaticarpa</i>	<i>M. luisana</i>
Size (mm)	Length	3.0-4.4	2.6-3.5
	Width	2.0-3.0	1.8-2.4
	Thickness	0.01-0.02	2.0-2.7
Mature seed weight (gr) (Mean ± SE)		0.008 ± 0.0004 <sup>a</sup>	0.00864 ± 0.0006 <sup>a</sup>
Production (number)		9,891	12,276
Infested seeds (%)		11.9	48.7
Non-infested seed production (%)		88.1	51.3

Values followed by the same letter are not significantly different ( $P < 0.05$ ).

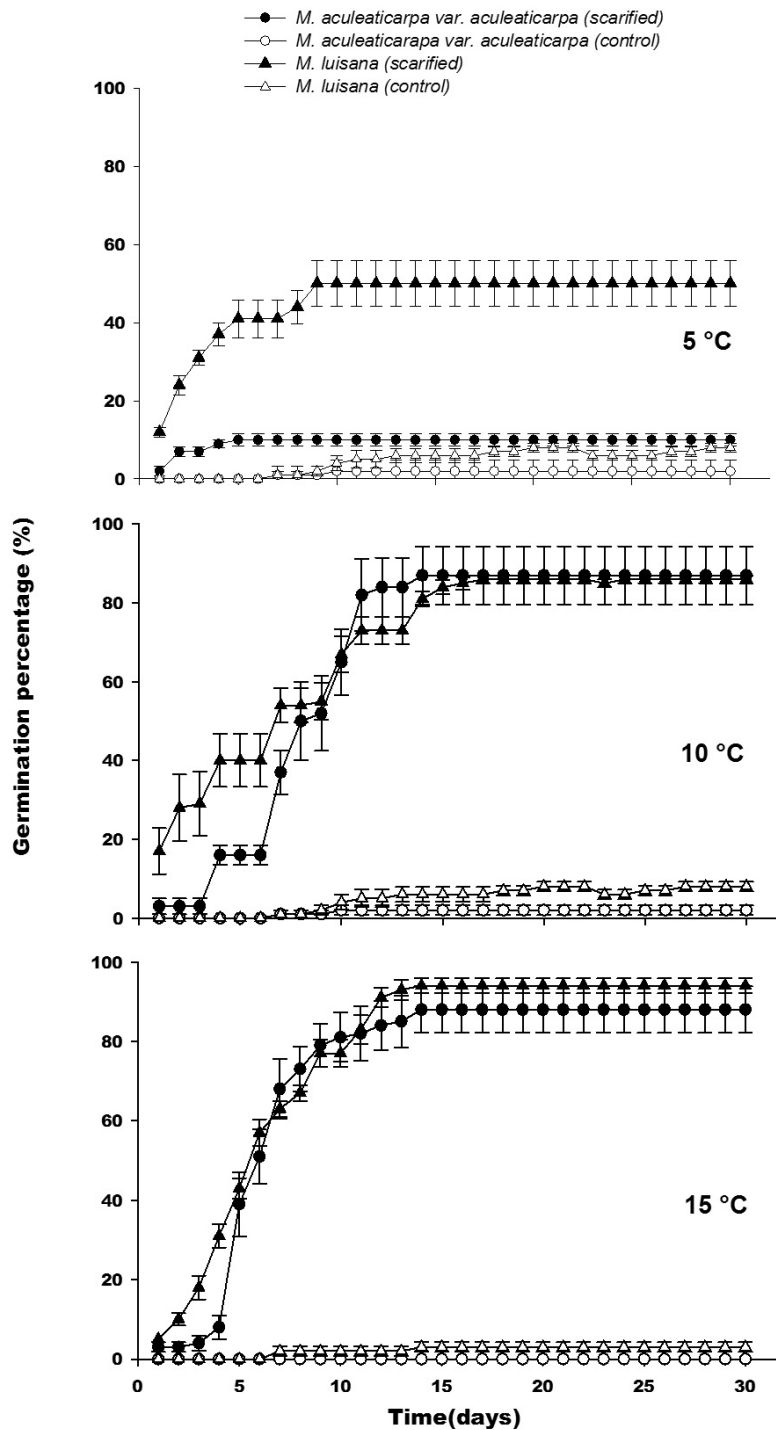


Figure 1. Seed Germination Percentage (PG  $\pm$  SE) of two *Mimosa* taxa along a temperature gradient 5 – 15 °C.

germination was also achieved at 15 °C, but seeds required more days (SR = 5) to reach this percentage (Table 4). In *M. luisana*, the highest percentage of germination (100 %) was reached at 30 °C; whereas, at higher or lower temperatures, the PG decreased (Figure 2; Table 4).

The effect of the interaction between the two factors, taxon and temperature, on the PG was significant ( $P < 0.001$ ),

indicating that the germinative response to temperature depended on the taxon. There was a decrease in the PG at extreme temperatures, 5 and 45 °C (Figures 1-3).

In relation to the half of seed germination ( $G_{50}$ ), the taxon factor presented an interaction with the temperature factor ( $P < 0.001$ , Table 3), indicating that seed germination response ( $G_{50}$ ) to temperature depended on the taxon.

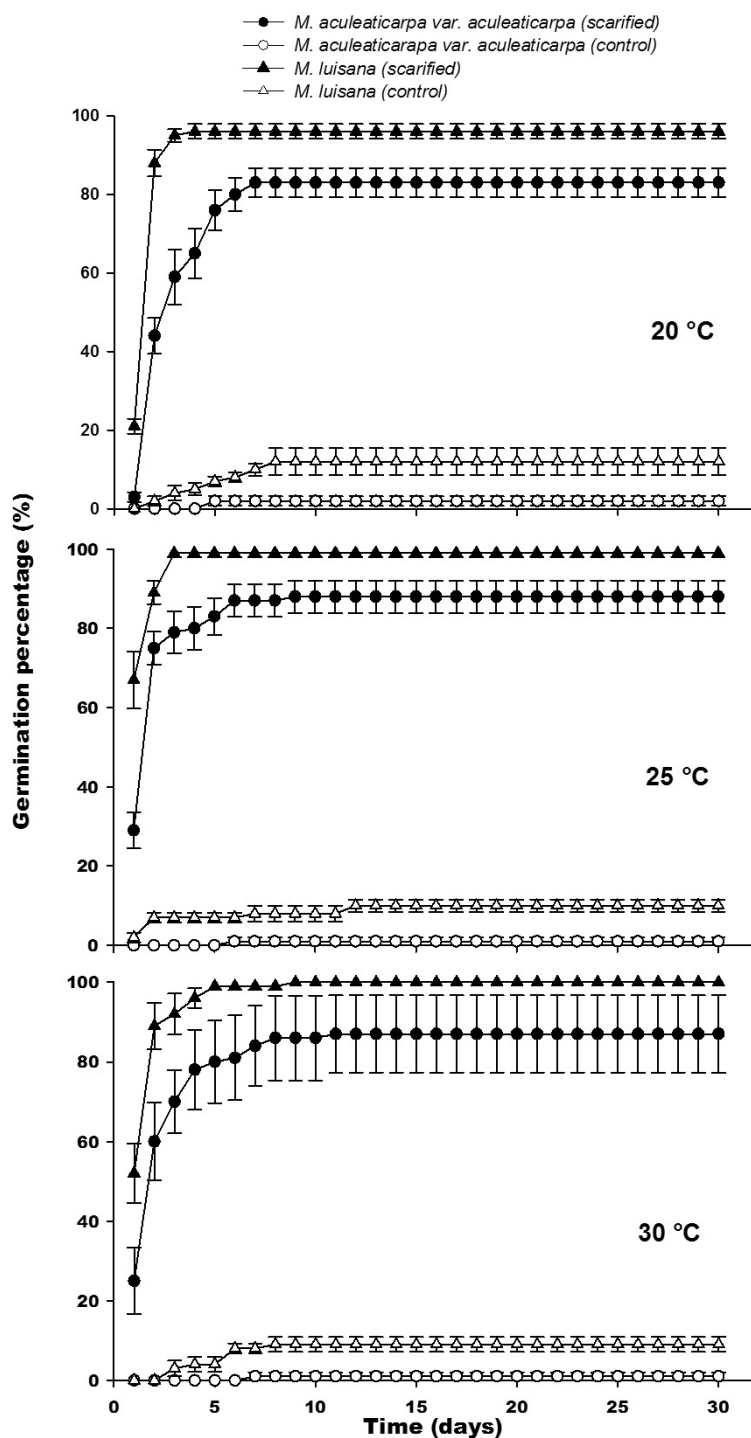


Figure 2. Seed Germination Percentage (PG  $\pm$  SE) of two *Mimosa* taxa along a temperature gradient 20 – 30 °C.

Although at 20 °C, *M. aculeaticarpa* var. *aculeaticarpa* reached 50 % of seed germination on the second day after sowing, at 25 and 30 °C, the 50 % of seed germination was exceeded on the second day after sowing. However, more days were needed to reach 50 % of germination at temperatures below 20 °C and above 35 °C. Moreover, only 35 % of seed germination was reached at 45 °C. In the case

of *M. luisana*, 50 % of seed germination was attained at 30 °C, the first day after sowing, and this percentage was exceeded at temperatures of 25, 35, and 40 °C; however, the total germination percentage decreased to less than 27 % at 45 °C (Figures 1-3).

Furthermore, the results showed that seeds of *M. aculeaticarpa* var. *aculeaticarpa* and *M. luisana* germinated



**Table 2.** Effect of seed scarification on the percentage of germination of two taxa of genus *Mimosa*.

Taxon	Germination (%)	
	Unscarified (Mean ± SE)	Scarified (Mean ± SE)
<i>M. aculeaticarpa</i> var. <i>aculeaticarpa</i>	1.22 ± 0.22 <sup>Aa</sup>	70.0 ± 8.35 <sup>Bb</sup>
<i>M. luisana</i>	6.33 ± 1.46 <sup>Aa</sup>	79.6 ± 8.31 <sup>Bb</sup>

Values followed by the same letter are not significantly different in columns (lower case) and rows (upper case),  $P < 0.05$ .

along the gradient of temperature (5-45 °C). However, the optimum temperature (OT) for seed germination for both taxa was determined at 25 °C (Table 4).

**Temperature effect on the germination rate (GR).** The Germination Rate (GR) was significantly affected by the taxon and the temperature factors ( $F = 9.9$ ;  $P < 0.001$ , Figure 4A, Table 3). GR increased significantly when the temperature reached 30 °C. In average, three seeds of *M. aculeaticarpa* var. *aculeaticarpa* germinated/day at 25 °C; whereas, six seeds of *M. luisana* germinated/day at the same temperature. However, the GR of seeds of both, *M. aculeaticarpa* var. *aculeaticarpa* and of *M. luisana*, decreased at temperatures above 30 and 35°C, respectively. In the case of *M. aculeaticarpa* var. *aculeaticarpa*, the GR increased at 45 °C; although, only 35 % of seed germination was recorded (Figure 4A).

Regarding the saturation rate (SR), a significant interaction between the two factors, taxon and temperature, was also observed ( $F = 4.38$ ;  $P < 0.001$ , Table 3), indicating that the SR depended on the taxon. *Mimosa aculeaticarpa* var. *aculeaticarpa* required numerous days to reach the PG; whereas, *M. luisana* required even more days to reach it at 15 °C (Figure 4B).

**Discussion**

The overall results obtained in this study are relevant under the context of possible temperature and precipitation fluctuation. In this case, this study tries to elucidate the influence of both temperature change and seed scarification on *Mimosa* seeds germination:

**Table 3.** Two-way ANOVA to analyze the effect of the temperature on the percentage of seed germination of two taxa of genus *Mimosa*.

	DF	Germination (%)	G <sub>50</sub> (d)	GR (seeds/d)	SR (d)
		F	F	F	F
Taxon (S)	1	20.65 <sup>ns</sup>	19.25 <sup>***</sup>	12.54 <sup>***</sup>	9.27 <sup>**</sup>
Temperature (T)	8	50.74 <sup>ns</sup>	49.95 <sup>***</sup>	17.71 <sup>***</sup>	68.04 <sup>***</sup>
S x T	8	3.38 <sup>***</sup>	3.62 <sup>***</sup>	9.92 <sup>***</sup>	4.38 <sup>***</sup>

G<sub>50</sub>=Half Germination Rate; GR=Germination Rate; SR=Saturation Rate; ns=not significant; \*\* $P < 0.01$ ; \*\*\* $P < 0.001$

**Table 4.** Total Percentage of seed Germination (PG) and Saturation Rate (SR) of two taxa of genus *Mimosa* under different temperatures, in a laboratory assay.

Temperature °C	Parameter/Taxon	<i>M. aculeaticarpa</i> var. <i>aculeaticarpa</i> (Mean ± SE)	<i>M. luisana</i> (Mean ± SE)
5	PG (%)	10 ± 1.58 <sup>Ab</sup>	50 ± 5.92 <sup>Bb</sup>
	SR	1	1
10	PG (%)	87 ± 7.34 <sup>Aa</sup>	86 ± 1 <sup>Aa</sup>
	SR	7	5
15	PG (%)	88 ± 5.83 <sup>Aa</sup>	94 ± 1.87 <sup>Aa</sup>
	SR	5	6
20	PG (%)	83 ± 3.74 <sup>Aa</sup>	96 ± 1.87 <sup>Aa</sup>
	SR	3	2
25	PG (%)	88 ± 4.06 <sup>Aa</sup>	99 ± 1 <sup>Aa</sup>
	SR	2	1
30	PG (%)	87 ± 9.69 <sup>Aa</sup>	100 ± 0 <sup>Aa</sup>
	SR	2	1
35	PG (%)	72 ± 8.30 <sup>Aa</sup>	81 ± 4.30 <sup>Aa</sup>
	SR	2	1
40	PG (%)	69 ± 7.31 <sup>Aa</sup>	84 ± 5.09 <sup>Aa</sup>
	SR	3	1
45	PG (%)	35 ± 6.12 <sup>Ab</sup>	27 ± 1.22 <sup>Ab</sup>
	SR	1	1
Optimum Temperature (°C)		25	25

Values followed by the same letter are not significantly different in columns (lower case), and a Tukey HSD to analyze the effect of temperature on the percentage of seed germination (upper case),  $P < 0.05$ .

**Seed size, weight and production.** Mature *Mimosa* seeds showed a significant size and weight variation even coming from the same mother plant. Both the small size of the seeds and their high production are intrinsic characteristics that lead to potentially more competitive plants, such as *Mimosa tenuiflora* (Willd.) Poir. (Camargo-Ricalde and Grether, 1998) and *Mimosa pigra* L., which are considered as pioneer or invasive species (Creager, 1992; Lonsdale, 1999).

On the other hand, predation of *Mimosa* seeds by bruchids (Insecta, Coleoptera), regardless of the size, could reduce their potential germination (Fontúrbel, 2002; Romero-Nápoles et al., 2005). In this study, the percentages of infested seeds were consistent with those reported by Orozco-Almanza et al. (2003) and Camargo-Ricalde et al. (2004), who found that the species of *Mimosa* had a high percentage of bruchid-infested seeds (30-75 %), reducing the number of viable seeds and, consequently, the number of seedlings for posterior recruitment.

In this study, seeds of *M. luisana* were infested by three different bruchid species; this *Mimosa* species had the highest percentage of parasitized seeds (48 %), as well as the highest GR; meanwhile, *M. aculeaticarpa* var. *aculeaticarpa* registered the presence of two bruchid species, which infested 11 % of the seeds, showing a lower percentage of

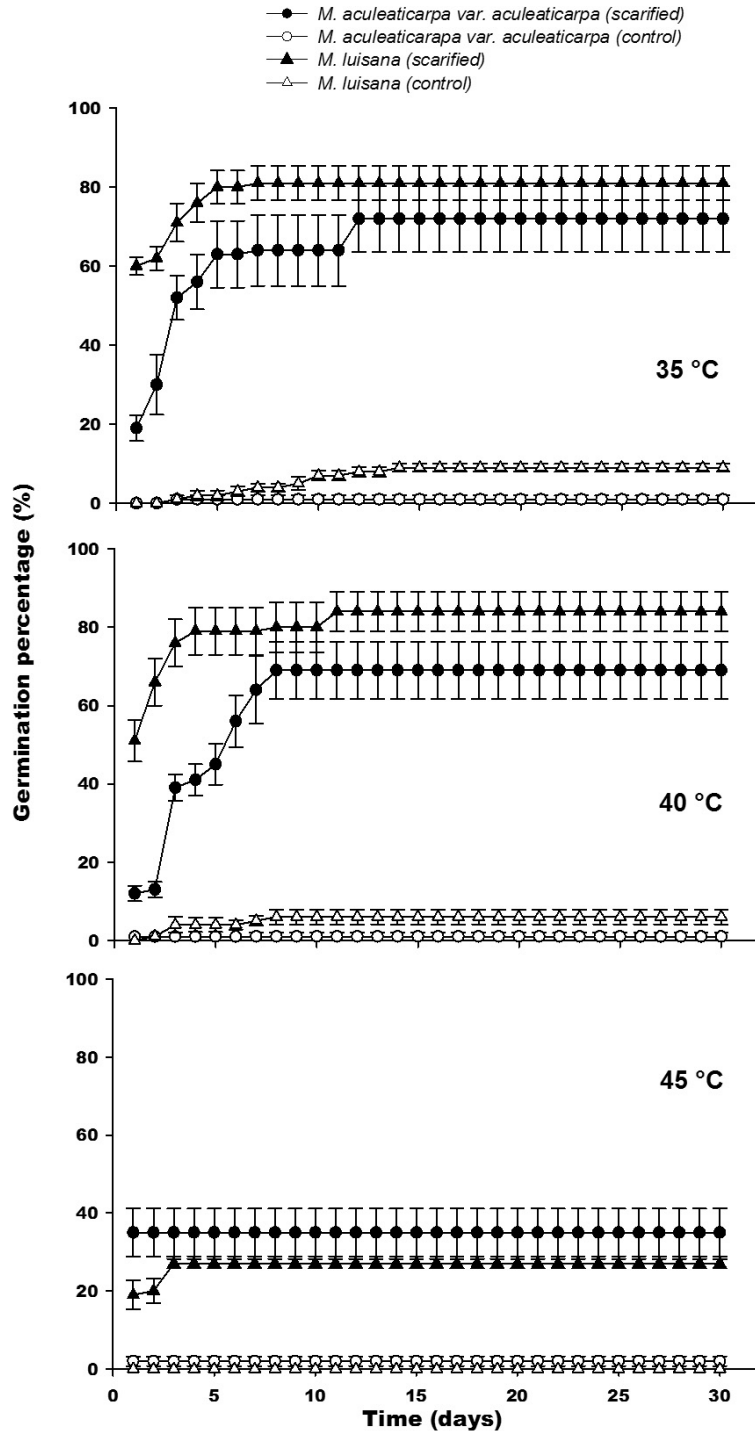
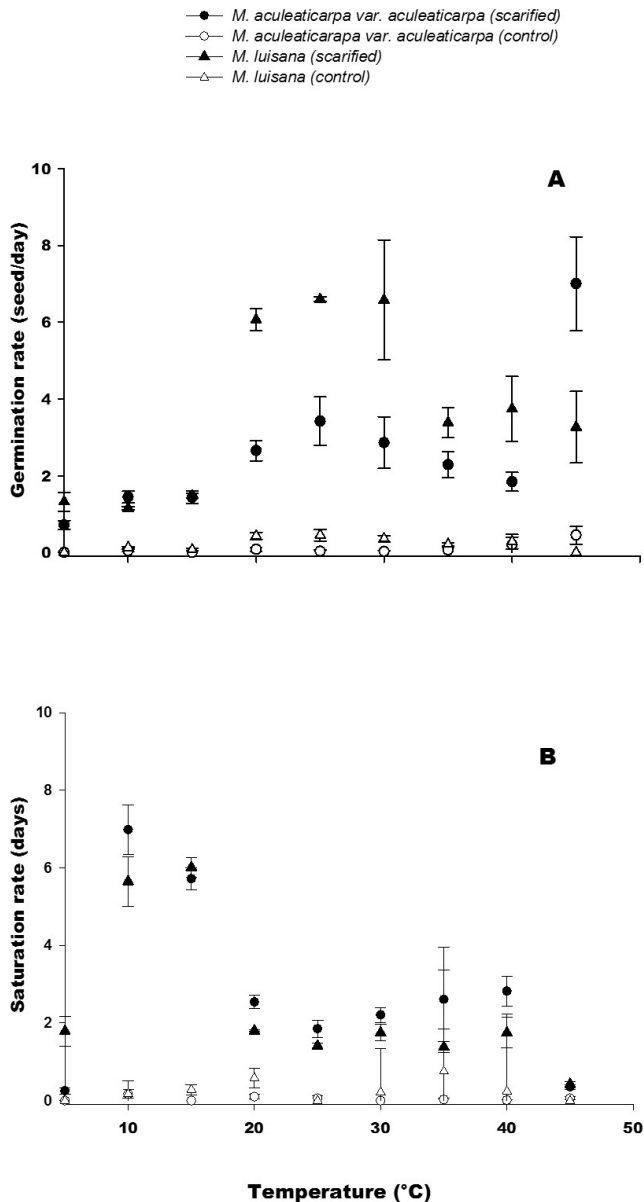


Figure 3. Seed Germination Percentage (PG  $\pm$  SE) of two *Mimosa* taxa along a temperature gradient 35 – 45 °C.

seed germination (PG), and suggesting that the presence of more than one bruchid species per plant, may be related to a reproductive strategy (Westoby *et al.*, 1992): a high percentage of bruchid-infestation leads to a high GR. It is possible that in nature, bruchids act as the “natural” seed mechanical scarifiers, as it happens in other plant species with a hard seed coat (Takakura, 2002).

*Effect of seed scarification on germination.* In general, legumes produce seeds with a waterproof coat that causes physical dormancy (Auld, 1996; Lopez *et al.*, 1999; Van Klinken and Flack, 2005), which restricts seed germination under natural conditions (D’Aubeterre *et al.*, 2002). According to Lonsdale (1993), Gehan-Jayasuriya *et al.* (2013), and Van Klinken and Goulier (2013), seeds of *Mi-*



**Figure 4.** A. Germination Rate (GR -  $\pm$  SE) of two *Mimosa* taxa along a temperature gradient; B. Saturation Rate (SR -  $\pm$  SE) of two *Mimosa* taxa along a temperature gradient.

*Mimosa* species present physical dormancy and, because the seed coat has one or more impermeable layers of palisade cells, a physiological dormancy is also possible (Baskin *et al.*, 2000). Therefore, scarification is necessary to promote seed germination (Camargo-Ricalde and Grether, 1998; Gehan-Jayasuriya *et al.*, 2013). Different seed scarification methods have been documented for *Mimosa* species (De Souza-Araújo and De Castro-Andrade, 1983; Camargo-Ricalde and Grether, 1998; Martínez-Pérez *et al.*, 2006; Silveira and Fernandes, 2006). However, De Souza-Araújo and De Castro-Andrade (1983), Camargo-Ricalde and Grether (1998), González-Castañeda *et al.* (2004), Mar-

tínez-Pérez *et al.* (2006), Chauhan and Johnson (2008), and Gehan-Jayasuriya *et al.* (2013), have demonstrated that mechanical scarification is the most appropriate technique for breaking seed dormancy of *Mimosa* species, since mechanically scarified seeds are able to reach the highest germination rates (GR) in comparison to other methods that, positively, damage the embryo and seedling development (*i.e.* chemical scarification, heat shock, etc.). In this study, mechanically scarified seeds reached high germination percentages (over 70 %) for both taxa; while, unscarified seeds only achieved 0-12 %. In nature, even if seed germination is low, *Mimosa* seed scarification occurs by different mechanisms such as changes in daily temperature, incomplete predation by insects as bruchids (Orozco-Almanza *et al.*, 2003; Camargo-Ricalde *et al.*, 2004), damage caused by soil microorganisms, abrasion by soil particles and fire (Baskin *et al.*, 1998; Taylor, 2005), and animal ingestion (*i.e.* goats, Baraza and Valiente-Banuet, 2008; Giordani, 2008).

However, it is important to mention that control seeds (unscarified seeds) should not be manipulated because the seed coat could be damaged, biasing the results. For instance, Martínez-Pérez *et al.* (2006) soaked control seeds of four species, including *M. aculeaticarpa*, in water/24 h, reaching *ca.* 60 % germination; almost the same germination behavior as the treated seeds from this study. Thus, control seeds should not be manipulated in any way.

Different assays (*i.e.* Camargo-Ricalde and Grether, 1998; Martínez-Pérez *et al.*, 2006; Leal and Biondi, 2007) confirm that *Mimosa* species have fast seed germination, which takes place within the first-fourth day after sowing; however, germination rate (GR) is related to temperature. This characteristic is also intrinsic to pioneer and invasive species (Lonsdale, 1999), and an important feature for plants with ecological restoration potential such as *M. luisana* (Camargo-Ricalde *et al.*, 2004) and *Mimosa aculeaticarpa* Ortega var. *biuncifera* (Benth.) Barneby (Pavón *et al.*, 2011).

**Temperature effects on seed germination.** *Mimosa* is a neotropical genus (Barneby, 1991; Simon *et al.*, 2011), growing in places where temperature oscillates between 0 and 35 °C (Cavalcante and Perez, 1995); temperatures that are consistent with this and other studies around the world (*i.e.* Moreno-Casasola, 1973; Parra, 1984; Creager, 1992; Ferreira *et al.*, 1992; Camargo-Ricalde and Grether, 1998; Camargo-Ricalde, 2000; Ursulino-Alves *et al.*, 2002; Orozco-Almanza *et al.*, 2003; Camargo-Ricalde *et al.*, 2004; Rivera-Aguilar *et al.*, 2005; Silveira and Fernandes, 2006; Cruz-Medina and Orozco-Almanza, 2010), which have defined ranges and optimum seed germination temperatures of 25 *Mimosa* species. Hence, *Mimosa* seeds are able to germinate between 0 and 45 °C; though, the optimum temperature (OT) registered for the two taxa studied here is 25 °C.

In this context, it is important to note that Cavalcante



and Perez (1995) suggested that tropical species can tolerate high temperatures, usually  $\geq 35$  °C. On the other hand, when seeds were exposed to cold temperatures ( $\leq 5$  °C), germination took place since *Mimosa* species did not slow their metabolic rate (Hendricks and Taylorson, 1976).

In general, in *Mimosa* species, temperature is not a limiting factor for seed germination, showing, as well, a fast GR, a high PG, and a fast growth (Camargo-Ricalde and Grether, 1998). These characteristics may explain why *Mimosa* species may be dominant or co-dominant elements within the plant communities where they occur; for instance, within arid and semi-arid ecosystems (Camargo-Ricalde *et al.*, 2002). Since ecosystems could expand or contract in reaction to plant species tolerance to an increase or decrease in temperature, the response of seed germination to scarification and temperature fluctuation is important to determine (Parmesan and Yohe, 2003). In this case, as both taxa of *Mimosa* are able to germinate from 5 to 45 °C, their populations may possibly be able to expand or contract in accompaniment with the ecosystems where they occur (*i.e.* temperate forest and arid tropical scrub).

*Effect of temperature on the germination rate (GR) and the saturation rate (SR).* Both taxa registered a high GR: 3.4 seeds/day for *M. aculeaticarpa* var. *aculeaticarpa* (25 °C), and 6.6 seeds/day for *M. luisana* (25-30 °C). Although, Carvalho and Nakagawa (2000) suggested that higher temperatures made seeds germinate faster due to an increase in water absorption speed and metabolic reactions. However, in the case of *M. aculeaticarpa* var. *aculeaticarpa*, when temperature was above 25 °C, the number of germinated seeds decreased daily; although, the GR increased at 45 °C. According to Baskin *et al.* (2000), this behavior may be the result of a temperature pulse that breaks the physiological dormancy.

Furthermore, when studying *Mimosa hostilis* (C.Mart.) Benth. (= *Mimosa tenuiflora*), De Souza-Araújo and De Castro-Andrade (1983) reported that the GR of scarified seeds showed a direct relationship with the PG, and these results are consistent with the results presented here.

The unscarified seeds of both *Mimosa* taxa had a GR of 0-0.45 seed/day. The highest value was achieved at 45 °C for *M. aculeaticarpa* var. *aculeaticarpa*, and 20-25 °C for *M. luisana*.

Regarding the SR, both *Mimosa* taxa studied required one to two days to germinate at 25 °C, after sowing, reaching a high germination percentages; whereas, the number of days for seed germination increases at temperatures under 25 °C, due to a decrease in seed metabolism (Amaral and Paulilo, 1992). The study carried out by Pavón *et al.* (2011) on the germination of scarified seeds of *M. aculeaticarpa* var. *biuncifera*, confirmed the inverse relationship between GR and SR: while GR increased, SR decreased. However, this inverse relationship was not found when comparing

unscarified seeds, where no significant differences were found neither among temperatures nor species.

Finally, if part of the success of a plant species is focused on its tolerance to the temperature factor, the overall results suggest that both *Mimosa* taxa may be able to succeed under different thermal scenarios. However, more studies are needed to understand how other climatic factors, such as different levels of precipitation and CO<sub>2</sub> concentrations, may affect the seed germination behavior of plant species.

## Conclusions

Seeds of *Mimosa*, like those of other legumes, have a hard and impermeable coat. To germinate, these seeds require scarification, and mechanical scarification is the best method, as neither the embryo axis nor the seedling is damaged. A comparison between healthy seeds and those preyed by bruchids is also relevant, since these insects are considered natural controls of *Mimosa* species populations.

Our hypothesis was confirmed: seeds of the two *Mimosa* taxa studied germinated according to the typical temperatures to their habitats, and both taxa showed tolerance to a wide thermal scenario. Furthermore, in this study, because seeds of both *Mimosa* taxa are capable to germinate under a wide range of temperatures, their populations may possibly be able to expand or contract in accompaniment with the ecosystems where they occur.

Currently, both *Mimosa* taxa may be used in ecological restoration programs. As Mexican native taxa, they favor successional processes leading to the restoration of their original ecosystems, respectively. We propose that: (i) In temperate forests (Mexican mesic ecosystems), *M. aculeaticarpa* var. *aculeaticarpa* would restore environments where the temperature is never below 5 °C or above 45 °C, and (ii) In the arid tropical scrub (dry ecosystem), *M. luisana* seeds would germinate in a wider range of temperatures, suggesting that this narrow endemic species is a good option for the restoration of the semi-arid ecosystem at the Valley of Tehuacán-Cuicatlán, Mexico.

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