*International Research Journal of Environmental Sciences*_____ ISSN 2319–1414
Vol. **6(4),** 13-17, April **(2017)** *Int. Res. J. Environmental Sci.*

Germination of *Bursera bipinata (DC.) Engl* and *Bursera glabrifolia (H.B.K.) Engl* under pre-germinative treatments

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Available online at: www.isca.in, www.isca.me
Received 29th January 2017, revised 6th April 2017, accepted 20th April 2017

Abstract

The Bursera family has been hardly studied in Mexico even when it is of major ecological and economic importance, for regeneration including preservation of biodiversity in deciduous tropical forests and the use for wood and resins by the human communities dwelling therein. This study was conducted in a greenhouse of Colegio de Postgraduados, campus Montecillo, Estado de México, to assess the seed germination of Bursera glabrifolia and Bursera bipinatausing 240 seeds from each species, with 60 seeds per treatment. Pre-germinative treatments in the study involved the soaking of seeds under different conditions: i. 95% sulphuric acid for 5 minutes; ii. hot water at 80ºC for 3 minutes; iii. 300 ppm Gibberellin AG5 for 24 h.; iv. water at 20 degrees as the control group. The substrate used was constituted by25%agrolite, 25% vermiculite and 50% peatmoss. Three seeds were putper 220 CC plastic tree tube. Three months after seeding the germinated seeds were counted. The largest number of germinated seeds was obtained with the gibberellin AG5 treatment, and the second largest was obtained from the control group. Treatments with sulphuric acid and hot water had a low effect on seed germination.

Keywords: Germination, *Bursera bipinata, Bursera glabrifolia,* Pre-germinative treatments, Dry tropical forests, Deforestation.

Introduction

Due to the serious deforestation of the dry tropical forests and lowland deciduous forests of Mexico there is an urgent need to propagate their native species. Around 70% of the lowland forest area has deteriorated in the last years, and 50% of this ecosystem is altered[1].

Bursera family includes around 100 species exclusively distributed in the American continent from the south west of United States and to the north of Peru and Brazil, including isolated regions like the West Indies and the Galapagos Islands.

From the total of species in the *Bursera* family, at least 80% of them live naturally in Mexico and are mainly distributed along the Pacific coast. The highest diversity of species in the *Bursera* genre is found in the lowland deciduous forests of Oaxaca, Guerrero and Chiapas[2].

Studies on seed germination of *Bursera* are scarce, in spite of the major relevance of these species not only from the ecological viewpoint but also from the economic viewpoint (regional artcraft production).

The propagation and reintroduction of species like *Bursera bipinata* and *Bursera glabrifolia* are needed to restore the composition and structure of natural communities, and to meet the demand of resin or incense, used as traditional medicine and for ceremonial purposes[3] by the human groups in the region.

Both species, *Bursera glabrifolia* and *Bursera bipinata,* have been ancestrally used by the inhabitants of the regions where they naturally grow. The latter is used in the manufacturing of wooden artcraft (alebrijes) and the former is used to extract the resin known as copal.

In the last few years both species have been over exploited, which, in addition to their scarce or zero natural regeneration, has seriously diminished their natural populations[4,5].

In consideration of the stated above, this study assessed the seed germination of *Bursera glabrifolia* and *Bursera bipinata* under four pre-germinative treatments.

Materials and methods

The study was carried out in the nursery of the Forestry Sciences Graduate Unit (Posgradoen Ciencias Forestales) of Colegio de Postgraduados campus Montecillos, Estado de México, located at latitude 19º17' N and longitude 98º54' W, at 2,240 m. Climate in this location is temperate sub humid with precipitation during the summer with an means annual rainfall of 750mm and an average temperature of 15.5ºC[6].

The seeds of *Bursera bipinata* and *B. glabrifolia* were donated by Instituto Tecnológico del Valle de Oaxaca (Technological Institute of the Oaxaca Valley), located in Ex-Hacienda de Nazareno, Santa Cruz Xoxocotlan, in the region of Valles Centrales (Central Valleys) of Oaxaca (latitude 17°02' N, longitude 96°44' W, at1,530 m.)[7].

Before establishing the experiment, the germplasm used was tested in the seed lab of División de Ciencias Forestales (DICIFO, for the Spanish acronym of Forestry Sciences Division) of Universidad Autónoma de Chapingo. The test methodology was based on[8]:

Morphological characterisation of the seed: Twenty five seeds of each species with four replications were used for the analysis, making a total of 100 seeds. In every seed the parameters of width (in its widest portion), total length, average thickness and total weight were measured.

Purity: This assessment was made to identify seed quality in the lot, stated as the percentage of clean seed. In this analysis 25 sample seeds were chosen, weighted and cleaned; impurities were removed and seeds were weighted again. The average value was then obtained using this formula:

$$P = (PSL/PSI)\ (100)$$

Where: PSL is the dry weight with no impurities and PSI is the dry weight with impurities.

Water content: This test was made to define the moisture content of the seeds, which was defined on the basis of dry weight (CHo). Two sub samples of 25 seeds were taken, one from each species. Sub samples were weighted to get the fresh weight (PF), they were put in Petri dishes and were introduced in the drying furnace at 70ºC for three days until getting a constant weight, thus getting the anhydrous weight (PO).

To get the value of moisture content the following equation was used:

$$CHo = ((PE-PO)/PO)\ (100).$$

Viability: This test indicates the number of seeds with a live embryo in the lot. The test a was removed from 25 seeds and a longitudinal cut was made on them to expose the embryo and then they were immersed in tetrazolium chlorine for 24 hours, in a dark environment inside Petri dishes coated with aluminium foil. Later on, the seeds that got a pinkish colour, the sign that their embryos are viable to germinate, were counted.

Germination: To define the germination percentage, 20 seeds of each species were set aside. Each group of seeds was deposited in an air tight plastic box with a felt pad humidified with distilled water in the bottom. The boxes were put in a germination chamber at a constant temperature of 30ºC with no light for two months, and the seeds that germinated every week were counted.

The completely randomized block design was used as the experimental design, where the block is the species and the treatments were randomly distributed.

The pre-germinative treatments for each one of the species consisted in: i. soaking 60 seeds for 24 hours in 300 ppm of hormone (Gibberellin AG5); ii. soaking 60 seeds in 95% sulphuric acid for 5 minutes; iii. soaking 60 seeds in hot water at 80ºC for 3 minutes and; iv. soaking 60 seeds in plain water at temperature of 20 degrees for 24 h. (control group).

It is important to note that the three former treatments went through soaking with water at room temperature for 24 h. after their pertinent treatment was applied.

Three seeds of each species were seeded per treatment in 220 CC plastic tree tubes in a soil mixture composed of 50% peat moss, 25% agrolite and 25% vermiculite. The 220 CC plastic tree tubes were put in crates placed on top of Tables and the treatments were randomly arranged. The plastic tree tubes were watered everyday using a hand held shower. Three months after seeding the germination experiment was finished and the number of germinated plants was counted.

Statistical analysis: The statistical analysis of the information was made using the SAS® statistical package with the ANOVA procedure to identify the effect of treatments and to get the comparison of measurements through the Tukey test.

Results and discussion

Seed morphology: ***Bursera glabrifolia:*** Under normal storage conditions, the seeds of this species contain 9% of humidity and weight 0.053 gr, on average, yielding a total of 469,043 seeds per kilogram. Their average length is 5.52mm, the average thickness is 4.17mm and the average widest part of the seed measures 5.01mm (Table-1).

The viability percentage in the lot was good (76%), which was not the case of the germination percentage that showed low values (15.35%), thus indicating that this particular species has a natural difficulty to germinate (Table-1).

Burserabipinata: The seeds in this species are smaller than those of the preceding one; they contain 8% of humidity and weight 0.0230 gr, with a total of 781,250 seeds per kilogram as an average. Their average length is 4.78mm, their average thickness is 3.03mm and the average measurement of the seed widest part is 3.77mm (Table-1).

The viability percentage in the lot was (76%), but the germination percentage of this species had low values (9.86%), thus indicating that this particular species has a natural difficulty to germinate (Table-1).

Table-1: Analysis of *Bursera glabrifolia* and *B. bipinata*seeds.

Species	Seeds in 1 kg	Purity (%)	Water content	Viability (%)	Germination (%)	Length (mm)	Width (mm)	Thickness (mm)
B. glabrifolia	469,043	89,7	9,15	76	19,73	5,52	5,01	4,17
B. Bipinata	781,250	99	8	76	13,15	4,78	3,77	3,03

Germination of seeds from both species under the pre-germinative treatments: The statistical analysis indicates that, with a 5% level of significance ($p < 0.05$), gibberell in AG5, applied as pre-germinative treatment, had an effect in the germination of seeds of *Bursera bipinata and Bursera glabrifolia* (Table-2).

Figure-1 shows the number of germinated seeds per pre-germinative treatment, considering both species. The largest number of germinated seeds corresponds to those treated with gibberellin AG5, with a significant difference over the seeds treated with sulphuric acid, hot water and water at room temperature (the control group).

Figure-2 shows the number of germinated seeds per species, where it can be noted that, on average, 15.35% of germinated seeds corresponds to *Bursera glabrifolia* and 9.86% correspond to *Burserabipinata*.

Table-2: Analysis of variance of the germination of *Bursera glabrifolia and B. bipinata* under the different pre-germinative treatments.

Source of variation	Degrees of freedom	Level of significance
Product (gibberellin AG5)	3	0.0168
Species	1	0.1394

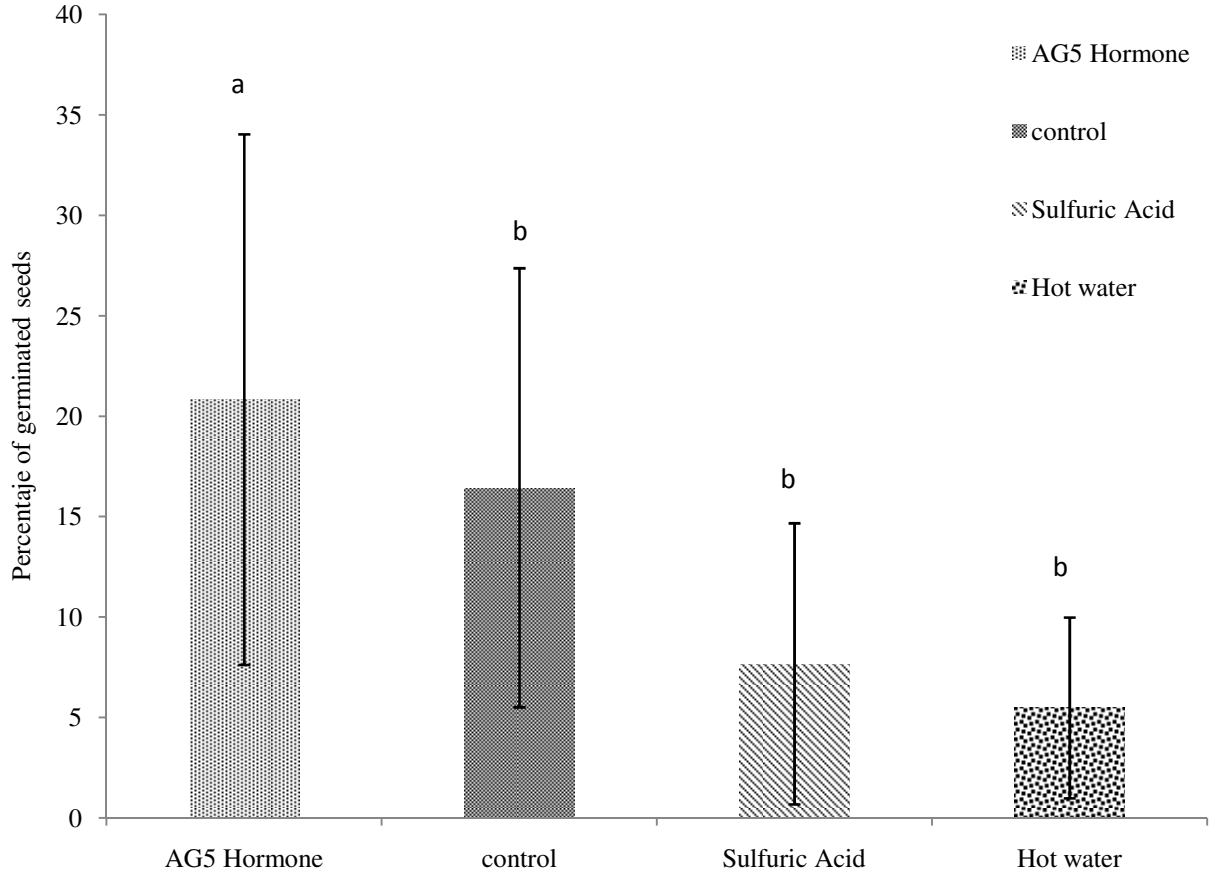

Figure-1: Number of germinated seeds in each pre-germinative treatment. Equal letters mean lack of significant differences according to the Tukeytests (α=0.05).

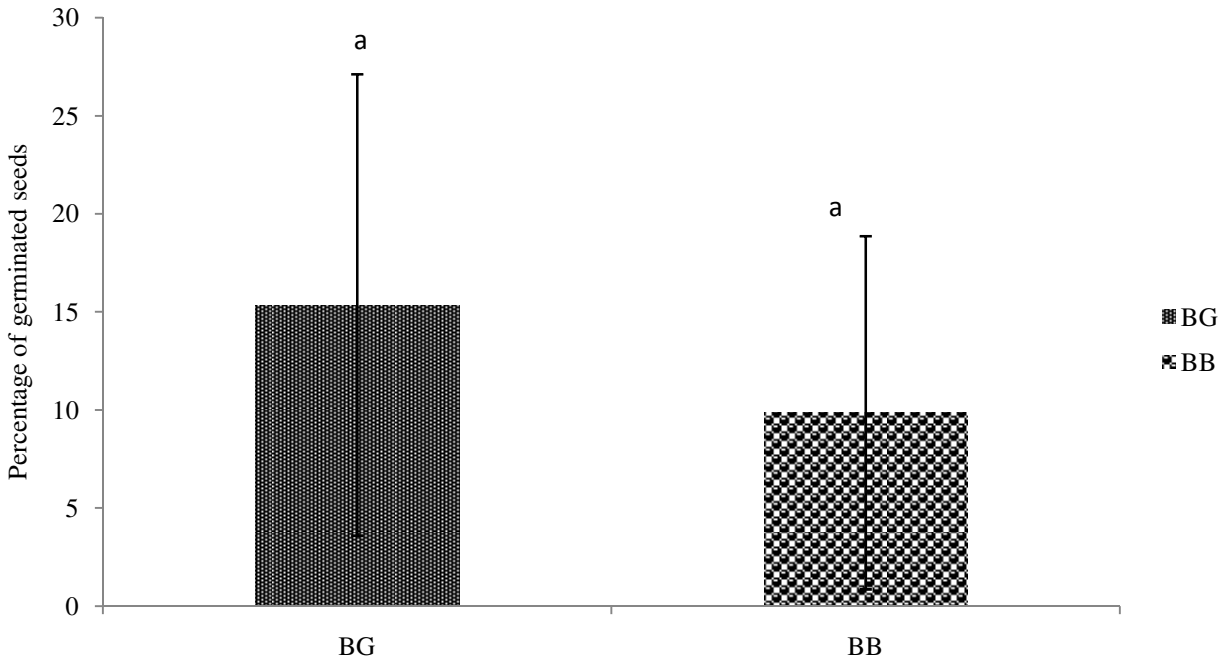

Figure-2: Number of germinated seeds per species. Equal letters mean absence of significant differences according to Tukey tests (α=0.05).

Discussion: An analysis was made on the seeds of *Bursera glabrifolia* and *Bursera bipinata,* which includes their morphological characterization and their germination potential. The results obtained are similar to those reported in past studies. Nonetheless, the studies on the seeds of these species are scarce.

A study made by Hernández-Pérez[9] shows that the distribution and the number of seeds produced by the *Bursera* family mostly depends on the climate conditions where the species grow. A different study conducted by Ortiz-Pulido y Pavón[10] indicates that ground slope and the planting orientation also have an influence on seed production of the *Bursera* genre. Therefore, it is very difficult to define and standardize the morphological features of each species due to their wide distribution and the environmental conditions in the sites where they grow.

The germination percentage of the *Bursera glabrifolia* and *Bursera bipinata* seeds was 19.73 % and 13.5%, respectively. These figures are low when contrasted with the findings of Bonfil[11], who reported germination percentages of 30 to 60% for *B. glabrifolia, B. bipinata, B. copallifera and B. bicolor.*

The treatment with the largest germination percentage was that of gibberellin AG5, because, according to Salisbury and Roos[12], this hormone has properties that promote germination, promote the elongation and emergence of radicles, and produces hydrolytic enzymes during germination. Orantes-García[13], also found that the seeds of *Bursera bipinata* treated with gibberellin AG5 had a larger germination percentage. Similar results in *B. copallifera* were found by Bonfil[11], with the use of cytosine (150 ppm of 6-Benzylaminopurine).

The second best treatment for both species was the control one. This result agrees with Latsague[14], who report that, even if the seed is not submitted to chemical substances, the mere fact of soaking seeds in water at room temperature for 24 hours increases its germination capacity. Bonfil[11] found that this type of management (control treatment) in one of their germination experiments with 6 species of *Bursera* had similar results to those obtained in this study.

With the pre-germinative treatments of sulfuric acid and hot water there was a low germination percentage in the seeds of *Bursera glabrifolia and B. bipinata*. This can be due to the inhibition of the germination capacity or because they had no influence on it. Similar results, although in other species, were found by Bilbao and Matías[15] who treated seeds of *Cenchrus ciliaris* cv. Biloela with 95% sulfuric acid and hot water at 70°C and found that said pre-germinative treatments did not show any influence on the germination of seeds at any time period of exposure.

On top of that, the acid caused the embryos of most of the treated seeds to rot. This is also confirmed by Narbona[16], in their germination study on strawberry tree (*Arbutus unedo* L.) seeds. In other species of *Bursera*, Nargaraja and Farooqi[17] reported that these two pre-germinative treatments promote the germination of recently harvested seeds and their effect is reduced in stored seeds.

Conclusion

The morphological description of *Bursera glabrifolia* and *B. bipinata* seeds in this study provides new information on the *Bursera* species from the state of Oaxaca.

The seeds of *Bursera glabrifolia* and *Bursera bipinata* had a better germination rate when 300 ppm gibberellin AG5was applied (20.83%); although the seeds in the control treatment also presented a good germination percentage (16.44%). The sulfuric acid and hot water treatments had a lower effect on seed germination, 7.6% and 5.4%, respectively.

It is recommended to conduct other studies on the same species that consider the time of seed storage and different regions of natural distribution in order to validate the results obtained herein.

References

1. Trejo I. and Dirzo R. (2000). Deforestation of seasonally dry tropical forest: a national analysis in Mexico. *Biological Conservation*, 94(2), 133-142.

2. Cházaro B.M., Mostul B.L. and García L.F. (2010). Mexican copals (Bursera spp.). *Bouteloua*, 7, 57-70.

3. Loeza-Corte J.M., Díaz-López E., Campos-Pastelín J.M. and Orlando-Guerrero J.I. (2013). Efecto de lignificación de estacas sobre enraizamiento de Bursera morelensis Ram. y Bursera galeottiana Engl. en la Universidad de la Cañada en Teotitlán de Flores Magón, Oaxaca, México. *Ciencia Ergo Sum*, 20(3), 222-226.

4. Andrés-Hernández A.R. and Espinosa-Organista D. (2002). Morfología de plántulas de Bursera Jacq. ex L.(Burseraceae) y sus implicaciones filogenética. *Soc. Bot. México,* 70, 5-12.

5. Barrales-Alcalá B.A. (2009). Bursera copallifera initial establishment of three sites with different degrees of disturbance.Professional thesis (Biology). Faculty of Sciences, Universidad Nacional Autónoma de México. México, D. F. México., 41.

6. Reyes Reyes J., Aldrete A., Cetina A.V.M. and López U.J. (2005). Producción de plántulas de Pinus pseudostrobus Var. apulcensis en sustratos a base de Aserrín. *Chapingo Journal, Forestry and Enviromental science series* 11(2), 105-110.

7. Ruiz L.J., Azcona C.M.I. and Velasco V.V.A. (2008). Risk factors and blood lead levels in undergraduate students. *Naturaleza y Desarrollo*, 6(1), 26-32.

8. Rodríguez T.D.A. (1993). Analysis of forest seeds. Universidad Autónoma Chapingo. División de Ciencias Forestales, 48.

9. Hernández-Pérez E., González-Espinosa M., Trejo I. and Bonfil C. (2011). Bursera gender distribution in the state of Morelos, Mexico and its relationship with climate. Mexicana de Biodiversidad, 82, 964-976.

10. Ortiz-Pulido R. and Pavón N.P. (2010). Influence of slopeorientationon sex ratio and sizedistribution in a dioeciousplant Bursera fagaroides var. purpusii., *PlantEcology*, 208(2), 271-277.

11. Bonfil S.C., Cajero L.l. and Evans R. (2008). Seed germination of six species of Burseras from central Mexico. *Agrociencias,* 42(7), 82-834.

12. Salisbury F. and Ross C. (2000). Physiology of plants. Plant development and environmental physiology. Edition Thomson Paraninfo. Spain, 3, 988.

13. Orantes-García C., Pérez F.M.A., Rioja P.T.M. and Garrido E.R. (2013). Viability and seed germination of three tree species native to the rainforest, Chiapas, Mexico. Polibotánica, 36, 117-127.

14. Latsague V.M., Sáez D.P. and Coronado A.L. (2010). Pregermination treatments of Myrceugenia exsucca (Myrtaceae). *BOSQUE*, 31(3), 243-246.

15. Bilbao B. and Matías C. (1979). Effect of different methods of scarification on germination of seeds Cenchrus ciliaris cv. Biloela. *Pastos y Forrajes,* 2(2), 225-238.

16. Narbona E., Arista M. and Ortiz P.L. (2003). Germinación de las semillas del madroño (Arbutus unedo L., Ericaceae). *ActaBotánicaMalacitana,* 28, 73-78.

17. Nargaraja C. and Farooqi A. (1989). Studies on the seed germination as influenced by various pre treatments in Bursera. *Indian Perfumer,* 33(1), 48-53.