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# Germinação e vigor de sementes de *Encholirium spectabile* em função da região geográfica, substrato e posição de semeadura

# Germination and vigor of *Encholirium spectabile* seeds according to geographical region, substrate and sowing position

# Germinación y vigor de semillas de *Encholirium spectabile* en función de la región geográfica, sustrato y posición de siembra

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**Resumo**

Dado o potencial de exploração comercial de *E. spectabile* e visando coibir o extrativismo predatório, torna-se relevante o desenvolvimento de pesquisas que orientem seu cultivo. Este estudo teve como objetivo avaliar o efeito de diferentes substratos e posições da semente na semeadura sobre a germinação e o vigor de sementes de duas regiões geográficas. O delineamento experimental foi inteiramente casualizado em esquema fatorial 2 x 4 x 2 (sementes de duas regiões geográficas: Serra Talhada-PE e Graça-CE, ambas no Brasil; quatro substratos: papel mata-borrão, areia, vermiculita e fibra de coco; e duas posições de semeadura: sobre e entre o substrato). A semeadura em papel mata-borrão, areia, vermiculita e fibra de coco, em geral, foi favorável a germinação e ao índice de velocidade de germinação das sementes das duas regiões geográficas. A semeadura entre a fibra de coco foi prejudicial à germinação e ao desenvolvimento radicular, independentemente da região geográfica das sementes. No entanto, a semeadura entre e sobre a fibra de coco e entre areia favoreceu o crescimento da parte aérea. As melhores combinações para produção de massa seca foram semear sobre papel e areia. Os substratos, papel mata-borrão, areia, fibra de coco e vermiculita são favoráveis à germinação das sementes de *E. spectabile*, independentemente das regiões geográficas, desde que a semeadura seja feita sobre os substratos.

**Palavras-chave:** Bromeliaceae; Macambira-de-flecha; Produção de mudas; Semiárido.

## Abstract

Given the potential for commercial exploitation of *E. spectabile* and aiming to curb predatory extractivism, the development of researches that guide its cultivation becomes relevant. This study aims to evaluate the effects of different substrates and seed positions at sowing on the germination and vigor of seeds from two geographical regions. The experiment design was completely randomized in a 2 x 4 x 2 factorial design (seeds from two geographic regions: i.e., Serra Talhada-PE and Graça-CE, both in Brazil; four substrates: blotting paper, sand, vermiculite and coconut fiber: and two sowing positions: over and in between the substrate). Sowing on blotting paper, sand, vermiculite and coconut fiber, in general, was favorable to germination and germination speed index for seeds from both geographic regions. Sowing in between coconut fiber was detrimental to germination and root development regardless of the seed geographical region. However, sowing in between and over coconut fiber and in between sand favored shoot growth. The best combinations for dry matter production were sowing over paper and sand. The substrates blotting paper, sand, coconut fiber and vermiculite are favorable to *E. spectabile* seed germination regardless of geographic regions, provided that the sowing is made over the substrates.

**Keywords:** Bromeliaceae; ‘Macambira-de-flecha’; Seedling production; Semiarid.

## Resumen

Dado el potencial para la explotación comercial de *E. spectabile* y con el objetivo de frenar la extracción depredadora, el desarrollo de la investigación para guiar su cultivo se vuelve relevante. En el presente estudio se evaluó el efecto de diferentes sustratos y posiciones de siembra sobre la germinación y el vigor de semillas oriundas de dos regiones geográficas. Se utilizó un diseño completamente aleatorizado, en arreglo factorial 2 x 4 x 2 (semillas de dos regiones geográficas, es decir, Serra Talhada-PE y Graça-CE, ambas en Brasil; cuatro sustratos: papel secante, arena, vermiculita y fibra de coco; y dos posiciones de siembra: sobre y entre el sustrato). La siembra sobre papel secante, arena, vermiculita y fibra de coco, en general, fue favorable a la germinación ya la velocidad de germinación para las semillas de ambas regiones. La siembra en fibra de coco fue perjudicial para la germinación y el desarrollo radicular, independientemente de la región geográfica de las semillas. Sin embargo, la siembra entre y sobre fibra de coco y en arena favoreció el crecimiento de la parte aérea. Las mejores combinaciones para la producción de masa seca fueron la siembra sobre papel y arena. Los sustratos papel secante, arena, fibra de coco y vermiculita son favorables a la germinación de semillas de *E. spectabile*, independientemente de las regiones geográficas, siempre que la siembra sea sobre los substratos.

**Palabras clave:** Bromeliaceae; 'Macambira-de-flecha'; Producción de plantulas; Semiárido.

## 1. Introduction

Drought is a climatic condition affecting several inhabited continents. It challenges the people's ability to produce food for both human and animal consumption. Thus, the agricultural exploitation of drought-resistant plant species that serve as a food source has become a reality in several regions of the world. In this perspective, the bromeliaceae *Encholirium spectabile* Mart. ex Schult. & Schult. f. stands out as resistant to drought. It is still used as food in the semiarid region of Caatinga, a Brazilian biome (Nascimento et al., 2012).

The identification of health-enhancing phytosterols such as campesterol and β-sitosterol and the absence of compounds with a significant toxicity in leaves of both *E. spectabile* and *Bromelia laciniosa* Mart. ex Schult. f. highlight these species as promising food sources in seasons and regions where there is a frequent water stress, confirming the importance of such plants as food (Juvik et al., 2017). *E. spectabile* has a nutritional value close to that of rice seeds (*Oryza sativa* L.). The plant part used as food is the basal portion of leaves, which presents 28.7% of carbohydrates, 0.7% of proteins and 0.8% of lipids. Dry leaves are also used for the preparation of flour (Nascimento et al., 2012).

*E. spectabile* is a xeromorphic species belonging to the family Bromeliaceae, subfamily Pitcairnioideae, popularly known as "macambira-de-flecha" (Ferrari et al., 2016). It has a restricted distribution to the Brazilian territory, occurring frequently in rocky outcrops. It grows generally exposed to high sunlight rates in the Caatinga, Cerrado and Mata Atlântica domains (Carvalho et al., 2010).

Recently, the pharmacological potential this bromeliaceae has been emphasized. Studies have evidenced that the ethanolic extract of *E. spectabile* has a gastroprotective activity against induced damages in the gastric mucosa. This suggests that this extract may activate cytoprotective mechanisms that increase the release of prostaglandins (Carvalho et al., 2010). Moreover, by the evaluation of crude extracts of leaves of *E. spectabile*, Santana et al. (2012) found antioxidant and antibacterial properties. Oliveira Júnior et al. (2013) investigated the presence of phenolic compounds and flavonoids in dry extracts of *E. spectabile*. They may act as natural sources of antioxidant agents, evidencing the possibility of using these extracts as a sunscreen in pharmaceutical preparations.

Due to the potential for a commercial exploitation of *E. spectabile* curbing predatory extractivism, the development of researches that guide its cultivation in an economically sustainable way becomes relevant. However, studies on germination, vigor and establishment of *E. spectabile* seedlings are still incipient in face of the importance of knowledge on the germinative behavior of seeds for the perpetuation and preservation of this species.

Germination is affected by internal factors, such as seed viability and longevity, and external factors, such as water availability, oxygen, temperature, luminosity and substrate (Carvalho & Nakagawa, 2012). Substrate directly affects germination due to its structure, water retention capacity, propensity to proliferation of pathogens and aeration, which may favor or hinder seed germination. According to the Rules for Seed Analysis (Brazil, 2009), seed size, water availability requirement, sensitivity to light and the benefits of choosing the right substrate, which can provide ease for conducting evaluations, should be taken into account. Upon evaluating the seed germination of two bromeliads on different substrates, Estevan et al. (2010) found that the substrates sphagnum, washed river sand and carbonized rice bark presented the best results for the germination of *Dyckia pectinata* Smith & Reitz and *Billbergia zebrina* (Herbert) Lindley.

Considering that *E. spectabile* may be commercially exploited to meet the demands of the pharmaceutical, cosmetics and food industries, studies on its cultivation are needed, especially studies on factors that may affect the seed germination process and the initial establishment of seedlings. In this perspective, this study aims to evaluate the effects of different substrates and seed positions at sowing on the germination and vigor of *E. spectabile* seeds from two geographical regions.

## 2. Material and Methods

The experiment was carried out at the Federal Rural University of Pernambuco/Serra Talhada Academic Unit (UFRPE/UAST), Pernambuco/Brazil, during September and October

2014. Seeds of *E. spectabile* were manually extracted from mature fruits collected from ten matrices between September and October 2013 in the municipalities of Graça-CE (4º02'48" S and 40º44'59" W, altitude 171 m) and Serra Talhada-PE (7°59'9" S and 38°17'45" W, altitude 430 m). Seeds were then benefited. Seeds poorly formed and/or attacked by insects were discarded. The remaining seeds were homogenized in size using sieves of mesh sizes ≥2.00 and ≥4.00 mm. Then, they were stored in plastic containers with a lid under average environment temperature (25°C) and relative humidity (59.9%) for ten months.

The seed water content of both geographic regions was measured by the oven method at 105 ± 3ºC for 24 hours (Brazil, 2009) with four replicates of 0.4 g of seeds. The results were expressed as percentage on wet basis.

The experiment was completely randomized in a 2 x 4 x 2 factorial design (seeds from two geographic regions, four types of substrates and two sowing positions) with four replicates of 50 seeds per treatment. Seeds were sown in transparent "gerbox" plastic boxes (11 x 11 x 3.0 cm) previously washed and sterilized using 70% alcohol. The sowing was performed over and in between the following substrates: blotting paper, sand, vermiculite and coconut fiber.

The substrates sand, vermiculite and coconut fiber were previously sterilized in an oven at 200ºC for two hours and moistened with 396, 675 and 542.25 mL of distilled water, respectively, according to the retention capacity of each substrate. The substrate paper was sterilized in an oven at 105ºC for two hours and moistened at 2.5 times the dry weight of the paper, corresponding to 203 mL of distilled water (Brazil, 2009).

Then, the properly capped boxes were packed inside plastic bags and kept in an air-conditioned room under constant lighting. The mean temperature (25°C) and air relative humidity (46.81%) inside the room were monitored daily using a digital thermo-hygrometer.

In order to evaluate the effects of treatments on germination and seed vigor, a germination percentage (G%) analysis was performed. At the end of the experiment, the number of germinated seeds was counted. Germinated seeds were those that developed the primary root and had a fully expanded cotyledon sheath, a criterion adopted to characterize normal seedlings. The germination speed index (GSI) was calculated through a daily count of seedlings emerged during the germination test according to Maguire (1962). The mean germination time (MGT) was determined together with the germination test. The result was expressed in days according to Labouriau and Valadares (1976). The germination synchronization index or uncertainty ($\bar{E}$) was calculated according to Labouriau (1983).

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Normal seedlings of each repetition were measured using a digital caliper after the germination test. The shoot length (from the base to the apex of the first leaf) and the root system length (from the base to the end of the primary root) were measured. The results were expressed in mm seedlings$^{-1}$.

The seedlings were packed in pre-identified Kraft® paper bags and placed in a forced air circulation oven at 80°C for 24 hours to determine seedling dry matter (SDM). Subsequently, the seedlings were placed in a desiccator, and then the samples were weighed in an analytical scale with a precision of 0.0001 g according to recommendations of Nakagawa (1999). The results were expressed as mg seedling$^{-1}$.

Data were submitted to analysis of variance by F test. The means were compared by Tukey test at 5% probability using the software ASSISTAT, version 7.7 (Silva & Azevedo, 2016).

## 3. Results and Discussion

Seed water contents of *E. spectabile* were around 9.0% for seeds from Serra Talhada-PE and 9.9% for seeds from Graça-CE. They are within the acceptable limits for obtaining consistent results, which is indispensable for the standardization of evaluations (Alves et al., 2012). Germination started on the fifth day after sowing. It was monitored until the 30th day, when all substrates stabilized. Table 1 shows the mean *E. spectabile* seed germination values. We found that the seeds from Graça-CE distributed over paper and coconut fiber and seeds from Serra Talhada-PE sown in between paper presented the highest germination percentages.

**Table 1.** Germination (%) and seed germination speed index (GSI) of *Encholirium spectabile* Mart. former Schult. & Schult. f. seeds in function of geographical regions, substrates and sowing positions.

| Geographical region x Substrate | Sowing Position | | | |
|---|---|---|---|---|
| | Germination (%) | | GSI | |
| | Over | In between | Over | In between |
| Graça-CE x Paper | 96.0 Aa | 96.0 Aab | 8.47 Aa | 7.63 Bab |
| Grace-CE x Sand | 84.0 Ab | 90.0 Aabc | 6.73 Abc | 6.70 Abc |
| Grace-CE x Vermiculite | 88.0 Aab | 84.0 Ac | 6.09 Ac | 5.86 Acd |
| Grace-CE x Coconut fiber | 95.0 Aa | 71.0 Bd | 7.40 Aab | 3.76 Bf |
| Serra Talhada-PE x Paper | 94.0 Aab | 98.0 Aa | 8.27 Aa | 8.77 Aa |
| Serra Talhada-PE x Sand | 94.0 Aab | 88.0 Abc | 8.19 Aa | 6.84 Bbc |
| Serra Talhada-PE x Vermiculite | 94.0 Aab | 67.0 Bd | 8.21 Aa | 5.08 Bde |
| Serra Talhada-PE x Coconut fiber | 90.0 Aab | 68.0 Bd | 7.59 Aab | 3.83 Bef |
| CV (%) | 5.19 | | 8.44 | |

Means followed by the same uppercase letters in lines and lowercase letters in columns do not differ by Tukey test at 5% probability. Source: Authors.

This result differed significantly from seeds from Graça-CE sown in between coconut fiber and seeds from Serra Talhada-PE sown in between vermiculite and coconut fiber, which presented the lowest germination percentages. The decrease in germination when we sowed seeds in between vermiculite and coconut fiber may be related to the seeds' density and particle size. Associated with seed size, it provided an obstacle to the germination process.

In *Luehea divaricata* Mart. seeds, Schulz et al. (2013) concluded that sowing over paper provided the highest percentage of germination when the authors evaluated them at different temperature combinations. Guedes et al. (2009) also obtained the same result. Upon studying the germination of *Cereus jamacaru* DC seeds, the authors verified that paper roll was the most suitable medium for the evaluation of seed viability and vigor.

The germination speed index results (Table 1) evidenced that there was a delay in the number of seedlings emerged per day regardless of the geographical region of *E. spectabile* seeds when sown in between coconut fiber and vermiculite. On the other hand, the best indexes were obtained by seeds from Graça-CE sown over blotting paper and by seeds from Serra Talhada-PE sown over blotting paper, sand and vermiculite, with an average of approximately eight seedlings per day. Azerêdo et al. (2010) obtained similar results. The authors found that the best germination speed indexes were obtained when seeds of *Brassica oleraceae* var. *capitata* L. were sown over blotting paper.

These results corroborate the results found by Estevan et al. (2010). By evaluating the germination of two bromeliads on different substrates, the authors found that coconut fiber provided the lowest germination speed indexes, with a mean of 3.52 seedlings per day for *Dyckia pectinata* Smith & Reitz seeds and 3.97 seedlings per day for *Billbergia zebrina* (Herbert) Lindley seeds. These results were statistically lower than the other substrates (washed river sand, carbonized rice husk, sphagnum and sawdust).

Sowing over blotting paper, sand, vermiculite and coconut fiber was generally the most favorable germination procedure. Consequently, the germination speed index of *E. spectabile* seeds from both geographic regions was the highest since there was no need to break the physical barrier imposed by the substrate layer on seeds, which are small (varying between 1 and 4.6 mm in length) and germinate on rocky outcrops in their natural habitat. This corroborates the findings of Guimarães et al. (2014). By evaluating the effects of sowing depth (0.0 cm over and 1.0 cm in between different substrates: soil, sand, sand + soil, soil + manure, and soil + sand + manure), the authors found that sowing over them favored the growth and development of *Solanun sessiliflorum* Dunal seedlings.

For the variables mean germination time, germination synchronization index and shoot length, there was no significant interaction between geographic regions, substrates and sowing positions. However, there was an interaction between substrates and sowing positions.

Regarding geographic regions, seeds from Serra Talhada-PE presented the shortest mean germination time, with approximately 1.68 day of interval for the appearance of other normal seedlings from the day on which the first seedling appeared. It differed significantly from the mean germination time of seeds from Graça-CE, which required an interval of 1.85 days to germinate (Table 2).

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**Table 2.** Mean germination time (MGT) and germination synchronization index (GSI) of *Encholirium spectabile* Mart. former Schult. & Schult. f. seeds in function of geographical regions, substrates and sowing positions.

| Geographical region | MGT (days) | | GSI | |
|---|---|---|---|---|
| Graça-CE | 1.85 a | | 2.28 a | |
| Serra Talhada-PE | 1.68 b | | 2.05 b | |
| | Sowing position | | | |
| Substrate | MGT (days) | | GSI | |
| | Over | In between | Over | In between |
| Blotting paper | 1.50 Aa | 1.56 Ab | 1.58 Ab | 1.82 Ac |
| Sand | 1.56 Aa | 1.76 Ab | 1.82 Bab | 2.45 Ab |
| Vermiculite | 1.67 Aa | 1.88 Ab | 1.84 Bab | 2.54 Ab |
| Coconut fiber | 1.62 Ba | 2.55 Aa | 2.13 Ba | 3.15 Aa |
| CV (%) | 15.79 | | 15.99 | |

Means followed by the same uppercase letters in lines and lowercase letters in columns do not differ by Tukey test at 5% probability. Source: Authors.

By studying the influence of geographic regions and different substrates on the germination of *Pseudobombax longiflorum* seeds (Mart. et Zucc. A. Robyns), Ladeia et al. (2012) found that the mean germination time ranged from 10.6 to 16.5 days for seeds from Rondonópolis-MT, Brazil, and from 14.7 to 18.1 for seeds from Cuiabá-MT, Brazil. The authors noted that seeds from Rondonópolis germinated faster in sand, with a mean time of 10.6 days; seeds from Cuiabá germinated faster in black soil with a mean time of 14.4 days.

Based on the interaction between substrates and sowing positions (Table 2), the sowing in between coconut fiber increased the mean germination time of seeds. They took about 2.55 days to germinate compared to the germination time of seeds sown over coconut fiber and in between blotting paper, sand and vermiculite. The mean germination time taken by seeds sown over blotting paper, sand and vermiculite did not differ statistically among themselves.

Seeds from Serra Talhada-PE presented a higher germination synchrony than seeds from Graça-CE (Table 2). However, upon evaluating the synchronization indexes obtained from interactions between substrates and sowing positions, the best indexes were obtained when seeds were sown over substrates.

The interval of days between the germination of one seed and another seed and the synchrony with which it occurs is essential for obtaining a stand with uniform seedlings. Thus, choosing the proper substrate and the position in which seeds are sown is important since a fast and uniform germination is essential in a short time interval for the production of seedlings.

Based on the results of Table 3, there was no significant difference between seedling shoot length and seed geographical regions since the average values were 9.96 and 9.94 mm seedling$^{-1}$ for seedlings from Graça-CE and Serra Talhada-PE, respectively.

**Table 3.** Shoot length (SL) of seedlings from *Encholirium spectabile* Mart. former Schult. & Schult. f. seeds in function of geographical regions, substrates and sowing positions.

| Geographical region | SL (mm seedling$^{-1}$) | |
|---|---|---|
| Graça-CE | 9.96 a | |
| Serra Talhada-PE | 9.94 a | |
| Substrate | Sowing position | |
| | Over | In between |
| Paper | 6.00 Ac | 6.64 Ac |
| Sand | 10.93 Ab | 12.00 Aa |
| Vermiculite | 9.86 Ab | 9.37 Ab |
| Coconut fiber | 13.00 Aa | 11.80 Aa |
| CV (%) | 12.12 | |

Means followed by the same uppercase letters in lines and lowercase letters in columns do not differ by Tukey test at 5% probability. Source: Authors.

The combinations between substrate and sowing positions more favorable to shoot growth were verified upon sowing *E. spectabile* seeds over and in between coconut fiber and sand. The sowing over and in between blotting paper did not differ in terms of shoot growth. However, there was a delay in seedling shoot development in relation to the other substrates evaluated.

By evaluating the growth of *Nidularium minutum* Mez. (Bromeliaceae) seedlings on different substrates, Silva et al. (2013) verified that shoot length was higher in seedlings grown over the substrates coconut fiber and vermiculite at 191 days of cultivation. Souza et al. (2007) observed that *Adenanthera pavonina* L. seedlings with longer lengths were from sowing in between and over coconut fiber and over vermiculite.

As for root system length, there was no interaction among the three evaluated factors. However, there were double interactions between geographic regions and substrates and substrates and sowing positions (Table 4).

**Table 4.** Root system length (RSL) of seedlings from *Encholirium spectabile* Mart. former Schult. & Schult. f. seeds in function of geographical regions, substrates and sowing positions.

| Geographical region | Substrate RSL (mm seedling$^{-1}$) | | | |
|---|---|---|---|---|
| | Paper | Sand | Vermiculite | Coconut fiber |
| Graça-CE | 7.87 Ab | 6.64 Aba | 7.15 Aba | 5.84 Ba |
| Serra Talhada-PE | 9.99 Aa | 6.01 Ba | 6.18 Ba | 5.81 Ba |

| Substrate | Sowing position | |
|---|---|---|
| | Over | In between |
| Paper | 10.82 Aa | 7.04 Ba |
| Sand | 6.10 Ab | 6.54 Aa |
| Vermiculite | 6.59 Ab | 6.74 Aa |
| Coconut fiber | 5.76 Ab | 5.89 Aa |
| CV (%) | 17.43 | |

Means followed by the same uppercase letters in lines and lowercase letters in columns do not differ by Tukey test at 5% probability. Source: Authors.

The initial development of seedlings as evaluated by root system length (Table 4) was satisfactory for seeds from Serra Talhada-PE sown over blotting paper: approximately 9.99 mm seedling$^{-1}$. It was statistically higher than seedling root length of seeds from Graça-CE. The substrate based on coconut fiber affected the growth of the seedling root system independently of the region of origin of seeds.

Regarding the interaction between substrates and sowing positions, there was no significant difference between the substrates sand, vermiculite and coconut fiber when seeds were sown over and in between substrates. However, seedlings from seeds sown over paper reached the highest value for root system length: approximately 10.82 mm seedling$^{-1}$. It differed significantly from the mean root length of seedlings from seeds sown in between paper and sand, vermiculite and coconut fiber. In *Myracrodruon urundeuva* Fr. All. seedlings, Pacheco et al. (2006) observed that the sowing positions in between and over sand and in between vermiculite and coconut fiber provided the best development of the root system.

Regarding the accumulation of seedling dry matter (Table 5), the highest value was observed for seedlings from Serra Talhada-PE seeds sown over both paper and sand: 0.1643 mg seedlings$^{-1}$. It differed statistically from the dry matter obtained by seedlings from seeds sown in between paper and sand (0.0239 and 0.11172 mg seedlings$^{-1}$, respectively) and from the dry matter obtained by seedlings from seeds from Graça-CE, whose sowing in between sand resulted in 0.1448 mg seedlings$^{-1}$.

**Table 5.** Seedling dry matter (SDM) from *Encholirium spectabile* Mart. former Schult. & Schult. f. seeds in function of geographical regions, substrates and sowing positions.

| Geographical region x Substrate | Sowing Position SDM (mg seedling$^{-1}$) | |
|---|---|---|
| | Over | In between |
| Graça-CE x Paper | 0.0225 Ac | 0.0197 Ab |
| Grace-CE x Sand | 0.1060 Bb | 0.1448 Aa |
| Grace-CE x Vermiculite | 0.0558 Ac | 0.0530 Ab |
| Grace-CE x Coconut fiber | 0.0384 Ac | 0.0275 Ab |
| Serra Talhada-PE x Paper | 0.1643 Aa | 0.0239 Bb |
| Serra Talhada-PE x Sand | 0.1643 Aa | 0.1172 Ba |
| Serra Talhada-PE x Vermiculite | 0.0508 Ac | 0.0335 Ab |
| Serra Talhada-PE x Coconut fiber | 0.0445 Ac | 0.0245 Ab |
| CV (%) | 32.37 | |

Means followed by the same uppercase letters in lines and lowercase letters in columns do not differ by Tukey test at 5% probability. Source: Authors.

By evaluating the effects of different temperatures and substrates on the germination of *Poincianella pyramidalis* (Tul.) LP Queiroz seeds, Lima et al. (2011) found that the substrates sand and paper were the most efficient in allocating dry matter in seedlings. On the other hand, *Crescentia cujete* L. seeds resulted in vigorous seedlings regarding the accumulation of dry matter when sown over the substrates sand and vermiculite (Azevedo et al., 2010).

## 4. Conclusion

The substrates blotting paper, sand, coconut fiber and vermiculite are favorable to *E. spectabile* seed germination regardless of geographic regions, provided that the sowing is made over the substrates.

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