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Temperature and light quality influence seed germination of two biquinho pepper cultivars

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Abstract

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Biquinho pepper crop is becoming an alternative source of income in many properties that work in horticultural sector. Generally speaking, peppers are grown all over the world, however, the cultivation of biquinho pepper comes in increasing ascent due to its low pungency. In this way, researches related to agronomic aspects are scarce and demands for research need to be fulfilled. The aim of this study was to determine the best temperature and light quality for seed germination of two biquinho pepper cultivars [BRS Moema (Moema) and Airatema Biquinho Amarela (Airatema)]. The experiment was divided into two, where different temperatures were tested (15, 20, 25, 30 and 35°C) by sowing on Germitest® paper (Experiment I); and different light quality (no light, white, red, far red, green and blue) germination in Gerbox® (Experiment II), on seed germination of two biquinho pepper cultivars. The temperature was 25°C for the best germination of biquinho pepper. Ambient conditions with light are favorable for germination of biquinho pepper seeds however, the luminous environments in green, red and far red colors influence positively germination of two biquinho pepper cultivars.

Keywords: Capsicum chinense; emergence; physiology; environmental conditions

Introduction

The *Capsicum* genus is native to tropical regions of American continent characterized by wide diversity, with about 30 species of peppers, among these are: *Capsicum chinense*, *Capsicum annum*, *Capsicum frutescens*, *Capsicum pubescens* and *Capsicum baccatum* (Costa et al., 2015). The peppers, rich in capsaicin, have antioxidant and antimicrobial properties (Moraes et al., 2013), being widely used in regional cuisine in several countries around the world, as well as in the food, pharmaceutical and cosmetic industries (Neitzke et al., 2015).

Pepper cultivation of *Capsicum* genus is of great importance, since besides economic and social impact in agribusiness, it is also a great source of employment and income (Domenico et al., 2012), making up the segment of large export industries.

Discovery and exploitation of new types of pepper has presented a dynamism to consumer market, as it generates development of several products with high added value (Neitzke et al., 2015). Among different types of pepper, it stands out small pepper, initially used as an ornamental plant, is currently an alternative to producers because of high aromatic content, crispness and absence of pungency, small and colorful fruits, which makes it highly appreciated *in natura* (Garruti et al., 2013; Heinrich et al., 2015)

Pepper culture is implanted by direct sowing seeds in trays, and after about 90 days of sowing, seedlings are transplanted into the soil. However, germination is considered the most critical phase of plant's life cycle, since it is a main component of seedling establishment and survival, as well as presenting high vulnerability to environmental and/or biological stresses (Rajjou et al., 2012). Seed germination begins with water uptake, imbibition and elongation of the radicle through structures surrounding the embryo, ending at beginning of seedling growth, which is a crucial process for establishing the crop (Hernández-Verdugo et al., 2001).

Among determinant factors on germination, it was highlight air temperature and light (Ortega-Baes & Rojas-Aréchiga, 2007). The air temperature acts on velocity of water absorption and on biochemical reactions in seed (Alves et al., 2011; Gomes et al., 2016), highlighting the minimum, optimal and maximum points within a temperature range, in which the seeds will express germinative potential (Cetnarski Filho & Carvalho, 2009). The light acts on development of vegetable, regulating metabolism, with beneficial action in stimulating the hormones and enzymes synthesis (Vieira et al., 2010), besides other aspects that occur through luminous signals capture that are regulated by photoreceptors called phytochromes (Neff, 2012). Depending of species, seeds may show variable sensitivity by light, so they may be negative, neutral or positive photoblastics (Sousa et al., 2008).

Different temperatures, combined with different luminous spectra, are important environmental factors as triggers of optimal germination. These factors are variable with species and knowledge about specific requirements of each plant allows guiding research on seed germination and direct sowing in the field (Carvalho & Nakagawa, 2000). For species of pepper *Piper hispidinervum* and *Piper aduncum*, temperature of 25°C with white light incidence produced better results, with higher percentage and germination speed values (Bergo et al., 2010). However, little is known about factors that influence germination of biquinho pepper (*Capsicum chinense*). Thus, the aim of this study was to evaluate temperature and light quality influence on seed germination of two biquinho pepper cultivars.

Material and Methods

The experiments were carried out in the Laboratory of Tissue Cultures and Aromatic Extractives of Federal University of Santa Maria, Campus of Frederico Westphalen, from September to November, 2016. The experiment was divided in two, and for Experiment I, were tested different temperatures for seeds germination of two biquinho pepper cultivars by sowing on Germitest® paper, and for Experiment II, the different light quality were tested for seeds germination at temperature determined in the Experiment I.

Experiment I

The experiment was conducted in a completely randomized design, in a 2 x 5 factorial scheme, consisting of two biquinho pepper cultivars ((*Capsicum chinense)* BRS Moema (Moema) and Airatema Biquinho Amarela (Airatema)), and five air temperatures (15, 20, 25, 30 and 35°C), totaling ten treatments, four replicates per treatment, the experimental unit being composed of 50 seeds per replicate. The seeds were seeded on two sheets of Germitest® paper with dimensions of 28 × 38 cm rolled and moistened with distilled water in the amount of 2.5 times the paper mass, following methodology by Regras para Análise de Sementes (2009), and individualized in plastic bags for humidity maintenance, and kept for 14 days in a BOD chamber with photoperiod of 12 light hours, in each of evaluated temperatures.

Experiment II

The experiment was conducted in completely randomized design, in a 2 x 6 factorial scheme, being two cultivars ((*Capsicum chinense)* BRS Moema (Moema) and Airatema Biquinho Amarela (Airatema)) and six light quality (no light, white, red, far red, green and blue), totaling 12 treatments, with four replicates each and the experimental unit composed of 50 seeds per replicate. The seeds were stored in transparent plastic boxes with dimensions of 11 × 11 × 3.5 cm, containing two sheets of Germitest® paper in dimensions of boxes, and moistened with distilled water in the amount of 2.5 times the paper mass, following methodology by Regras para Análise de Sementes (2009), after planting Gerbox® type boxes were kept for 14 days in a BOD chamber with light photoperiod of 12 hours and kept at a constant temperature of 25°C.

To provide different light environments, the Gerbox® was wrapped using cellophane-type paper. To match temperature and light conditions of each treatment, transparent cellophane paper was used for white quality; for red, green and blue light, cellophane papers of same colors were used, for far red a blue cellophane paper was used between two red colored sheets, and for dark environment, Gerbox® were covered with aluminum paper.

Evaluated traits in Experiment I and II

To evaluate cultivars and temperatures effects on seed development, germination test was realized according to Regras para Análise de Sementes (2009). Traits analyzed for both experiments were: germination percentage at 7 days (7DAS), germination percentage at 14 days (14DAS), abnormal seedlings, and dead seeds. For this, first evaluation was carried out seven days after sowing (7DAS), in which the beginning of the germination in seeds with initial radicle emission was considered. At 14 days (14DAS) were evaluated germination percentage, abnormal seedlings and dead seeds.

For germination percentage, germinated seeds were considered as normal seedlings, those with long primary root and presence of root hairs, aerial part with straight, thin and elongated hypocotyl, uniform cotyledons and green coloring. Abnormal seedlings were considered to be those with structures that were damaged, deformed, deteriorated or had no normal plant characteristics. Those seeds that did not germinate at the end of experiment were considered dead seeds.

Variance Analysis

In this way, variance analysis was performed to verify cultivars and temperatures effects (Experiment I) and cultivars and light qualities effects (Experiment II), respectively for germination percentage at 7 days (7DAS), germination percentage at 14 days (14DAS), abnormal seedlings and dead seeds. Regression analysis was performed for quantitative factor (temperature), and for qualitative factor (light quality), means were compared by Tukey test, at 5% error probability, using Genes statistical program (Cruz, 2013).

Results and Discussion

Experiment I – Temperature influence on biquinho pepper cultivars

There was significant interaction ($p < 0.05$) for the factors temperature X cultivar for all traits evaluated: germination percentage at 7 days, germination percentage at 14 days, abnormal seedlings, and dead seeds. At the first count, at 7DAS (Fig. 1A), seeds had a higher germination percentage when kept at a temperature of 25°C, observing that at 15°C there was no response, independently of cultivar (Fig. 2). This result reflected germination of normal seedlings, with the highest germination percentage at 14 days at 25°C for both red and yellow biquinho pepper cultivars, with percentages of 75 and 59.5% respectively (Fig. 2B). However, there was a reduction in germination at a temperature of 30°C, and the Moema cultivar showed germination higher than Airatema cultivar ($p < 0.05$). At the temperature of 35°C, both cultivars had a low germination percentage.

Analyzing abnormal seedlings percentages (Fig. 1C), it was observed the lowest values of abnormal plants were recorded at temperatures of 15 and 25°C. At temperatures of 20 and 30°C, Moema cultivar presented larger amounts of abnormal plants compared to Airatema cultivar (Fig. 2C).

Among evaluated temperatures it is observed that 25°C excelled, showing the smaller abnormal seedlings percentages for both cultivars. It is also observed that at temperatures of 20°C and 30°C, Airatema cultivar had lower abnormal seedlings percentages than Moema cultivar ($p < 0.05$), and value obtained at 30°C was due to the fact of the lower germination at this temperature, while at 20°C the Airatema cultivar presented a better response at this temperature compared to Moema cultivar (Fig. 2C).

The lowest dead seeds percentage was observed in Moema cultivar at temperatures of 25°C and 30°C, showing values of 5.5 and 14.5% respectively (Fig. 1D). Comparing cultivars, it was observed that Moema cultivar showed the lowest seed mortality. Both for Moema and Airatema cultivar it is observed at temperature of 25°C there was a lower number of dead seeds. At temperature of 15°C the worst results were obtained, observing values of 90 and 100% of dead seeds for Moema and Airatema cultivars, respectively (Fig. 2D).

Experiment II – Light quality influence on germination of biquinho pepper

There was significant interaction for cultivar x luminosity, for all analyzed traits (germination percentage at 7 days, germination percentage at 14 days, abnormal seedlings, and dead seeds).

For germination percentage at 7 days after sowing (DAS), it was observed that Airatema cultivar was significantly lower than Moema in white, blue, green light and no light, demonstrating a lower germinative potential (Fig. 3). However, for both red and far red light, Airatema cultivar was significantly higher when compared to Moema (Fig. 3A).

For germination percentage at 14 days, it was possible to observe that blue light provided the lowest results for Moema cultivar. Other light quality did not differ among them-

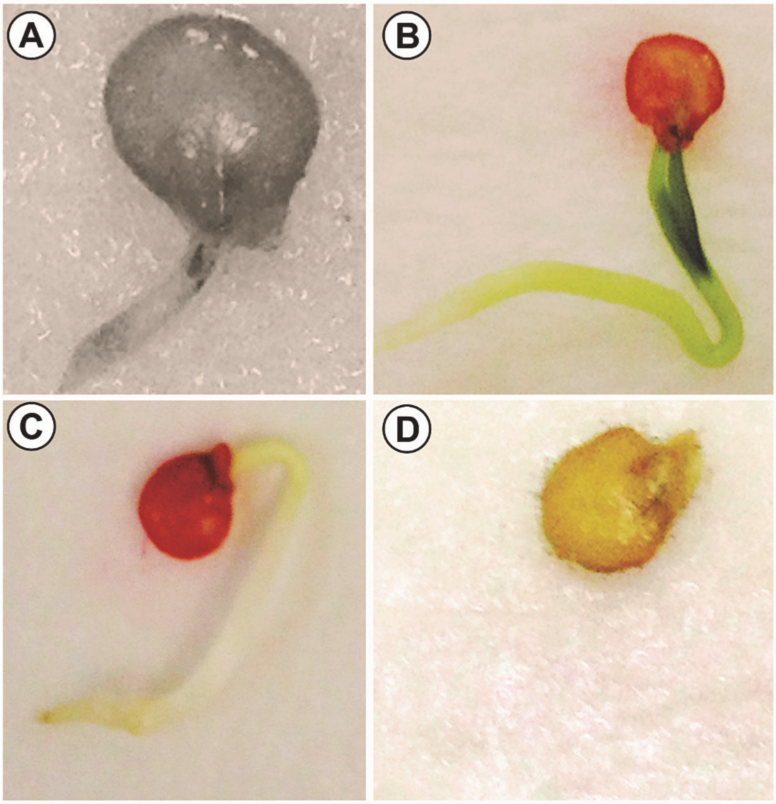

Fig. 1. Traits (A) germination percentage at 7 days (7DAS), (B) germination percentage at 14 days (14DAS), (C) abnormal seedlings and (D) dead seeds of biquinho pepper cultivars [(*Capsicum chinense*) BRS Moema (Moema) and Airatema Biquinho Amarela (Airatema)] sown at different air temperatures

selves ($p < 0.05$), presenting germination potential for this cultivar. When comparing cultivars within each light quality, there was no significant difference between cultivars only for the blue light (Fig. 3B).

For abnormal seedlings percentage, it was observed that Moema cultivar, environment with no light presented highest value when compared to other light quality, since it showed 61% of abnormal seedlings ($p < 0.05$). For Airatema cultivar, a smaller abnormal seedlings percentage in blue and red light was observed, with 12 and 23% ($p < 0.05$). When comparing cultivars within each level of light quality, it was possible to observe Moema was significantly superior to Airatema only for no light, and did not differ for red and blue light quality ($p < 0.05$) (Fig. 3C).

The lowest dead seeds percentage was observed in Moema cultivar with values of 4 to 8%, did not differing among different light quality. For Airatema cultivar, it was observed higher dead seeds percentages in no light, white and blue, and did not differ significantly among them (21, 16, 12 and 19%, respectively). When evaluating cultivars within each level of light quality, Airetama cultivar was significantly superior to Moema in all lights (Fig. 3D).

It is desired that seed germination be uniform, in order to ensure rapid establishment field after sowing (Rajjou et al., 2012). However, several factors can prevent or hinder this process, we can cite temperature; and this, when appropriate, promote oxygenation and greater expression of physiological potential of seeds (Martins et al., 2008). Temperatures considered not optimal for specie cause late emergence seeds, or seedlings emergence without uniformity.

Carter and Vavrina (2001) evaluated different temperatures (20, 25, 30, 35 and 40ºC) in a study with several *Capsicum annum* cultivars, demonstrating that at 35 and 40ºC there was reduction in seed germination, values until 1%, due to term inhibition. (Nascimento, 2005), also indicated a inhibitory effect on germination potential and vigor in *Solanum lycopersicum*, *Solanum melongena*, *Citrullus lanatus* and *Cucumis melo*, when conditioned at 10, 15 and 17ºC.

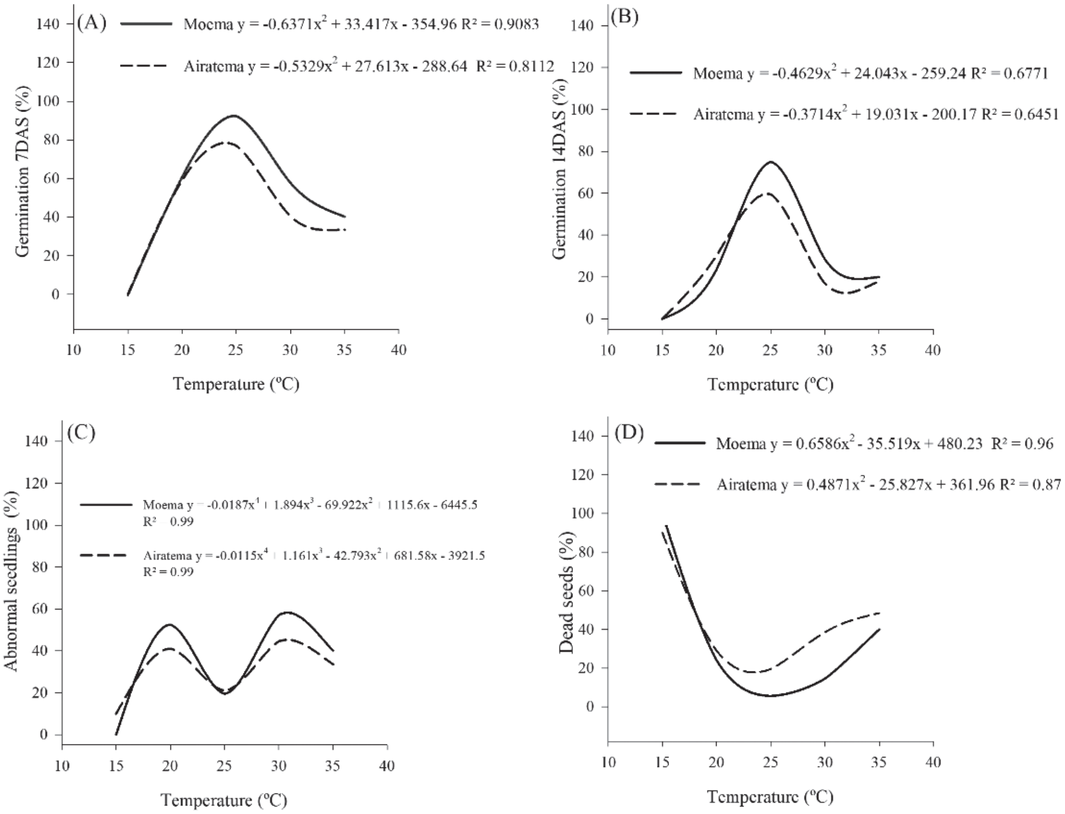

Fig. 2. Regression analysis for (A) germination percentage at 7 days (7DAS), (B) germination percentage at 14 days (14DAS), (C) abnormal seedlings and (D) dead seeds analyzed during seeds germination of biquinho pepper [(*Capsicum chinense*) BRS Moema (Moema) and Airatema biquinho (Airatema)] on different temperatures

Guedes et al. (2011) report temperature importance in water absorption and biochemical reactions regulating germination. High temperatures are very aggressive to germination and seedling development, because this growth stage is most sensitive to high temperatures (Yin et al., 2003). High temperature effect on germination is evil because of increased fluidity of membrane lipids, which modifies composition and structure causing rupture of biological membranes, which leads to loss function, thus accelerating seeds respiration (Alves et al., 2015). Results found in this work for Moema and Airatema cultivars, corroborate that temperature regulates seeds germination of biquinho pepper; because it influences water absorption rate and metabolic reactions of seed (Alves et al., 2015).

Nascimento and Aragão, (2002) showed, in *Cucumis melo* seeds, the increase in temperature results in greater water absorption, and thus potentiates germination process. Extreme temperatures cause imbalance in cell enzymatic and metabolic activity, which is reflected in higher endosperm resistance of seed (Watkins & Cantliffe, 1983), and thus, slow root protrusion, high germination of abnormal seedlings, unviability and death seeds, as these factors lead to damages in vital organs of seedlings (Maekawa et al., 2010). As well as very high temperatures, low temperatures may also cause destabilization in cell membranes (Guan et al., 2009), due to reduced fluidity of membrane lipids, requiring higher activation energy to perform biochemical processes during germination (Oliveira et al., 2012). Thus, for germination of biquinho pepper, there is optimum temperature, and below or above this, germination process is negatively influenced.

The best results observed in present work, at temperature of 25ºC, probably occurred due to better enzymatic activity and activation of seed metabolic mechanisms; these factors are important in embryo development, providing better vigor and greater germination uniform (Almeida et al., 2012). Another factor associated with seed germination is light quality, and many studies have been carried out to study effects on plant morphophysiological development (Hideg et al.,

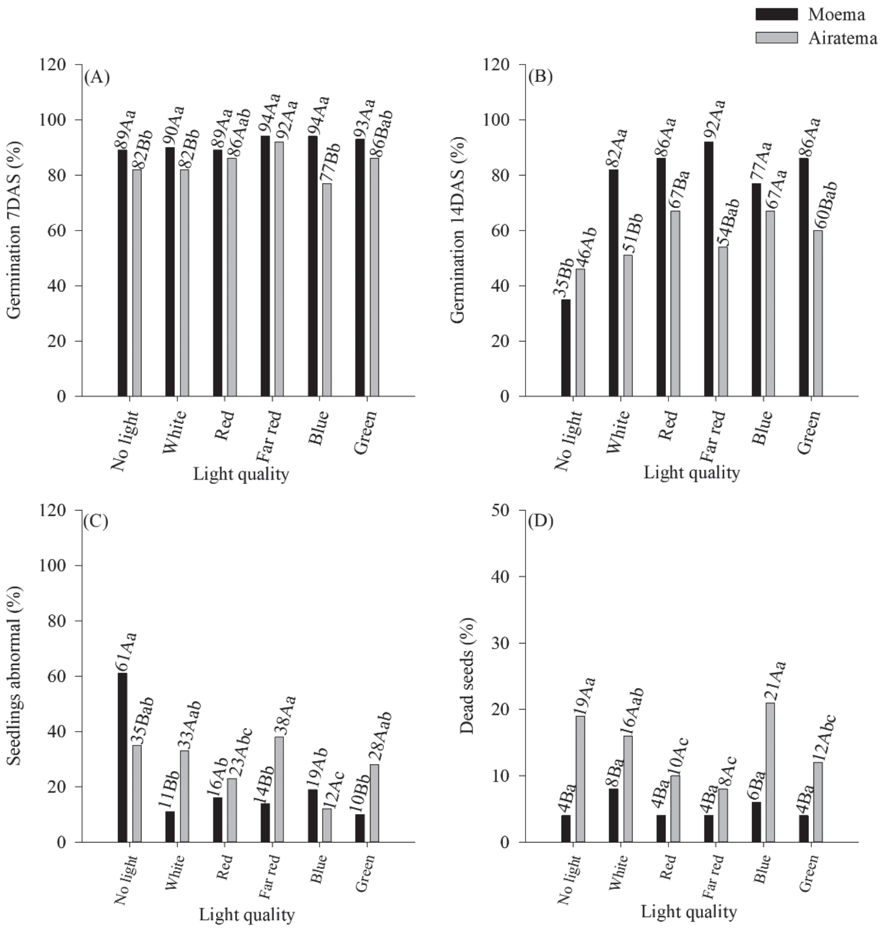

Fig. 3: Germination percentage at 7 days (A), germination percentage at 14 days (B), abnormal seedlings (C) and dead seeds (D) evaluated during seeds germination of biquinho pepper [(Capsicum chinense) BRS Moema (Moema) and Airatema biquinho amarela (Airatema)] on different light quality; Upper case letters differ from cultivars and lower case letters differ from light quality by Tukey's test at 5% probability

2013); this because depending plant species, quality and light spectra effects may be easily viewed (Rajjou et al., 2012), as regulate various routes related to plant development as well as influence the acclimatization (Hoffmann et al., 2015).

In study realized by (Galindo et al., 2012) with seeds of *Crataeva tapia*, authors observed that vigor and germination were not inhibited by different light conditions and not even in no light, suggesting, in this case, that seeds showed a non-photoblastic response, that is, no influenced by presence or no light. For *Solanum sessiliforum*, (Stefanello et al., 2008) observed interference of different light quality on speed and germination percentage, observing higher percentages in red and far red light conditions. In seeds of *Murdannia nudiflora*, (Ferraresi et al., 2009) observed a decrease in germination percentage under green, blue and no light conditions, presenting higher values when seeds were submitted to white and red light, considering a preferential photoblastic response, since they reported germination both in presence and no light.

In the present work, results demonstrated that Moema and Airatema cultivar have high light dependence, showing a positive photoblastic response. These results point to a possible interference of light quality (Yamashita et al., 2011), controlled by phytochrome (Shinomura, 1997), which is photoreceptor pigment responsible for light signals absorption (Galindo et al., 2012). Phytochrome inhibits or stimulates germination according to light quality (Dissanayake et al., 2010), associating with flow control and cell membrane permeability (Yamashita et al., 2011), thus regulating growth capacity of seed the embryo (Casal & Sánchez, 1998).

The decrease in germination and high abnormal seedlings percentage for Moema cultivar after the longest exposure time under no light conditions, possibly occurred due to absence of red and far red light ratio; as reported also by (Godoi & Takaki, 2005). This is because red and far red proportions are detected by phytochromes (Stamps, 2009); and, second (Leite & Takaki, 2001), phytochrome absorption in Pr form (red) causes photochemical conversion and physiologically active Pfr (far red), which stimulates seeds germination. In pepper, spectral manipulation may trigger a wide range of physiological responses, while efficiency of light-dependent processes, and a number of biochemical reactions in photosynthesis, have a great productive impact (Ilić & Fallik, 2017).

Conclusions

For seeds germination of biquinho pepper Moema and Airatema cultivars is recommended temperature of 25ºC.

Light quality in green, red and far red colors influence positively on seeds germination of biquinho pepper Moema and Airatema cultivars.

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