




Temperatures and light regimes in the germination of *Areca vestiaria* and *Areca triandra* seeds¹

Antonio Maricélio Borges de Souza^{2*} , Kássia Barros Ferreira², Marcos Vieira Ferraz²,
Larissa Benetasso Chioda², Kathia Fernandes Lopes Pivetta²

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ABSTRACT

Propagation of palm trees is mainly by seeds and, in general, germination is uneven and slow, caused by several factors, such as temperature and light. We aimed to evaluate the effect of different temperature conditions and light regimes in the germination of *Areca vestiaria* and *Areca triandra* seeds. Two experiments were carried. The experimental design was completely randomized with a 5×2 factorial (temperature \times light), with four replicates per treatment for the species *A. vestiaria* and five replications for *A. triandra*, with 25 e 20 seeds, respectively. The temperatures tested were 25, 30, 35, 20-30 and 25-35 °C and the light regimes were presence and absence of light. The germination percentage and germination speed index were evaluated. Analysis of variance was performed using the F Test, with subsequent comparison of the means using the Tukey test ($\alpha = 0.05$). For both species, there were significant differences between the temperature conditions for the studied characteristics. It was concluded that temperatures of 25 °C, 30 °C, 25-35 °C and 20-30 °C are indicated for germination of seeds of the species *Areca vestiaria* and, 25-35 °C for *Areca triandra*, being the seeds classified as neutral photoblastics.

Keywords: Arecaceae; ornamental plants; photoblastia; palm trees; thermal stress.

INTRODUCTION

Palm trees have great variability in species richness, phylogenetic composition and life forms because they are adapted to a variety of climates and soils (Eiserhardt *et al.*, 2011). The Arecaceae include approximately 2700 species in 240 genera (Lorenzi *et al.*, 2010). Due to its exuberance, beauty, size and crown that stand out in nature, they have great ornamental potential, providing remarkable harmony in the landscape compositions. Due to these characteristics, all palm trees are considered ornamental, although some are widely used and others are unknown (Costa *et al.*, 2018).

Areca vestiaria Giseke, originally from eastern Indonesia, is a cespitose palm tree, occasionally solitary, monoecious, forming thin or compact clumps and quite attractive due to the unique color of the leaf sheath in the palm heart region. *Areca triandra* Roxb. ex Buch-Ham., originally from Southeast Asia, East India,

Sumatra, Borneo and the Philippines, is a cespitose palm with detached nodes and internodes similar to bamboo, sometimes with adventitious roots at the base, as well as flowers with a characteristic scent of lemon (Lorenzi *et al.*, 2004).

The propagation of palm trees is mainly by seeds and in general they have uneven and slow germination, caused by several factors (Meerow & Broschat, 2015). Temperature, humidity, light and oxygen are considered fundamental for its germination (Carvalho & Nakagawa, 2012). For Peske *et al.* (2012), the ability of seeds to germinate under a wide range of environmental conditions ensures the survival and regeneration of species.

Germination is dependent on well-defined temperature limits, characteristic for each species (Bewley *et al.*, 2013), considering that, at optimal temperature, the percentage of germination is higher and, in less time,

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² Universidade Estadual Paulista, Faculdade de Ciências Agrárias e Veterinárias, Departamento de Ciências da Produção Agrícola, Setor de Produção Vegetal, Jaboticabal, São Paulo, Brazil. maricelio_@hotmail.com; kassiaferreira@gmail.com; ferrazmarcos@yahoo.com.br; larissabchioda@hotmail.com; kathiaflpivetta@hotmail.com

*Corresponding author: maricelio_@hotmail.com

already at maximum and minimal temperatures, there may be a lower percentage of germination or death of the embryo (Carvalho & Nakagawa, 2012). Temperature interferes in the dynamics of water absorption, as well as in the limits and speed of biochemical reactions and, also, in the physiological processes that determine the germination of seeds (Marcos Filho, 2015).

Regarding the light stimulus in the germination process, the seeds that need light are called positive photoblastics, while those that germinate better in the absence of light are negative photoblastics and, when there is no light interference in the germination, the seeds are considered neutral photoblastics (Nogueira *et al.*, 2014). The light participates in the activation of the seeds and is linked to phytochrome, which is the pigment responsible for capturing the light signals from the environment (Brancaion *et al.*, 2012), which can have a stimulating or inhibiting effect on germination, depending on the light wavelength at which it was submitted (Carvalho & Nakagawa, 2012).

Temperature and light conditions can be controlled and, thus, to contribute to improving the percentage, speed and synchronization of germination. In view of the need to determine which conditions are most suitable for seed germination for the establishment of a reliable protocol, especially for species of the Arecaceae Family.

The objective of this study was to evaluate the effect of different temperature conditions and light regimes on the germination of *Areca vestiaria* and *Areca triandra* seeds.

MATERIAL AND METHODS

The fruits of *Areca vestiaria* were harvested from parent plants grown on a property located in Brumadinho city, Minas Gerais State (20°7'26.86" S and 44°13'7.63" O), on October 13th, 2019 and from the species *Areca triandra* from plants grown at the Experimental Nursery of Ornamental and Forest Plants at São Paulo State University (Unesp) (21°15'2" S and 48°16'47" O), Universidade Estadual Paulista, Jaboticabal, São Paulo State, on February 12th, 2020. Six plants matrices were randomly selected for each species. The fruits of both species were harvested ripe, evidenced by the red color of the epicarp and loosening of the bunches (Lorenzi *et al.*, 2004).

Two experiments were carried out at the Laboratory of Horticultural Seeds, located in the Department of Agricultural Sciences, Plant Production Sector, FCAV/UNESP, Jaboticabal, SP. The experiments were carried out separately for each species, at different times. In the laboratory, the fruits were pulped (removal of the epicarp and mesocarp) through manual friction with a steel mesh sieve (6 mm) (Beckmann-Cavalcante *et al.*, 2012). Fruit

depulping was done three days after harvest for *A. vestiaria* and, for *A. triandra*, seed extraction was done on the day of harvest. For both, sowing was carried out on the day after depulping. The diaspores subsequently underwent asepsis by soaking in sodium hypochlorite solution (2%) for 10 minutes, then they were rinsed with running water. This process aims to reduce fungal infestation in seeds (José *et al.*, 2012).

For both, the experimental design was completely randomized with a 5 × 2 factorial, with four replicates per treatment for the species *A. vestiaria* and five for *A. triandra*, with 25 and 20 seeds, respectively, totaling 1000 seeds for each species. The combinations of temperatures were 25, 30, 35, 20-30 and 25-35 °C and two light regimes, it being in the presence: photoperiod with a regime of 8 hours with light and 16 hours without light; and total absence of light. At alternate temperatures, those higher (30 °C and 35 °C) corresponded to the daytime temperature, while the lowest (20 °C and 25 °C) to the night. The number of repetitions and seeds in each treatment differed between species because *A. triandra* has larger seeds than *A. vestiaria*, thus needing more repetitions.

Transparent and black plastic boxes with lids (11 × 11 × 3 cm) were used for the treatments of presence and absence of light. In the presence of light, the lamps were turned on and off automatically for a photoperiod of 8 hours of light being in accordance with what was proposed by Luz *et al.* (2014). The substrate medium-sized expanded vermiculite (Batista *et al.*, 2016; Costa *et al.*, 2018; Almeida *et al.*, 2018), maintained at 100% of its water-holding capacity. The substrate was slowly saturated with water, until the interruption of its drainage was observed to determine the of water retention. The boxes were wrapped in transparent plastic bags and placed in a germination chamber of the Biochemical Oxygen Demand (B.O.D), with their respective temperature and photoperiod conditions for each treatment. The germination chambers used had 4 daylight white fluorescent lamps with 20 watts each (ELETROLAB®, model EL202/4).

The water content, germination and germination speed index were evaluated. The germination evaluation was performed daily, considering that the seeds that emitted the germinative bud were considered germinated, until germination stabilization for all treatments. The stabilization of the germination process was observed at 52nd days for *A. vestiaria* and at 105th days for *A. triandra*. For the treatment with total absence of light, the evaluations were carried out in a darkroom, using a flashlight (20 watts) covered with two sheets of green cellophane paper (Coelho *et al.*, 2012). In this case, the light was turned on only at the time of evaluation of the

experiments. In order to maintain the randomness of the experiments, at each evaluation the positions of the boxes were changed at random.

To determine the water content of the seeds, for both experiments, two subsamples with 10 seeds each were separated, using the greenhouse method at 105 ± 3 °C, for 24 hours (Brasil, 2009). This method consists of extracting water from the seeds in the form of steam by applying heat under controlled conditions, and the results are expressed as a percentage. The water contents found in the seeds were 43.09% and 41.01% respectively, for *A. vestiaria* and *A. triandra*. To evaluate the Germination Speed Index (GSI), the formula established by Maguire (1962) was used, based on the values of seed count germinated daily, where $[GSI = (G1/N1) + (G2/N2) + (Gn/Nn)]$, where GSI = Germination Speed Index; G1, G2 and Gn = number of seeds germinated in the first, second and last counts; N1, N2 and Nn = number of days elapsed after sowing in the first, second and last count. The percentage of germination was evaluated at the end of the experiments, using the formula proposed in the Rules Manual for Seed Analysis (Brasil, 2009), considering the formula $[\%G = (N/A) \times 100]$, where %G = germination percentage; N = number of germinated seeds; A = total number of seeds placed to germinate. Results are expressed as a percentage.

The data were submitted to Shapiro-Wilk normality tests and Levene homogeneity of variances ($p > 0.05$), with the germination percentage values previously transformed into arc sine $(x/100)^{1/2}$, due to the failure to meet the assumptions for the variable. Then, the analysis of variance (ANOVA) was performed by the F Test ($p \leq 0.05$) and the comparison of the averages by the Tukey test at the significance level of 5% ($\alpha = p \leq 0.05$), using the statistical software AgroEstat® version 1.1. 0.711 (Barbosa & Maldonado Júnior, 2015). Scatter plots were built with the aid of Microsoft Excel® software version 2016 for better visualization of germination over time.

RESULTS AND DISCUSSION

The interaction between temperature and light was not significant for evaluated with the two species (Table 1). For *A. vestiaria*, the temperature had significant effect for all characteristics evaluated, with higher germination percentages in the temperatures of 25; 30; 20-30 and 25-35 °C, when compared to 35 °C. For the GSI characteristic, it is observed that the temperature of 35 °C differed statistically only from the temperatures of 30 °C and 20-30 °C. It is possible to notice that the germinative process of the seeds submitted to the presence of light was similar to those kept in the continuous dark, not differing statistically its averages for percentage of germination and GSI (Table 1).

It was also observed (Table 1) that the germination percentages are low (below 40%). Several factors may have influenced the results, for instance, the time from harvest to sowing, the form of storage and transport that differed between species, resulting in a possible loss of viability for *A. vestiaria* seeds. According to the literature for palm trees, in general, sowing should be done soon after the fruit is harvested. Furthermore, the differences in genotype, origin and genetic load of the matrices during fruit filling and the water replacement rate, which may have been different in the studies, may have a direct influence on the physiological quality and viability of the seeds, as reported by Beckmann-Cavalcante *et al.* (2012). However, for the species of this research, studies on the physiological maturity of its seeds were not found in the literature.

Although it has a range of temperature for germination, it was found that the constant of 35 °C negatively affected the germination process, as it promoted a lower value for the percentage of germination and GSI for *A. vestiaria* (Table 1). Marcos Filho (2015) reports that high temperatures can reduce the percentage of germination, and the number of seeds that manage to germinate drops quickly, basically as a result of the effects on enzyme activity and restrictions on oxygen access. In general, the seeds show variable behavior and there is no optimum and uniform temperature for the germination of all species (Bewley *et al.*, 2013), as observed for both species in this study, which demonstrated a wide variation in thermal need for seed germination. Palm seeds generally require high temperatures for fast and uniform germination, considering temperatures in the range from 21 to 38 °C as acceptable and 29 to 35 °C as more favorable (Meerow & Broschat, 2015).

Regarding the species *A. triandra*, for germination percentage, there was an effect only for the temperature conditions tested (Table 1). It is also noteworthy that there was no germination at a temperature of 25 °C in both assigned light regimes (Table 1), which demonstrates the sensitivity of the seeds to these conditions, and can be considered as an adaptive characteristic capable of regulating germination in non-ideal times, even being designated as critical for this species. Low temperatures compromise essential pathways for germination, so that the reorganization of the cell membrane system can become slower, influencing water absorption and, consequently, reducing the biochemical and physiological reactions that determine the vigor and germination of seeds (Flores *et al.*, 2014; Carvalho & Nakagawa, 2012).

As in the present work, other palm species have the light factor as a non-limiting factor for the germination

of their seeds, thus the seeds are indifferent to light in their germination process, namely, *Copernicia alba* Morong ex Morong & Britton (Masetto *et al.*, 2012), *Syagrus coronata* (Mart.) Becc. (Porto *et al.*, 2018), *Euterpe precatória* Mart. (Costa *et al.*, 2018) and *Mauritia flexuosa* L. f. (Almeida *et al.*, 2018). In many species, the presence of light favors the germination of the seeds, while in others, the germinative behavior of the seeds is favored in the absence than in the presence of light (Melo *et al.*, 2018). Thus, it can be inferred that both species of this research are classified as neutral photoblastics, since there was germination regardless of the presence or absence of light, being in accordance with what was proposed by Bewley *et al.* (2013) and Marcos Filho (2015).

It is observed that for the specie *A. triandra* the alternating temperature of 25-35 °C reached higher values for the characteristics germination percentage (76.76%) and GSI (0.2678), differing statistically from the others (Table 1). Germination in alternating and constant temperature regimes evidence the species' adaptation to the natural thermal fluctuations of the environment, which gives the seedlings greater ability to establish in the field, making them capable of withstanding adverse environmental conditions (Guedes *et al.*, 2010). However, the difference between high and low temperatures must be equal to or greater than 10 °C (Baskin & Baskin, 2014), thus simulating natural thermal variations. For Marcos Filho (2015) alternating temperatures favor, mainly, the germination of species that have not undergone an intense domestication process.

There are species that present higher percentages of germination when under alternating temperatures, and this is indicative of adaptive capacity to variations in the temperature of the environment (Martins *et al.*, 2010),

an event observed for the species *A. vestiaria* and *A. triandra* at temperatures of 20-30 and 25-35 °C, respectively (Table 1). This is due to the similarity with the environmental germination where some species develop, in which the daytime temperatures are higher than the nighttime temperatures (Carvalho & Nakagawa, 2012). Additionally, knowledge about the response of seeds to temperature is essential, as it makes it possible to understand the range of tolerance of species in relation to temperature, as well as the climatic conditions in which crops can germinate and establish themselves properly (Motsa *et al.*, 2015).

Some species of palm trees also showed satisfactory results in the germination process of their seeds when under conditions of alternating temperatures, for example, *Phoenix canariensis* hort. ex Chabaud, 20-30 °C (Pimenta *et al.*, 2010); *Copernicia alba* Morong ex Morong & Britton, 20-30 °C (Masetto *et al.*, 2012); *Bactris maraja* Mart., 26-40 °C (Rodrigues *et al.*, 2014); *Livistona rotundifolia* (Lam.) Mart., 25-35 °C (Viana *et al.*, 2016) and *Mauritia flexuosa* L. f., 20-30 °C (Almeida *et al.*, 2018).

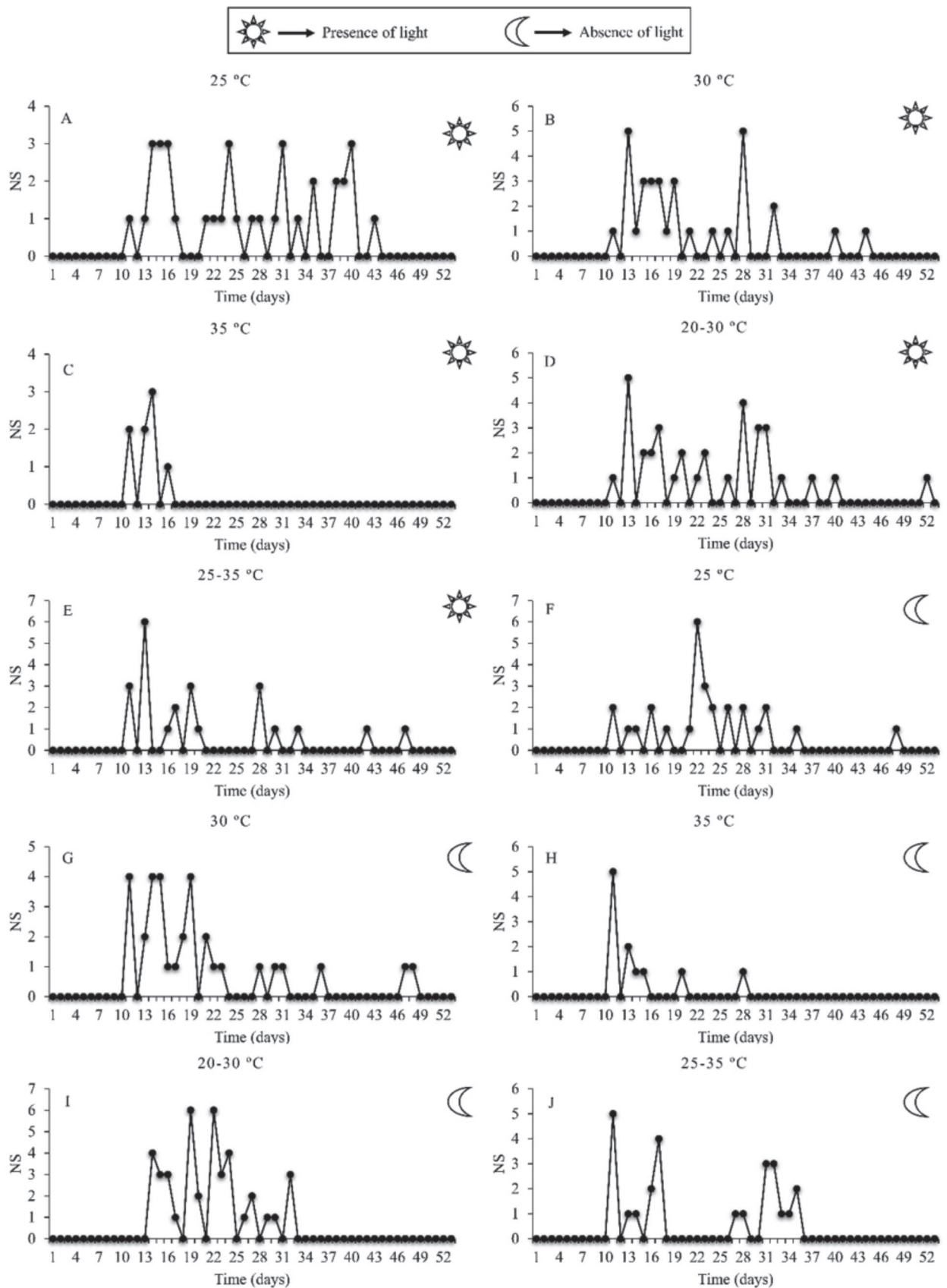
However, others germinated better at constant temperatures, such as *Archontophoenix alexandrae* H. Wendl. & Drude, 25 °C (Teixeira *et al.*, 2011); *Oenocarpus bacaba* Mart., 30 °C (José *et al.*, 2012); *Sabal mauritiiformis* (H. Karst.) Griseb. ex. H. Wendl., 30 °C (Luz *et al.*, 2014); *Euterpe precatória* Mart., 20 °C (Costa *et al.*, 2018); *Syagrus coronata* (Mart.) Becc., 25 °C (Porto *et al.*, 2018).

The germination of the seeds of *A. vestiaria* (Figure 1) and *A. triandra* (Figure 2) responded heterogeneously to all temperatures in the conditions of presence and absence of light, with no expressive single peak, characterizing a polymodal behavior. The results may be

Table 1: Comparison of means for Germination percentage (G%) and germination speed index (GSI) of *Areca vestiaria* Giseke and *Areca triandra* Roxb seeds. ex Buch.-Ham. seeds, submitted to five temperatures and two light regimes

Averages	<i>Areca vestiaria</i>		<i>Areca triandra</i>	
	G% ¹	GSI ²	G% ¹	GSI ²
Light	30.74 a	0.3487 a	60.65 a	0.2067 a
Dark	29.75 a	0.3687 a	59.69 a	0.2043 a
DMSs (5%)	4.70	0.09	8.26	0.03
25 °C	34.32 a	0.3703 ab	0.0 c	0.0 c
30 °C	34.05 a	0.4397 a	53.62 b	0.1960 b
35 °C	16.40 b	0.1805 b	56.83 b	0.1900 b
20-30 °C	37.19 a	0.4583 a	53.49 b	0.1683 b
25-35 °C	29.27 a	0.3447 ab	76.76 a	0.2678 a
DMSs (5%)	10.56	0.21	15.54	0.07
CV (%)	24.08	41.81	21.31	28.20

¹ Data transformed to arc sine $(x/100)^{1/2}$; ² Untransformed data; Means followed by the same letter do not differ in the column, by the Tukey Test, at 5% probability. DMSs (%): minimum significant difference and CV (%): coefficient of variation, expressed as a percentage.



NS – Number of germinated seeds day⁻¹, over 52 days.

Figure 1: Distribution of seed germination of *Areca vestiaria* Giseke submitted to five temperatures in the presence (A, B, C, D and E) and absence (F, G, H, I and J) of light.

related to the one proposed by Meerow and Broschat (2015), in which palm seeds in general have uneven and slow germination, caused by several factors, such as seed maturation stage, presence of mechanical dormancy caused by structures of the fruit, such as the rigid endocarp, which provide resistance to the embryo's expansion. The presence of the pericarp (epicarp and mesocarp) is a limiting factor in seed germination of some palm trees species (Teixeira *et al.*; 2011; Beckmann-Cavalcante *et al.* (2012); Pinto *et al.*, 2012; Pereira *et al.*, 2014). Based on these considerations, it was decided to remove these structures before starting the germination tests, in order not to interfere in the germination process.

For *A. vestiaria*, germination started on the 11th day for all temperatures and light regimes (Figure 1). The stabilization in the presence of light occurred at 16th; 43rd; 44th; 47th and 52nd day for temperatures of 35; 25; 30; 25-35 and 20-30 °C, respectively. Larger peaks were noted on the 14th; 15th; 16th; 24th; 31st and 40th for the temperature of 25 °C with 3 germinated seeds (Figure 1A). For the temperature of 30 °C, it occurred at 13th and 28th days with 5 seeds (Figure 1B). For 35 °C it occurred on the 14th day with 3 seeds (Figure 1C). The alternating temperature of 20-30 °C reached higher peaks on the 13th and 28th days with 5 and 4 germinated seeds, respectively (Figure 1D), and 25-35 °C on the 13th day with 6 seeds (Figure 1E).

In the absence of light, germination stabilization for *A. vestiaria* occurred on the 28th; 32nd; 35th and 48th days for temperatures of 35; 20-30; 25-35; 25 and 30 °C, respectively. Larger peaks were noted on the 22nd day for a temperature of 25 °C with 6 germinated seeds, 11th (Figure 1F); 14th; 15th and 19th day for 30 °C with 4 seeds (Figure 1G). For 35 °C, it occurred on the 11th day with 5 seeds (Figure 1H), 20-30 °C on the 19th and 22nd days with 6 seeds (Figure 1I), and 25-35 °C on the 13th and 17th days, with 5 and 4 seeds (Figure 1J).

For the species *A. triandra*, in the presence of light, temperatures of 30 °C and 20-30 °C started the germination process on the 47th day (Figure 2A; 2C), 35 °C on the 49th and 25-35 °C on the 45th day (Figure 2B; 2D). The germination stabilization occurred on the 96th; 103rd and 105th day for temperatures of 25-35; 35; 30 and 20-30 °C, respectively. Larger peaks were observed on days 63rd; 65th; 84th and 96th for temperatures of 35 °C; 30 °C; 20-30 °C and 25-35 °C with 9; 12; 6 and 16 germinated seeds, respectively.

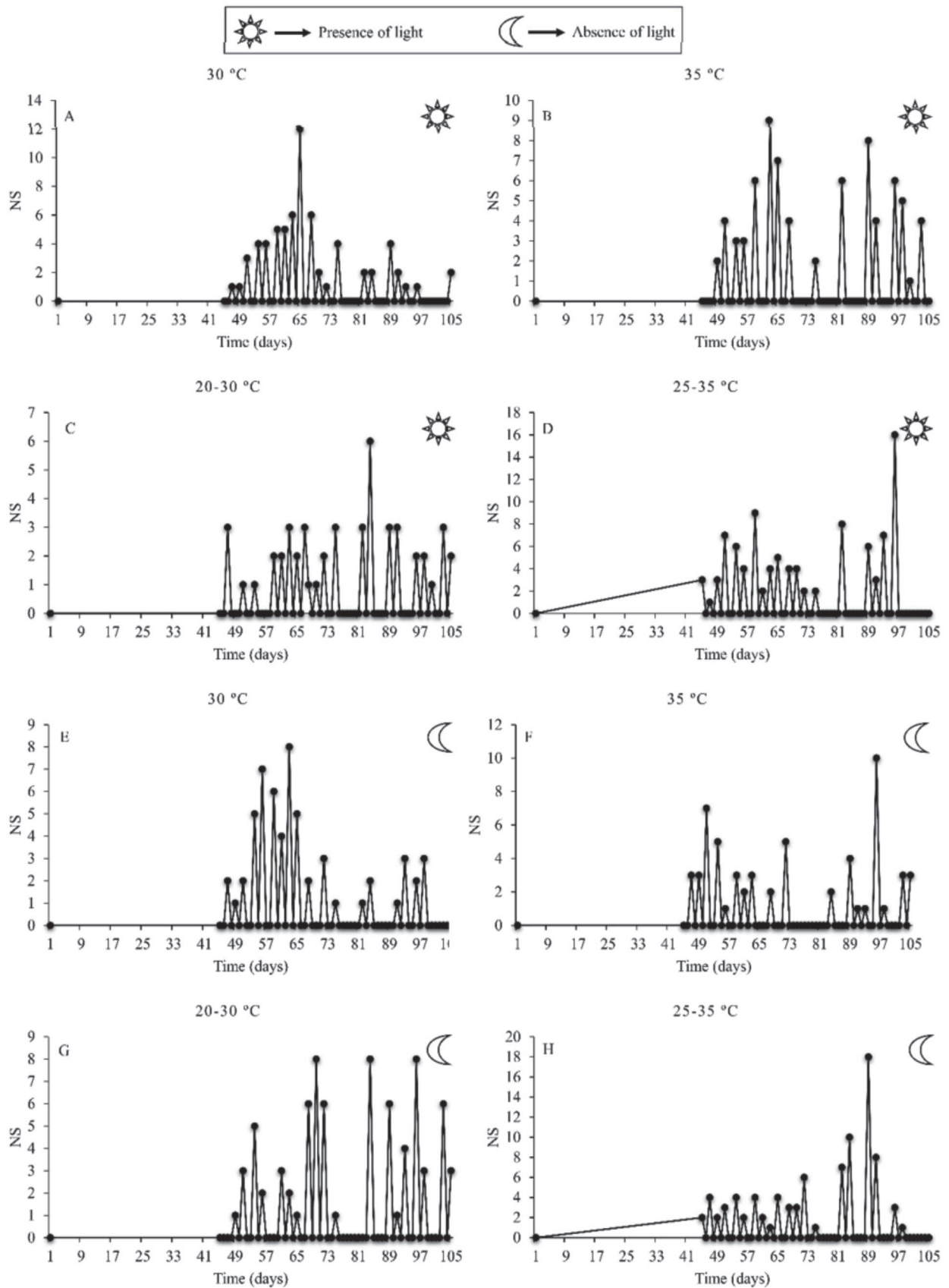
In the absence of light, the germination process for *A. triandra* started on the 45th day with an alternating temperature of 25-35 °C (Figure 2H), after the 47th day for 30; 35 and 20-30 °C (Figure 2E; 2F; 2G). Stabilization occurred at 98th days at 30 °C and 25-35

°C and at 105th at 35 °C and 20-30 °C. With regard to the greatest germination peaks, these occurred at 63rd; 96th and 89th days for temperatures of 30; 35 and 25-35 °C, therefore the alternating temperature of 20-30 °C reached peaks equal to 70th, 84th and 96th days. The number of germinated seeds for each peak was 8; 10; 8 and 18, respectively, for 30; 35; 20-30 and 25-35 °C. During the evaluations, the presence of fungi was observed, which possibly caused deterioration in the seeds, preventing and even delaying the germination process of the seeds that remained viable. Fungi cause decay and heating of the seed mass, resulting in an increase in the respiratory rate and production of mycotoxins, as pointed out by Carvalho & Nakagawa (2012). Thus, in the irrigations, distilled water with 0.2% nystatin was used to minimize contamination by fungi and were carried out whenever there was a need to replace water in the substrate, according to Luz *et al.* (2012).

The species *Oenocarpus bacaba* Mart. studied by José *et al.* (2012) required only 30 days for full germination. Viana *et al.* (2016) researching *Livistona rotundifolia* (Lam.) Mart., observed that this species needed 39 days. The species *Euterpe oleracea* Mart. it required 75 days, accordingly to Gonçalves *et al.* (2010), and *Dypsis lutescens* (H. Wendl.) Beentje & J. Dransf required 59 days (Pêgo & Grossi, 2016), corroborating with the exposed by Meerow and Broschat (2015).

The heterogeneity of germination within the same seed lot distributes the establishment of individuals over time and, with this, the population survives the phases inadequate to its development in the field (Carvalho & Nakagawa, 2012). However, the greater the number of days to emerge and the seedling to remain in the early stages of development, the greater the vulnerability to environmental conditions (Marcos Filho, 2015). However, for the producer who seeks to germinate quickly and homogeneously, it ends up becoming an obstacle, compromising his production, especially if it is on a large scale.

The results indicate that the species used in this research, even belonging to the same genus, have different mechanisms related to the germination process of their seeds, where *A. vestiaria* shows lower germination percentage compared to *A. triandra*. The temperature variation in the germination process of both species can be directly related to the breeding place of the matrices, as it is, according to Köppen (1948), for the region of Brumadinho, MG, average annual temperatures of 20.5 °C and, for Jaboticabal, SP, of 32.5 °C com. According to the same author, both regions have a humid subtropical climate, with a cold, dry winter and a hot, rainy summer.



NS – Number of germinated seeds day⁻¹, over 105 days.

Figure 2: Distribution of seed germination of *Areca triandra* Roxb. ex Buch.-Ham. submitted to four temperatures in the presence (A, B, C and D) and absence (E, F, G and H) of light.

For both species, varying temperatures (constant and alternating) to germination were shown, which may be indicative of adaptability to thermal fluctuations in the environment, thus making them capable of withstanding different climatic conditions. According to Silva *et al.* (2014), seeds of the same species can express diversified germinative behavior as a function of temperature and photoperiod, since within the same species there are variations between individuals as a function of the environment and their genetic constitution, corroborating the results obtained. However, the species *A. triandra* seems to present dormancy due to the prolonged period for the beginning of germination, thus requiring further studies dealing with the methods of overcoming dormancy, but dormancy should always take into account its effective cost and ease of execution.

CONCLUSION

Temperatures of 25 °C, 30 °C, 25-35°C and 20-30 °C are indicated for germination of seeds of the species *Areca vestiaria* and, 25-35 °C for *Areca triandra*, under the presence or the absence of light regimes, and are classified as neutral photoblastics.

Seed germination time varied for both species, with stabilization on the 52nd day for *Areca vestiaria* and on the 105th day for *Areca triandra*.

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