

Germination of *Dypsis decaryi* seeds under salt stress

Germinação de sementes de *Dypsis decaryi* submetidas ao estresse salino

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Highlights

Dypsis decaryi is moderately tolerant to salt stress by NaCl and KCl in germination.

Saline conditions impair seed germination in *Dypsis decaryi*.

Abiotic stresses interfere with seed germination in *Dypsis decaryi*.

Abstract

The triangle palm (*Dypsis decaryi*), a species native to Madagascar, possesses various ornamental characteristics that make it highly valued in the international market. Seed propagation is the primary method for its cultivation, and this process is influenced by factors such as sowing time and salinity. In this study, we aimed to investigate the germination behavior of *D. decaryi* seeds under different saline concentrations of sodium chloride (NaCl) and potassium chloride (KCl), in two distinct periods. The experiment followed a completely randomized design, employing a $2 \times 2 \times 5$ factorial arrangement corresponding to two seasons of experimentation (summer and winter), two salt types (NaCl and KCl), and four salt concentrations (25, 50, 75, and 100 mM), in addition to a control group without salt. Each treatment consisted of four replications, with 25 seeds per replication. The evaluated parameters were germination percentage, germination speed index, and mean germination time. No significant effects were observed when analyzing the salt types individually, the interaction between seasons and salts, or the interaction between all three factors. However, during the summer season, the seeds exhibited higher mean germination percentages and germination speed indices and shorter mean germination times, regardless of the salt used. A high germination percentage (73.79%) was observed at the highest salt concentration tested for both NaCl and KCl. Consequently, we conclude that *D. decaryi* is tolerant to salinity, as simulated by KCl and NaCl, during the seed germination process. Sowing time, salt concentrations, and the interaction between these variables influenced germination.

Key words: Abiotic Stress. Arecaceae. KCl. NaCl. Ornamental plant. Plant propagation.

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Resumo

A palmeira triângulo (*Dypsis decaryi*), nativa de Madagascar, apresenta diversas características de interesse ornamental, tornando-a uma espécie importante para o mercado internacional. Seu principal método de propagação é por semente, que é influenciado por vários fatores como época de semeadura e salinidade. Propôs-se avaliar o comportamento germinativo de sementes de *D. decaryi* em diferentes concentrações salinas de cloreto de sódio (NaCl) e cloreto de potássio (KCl) em duas épocas. O delineamento experimental foi o inteiramente casualizado; o experimento foi realizado em esquema fatorial 2 x 2 x 5, sendo duas épocas de condução do experimento (verão e inverno), dois tipos de sais (NaCl e KCl) e quatro concentrações salinas (25, 50, 75 e 100 mM) e ausência de sais - controle; foram quatro repetições e 25 sementes por parcela. Avaliou-se porcentagem de germinação, índice de velocidade de germinação e tempo médio de germinação. Não houve efeito significativo de forma isolada entre os tipos de sais, para interação de épocas e sais e, entre os três fatores avaliados. Observou-se que no verão as sementes apresentaram maiores médias de porcentagem de germinação, maior índice de velocidade de germinação, e menor tempo médio de germinação para ambos os sais. Foi possível observar alta porcentagem de germinação (73,79%) na maior concentração salina avaliada para ambos os sais. Concluiu-se que a espécie se mostrou tolerante à salinidade simulada por KCl e NaCl, durante o processo de germinação de sementes, o qual foi influenciado pela época de semeadura, concentrações salinas e interação entre épocas e concentrações.

Palavras-chave: Areaceae. Estresse abiótico. KCl. NaCl. Planta ornamental. Propagação vegetal.

Introduction

Dypsis decaryi (Jum.) Beentje & J. Dransf., popularly known as the triangle palm, is a species originating from the savannas of southern Madagascar. It is categorized as vulnerable in its natural habitat, exhibiting a continuous drop in the number of adult individuals, primarily due to the advancement of agriculture and the harvesting of its fruits for the international market of ornamental plants. This species possesses distinct features, such as the tristichous arrangement of its grayish leaves and erect columnar stem (Lorenzi et al., 2004), which contribute to its significant landscape appeal, as recognized by the International Union for Conservation of Nature [IUCN] (2012).

Seeds are the primary means of propagating palm trees. However, successful germination and development of each

species depend on specific conditions (Norsazwan et al., 2020). Given the variability of environmental factors, it is crucial to investigate the effects of abiotic stresses, including water restriction, temperature fluctuations, and salinity, on seed germination (Jiménez-Alfaro et al., 2018).

Salinity poses a significant challenge in global agriculture due to the excessive use of synthetic fertilizers, groundwater irrigation, and aquifer salinization. Thus, there is a growing need to identify plants that are better adapted to saline conditions (Mukhopadhyay et al., 2021).

Salt stress can increase the production of reactive oxygen species, leading to lipid peroxidation of cell membranes, loss of cellular functions, and ultimately seed death (Liang et al., 2018). However, the specific effects of salt stress depend on factors such as the types of salts, their concentrations,

the duration of exposure, crop management practices, and the species involved (Arif et al., 2020).

Sowing time is another crucial factor that influences seed germination and subsequent seedling development. Each species has specific temperature limits that affect germination (Batlla & Benech-Arnold, 2015) and interact with other abiotic factors (Balfagón et al., 2020).

Therefore, the objective of this study was to assess the germination behavior of *Dypsis decaryi* seeds subjected to salt stress induced by the presence of NaCl and KCl at different time periods.

Material and Methods

The study was conducted at the Laboratory of Horticultural Plant Seeds, Department of Agricultural Production Sciences, Plant Production Unit, Faculty of Agricultural and Veterinary Sciences (UNESP/FCAV), Jaboticabal Campus (SP), Brazil. The fruits of *Dypsis decaryi* were collected from five mother plants located in Jaboticabal - SP, Brazil (coordinates 21°15'19" S, 48°19'21" W, 615 m altitude) during two seasons: summer and winter. The fruits were harvested when they started detaching from the clusters and exhibited typical signs of maturation, indicated by a whitish-yellow color of the epicarp (Lorenzi et al., 2004).

A completely randomized experimental design was employed. The treatments were arranged in a 2 × 2 × 5 factorial design, consisting of two seasons (summer and winter), two types of salts (sodium chloride [NaCl] and potassium chloride [KCl]), and four salt concentrations (25, 50, 75, and

100 mM), in addition to a control treatment without salts. Four replications were used, and each plot contained 25 seeds. The NaCl amounts used were 1.46, 2.92, 4.98, and 5.84 g L⁻¹, while for KCl, the amounts used were 1.86, 3.72, 5.60, and 7.45 g L⁻¹, to obtain the desired concentrations. The electrical conductivity of the salt solutions was measured using a pocket conductivity meter (HI98304 DiST®4, Hanna®).

Upon harvesting, the fruits were immediately transported to the laboratory and pulped by manually removing the epicarp and mesocarp using a steel mesh sieve (6 mm). The diaspores (seeds) were then subjected to asepsis by immersing them in a 2% sodium hypochlorite solution for 10 min, followed by rinsing with running water.

After pulping, the initial water content of the seeds was determined. The water content was found to be 43.42% in summer and 49.61% in winter, determined by oven-drying 10 seeds at 105 ± 3 °C for 24 h, with two replicates (Ministério da Agricultura Pecuária e Abastecimento [MAPA], 2009).

Sowing was then conducted in transparent plastic boxes ("gerbox") measuring 11 × 11 × 3 cm, filled with vermiculite of medium particle size as the substrate. The vermiculite was previously moistened with NaCl and KCl solutions to simulate salt stress. Sowing for the summer season was performed on February 18, and for the winter season on July 24, 2021. The substrate was maintained at 100% of its water retention capacity. The choice of vermiculite as the substrate was based on previous studies that found no significant difference in germination between vermiculite and sand for *Dypsis decaryi* seeds. Vermiculite is also

a commonly used substrate in germination tests for species of the Arecaceae family (J. K. Rodrigues et al., 2014; Batista et al., 2016; A. G. Rodrigues et al., 2016). The boxes containing the seeds were placed on laboratory benches under ambient conditions, with average temperatures ranging from 22.1 °C

in winter to 25.5 °C in summer, which have been reported to promote high germination rates in triangular palm seeds (Luz et al., 2008). Temperature (°C) and relative humidity (%) data were recorded daily using a thermo-hygrometer (Simpla TH01, AKSO®) (Table 1).

Table 1
Minimum, average, and maximum temperatures (°C) and relative humidity (%) during the experiments. Jaboticabal - SP, Brazil, 2021

Season	minT	avgT	maxT	RH
	°C			%
Summer	24.4	25.5	27.1	57.3
Winter	19.2	22.1	24.9	49.3

minT = minimum temperature; avgT = average temperature; maxT = maximum temperature; and RH = relative humidity.

Germination was evaluated daily after the start of the experiment. Seeds that produced a germination bud until germination stabilization (observed at 40 days, for both seasons) were considered germinated (A. G. Rodrigues et al., 2016). From the collected data, the following parameters were calculated: germination percentage (%G), using the formula specified in the Manual of Rules for Seed Analysis (MAPA, 2009); Germination Speed Index (GSI), based on the formula developed by Maguire (1962) using the daily count of germinated seeds; and mean germination time (MGT), according to the formula proposed by Labouriau (1983).

Statistical analysis was performed using AgroEstat® statistical software version 1.1.0.711 (Barbosa & Maldonado, 2014). When significant effects of season and salt type were observed, means were compared using Tukey's test at a significance level of 5% ($p \leq 0.05$). Polynomial regression

analysis was conducted to examine the behavior of the variables with increasing salt concentrations when significant differences were detected between the concentrations. The germination percentage data were transformed into arcsine $(x/100)^{1/2}$ for statistical analysis purposes. Germination distribution over time was also plotted graphically.

Results and Discussion

Sowing time significantly influenced germination percentage (%G), with seeds sown in the summer showing a higher percentage of germination. Seeds sown in the summer also exhibited faster germination, as indicated by the higher germination speed index (GSI) and lower mean germination time (MGT). The type of salt (NaCl and KCl) did not have a significant isolated effect on the analyzed variables (Table 2).

Table 2

Germination percentage (%G), germination speed index (GSI), and mean germination time (MGT) of *Dypsis decaryi* (Jum.) Beentje & J. Dransf. seeds sown in two seasons, under different NaCl and KCl saline concentrations. Jaboticabal - SP, Brazil, 2021

Season	%G ¹	GSI ²	MGT (days)
Summer	82.12 a	2.06 a	12.08 b
Winter	78.00 b	1.34 b	20.98 a
Salt			
NaCl	79.06 a	1.72 a	16.44 a
KCl	81.06 a	1.68 a	16.63 a
CV (%)	9.02	8.28	9.28

¹Data transformed into arcsine $(x/100)^{1/2}$; ²Untransformed data; means followed by the same letter do not differ from each other in the column, by Tukey's test, at 5% probability. CV (%): coefficient of variation, expressed as a percentage.

During the summer season, the average temperature and relative humidity were 25.5 °C and 57.3%, respectively, while in winter, they were 22.1 °C and 49.3%, indicating milder and drier conditions in the latter season (Table 1). Similar to the findings of this study with *D. decaryi*, other palm species of tropical origin, such as *Attalea vitrivir* Zona (babassu palm) (Neves et al., 2013) and *Bactris maraja* Mart. (marajá palm) (J. K. Rodrigues et al., 2014), have also shown better germination at higher temperatures above 25 °C, while lower temperatures negatively affect germination. Previous studies on *D. decaryi* by Luz et al. (2008) also reported high germination percentages (above 80%) in several ranges of controlled temperatures above 25 °C and higher GSI at ambient temperatures ranging from 24 to 29 °C.

Seeds germinate slowly at lower temperatures due to reduced metabolic activity, which delays the germination process. Extreme temperatures, whether too high or too low, can result in complete inhibition of germination due to loss of seed viability (A. G. Rodrigues et al., 2016). The higher

germination percentage observed during the summer season in this study suggests a correlation with high temperatures and relative humidity levels throughout the days (Table 1). In contrast, during the winter season with lower average temperature and relative humidity, the germination process slows down and shows dispersed germination over time, which can be considered an adaptive trait of the species (Jaganathan, 2021).

Regarding the NaCl and KCl concentrations and their effect on %G, there was a significant effect in both seasons, with a decrease in %G as the salt concentrations increased. The highest salt concentration (100 mM) resulted in an 11.03% reduction in germination compared to the control treatment without salts. However, even at the highest salt concentration, a high germination percentage (73.79%) was still observed, indicating tolerance to salinity during germination (Figure 1A). Similar findings were reported for *Phoenix dactylifera* 'Mabroom' (date palm), which exhibited germination rates above 90% at the highest NaCl concentration (150 mM) (Al-Qurainy et al., 2020).

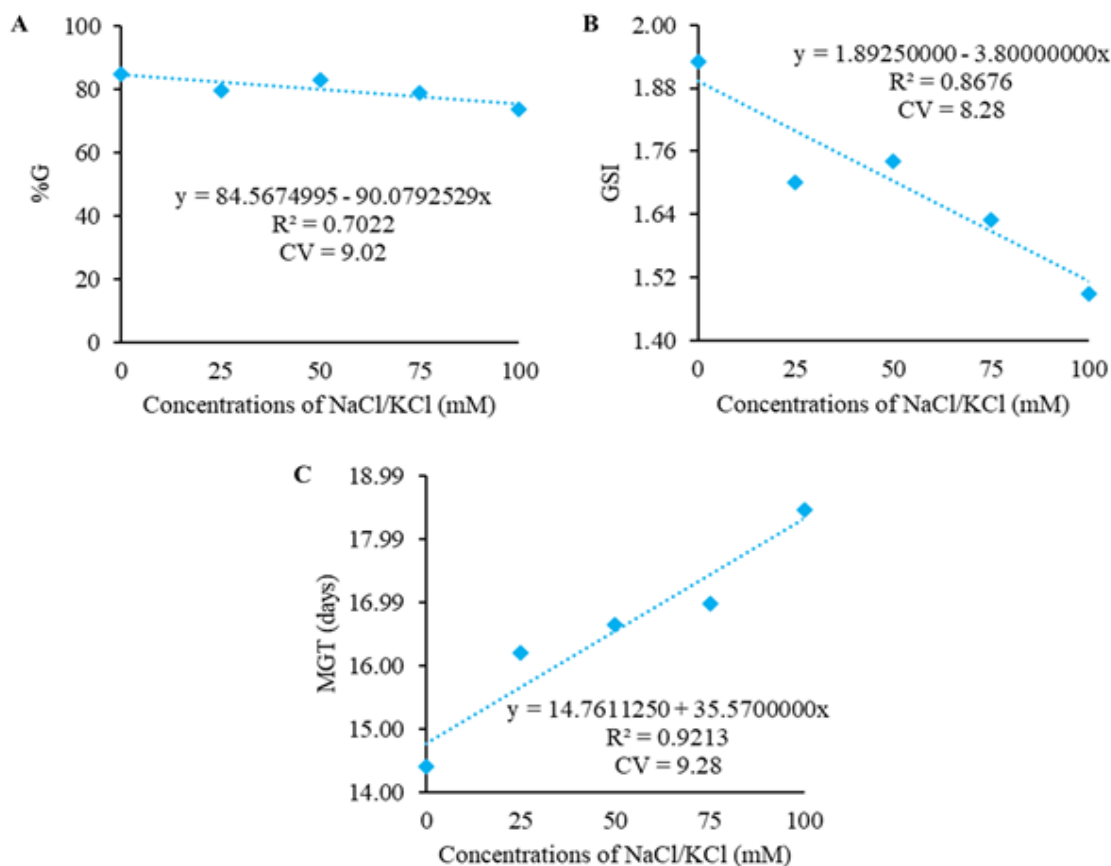


Figure 1. Germination percentage - %G (A); Germination Speed Index – GSI (B); and mean germination time – MGT (C) of *Dypsip decaryi* (Jum.) Beentje & J. Dransf. seeds sown in two seasons, as a function of increasing NaCl and KCl concentrations. Jaboticabal - SP, Brazil, 2021.
* - Significant at $p \leq 0.05$ by the F test.

The GSI of the seeds decreased by 22.79% with increasing NaCl and KCl concentrations (Figure 1B). Additionally, MGT increased by 28.19% at the 100 mM concentration compared to the control treatment (Figure 1C), suggesting that higher salt concentrations induced a delay in seed germination.

The interference with the seed germination process due to high salt concentrations is attributed to reduced osmotic potential, which delays water absorption by the seeds and increases the concentration of ions such as sodium and

chlorine to toxic levels in the embryo (Van Zelm et al., 2020). The impact of high salinity can be further influenced by temperature variations. In *Limonium mansanetianum* (stative) seeds, a 5 °C decrease in temperature from the optimum level for germination of the species resulted in reduced final germination at all studied salt concentrations (Fos et al., 2021). Similarly, *Limonium supinum* (stative) exhibited decreased or inhibited germination at the NaCl concentrations of 100, 200, and 400 mM with a temperature increase of 5 to 10 °C above the optimal germination temperature (Melendo & Giménez, 2019).

Salinity stress reduces the osmotic potential of the medium, prolonging water absorption time and leading to a longer emergence of the germinal bud and primary root. Therefore, the decrease in GSI and %G and the increase in MGT (Figure 1) can be attributed to the physiological drought caused by increased salt concentration (Jacob et al., 2020). However, the response to salt stress can vary between species, types of salts, and their concentrations, highlighting the importance of studies to determine tolerance levels and survival capacity in environments with these characteristics (Shahid et al., 2020).

Similar to *D. decaryi*, other palm species, such as *Carpentaria acuminata* (carpentaria palm) and *Ptychosperma elegans* (solitaire palm), have shown tolerance to salinity during the seed germination process (Batista et al., 2016). Moreover, species from other plant families, including *Festuca arundinacea* (tall fescue) and *Phleum pratense* (Timothy grass), have exhibited similar patterns (Sharavdorj et al., 2021). Understanding salinity tolerance is crucial for developing production strategies in regions affected by salt stress, as over 20% of irrigated land and 8.7% of global soils are estimated to be impacted by salt stress as reported by the Food and Agriculture Organization [FAO] (2021).

Analysis of the germination distribution over time revealed variations in germination patterns between the studied periods. For NaCl treatments, germination initiation occurred on the 8th day of evaluation, with a pronounced peak between days 11 and 13 during the summer season. In the winter season, germination started on the 9th day for saline treatments but showed

uneven distribution over the days (Figure 2). Similar germination distribution patterns were observed for the KCl treatments (Figure 3).

In a study that did not involve the use of saline solutions, Luz et al. (2008) observed a germination peak for *D. decaryi* seeds at room temperature on the sixth day of evaluation. Under similar conditions, Bao et al. (2010) reported the initiation of germination only on the tenth day. These variations may be attributed to climatic conditions, genetic factors, the origin of the fruit matrices, and temperature conditions during the experiments (Saatkamp et al., 2019).

The electrical conductivity values obtained for NaCl treatments were 3.14, 5.44, 7.62, and 10.71 dS m⁻¹ for the concentrations of 25, 50, 75, and 100 mM, respectively. For KCl treatments, the electrical conductivity values were 3.53, 6.32, 9.31, and 11.47 dS m⁻¹ for the same concentrations. Based on the results of this study, *D. decaryi* seeds demonstrated moderate tolerance to the stress caused by NaCl and KCl, as they were able to germinate under the highest saline concentration evaluated (100 mM), corresponding to electrical conductivity values of 10.71 and 11.47 dS m⁻¹ for NaCl and KCl, respectively, which are considered highly saline solutions (Srivastava et al., 2019).

However, due to the sensitivity of *D. decaryi* seeds to desiccation (Batista et al., 2016), water restriction associated with lower temperatures can directly affect seed germination, leading to lipid peroxidation and seed deterioration, thereby hindering successful germination (Ibrahim, 2016) and subsequent seedling production.

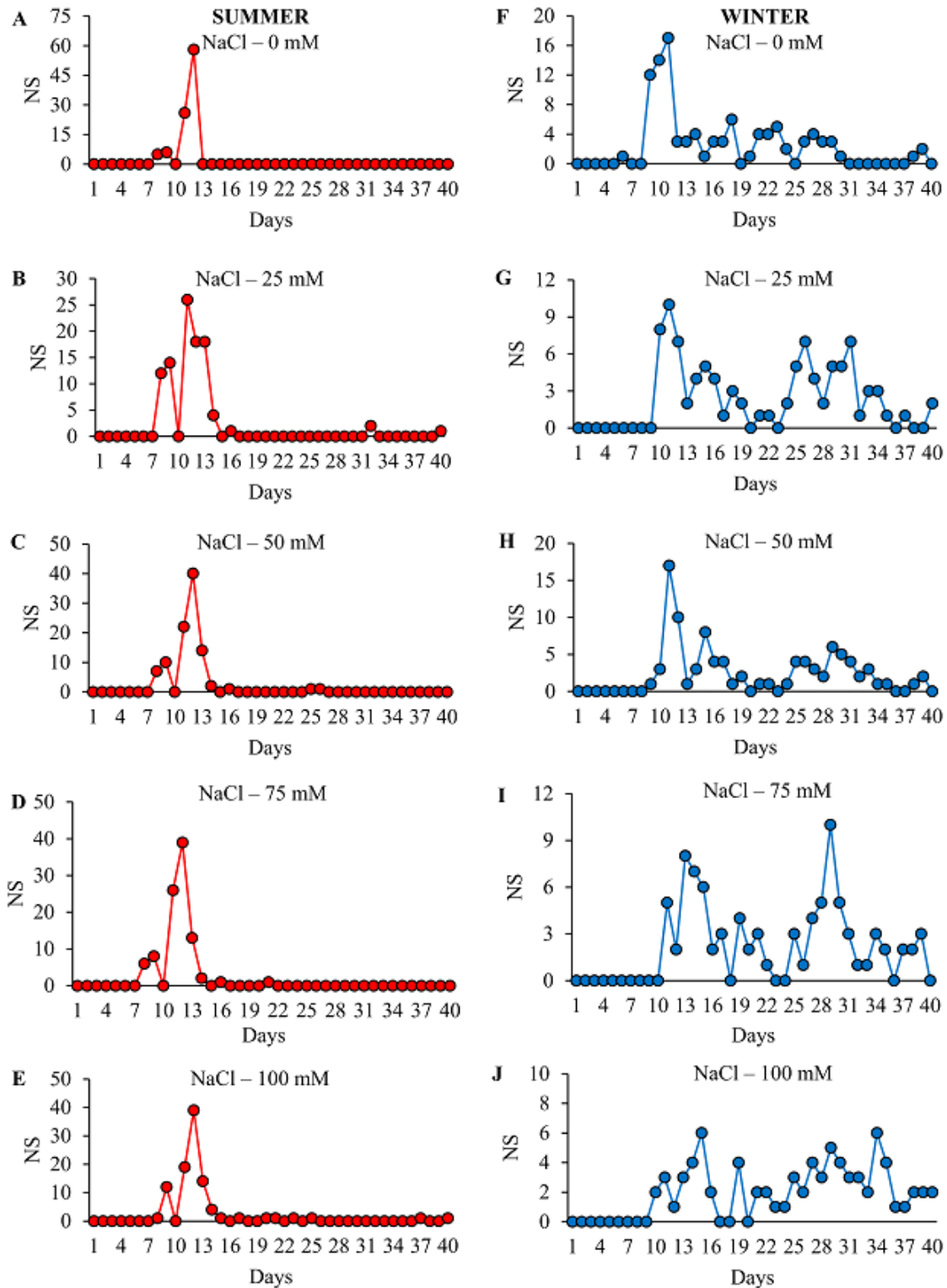


Figure 2. Seed germination distribution of *Dypsis decaryi* (Jum.) Beentje & J. Dransf. sown in two seasons, under different NaCl saline concentrations. Jaboticabal - SP, Brazil, 2021. NS – Number of germinated seeds day⁻¹, over 40 days.

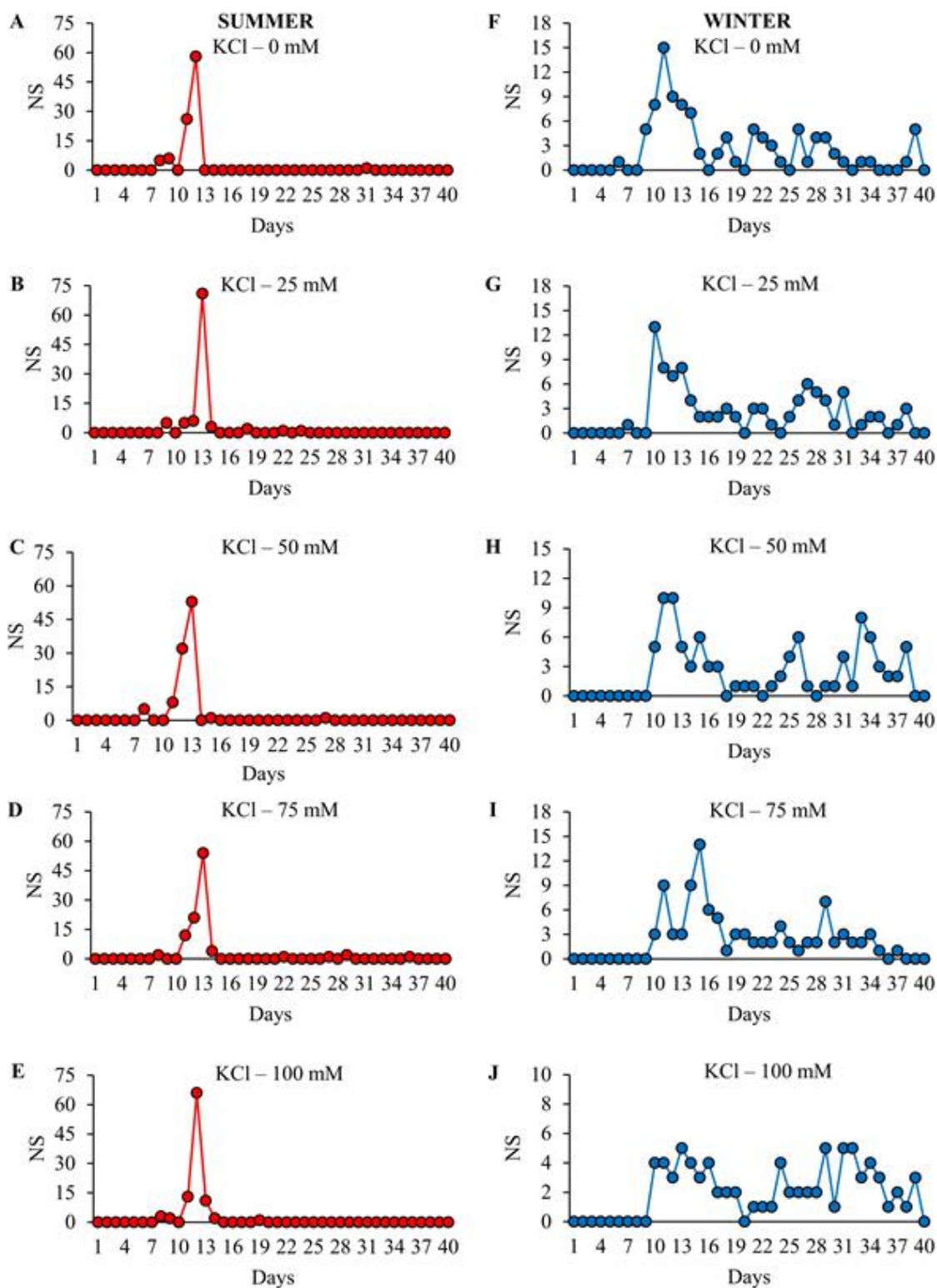


Figure 3. Seed germination distribution of *Dypsis decaryi* (Jum.) Beentje & J. Dransf. sown in two seasons, under different KCl saline concentrations. Jaboticabal - SP, Brazil, 2021. NS – Number of germinated seeds day⁻¹, over 40 days.

Conclusions

Dypsis decaryi exhibited a moderate level of tolerance to salinity stress induced by NaCl and KCl during the seed germination process.

The summer season was found to be favorable for seed germination, as indicated by higher mean germination percentages and germination speed indices and shorter mean germination times observed under both salt treatments.

The germination peak for both salts occurred between days 11 and 13 in the summer season. In contrast, during the winter season, there was an uneven distribution of seed germination over the evaluation period.

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