Acta Scientiarum
http://periodicos.uem.br/ojs
ISSN on-line: 1807-8664
Doi: 10.4025/actascitechnol.v46i1.67282

STATISTICS

Lifetime of forage cactus submitted to different water and saline levels

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ABSTRACT. Forage cactus is mostly grown in hot climates, where water deficit is quite common, consequently they end up suffering from saline stress, as the waters of these regions have high concentrations of salts. The buildup of salts in the soil promotes an unfavorable environment for the development of plants, as it negatively alters the growth of crops, reducing the productivity of forage plants in agricultural areas. The objective was then to evaluate the lifetime of *Nopalea cochenillifera* cactus under water and saline stress conditions. This research was conducted from September 2021, to August 2022, in the forage farming sector of the Federal Rural University of Pernambuco. The design used was completely randomized, in a factorial scheme, being composed of two clones of cactus (Clones Giant and Little Sweet), two levels of water irrigation (0.5 and 1L weekly), five levels of saline stress (0, 2, 4, 6 and 8 dS m^{-1}), with four replications. The lifetime of forage cactus was evaluated using the statistical methodology of survival analysis by Kaplan-Meier method. The plant mortality rate of *N. cochenillifera* clone Giant Sweet at 257 days after planting was of 20%, while the clone Little Sweet showed a mortality rate of 15%. When irrigated with 0.5 liter of water, the plants of the clone Giant Sweet presented a mortality rate of 5.0%, whereas the clones Little Sweet showed no mortality. When using 1.0 liter of irrigation water, the clones Giant and Little Sweet showed a mortality rate of 35.0 and 30.0%, respectively.

Keywords: Survival; *Nopalea cochenillifera*; salinity.

Received on January 26, 2023.
Accepted on February 05, 2024.

Introduction

Forage cactus (*Nopalea* spp. and *Opuntia* spp.), can contribute to increasing agricultural biomass yield by improving efficient use of local natural resources (Costa, Oliveira, Caraciolo, Lucena, & Leite, 2022). Cacti offer alternative forage in semiarid regions, due to their high phytomass production and energy values, the richness of their non-fibrous carbohydrate content, large water reserves, and easiness of propagation (Freire et al., 2018).

Water stress promotes a series of damages to plant metabolism, such as reduced photosynthesis, reduced cell division, less cell differentiation and fewer cells, compromising the expansion of plant tissues in plants, greater energy expenditure and production of reactive oxygen species (Farooq, Hussain, Ul-Allah, & Siddique, 2019; Tardieu, Granier, & Muller, 2011).

Knowledge of how forage plants perform under water stress is therefore valuable to understanding the effect of the “dry” period on forage production, and therefore enabling the development of viable cultivation improvement practices (Araújo Jr. et al., 2019), these plants may develop mechanisms of tolerance or even adaptation to such conditions (Almeida et al., 2021). Forage cactus stands out as a promising solution to these difficulties, being tolerant of both water and salt deficit (Freire et al., 2018).

Given the above, the study of the lifetime of these plants can provide useful information for agriculture, the knowledge of phenological events measured on a temporal scale (lifetime, time to flowering, time to harvest, etc.) is of fundamental importance, and useful for the efficient management of crops. The lifetime of forage plants provides very useful information in the cultivation and management of natural populations, and they have great biological importance, as they enable the viability of the cultivation of a species by evaluating the growth rate.

Survival analysis is still not widespread in agricultural sciences, this type of analysis presents a series of advantages over traditional approaches, as it allows comparing the pattern of occurrence of phenological events over time; makes it possible to estimate the probability of occurrence of events at specific intervals, which are important for planning management or marketing activities and provides information on the percentage of phenological events; in addition to not requiring data normality or homogeneity of variances (Gienapp, Hemerik, & Visser, 2005).

In survival analysis, the response variable is, in most cases, the time until a certain event occurs. his time is called failure time and can be the time until the death of the individual under analysis or any other event of interest (Mazucheli, Oliveira, Peralta, & Emanuelli, 2018). A characteristic resulting from these studies is, then, the presence of incomplete or partial observations. These observations, called censure, can occur for a variety of reasons, including the death of a plant during the study and the non-occurrence of the event of interest until the end of the research (Lucena et al., 2020).

Studies on the lifetime of forage plants under water deficit are still rare, even though this is one of the most frequent stresses, however Lucena et al. (2020) evaluated the lifetime of *Pennisetum glaucum* under water and saline stress conditions through survival analysis. Given the above, this study aimed to analyze the lifetime of cactus forage of *Nopalea cochenillifera* Giant Sweet and Little Sweet clones submitted to different water and saline regimes using survival analysis.

Material and methods

This research was carried out from September 2021 to August 2022, in the forage farming sector (GEFOR) of the Federal Rural University of Pernambuco (UFRPE), Academic Unit of Serra Talhada (UAST), located in the State of Pernambuco, Northeast region, Brazil, which was located in the following geographic coordinates (07° 57’ 01” S and 38° 17’ 53” E) at an elevation of 523 meters.

According to Koppen, the climate condition is a BSwh’ with a rainy season during the summer, starting in November and ending in April. The average annual rainfall is 632.2 mm, the average annual air temperature is 26°C, and the average air relative humidity is 60% (Lucena, Leite, Simões, Almeida, & Simplício, 2021).

The soil used in the experiment was collected at a depth of 0-20 cm and classified as Typical Haplic Cambisol Ta Eutrophic, as described by Empresa Brasileira de Pesquisa Agropecuária [EMBRAPA], (2013). The soil sample was analyzed by the soil fertility laboratory of the Empresa Pernambucana de Pesquisa Agropecuária (IPA) and was characterized by the following chemical attributes: pH (water) = 6.80; P (extractor Mehlich I) = 40 mg dm^{-3}; K^+ = 0.45; Ca^{2+} = 5.50; Mg^{2+} = 1.60; and Al^{3+} = 0.0 cmolc dm^{-3}.

The accumulated monthly rainfall throughout the experimental period was higher than the monthly average except for the months of August and September 2021. While for the month of October 2021 it rained 475% more than expected for the month, in November and December rained 163 and 246% more than expected for the month, respectively. In the first three months of 2022, it rained 191, 133 and 142% more than expected for the period, while the months of April (58%) and May (13%) rained below the monthly average and the months of June and July 2022 showed records above the monthly average with 232 and 170%, respectively Figure 1.

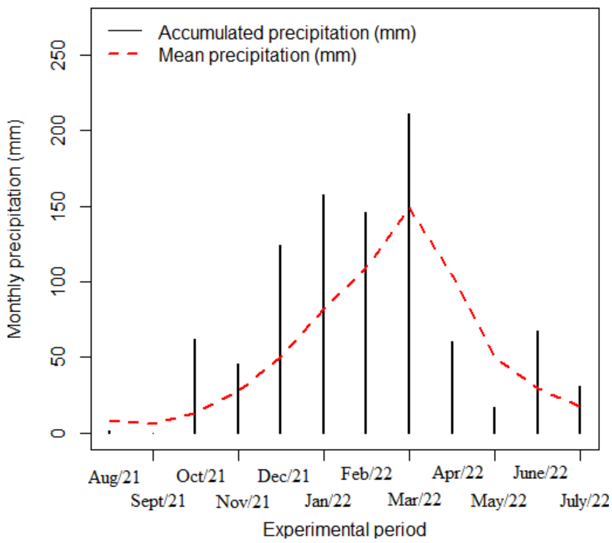

Figure 1. Monthly accumulated rainfall in the municipality of Serra Talhada during the experimental period.

The design used was in a factorial scheme, consisting of two forage cactus clones (Giant Sweet and Little Sweet), two levels of water replacement (0,5L and 1L) and five levels of salinity (0, 2, 4, 6 and 8 dS m^{-1}), with four replications, totalizing 80 sample units.

The plants received two levels of water, where they were irrigated weekly, one plant group received 0.5 liter and the other received 1.0 liter of irrigation water. Five levels of water salinity were used, resulting from different electrical conductivities (ECa) (0, 2, 4, 6 and 8 dS m^{-1}).

According to the classification of irrigation water in terms of salinity risk, proposed by Sales, Lopes, Meireles, Chaves, and Andrade (2014), CEa levels (0, 2, 4, 6 and 8 dS m^{-1}) are classified as C1 (low risk-0 dS m^{-1}), C3 (high risk- 2 dS m^{-1}) and C4 (very high risk- 4, 6 and 8 dS m^{-1}). To obtain the saline levels, sodium chloride (NaCl) salts were added to distilled water. In the laboratory, the levels of 2, 4, 6 and 8 dS m^{-1} were obtained through the concentrations of salts of (NaCl) corresponding to 2.32, 4.64, 6.96 and 9.28 g/L, respectively.

The cladodes seeds came from mother plants aged four years, free from damage and pests, avoiding phytosanitary complications, reducing mortality and low development of the crop. Posteriorly the cladodes seeds underwent a curing time of 15 days in full shade. Cladodes of forage cactus Giant Sweet and Little Sweet clones were planted in pots (capacity, 14.41 dm^3; 15 kg soil; one cladode per pot). Pots were arranged with a spacing of 0.5×0.5 m and placed on brick blocks to avoid direct contact with the underlying soil and facilitate the collection of drained water.

To carry out the planting, there was a process of randomization of the treatments (T1- 0 dS m^{-1} of salinity; T2- 2 dS m^{-1} of salinity; T3- 4 dS m^{-1} of salinity; T4- 6 dS m^{-1} of salinity; T5- 8 dS m^{-1} salinity) through the R-project software, where the position of the treatments was defined, according to the sampling scheme in Figures 2 and 3.

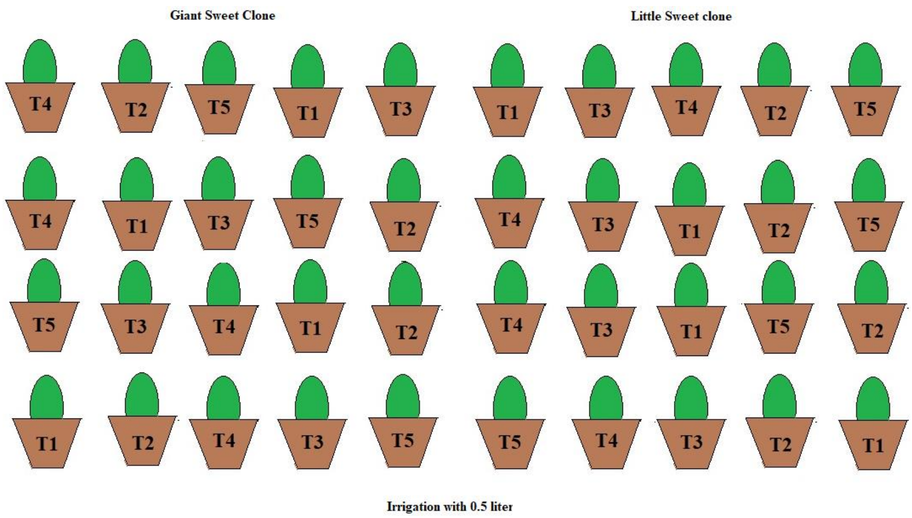

Figure 2. Sampling scheme of forage cactus irrigated with 0.5 liters of water. T1-0 dS m^{-1} salinity; T2- 2 dS m^{-1} salinity; T3-4 dS m^{-1} salinity; T4- 6 dS m^{-1} salinity; T5- 8 dS m^{-1} salinity.

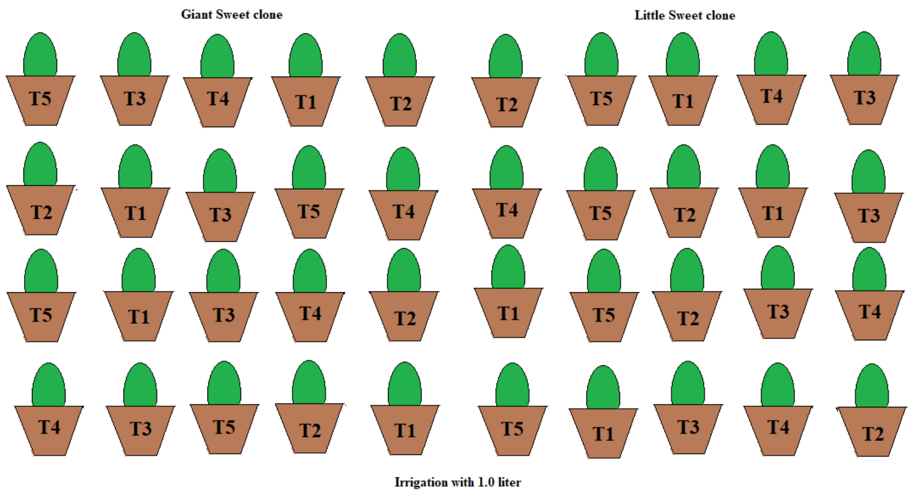

Figure 3. Sampling scheme of forage cactus irrigated with 1.0 liters of water. T1-0 dS m^{-1} salinity; T2- 2 dS m^{-1} salinity; T3-4 dS m^{-1} salinity; T4- 6 dS m^{-1} salinity; T5- 8 dS m^{-1} salinity.

The life-to-death time of forage cactus was evaluated. The evaluations were performed daily throughout the experimental period. The results were recorded in an Excel spreadsheet and the analysis were performed using the R-project software, version 2.13.1 for Windows (R Core Team, 2019).

To evaluate the lifetime of forage cactus, the statistical methodology of survival analysis was used. In survival analysis, the response variable is in most cases the time pending the occurrence of a certain event. This time is called failure time and can be the time until the death of the individual (plant) under evaluation or any other event of interest (Lucena et al., 2020).

The non-negative random variable T, usually continuous, which represents the time to failure, is usually specified in survival analysis by survival function or by the failure rate function (risk) (Lucena et al., 2020). The survival function is defined as the probability that an observation does not fail until a certain time t, that is, the probability that an observation will survive time t. The survival function is described by:

$$S(t)=P(T \geqslant t)$$

In contrast, the cumulative distribution function is defined as the probability that an observation will not survive time t, that is,

$$F(t)=1-S(t)$$

The survival function can be obtained through non-parametric estimators, being largely through the Kaplan-Meier estimator. The Kaplan-Meier estimator is an adaptation of the empirical survival function defined by:

$$S(t)=\prod_{j:\, t_j<t}\left(\frac{n_j-d_j}{n_j}\right)=\prod_{j:\, t_j<t}\left(1-\frac{d_j}{n_j}\right)$$

where, $t_1< t_2< \ldots < t_k$, the k times distinct and ordered of failure; d_j number of failures in t_j, $j=1,2,\ldots,k$ and n_j is the number of plants at risk in t_j.

The graph of S(t) versus time (t) is called the survival curve (Nesi, Shimakura, Junior, & Mio, 2015). Kaplan-Meier estimates the curve based on survival times, without having to assume a probability distribution, even when there are censored data in the set of observations (Lira, Foschini, & Rocha, 2020).

Results and discussion

Figure 4 shows that the clone of cactus Giant Sweet began to die at 25 days after planting (DAP). The mortality rate of *N. cochenillifera* clone Giant Sweet plants at 257 DAP is 20%.

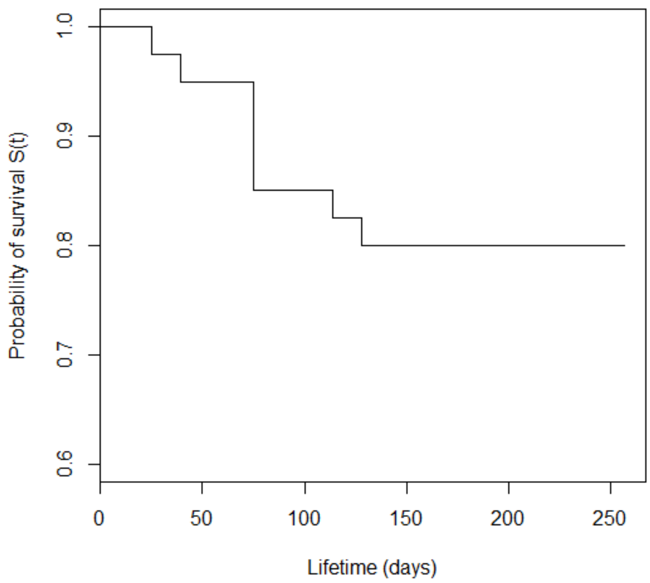

Figure 4. Lifetime of Giant Sweet cactus submitted to water and salt deficit.

N. cochenillifera clone Little Sweet plants began to die at 75 DAP, leading to a 15% mortality rate after 257 DAP, Figure 5.

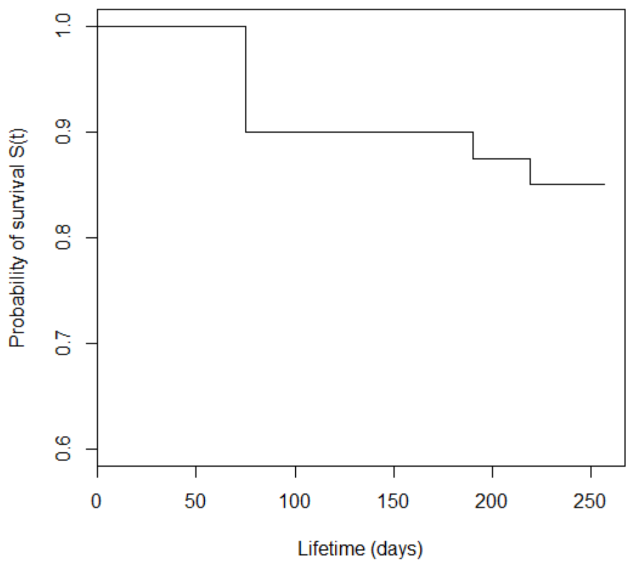

Figure 5. Lifetime of Little Sweet cactus submitted to water and salt deficit.

It was confirmed that when 0.5 liter of irrigation water was used weekly in the Giant Sweet cactus clones, only one plant (5.0%) died, whereas for the Little Sweet clone, the mortality rate was 0%. When using 1.0 liter of irrigation water, the Giant Sweet cactus clones presented a mortality rate of 35.0%, regardless of the salinity level evaluated, while the Little Sweet cactus clones presented a mortality rate of 30.0%, Table 1.

Table 1. Mortality of Giant and Little Sweet cactus as a function of different levels of irrigation and salinity.

Salinity level (dS m^{-1})	Mortality rate (%)			
	Giant Sweet		Little Sweet	
	0.5 L	1.0 L	0.5 L	1.0 L
0	5.0%	10.0%	0.0%	0.0%
2	0.0%	10.0%	0.0%	5.0%
4	0.0%	0.0%	0.0%	5.0%
6	0.0%	10.0%	0.0%	5.0%
8	0.0%	5.0%	0.0%	15.0%
Total by irrigation level	5.0%	35.0%	0.0%	30.0%
General total	20.0%		15.0%	

Cactus that received 1.0 liter of water weekly irrigation showed a mortality rate of 35.0% for the clone Giant Sweet and 30.0% for the clone Little Sweet, regardless of the applied salinity level, these results may have occurred due to the fact that in the experimental period the rainfall was much higher than expected for the months (Figure 1) causing an increase in the volume of water in the pots and consequently dilution of the salt applied to them.

One of the main factors causing stress in plants is water availability. Plants can suffer damage from either excess or lack of water. Excess water in the soils of the pots caused poor drainage, resulting in reduced respiration of the root system and absorption of soil solution, in addition to rotting of the roots, causing the death of the plants. Therefore, cactus mortality is due to excess water in the soil and not to salt, as it was diluted and did not directly influence palm mortality.

Through the Kaplan-Meier estimator, it was observed that the cactus of *N. cochenillifera* clone Giant Sweet, when irrigated weekly with 0.5 liters of water and without application of salt, started to die at 128 DAP and presented a survival rate of 75.0% after 257 DAP, while cactus that received 1.0 liter of irrigation water weekly and without salt application began to die at 75 DAP, and had a survival rate of 50.0% after 257 days of life, Table 2.

Cactus that received application of 2, 4, 6 and 8 dS m^{-1} of salinity and were irrigated with 0.5 liters of water weekly, presented survival of 100.0% at 257 days of life, while plants irrigated with 1.0 liter of water weekly showed survival of 50.0% when they received 2 and 6 dS m^{-1} of salinity, 75.0% for those who received 8 dS m^{-1} of salinity and 100.0% for those who received 4 dS m^{-1} of salinity.

Table 2. Survival of Giant Sweet cactus under different levels of irrigation and salinity.

Irrigation level of 0.5 liter per week				Irrigation level of 1.0 liter per week			
Lifetime	Exposed cactus	Dead cactus	S(t)	Lifetime	Exposed cactus	Dead cactus	S(t)
			Salinity level of 0 dS m^{-1}				
0	4	0	100.0	0	4	0	100.0
128	4	1	75.0	75	4	1	75.0
257	3	0	75.0	114	3	1	50.0
				257	2	0	50.0
			Salinity level of 2 dS m^{-1}				
0	4	0	100.0	0	4	0	100.0
257	4	0	100.0	75	4	2	50.0
				257	2	0	50.0
			Salinity level of 4 dS m^{-1}				
0	4	0	100.0	0	4	0	100.0
257	4	0	100.0	257	4	0	100.0
			Salinity level of 6 dS m^{-1}				
0	4	0	100.0	0	4	0	100.0
257	4	0	100.0	39	4	1	75.0
				75	3	1	50.0
				257	2	0	50.0
			Salinity level of 8 dS m^{-1}				
0	4	0	100.0	0	4	0	100.0
257	4	0	100.0	25	4	1	75.0
				257	3	0	75.0

S(t)-probability of survival by the Kaplan-Meier estimator.

Using the Kaplan-Meier estimator, it was observed that the cactus of *N. cochenillifera* clone Little Sweet, when irrigated weekly with 0.5 liters of water and regardless of the applied salinity level, showed a survival of 100.0%, respectively, after 257 days of life. The same was observed for cactus that received 1.0 liter of irrigation water weekly and did not receive salt. The plants irrigated with 1.0 liter of water weekly and received application of 2, 4, 6 and 8 dS m^{-1} of salt, presented survival of 75.0, 75.0, 75.0 and 25.0%, respectively after 257 days of life, Table 3.

Table 3. Survival of Little Sweet cactus under different levels of irrigation and salinity.

Irrigation level of 0.5 liter per week				Irrigation level of 1.0 liter per week			
Lifetime	Exposed cactus	Dead cactus	S(t)	Lifetime	Exposed cactus	Dead cactus	S(t)
			Salinity level of 0 dS m^{-1}				
0	4	0	100.0	0	4	0	100.0
257	4	0	100.0	257	4	0	100.0
			Salinity level of 2 dS m^{-1}				
0	4	0	100.0	0	4	0	100.0
257	4	0	100.0	75	4	1	75.0
				257	3	0	75.0
			Salinity level of 4 dS m^{-1}				
0	4	0	100.0	0	4	0	100.0
257	4	0	100.0	75	4	1	75.0
				257	3	0	75.0
			Salinity level of 6 dS m^{-1}				
0	4	0	100.0	0	4	0	100.0
257	4	0	100.0	219	4	1	75.0
				257	3	0	75.0
			Salinity level of 8 dS m^{-1}				
0	4	0	100.0	0	4	0	100.0
257	4	0	100.0	75	4	2	50.0
				190	2	1	25.0
				257	1	0	25.0

S(t)-probability of survival by the Kaplan-Meier estimator.

Plant responses to water deficit depend on the amount of water lost through transpiration, the rate of loss and the duration of the stressful condition. In addition, the effects of water stress still depend on irrigation management, and the shorter the interval between irrigation events, the smaller the effect of water deficit will be (Ismail, El-Nakhalwy, & Basahi, 2018). In other words, the irrigation management carried out in the present work, with a weekly irrigation shift together with the amount of precipitation that occurred in the experimental period, also influenced a shorter lifetime of the evaluated palm clones.

The lifetime of plant tissue may be related to the degree of damage or inhibition of vital metabolic processes in plants (especially those that require water), such as ion and nutrient transport functions, cell development and solute translocation (Zargar et al., 2017).

In addition, it should be considered that the first response of plants to water deficit (stomatal closure) leads to a lower absorption of CO_2, contributing to lower yields or making the survival of plants unfeasible. (Tardin et al., 2013). In addition, a lower assimilation of CO_2 promotes a reduction in the speed of the Calvin Cycle, resulting in a surplus of electrons from the photochemical part of photosynthesis, resulting in the production of reactive oxygen species (EROs) that promote cell death.

Conclusion

Cactus mortality occurs due to the large volume of rains during the plant cycle.

The *Nopalea Cochenillifera* clone Giant Sweet showed a 75% higher mortality rate than the clone Little Sweet, regardless of irrigation volume and salinity.

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