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Effects of the hydrogen potential and fungicide treatment on Pitaya seed germination

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ABSTRACT. The objective of the present study was to evaluate pitaya seed germination under different hydrogen potentials, with or without fungicide treatment. A completely randomized design was employed under a 3 x 13 x 2 factorial scheme, corresponding to three pitaya species (white pitaya, pitaya hybrid I, pitaya hybrid II), thirteen hydrogen potentials (3.0, 3.5, 4.0, 4.5, 5.0, 5.5, 6.0, 6.5, 7.0, 7.5, 8.0, 8.5, and 9.0) and two fungicide conditions (presence and absence), with four replications. The percentage of germination, germination speed index and mean germination time were evaluated. The data were subjected to ANOVA, and the means were compared using the Scott-Knott test (p < 0.05). The pH influenced the germination speed index in white pitaya. The mean germination time was affected by the pH levels for white pitaya and pitaya hybrid II. The fungicide did not increase the germination percentage in the evaluated species.

Keywords: Hylocereus undatus, H. costaricensis, carboxim+thiran, vigor, viability, physiological quality.

Potencial hidrogeniônico e fungicida na germinação de sementes de pitaya

RESUMO. O trabalho teve como objetivo avaliar a germinação de sementes de pitaya submetidas a diferentes potenciais hidrogeniônicos, com ou sem fungicida. Utilizou-se o delineamento experimental inteiramente casualizado, em esquema fatorial 3 x 13 x 2, correspondente a três espécies de pitaya (pitaya branca, pitaya híbrida I e pitaya híbrida II), treze potenciais hidrogeniônicos (3,0; 3,5; 4,0; 4,5; 5,0; 5,5; 6,0; 6,5; 7,0; 7,5; 8,0; 8,5 e 9,0) e dois níveis de fungicida (presença e ausência), com quatro repetições. Foram avaliados a porcentagem de germinação, o índice de velocidade e o tempo médio de germinação. Os dados foram submetidos à análise de variância e as médias comparadas pelo teste de Scott-Knott (p < 0,05). O pH influenciou o índice de velocidade de germinação da pitaya branca. O tempo médio de germinação foi alterado pelos níveis de pH para a pitaya branca e híbrida II. O fungicida não favoreceu a porcentagem de germinação das espécies avaliadas.

Palavras-chave: Hylocereus undatus, H. costaricensis, carboxim+thiran, vigor, viabilidade, qualidade fisiológica.

Introduction

The family Cactaceae (eudicotiledonea) consists of approximately 200 genera and 2000 species, found mainly in semi-desert areas in warm regions of Latin America (ARRUDA et al., 2005). Pitaya, which means scale fruit, is the name for both the cactus plant and its fruit which are grouped into four genera: *Stenocereus* Briton and Rose, *Cereus* Mill., *Selenicereus* (A. Berger) Riccob and *Hylocereus* Britton and Rose, which is considered by some researchers as the most beautiful member of this family (LE BELLEC et al., 2006; ZEE et al., 2004).

Pitayas are native to tropical and subtropical America and belong to the group of fruit trees considered promising for cultivation. Some decades ago, pitayas were little known, but they now occupy a growing niche in the exotic fruit market (LE BELLEC et al., 2006) and are sought for not

only for their exotic appearance but also for their organoleptic characteristics (ANDRADE et al., 2008a; SILVA et al., 2006). However, pitayas have been known and consumed since pre-Columbian times in their native countries (CRANE; BALERDI, 2005).

Colombia and Mexico are the main producers of these fruits, and *Hylocereus undatus* is the most cultivated species (ANDRADE et al., 2005; NERD et al., 2002). The fruit is eaten traditionally in Mexico, Vietnam, Colombia and Nicaragua (CALVALCANTE et al., 2011). However, the exotic fruit market has grown recently, triggering the interest both of the consumer and fruit grower because of the fruit´s nutritional therapeutic properties and aggregated commercial value (MOREIRA et al., 2011; SILVA et al., 2011). The pitaya is considered a nutritive fruit and can be

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consumed fresh or made into a range of industrial products. Some species are rich in antioxidants, showing medicinal properties that are favorable for cancer and diabetes prevention, in neutralizing toxic substances such as heavy metals and in reducing cholesterol and blood pressure. Additionally, they also contain phosphorus and calcium (GUNASENA et al., 2007).

The mean pitaya yield varies according to the edaphoclimatic conditions, cropping techniques and orchard age involved, ranging from 10 to 30 ton ha⁻¹ (LE BELLEC et al., 2006; VAILLANT et al., 2005). In Brazil, the southeast is the main producing region, especially in the city of Catanduva, São Paulo State, where fruit production occurs from December to May, and there is an annual mean yield of 14 ton ha⁻¹ (BASTOS et al., 2006). There is strong evidence that the central region of Brazil represents the greatest dispersion center for these species (JUNQUEIRA et al., 2010).

Pitayas can be propagated via seeds or plant structures. Their seeds show 83 to 95% viability (ELOBEIDY, 2006; ORTIZ, 2000), and different genetic information can be obtained from them, such as data on diversity, which can be used to select materials with desirable characteristics, for example, related to the yield, external appearance, pulp color and best adaptation to different edaphoclimatic conditions (ANDRADE et al., 2008b; SILVA et al., 2011).

The percentage of germination depends on internal and external factors related to the seeds, but the process will normally be completed if there is no restriction during germination stages (WAGNER JÚNIOR et al., 2007). Several factors are involved in and can influence the germination of seeds of any species, the best studied of which are water, oxygen, temperature, light, the substrate and health. In addition, the pH is a relevant factor in the germination process and seedling development in cultivated species, but this parameter has been little studied.

The Rules for Seed Analysis (BRASIL, 2009) recommend for most species that the pH of the substrate and water should be between 6.0 and 7.5, but there are no studies on this topic for cactus species. Additionally, the influence of the pH on germination has received little attention, although pH values lower than 3.0 and greater than 8.0 have been reported to inhibit the germination process (WAGNER JÚNIOR

et al., 2007). Therefore, assessing the effects of pH on seed germination can supply important information on the resistance of species in acid soils.

Chemical control of seeds is an essential measure for many species that are important in agricultural production, especially for the control of pathogens linked to seeds (MACHADO, 1988). Thus, pitaya seed germination may be influenced by the hydrogen potential and health of the medium.

There are few studies addressing pitaya species, and research is therefore urgently needed to obtain practical information that can be applied to crops under Brazilian edaphoclimatic conditions (CAVALCANTE et al., 2011). The objective of the present study was to evaluate pitaya seed germination under different hydrogen potentials, with or without fungicide treatment.

Material and methods

This study was conducted in the Seed Production and Technology Laboratory at the State University of Londrina (UEL), Paraná State, Brazil, which is located at 23° 23' S and 51° 11' W at a mean altitude of 566 m, in April and May 2012.

Seeds were obtained from ripe fruit of parent plants of *Hylocereus undatus* (white pitaya) and the hybrids *H. undatus* x *H. costaricensis* (pitaya hybrid I) and *H. costaricensis* x *H. undatus* (pitaya hybrid II), cultivated in the experimental area of the Agronomy Department.

At the point of ripenness, the pulp of the fruit was extracted by hand using a spoon and placed in a 2 L beaker with a solution consisting of water (1 L) and sugar (25 g L⁻¹). This mixture was then allowed to rest for 48 hours at room temperature to favor the fermentation process to facilitate seed extraction.

After the resting period, the solution was sieved under running water to eliminate pulp residues and retain the seeds, which were subsequently placed on filter paper and dried in the shade at room temperature for 48 hours.

A completely randomized block design was used under a 3 x 13 x 2 factorial design corresponding to the three pitaya species (white pitaya, pitaya hybrid I and pitaya hybrid II), 13 hydrogen potentials (3.0, 3.5, 4.0, 4.5, 5.0, 5.5, 6.0, 6.5, 7.0, 7.5, 8.0, 8.5, and 9.0) and two

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fungicide conditions (presence and absence), with four replications. Acidic solutions containing hydrochloric acid (HCl) and basic solutions containing sodium hydroxide (NaOH) were used to obtain the desired pH levels, which were prepared with distilled water and measured using a Jenway pH meter, model 3510. A sufficient quantity of each solution was prepared to set up and conduct the experiment and to rewet the blotting paper if there was evaporation from the initial material. The substrate was moistened according to the variation in its absorption capacity, that is, until saturation, without excess.

Fifty seeds were used per treatment, which were placed in plastic boxes (Gerbox® type) lined with blotting paper moistened to 2 ½ times the substrate dry matter content (BRASIL, 2009) with the solutions at the established pH levels. The treatments involving fungicide were applied using Carboxim+Thiram fungicides at a dose of 250 mL 100 kg⁻¹, and seeds were sprayed at the end of plot preparation.

The experiment was conducted in a biochemical oxygen demand chamber under a constant light and temperature of 25°C. Assessments were conducted daily for 12 days, when germination stabilized, to determine the germination percentage (%G), germination speed index (GSI), calculated according to the methodology described by Maguire (1962), and mean germination time in days (MGT), according to Lima et al. (2006). Seeds were considered germinated when they presented a root protrusion equal or greater than 2 mm.

The data were subjected to analysis of variance, and the means were compared using the Scott-Knott test (p < 0.05).

Results and discussion

The triple interaction between the factors species, hydrogen potential and fungicide was not significantly different among the analyzed variables. However, significant differences were observed for the germination percentage (%G) in the species and fungicide interaction factors. The species and hydrogen potential interactions were significant for the variables germination speed index (GSI) and mean germination time (MGT). The hydrogen potential and fungicide interaction was significantly different only for the MGT variable (Table 1).

Table 1. Germination percentage (%G), germination speed index (GSI) and mean germination time in days (MGT) for pitaya seeds subjected to different hydrogen potentials, with or without fungicide treatment. Londrina, UEL, 2012.

Causes of variation	Variables analyzed			
Causes of variation	%G	GSI	MGT	
Species (S)				
White pitaya	98.38	26.39	4.05	
Pitaya hybrid I	98.04	27.52	3.74	
Pitaya hybrid II	86.36	21.21	4.41	
Hydrogen potential (pH)				
3.0	94.75	25.31	3.95	
3.5	93.25	24.43	4.05	
4.0	93.92	24.50	4.16	
4.5	93.82	24.59	4.12	
5.0	94.44	24.47	4.20	
5.5	94.40	25.56	4.06	
6.0	95.58	23.26	4.56	
6.5	95.26	26.67	3.87	
7.0	93.40	26.16	3.79	
7.5	93.31	24.52	4.11	
8.0	94.47	25.68	3.88	
8.5	94.72	25.77	4.00	
9.0	94.07	24.58	4.08	
Fungicide (Fg)				
Presence	93.18	25.60	3.97	
Absence	95.34	24.47	4.16	
Value of F				
Species (S)	360.53*	132.97*	30.34*	
Hydrogen potential (pH)	0.94^{ns}	2.27*	2.28*	
Fungicide (Fg)	26.74*	11.22*	6.71 [*]	
S*pH	$0.50^{\rm ns}$	1.71*	1.55*	
S*Fg	15.37*	0.69^{ns}	$0.94^{\rm ns}$	
pH*Fg	0.69^{ns}	1.57 ^{ns}	1.94*	
S*pH*Fg	1.25 ^{ns}	1.29 ^{ns}	1.14 ^{ns}	
CV (%)	3.90	11.89	15.38	

^{ns}Non-significant, *Significant p < 0.05.

Table 2 shows that the interaction of the factors (species and fungicide) was partitioned for the germination percentage. For the species factor, white pitaya and pitaya hybrid I presented higher means both in the presence and absence of the fungicide. Regarding the fungicide effect, the absence of the fungicide increased the germination percentage of pitaya hybrid II.

Table 2. The partitioning of the species and fungicide interaction regarding the germination percentage of pitaya seeds subjected to different hydrogen potentials, with or without fungicide treatment, Londrina, UEL, 2012.

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Presence	Absence
98.08 A a	98.69 A a
97.82 A a	98.25 A a
83.65 B b	89.07 B a
	98.08 A a 97.82 A a

Means followed by the same uppercase letter in a column for the species effect and lowercase letter in a row for the fungicide effect do not differ by the Scott-Knott test at p < 0.05.

However, in a study using the Carboxim+Thiram fungicide for the control of fungi associated with peanut seeds, Bittencourt et al. (2007) obtained favorable results in terms of increases in the germination percentage and speed as well as seed health in the presence of the fungicide. Pereira et al. (2011) applied a germination test in soybean seeds and verified that

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treatment with Thiabendazole+Thiram or Carbendazin+Thiram increased the germination percentage.

Marini et al. (2011) reported a reduction in the germination rate of wheat seeds under increased doses of a Carboxim+Thiram fungicide, as did Arsego et al. (2006) in studies with irrigated rice seeds. This phenomenon was also observed in the present study for the pitaya hybrid II species, demonstrating the damaging effect of the fungicide on the germination rate. The absence of an effect of the fungicide in the other species can be explained by the high mean germination rates obtained in the assessed seed lots (97-98%), which contributed to abrogating the fungicide effect. Similar findings were reported by Tropaldi et al. (2010), who assessed the physiological quality and health of castor bean seeds treated with Carbendazin and Carboxim+Thiram fungicides and observed that the fungicides did not promote increases in seed germination, with a high percentage of initial germination recorded in the lots (90-95%).

Table 3 shows that the interaction of the factors (species and hydrogen potential) was partitioned for the germination speed index. The white pitaya species presented a lower GSI at pH 4.0, 5.0, and 6.0, while the pitaya hybrid I and hybrid II species did not present significant differences among different pH levels.

Table 3. The partitioning of the interaction of the species and hydrogen potential factors regarding the germination speed index for pitaya seeds subjected to different hydrogen potentials, with or without fungicide treatment. Londrina, UEL, 2012.

	Species		
Hydrogen potential	White pitaya	Pitaya hybrid I	Pitaya hybrid II
3.0	28.55 A a	25.86 A a	21.51 A b
3.5	25.82 A a	26.77 A a	20.70 A b
4.0	24.12 B b	27.98 A a	21.40 A b
4.5	25.92 A a	25.89 A a	21.95 A b
5.0	23.67 B b	27.93 A a	21.81 A b
5.5	27.54 A a	28.52 A a	20.63 A b
6.0	23.18 B b	27.63 A a	18.97 A c
6.5	30.19 A a	28.15 A a	21.68 A b
7.0	27.55 A a	29.10 A a	21.84 A b
7.5	27.35 A a	27.16 A a	19.06 A b
8.0	26.56 A a	27.62 A a	22.84 A b
8.5	26.25 A a	28.11 A a	22.94 A b
9.0	26.38 A a	27.01 A a	20.34 A b

Means followed by the same uppercase letter in a column for the hydrogen potential effect and lowercase letter in the row for the species effect do not differ by the Scott-Knott test at p < 0.05.

An effect of pH on seedling germination and development occurs in extremely acidic or alkaline mediums, and the recommended pH is in the 6.0-7.5 range. This range is ideal for the germination of most plant species (KERBAUY,

2004; LAYNEZ-GARSABALL; MÉNDEZ-NATERA, 2006) and favors biochemical and plant nutrition processes (LARCHER, 2000).

According to Wandscheer et al. (2011), only extremely acidic pH values (lower than three) or extreme alkaline values (over 11) can interfere in the germination process. For example, Therios (1982) observed that sweet almond seeds germinated in a wide pH range, between 2.8 and 8.4, without presenting significant differences in terms of the germination percentage. Similar responses were recorded in the present study for the pitaya hybrid I and pitaya hybrid II species.

Table 4 shows that the interaction of the factors (species and hydrogen potential) was partitioned for the mean germination time. Regarding the effect of the hydrogen potential, pH levels of 4.0, 5.0, 6.0, and 8.5 promoted a longer mean germination time for the white pitaya, while pH levels of 5.5, 6.0, and 7.5 delayed the germination process in pitaya hybrid II. However, the pH levels did not influence pitaya hybrid I with respect to the mean germination time.

Table 4. The partitioning of the interaction of the species and hydrogen potential factors regarding the mean germination time of pitaya seeds subjected to different hydrogen potentials, with or without fungicide treatment. Londrina, UEL, 2012.

Hydrogen potential	Species		
	White pitaya	Pitaya hybrid I	Pitaya hybrid II
3.0	3.67 A a	3.86 A a	4.33 A a
3.5	3.98 A a	3.81 A a	4.35 A a
4.0	4.47 B b	3.63 A a	4.38 A b
4.5	4.00 A a	4.10 A a	4.27 A a
5.0	4.65 B b	3.67 A a	4.28 A b
5.5	3.85 A a	3.65 A a	4.68 B b
6.0	4.80 B b	3.76 A a	5.13 B b
6.5	3.53 A a	3.66 A a	4.42 A b
7.0	3.70 A a	3.51 A a	4.15 A a
7.5	3.78 A a	3.78 A a	4.78 B b
8.0	3.90 A a	3.69 A a	4.05 A a
8.5	4.27 B a	3.65 A a	4.07 A a
9.0	3.99 A a	3.79 A a	4.45 A a

Means followed by the same uppercase letter in a column for the hydrogen potential effect and lowercase letter in a row for the species effect do not differ by the Scott-Knott test at p < 0.05.

Table 5 shows that the interaction of the factors (hydrogen potential and fungicide) was partitioned for the mean germination time. With respect to the hydrogen potential, a pH of 6.0 in the presence of the fungicide resulted in a longer germination time for the seeds, whereas in the absence of the fungicide, there was no significant difference detected. Regarding the fungicide effect, a shorter time was required for germination in the presence of the fungicide at pH levels of 6.5 and 7.5 and in the absence of the fungicide at a pH of 6.0.

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Table 5. The partitioning of the interaction of the hydrogen potential and fungicide factors regarding the mean germination time of pitaya seeds subjected to different hydrogen potentials, with or without fungicide treatment. Londrina, UEL, 2012.

II. 1	Fungicide		
Hydrogen potential	Presence	Absence	
3.0	3.77 A a	4.13 A a	
3.5	3.94 A a	4.15 A a	
4.0	4.15 A a	4.16 A a	
4.5	3.95 A a	4.30 A a	
5.0	4.03 A a	4.36 A a	
5.5	3.91 A a	4.21 A a	
6.0	4.98 B b	4.15 A a	
6.5	3.58 A a	4.16 A b	
7.0	3.68 A a	3.90 A a	
7.5	3.84 A a	4.39 A b	
8.0	3.73 A a	4.03 A a	
8.5	4.02 A a	3.97 A a	
9.0	4.05 A a	4.11 A a	

Means followed by the same uppercase letter in a column for the hydrogen potential effect and lowercase letter in a row for the fungicide effect do not differ by the Scott-Knott test at p < 0.05.

There are few available agronomic data on pitaya management and cropping. Therefore, new research should be conducted to develop techniques to introduce new varieties to increase the cropping area associated with these species in the context of Brazil.

Conclusion

The pH influenced the germination speed index in the white pitaya.

The mean germination time was affected by the pH levels for white pitaya and pitaya hybrid II.

The fungicide did not increase the germination percentage in the evaluated species.

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