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Selection as the best method of genetic improvement in high temperature germinated bell pepper cultivars

Seleção como melhor método de melhoramento genético em cultivares de pimentão germinadas em alta temperatura

La selección como mejor método de mejora genética en cultivares de pimiento germinados a alta temperatura

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Abstract
This work was carried out to estimate genetic parameters for the germination of seeds of four varieties of bell pepper to assist in choosing the most suitable genetic material for regions with high temperatures. The study covered two experiments, each comprising a germination temperature of 35.0 °C and an ambient temperature with an average of 19.6 °C, for the cultivars All big, Amarelo Alegria, Casca dura Ikeda and Yolo wonder. The design used was randomized, with eight replicates composed of 25 seeds each, totaling four treatments with 200 seeds per experiment. The germination percentage of the seeds was evaluated at temperatures of 19.6 °C and 35.0 °C, later used to obtain the genetic parameters. Bell pepper cultivars showed significant genetic variation to the detriment of the environmental one, showing that the genetic factor contributed more than the environmental factor to the studied correlation. Despite the low percentage of germination observed at 35.0 °C, the highest values were obtained for the cultivars All big, and Yolo wonder. The predominance of genetic and phenotypic influence on seed germination to the detriment of environmental impact, even in high temperatures, allows the continuity of bell pepper production in thermal conditions above 30.0 °C, mainly for the cultivars All big and Yolo wonder.
Keywords: *Capsicum annuum*; Correlations; Genetic variability; Genotypes; Selection.

Resumo
Este trabalho foi realizado com o objetivo de estimar parâmetros genéticos para germinação de sementes de quatro variedades de pimentão de forma a auxiliar na escolha do material genético mais apropriado para regiões com elevadas temperaturas. O estudo abrangeu dois experimentos, cada um compreendendo uma temperatura de germinação, 35.0 °C e temperatura ambiente com média de 19,6 °C, para as cultivares de pimentão All big, Amarela

alegria, Casca dura ikeda e Yolo wonder. Utilizou-se o delineamento inteiramente casualizado, com oito repetições compostas por 25 sementes cada, totalizando quatro tratamentos com 200 sementes, por experimento. Foi avaliada a porcentagem de germinação das sementes nas temperaturas de 19,6 e 35.0 °C, posteriormente utilizada para a obtenção dos parâmetros genéticos. As cultivares de pimentão demonstraram grande variação genética em detrimento da ambiental, mostrando que o fator genético contribuiu mais que o ambiental para a correlação estudada. Apesar da baixa porcentagem de germinação observada à 35.0 oC, os maiores valores foram obtidos para as cultivares All big e Yolo wonder. A predominância da influência genética e fenotípica sobre a germinação das sementes em detrimento da influência ambiental, mesmo que na presença de elevada temperatura, possibilita a continuidade da produção de pimentão em condições térmicas acima de 30.0 oC, sobretudo das cultivares All big e Yolo wonder.
Palavras-chave: *Capsicum annuum*; Correlações; Variabilidade genética; Genótipos; Seleção.

Resumen

Este trabajo se realizó con el objetivo de estimar parámetros genéticos para la germinación de semillas de cuatro variedades de chile dulce con el fin de ayudar en la elección del material genético más apropiado para regiones con altas temperaturas. El estudio comprendió dos experimentos, cada uno con una temperatura de germinación de 35,0 °C y una temperatura ambiente promedio de 19,6 °C, para los cultivares de pimiento All big, Amarela alegria, Casca dura ikeda y Yolo wonder. Se utilizó un diseño completamente al azar, con ocho repeticiones de 25 semillas cada una, totalizando cuatro tratamientos con 200 semillas por experimento. Se evaluó el porcentaje de germinación de semillas a temperaturas de 19.6 y 35.0 °C, utilizado posteriormente para obtener los parámetros genéticos. Los cultivares de pimiento morrón mostraron gran variación genética en detrimento del ambiental, mostrando que el factor genético contribuyó más que el ambiental a la correlación estudiada. A pesar del bajo porcentaje de germinación observado a 35,0 oC, los valores más altos se obtuvieron para los cultivares All big y Yolo wonder. El predominio de la influencia genética y fenotípica en la germinación de las semillas en detrimento de la influencia ambiental, incluso en presencia de alta temperatura, permite la continuación de la producción de pimiento en condiciones térmicas superiores a 30,0 oC, especialmente para los cultivares All big y Yolo wonder.
Palabras clave: *Capsicum annuum*; Correlaciones; Variabilidad genética; Genotipos; Selección.

1. Introduction

The bell peppers (*Capsicum annuum* L.) belong to the Solanaceae family. They are among the vegetables with the largest cultivated area in Brazil and the world (Cardozo et al., 2016), where about 732.5 tons of this vegetable were produced in 2018 (Fao, 2019). According to the 2017 Census of Agriculture, conducted by the Brazilian Institute of Geography and Statistics, Bahia is the fourth culture producer, contributing 8.9% of national production.

The temperature has a decisive influence on the germination of seeds of the genus *Capsicum* sp. (Teixeira et al. 2018), which can cause an imbalance in seedling formation processes. As the requirements of this climatic factor vary between cultivars and due to global climate changes and adverse factors that are causing the continuous increase in temperature, it is essential to study the germinative capacity of pepper seeds under different differences from those considered ideal.

Due to the socio-economic relevance of this vegetable, it is necessary to analyze the genetic parameters of pepper cultivars. Thus, aiming at competitiveness in consumer markets, it is essential to infer the genetic variability of cultivars through direct or indirect selection (Li et al., 2016), assisting in decision making.

The search for knowledge of viable conditions for seed germination and the initial development of plants, mainly emphasizing the effects of adverse temperatures, plays a fundamental role in scientific research and provides valuable information on species propagation (Stefanello et al., 2015). According to Dajoz (2005), such knowledge can allow clarifications about changes in the genetics of this culture under the influence of these conditions. Therefore, the selection of cultivars adapted to climatic variations and with desirable agronomic attributes. This is of great importance for the Brazilian Northeast, where most pepper producers, as also other familiar producers, have limited capital and cannot invest in sophisticated production technologies (Nunes et al., 2015).

Studies related to the estimation of genetic parameters for the germination of *C. annuum* at different temperatures are developing. It is still essential because it can establish and characterize cultural management strategies through them, highlighting the region and its climatic conditions. In this context, the study consisted of estimating the genetic parameters for

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the seed germination of four pepper cultivars to assist in choosing the most suitable genetic material for regions with high temperatures.

2. Methodology

The work comprised two experiments, evaluating genetic parameters for germination of four cultivars of All big (Treatment 1), Amarelo Alegria (Treatment 2), Casca dura Ikeda (Treatment 3), and Yolo wonders (Treatment 4), which are among the most commercialized in Brazil, at temperatures of 19.6 °C and 35.0 °C. The experiment was conducted in a completely randomized design, with eight replicates composed of 25 seeds each, totaling four treatments with 200 sources in each experiment. The average ambient temperature (19.6 °C) was obtained using an average of the daily temperatures, which varied between 16.7 °C and 21.1 °C, recorded by the National Institute of Meteorology (Inmet) the period of the experiment that comprised 14 days.

The seeds were placed to germinate on paper towels (Germitest) hydrated with demineralized water, in a volume 2.5 times the dry weight of the paper, and inside transparent plastic boxes called gearboxes. Then, the gearboxes were allocated at room temperature and in a germinator of Biochemical Oxygen Demand (B.O.D.), maintained at a constant temperature of 35.0 °C and a photoperiod of 12 hours.

Regarding germination, the first and second counts were performed at seven and 14 days after the experiment was implanted, in that order, and the percentage of germination at both temperatures was obtained, according to the criteria established by the Regulation of Analysis of Seeds (Brasil, 2009). The data obtained were submitted to the Lilliefors Normality, Cochran's Homogeneity, and analysis of variance tests. The means between cultivars, when significant, were compared using the Tukey test ($p < 0.05$) for each temperature, and the t-test ($p < 0.05$) was used to verify the influence of temperature on the germination percentage of each cultivar. The information from the analysis of variance was used to estimate genetic, phenotypic, and environmental variances, according to equations 1, 2, and 3, respectively.

$Vg = [(QMG – QMR)/r]$ Equation 1

$Vp = [QMG/r]$ Equation 2

$Ve = [QMR/r]$ Equation 3

Where: Vg is the genetic variance, QMG is the average square of the genotype, QMR is the middle square of the residue, r is the number of repetitions, Vp is the phenotypic variance, and Ve is the environmental variance. Subsequently, the genetic, phenotypic, and ecological variation coefficients were calculated using equations 4, 5, and 6.

$CVg = [(\sqrt{Vg}/\bar{x})*100]$ Equation 4

$CVp = [(\sqrt{Vp}/\bar{x})*100]$ Equation 5

$CVe = [(\sqrt{Ve}/\bar{x})*100]$ Equation 6

Being: CVg: coefficient of the genetic variation, Vg the genetic variance, $\bar{x}$ the average of the evaluated character, CVP the coefficient of phenotypic variation, Vp the phenotypic variance, CVe the coefficient of environmental variation, and Ve the ecological variation. The calculation of the variation index, also called quotient "b" or the relative coefficient of variation (CVR), was obtained by the ratio between the coefficients of genetic and environmental variation (CVg / CVe). According to equation 7, heritability was obtained by equation 8 and genetic gain as a percentage of the mean by equation 9.

$h2a (\%)= [(Vg/Vp)*100]$ Equation 7

$GA (\%)= [(i * \Delta p * h2a]$ Equation 8

GAM (%)= [(GA/$\bar{x}$) * 100] Equation 9

Where: h2a (%) is heritability in percentage in the broad sense, Vg the genetic variation, Vp the phenotypic variation, GA (%) the genetic gain, ia selection intensity (5%) = 2.06, Δpo standard deviation of the phenotypic variance = √("Ve"), and GAM (%) the genetic gain in the percentage of the average.

The correlation coefficients of genetic (rg), phenotypic (rp) and environmental (re) between the characters germination speed index and germination rate were estimated by equations 10, 11 and 12:

rg = [COVpxy/√("σ" ^"2" " px*" "σ" ^"2" "py")] Equation 10

rp= [PMpxy /√(" QMpx*QMpy")] Equation 11

re = [PMpxy /√(" QMrx*QMry")] Equation 12

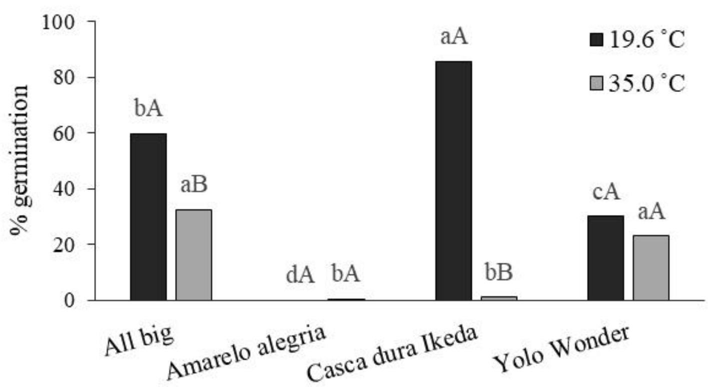
In these expressions, COVpxy refers to the genetic covariance of progenies between characteristics X and Y; px and py, for the genetic variances of progenies of the characters X and Y, respectively; PMp and PMR, for medium progeny and residual products; and QMp and QMr, for the mean squares of progenies and residues, respectively.

To verify the accuracy of the experiment, the accuracy of the selection of the pepper seed germination variables was calculated. For this purpose, the equation Exactness = (h^2a)1/2 was used, where h^2a is the heritability.

3. Results and Discussion

The germination percentage of the cultivars at room temperature varied by more than 80.0% and above 30.0% at 35.0 ºC. In the first experiment, the conditions in which the seeds were submitted favor the germination of the cultivar Casca dura Ikeda. In no second experiment, the highest germination percentage was observed for Yolo wonder and All big. The yellow pepper did not germinate at room temperature during the 14 days, which should be further studied to characterize the time required for the germination of this genotype under these conditions. When submitted to 35.0 ºC, the seeds of this cultivar germinated, however insignificantly (Figure 1).

Figure 1. Germination percentage (% germination) at 19.6 ºC and 35.0 ºC for four pepper cultivars. The same lower case letters do not differ by Tukey's test at p <0.05 at each temperature. Equal capital letters do not differ by t test <0.05 for each cultivar.



Source: Authors.

Although the cultivars All big and Yolo wonder have the highest values of germination percentage (between 23 and 33%) at 35.0 oC, such results are still considered low. Teixeira et al. (2018), studying the germination of peppers (Capsicum frutescens) at temperatures of 15, 25, and 35.0 oC, observed that soaking the seeds is hindered by the higher temperature through the deterioration of the membranes. This condition was evidenced by the high electrical conductivity obtained by this treatment in 24 hours (79.6 µS cm-1 g-1 seed), a value 35.7% higher than that observed at 25 oC. Because of the relatively close results of germination percentage at 35.0 oC obtained in the present study with those followed by these authors (53%), it can be inferred that high temperatures negatively influence the germination of seeds of pepper cultivars. Thus, initial studies of genetic improvement in adverse thermal conditions become relevant.

However, pepper cultivars showed high genetic variation at the expense of environmental variation (Table 1). Such results indicate that the environment had little influence on the evaluated characteristics and high genetic variability in the studied characters.

Table 1. Estimation of genetic parameters, genetic variation (V_g), environmental variation (V_e), phenotypic variation (V_p), genetic variation coefficient (CV_g), environmental variation coefficient (CV_e), phenotypic variation coefficient (CV_p), variation coefficient relative (CVr), heritability (h^2a%), gain by selection (GA), average gain by selection (GAM) for four pepper cultivars.

Estimation of genetic parameters	Germination (%)	
	---19.6 ºC---	---35.0 ºC---
V_g	135.07.55	247.03
V_e	7.20	11.05
V_p	1364.75	258.08
CV_g (%)	84.22	110.30
CV_e (%)	6.13	23.33
CV_p (%)	84.44	112.74
CVr	13.73	4.73
h^2a (%)	99.47	95.72
GA	7570.03	3167.65
GAM (%)	17302.93	22229.09
Accuracy	0.997	0.978

Source: Authors.

Differences in behavior were observed in terms of genetic variability with a variation of 1110.5% between temperatures, the lowest result being obtained at 35.0 ºC. Comparing the Vg parameters with the Ve, the genetic variation was more significant than the environmental variation in the two thermal conditions. Other studies with peppers have demonstrated the predominance of the genetic factor over the environment for the species, such as that by Jogi et al. (2015) on the genetic variability of peppers for yield and quality attributes. These results are of great importance for the producers of this vegetable, as they signal the possibility of continuing activities, given the increase in the environment's temperature.

The high values of the coefficient of genetic variation theoretically demonstrate that the existing genetic variability contributes significantly to the phenotypic expression of the characters (Silva et al., 2014). The high values of the coefficient of phenotypic variation are also desirable since CVP is one of the efficient ways of choosing genotypes to demonstrate the plant's genetic potential, primarily if this coefficient reflects the non-environmental element.

The low levels of the coefficient of environmental variation (CVe) presented by pepper cultivars for % germination at both temperatures reveal that the environmental factor minor influences the expression of this character. This becomes important because, according to González-Jara et al. (2011), the domestication of species can reduce their genetic variability, which did not occur for the cultivars studied, considering the target characteristics of this work.

Therefore, from the results obtained above and in conjunction with the CVg facts having been higher than CVe and CVP having also reached expressive values, it can be said that the best method of genetic improvement to be used in the studied genotypes corresponds to the selection. Such information is of excellent relevance o it indicates that even in unfavorable temperature situations (35.0 ºC), there is the existence of genetic variability in the material studied, enabling the application of one of the relatively simple methods of genetic improvement.

The coefficient of relative variation (CVR) was more significant than 1 for both temperatures. Such results reinforce the indication that the genetic variability between materials and selection occurs according to the breeding method indicated for the cultivars All big, Amarelo Alegria, Casca dura Ikeda and Yolo wonder, o it demonstrates that the environmental variation is less than the genetic variation concerning the average (Cunha and Lima, 2010).

The h^2a estimate, according to Thair (2011), was considered high, being above 99.0% at 19.6 ºC and more excellent than 95.0% for germination% at 35.0 ºC. Studies like Jogi et al. (2015) also reveal the propensity of peppers to present characteristics with high heritability since this condition, in the authors' work, occurred for the number of fruits per plant in the first harvest (98.2%); the total number of fruits per plant (95.6%); total yield (91.4%); fruit length (91.2%); fruit width (96.2%); stem length (81.0%) and chlorophylls a (95.5%), b (97.5%) and total (97.9%).

Heritability assumes immense relevance for the selection of cultivars. The high values found in the study indicate that the choice of superior genotypes has a high probability of expression of genetic characteristics in the next generation (Rashwan 2010). Aruah et al. (2012) state that high heritability in a broad sense indicates minimal environmental influence on phenotypic expressions. Therefore, from the development of the plants, it is possible to observe the progenies' behavior and select those that present the best characteristics of interest. The ability to maintain germinative power over generations by pepper seeds at high temperatures is a condition desired by the agricultural sector.

According to the classification proposed by Johnson et al. (1955), a high genetic gain was obtained for the germination percentages, and the calculated values were 7570.03 at room temperature and 3167.65 at 35.0 ºC. Earning by selecting selection guide improvement programs, predicting your success, choosing or discarding groups, and focusing your efforts on indicating the most important characters and making potential (Cruz et al., 2012). Thus, given the % germination for pepper reproduction, this study suggests a probable success in works of genetic improvement of the species with the possibility of improving the production of *C.annuum* seedlings.

Despite the relevance of the GA parameter, Cruz et al. (2012) consider the GAM to be more important because it represents a percentage gain, which allows observing if the selection by the characteristic of interest was efficient. Thus, the high average gain per selection observed at 35.0 ºC is a result of great interest in the production of pepper, o it represents the possibility of lower losses with non-germinated seeds.

The relationship between high heritability and high genetic gain indicates, according to Manggoel et al. (2012), a probable predominance of additive gene action. For this study, direct phenotypic selection for the evaluated characteristics is suggested since phenotypes are being expressed according to Janaki et al. (2015).

The genetic (rg), phenotypic (RP), and environmental (re) correlation coefficients found were, respectively, 0.5623, 0.4996, and 0.0455. Knowledge about correlation is of excellent importance o it demonstrates how the selection of a character interferes with the expression of other characters. Inbreeding programs, generally seeking to improve the main character, also seek to maintain or enhance the presentation of other characters simultaneously (Lopes et al., 2014).

The genetic correlation was more significant than the phenotypic, both of the same sign, showing that the genetic factor contributed more than the environmental factor to the studied correlation. This result suggests that a natural selection of the evaluated character will allow simultaneous genetic gains (Cruz et al., 2014). The low phenotypic value may be due to the interaction of genotypes with the environment.

The selective accuracy found in this study is over 97%, which according to Resende and Duarte (2007), is classified as very high. Such results indicate a close relationship between the actual and estimated values of the genetic parameters for the germination of pepper seeds at high temperatures (Pimentel et al., 2014). Thus, combined with the high heritability obtained, there is significant reliability in the possibility of making genetic improvement through selection, getting high genetic gains for pepper cultivars in adverse conditions such as temperatures of 35.0 ºC.

4. Conclusions

Despite the low percentage of germination observed at 35.0 ºC, the highest values were obtained for the cultivars All big, and Yolo wonder.

Pepper cultivars have high genetic variability for seed germination, and the high genetic gain makes it possible to improve the studied cultivars under the conditions evaluated through natural selection.

The predominance of genetic and phenotypic influence on seed germination to the detriment of the environmental effect, even in high temperatures, allows the production of pepper to continue under thermal conditions above 30.0 ºC.

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