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ELECTRICAL CONDUCTIVITY TEST FOR EVALUATION OF THE PEA SEED VIGOR

Edvan Costa da Silva[1*], Wagner Menechini[1], Carolina dos Santos Galvão[2],
Luís Augusto Batista de Oliveira[2], Natália Cássia de Faria Ferreira[3], Luciana Sabini da Silva[1]

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ABSTRACT - The electrical conductivity test is routinely used to differentiate the physiological quality of seed lots, and has stood out for its ease of execution, low cost, speed, repeatability, and straightforward interpretation of results. This study aimed to establish a methodology for the electrical conductivity test in pea seeds (*Pisum sativum* L.) involving seed quantity and seed imbibition periods. The experiment was carried out at the Multidisciplinary Laboratory of the State University of Goiás, the campus of Ipameri Campus, in 2018. Pea seeds (*Pisum sativum*) stored in the seed laboratory of the same institution were used for this experiment. The completely randomized experimental design with four replications in a 4x6 factorial scheme was used. Four quantity of seeds (25, 50, 75, and 100 seeds) and six imbibition periods (4, 8, 12, 16, 20, and 24 h) at 25ºC were evaluated. The electrical conductivity test was performed with four replicates of 25, 50, 75, and 100 seeds each, weighed on a precision scale (0.001g) and placed in disposable plastic cups, with a capacity of 200 mL, containing 75 mL of distilled water. The cups were then kept in a BOD chamber, set at 25°C, and the measuring was performed after 4, 8, 12, 16, 20, and 24 h of imbibition with a conductivity meter and results were expressed in μS cm^{-1} g^{-1}. There were no significant results for the interaction between the number of seeds and the imbibition time. Significant results were observed only for the individual effects of each factor. It is recommended to use 100 seeds of Maria pea, and 24 h for the electrical conductivity test.
Keywords: *Pisum sativum* L., conductivity meter, physiological potential.

TESTE DE CONDUTIVIDADE ELETRICA PARA AVALAIÇÃO DO VIGOR DE SEMENTES DE ERVILHA

RESUMO - O teste de condutividade elétrica é rotineiramente utilizado para diferenciar a qualidade fisiológica de lotes de sementes, e tem se destacado por sua facilidade de execução, baixo custo, rapidez, repetibilidade e fácil interpretação de resultados. O presente trabalho teve como objetivo estabelecer metodologia para o teste de condutividade elétrica em sementes de ervilha (*Pisum sativum* L.) envolvendo quantidade de sementes e períodos de embebição das sementes. O experimento foi conduzido no Laboratório Multidisciplinar da Universidade Estadual de Goiás (UEG), *Campus* Ipameri, em 2018. Foram utilizadas sementes de ervilha Maria, armazenadas no Laboratório de Sementes da mesma instituição. O delineamento experimental utilizado foi inteiramente casualizado, em esquema fatorial 4x6, constituído por quantidade de sementes (25, 50, 75 e 100 sementes) e períodos de embebição (4, 8, 12, 16, 20 e 24 h) a 25ºC, com 4 repetições. O teste de condutividade elétrica foi realizado com quatro repetições de 25, 50, 75 e 100 sementes cada, pesadas com precisão de 0,001 g e acondicionadas em copos de plástico descartáveis, com capacidade para 200 mL, contendo 75 mL de água destilada. Em seguida, os copos permaneceram em câmara BOD, regulada a 25°C, e a leitura foi realizada após 4, 8, 12, 16, 20 e 24 h de embebição, em condutivímetro, com resultados expressos em μS cm^{-1} g^{-1}. Não houve resultado significativo para a interação entre o número de sementes e o tempo de embebição, com resultado significativo apenas ao analisar o efeito individual de cada fator. Recomenda-se a utilização de 100 sementes de ervilha Maria, no período de 24 h para o teste de condutividade elétrica.
Palavras-chave: *Pisum sativum* L., condutivímetro, potencial fisiológico.

INTRODUCTION

The pea (*Pisum sativum* L., Fabaceae family) has its origin in the Middle East, and it is consumed all over the world as a vegetable and can be consumed as canned grains or as green grains. Until the mid-eighties in Brazil, this vegetable was practically imported, whereas, at present, all demand can be met by national production (IAC, 2011). Green pea is one of the consumers' choices, which is why this crop has become a production option to

[1]Ph.D. student(s) in Agronomy, State University of Western Paraná (Unioeste), 1777, Pernambuco street, Center, Zip-code 85960-000, Campus Marechal Cândido Rondon, Paraná, Brazil. E-mail: edvan_costa@outlook.com; wmmenechini@hotmail.com; luciana.sabini@hotmail.com. * Corresponding author.
[2]M.Sc. in Plant Production, State University of Goiás (UEG), Campus of Ipameri, Zip-code 75780-000. Ipameri, Goiás, Brazil. E-mail: carolgallvao@hotmail.com; luisaugusto-1993@hotmail.com.
[3]Ph.D. student, Graduate Program in Forest Sciences, University of Brasilia (UnB), Darcy Ribeiro University Campus, Asa Norte, Brasilia, Distrito Federal, Brazil. E-mail: natcassiadefaria@gmail.com.

respond to market demand, especially frozen products (EMBRAPA, 2012).

One of the main characteristics of physiological quality to be pointed out when planting a crop is the vigor of the seeds. The use of high vigor seeds is essential for all crops, to enable an appropriate plant population over a wide range of environmental field conditions encountered during emergence, and to provide an increase in production when the plant density is lower than requested (SCHEEREN et al., 2010).

Despite the effects of seed vigor on seedling emergence and initial seedling development, under unfavorable environmental conditions, these are well documented in the literature. However, the results obtained are not conclusive to the extent that these effects extend to the most advanced phenological stages and affect crop production. The results achieved on germination performed in laboratories usually approach the percentage of seedling emergence in the field under satisfactory environmental conditions after sowing; however, it is usually not determined when these circumstances reorient the most satisfactory circumstances (MIELEZRSKI, FILHO, 2012).

Of all the existing physiological quality tests, one of the most used is the electrical conductivity test, which has stood out for its simplicity of execution, repeatability, speed, easy interpretation of results, and low cost. It is based on the cause that cell membranes of the seeds are the last to be arranged during the maturation process. However, they are the first with signs of deterioration after the seeds reach the physiological maturity stage. During imbibition with dry seeds, rapid and intense electrolyte leaching occurs when the seeds come into contact with the water being balanced to the state of disorganization of the membranes, followed by a decrease in the loss of solutes, as the tissues are rehydrated until reaching stability (MACHADO et al., 2011).

The electrical conductivity test has achieved satisfactory results in the separation of seed lots with different levels of vigor for some crops, such as pepper (VIDIGAL et al., 2008), cubiu (PEREIRA and MARTINS FILHO, 2012), black oats (MENEZES et al. 2007, NOGUEIRA et al., 2013) and ryegrass (LOPES and FRANKE, 2010). Given the above, this study aimed to establish a methodology for testing electrical conductivity in pea seeds (*Pisum sativum* L.), involving the number of seeds and imbibition periods.

MATERIAL AND METHODS

The experiment was carried out at the Multidisciplinary Laboratory of the Universidade Estadual de Goiás (the State University of Goiás), Campus Ipameri, in 2018. Maria pea seeds (*Pisum sativum* L.) were used, stored in the Seed Laboratory of the same institution. The initial quality of the seeds was characterized (water content, first germination count, germination, accelerated aging, and 1000-seed weight).

The oven method at 105 ± 3°C was used to determine the water content according to the methodology prescribed in the Regras para Análise de Sementes - RAS (Rules for Seed Analysis) (BRASIL, 2009). Two sub-samples of 20 seeds were taken per treatment. The oven was first regulated at a temperature of 105 ± 3°C, and the containers were dried for 30 minutes, then weighed on a precision scale (0.001g) with their respective lids and then with the seeds. Samples remained in oven for 24 h, after this period the samples were removed and kept in a desiccator to reduce the temperature and they were weighed. The results were expressed in % of water content (wet basis). Germination test was conducted with four replications of 50 seeds per treatment, the seeds were placed on paper towels moistened with distilled water in the proportion of 2.5 times the weight of the dry paper and placed to germinate in B.O.D. at a constant temperature of 25°C, computing the percentage of normal seedlings on the seventh day. The evaluation was carried out following the RAS (BRASIL, 2009). The results were expressed as a percentage of normal seedlings per treatment (%). The first counting of germination test was conducted together with the germination test, computing the percentage of normal seedlings on the fourth day after the start of the test installation, and the results expressed as a percentage of normal seedlings (%).

For the 1000 -seed weight, eight replications of 100 seeds were used, which were counted manually and weighed on a precision scale (0.001 g), according to the RAS (BRASIL, 2009). For the accelerated aging test, 200 seeds were distributed on an aluminum screen, fixed in a gerbox-type plastic box, containing 40 mL of distilled water at the bottom. Boxes with seeds were closed with a lid and kept in B.O.D. at a constant temperature of 42ºC for 48 h. At the end of the accelerated aging test, the seeds were submitted to the germination test, and evaluation of the percentage of normal seedlings was performed on the 4th day after the start of the test. The results were expressed as a percentage of normal seedlings (%).

In the electrical conductivity test, four repetitions of 25, 50, 75, and 100 seeds each were used, weighed on a precision scale (0.001 g) and placed in disposable plastic cups (200 mL capacity), containing 75 mL distilled water. Then, the cups remained in a BOD chamber, regulated at 25° C, where the measuring (in conductivity meter - DIGIMED DM 31) was performed after 4, 8, 12, 16, 20, and 24 h of imbibition, the results being expressed in $\mu S\ cm^{-1}\ g^{-1}$.

The completely randomized experimental design with four replications in a 4x6 factorial scheme was used. Four quantity of seeds (25, 50, 75, and 100 seeds) and six imbibition periods (4, 8, 12, 16, 20, and 24 h) at 25ºC were evaluated. The data were subjected to analysis of variance, and the means were compared by the Tukey test at 5% probability. Also, the data were submitted to the regression analysis. The software Statistica (FERREIRA, 2011) was used in the analyses.

RESULTS AND DISCUSSION

The initial moisture content of the pea seeds was 13.8% (Table 1), thus not adjusting the initial water content, staying within the recommended limits for the evaluation of the electrical conductivity of common pea seeds, which would be between 10 to 14% (ISTA, 2006). The seeds also showed a good percentage for the first germination count and germination test, with 68 and 97%, respectively (Table 1). These results are corroborated by Machado et al. (2011), who evaluated ten lots of peas, and they observed in the best lot had 66% in the first germination count and from 97 to 100% in the germination teste.

For the accelerated aging test, 92% germination was obtained (Table 1). For 1000-seed weight, the value of 149.04 g was obtained (Table 1), which is also corroborated by the work of Machado et al. (2011), which for the best lots obtained values from 149.04 to 150.24 g.

TABLE 1 - Average data on water content (WC), germination test (GT), first count of germination test (FC), accelerated aging (AA), and 1000-seed weight (W1000) of Maria pea.

WC (%)	FC (%)	GT (%)	AA (%)	W1000 (g)
13.8	68	97	92	149.04

According to Table 2, there was no significant result for the interaction between the number of seeds and the imbibition time, with a significant result only for the individual effect of each factor.

TABLE 2 - Summary of analysis of variance for the interaction between the number of seeds and imbibition periods.

	DF	SS	SM	FC	p
Number of seeds (NS)	3	15056.9	5019.0	138.411	0.0000*
Periods of imbibition (PI)	5	4585.0	917.0	25.289	0.0000*
NS*PI	15	665.9	44.4	1.224	0.27449^{ns}
Error	72	2610.8	36,3		
Total	95	22918.6			

*Significant at 5% probability. ns = not significant.

There was a significant difference between treatments, with the treatment of 100 seeds (68.62) being the most sensitive to detect differences in seed vigor (Table 2a). Lopes et al. (2010) found that the number of ryegrass seeds did not provide significant differences in electrical conductivity. The smallest number of seeds saved time and material used in the test. Ataíde et al. (2012) also found that for the electrical conductivity test, the amounts of peanut seeds do not cause significant differences, only when the factors, imbibition time and water volume are combined (interaction between factors).

TABLE 2a - Average data for the electrical conductivity of Maria pea according to the number of seeds.

Number of seeds	Averages
25	33.17 a*
50	50.74 b
75	52.67 b
100	68.52 c

*Means followed by the same letter in the column do not differ by Tukey's test, at 5% probability.

There was a significant difference between treatments, with the 24 h treatment (61.61) being the most sensitive to detect differences in seed vigor (Table 2b) and the treatment with 4 h is the least sensitive.

TABLE 2b - Average data for electrical conductivity in pea seeds according to the imbibition period.

Imbibition period of seeds	Averages
4	39.84 a*
8	46.32 b
12	50.46 bc
16	54.49 c
20	54.94 c
24	61.61 d

*Means followed by the same letter in the column do not differ by Tukey’s test, at 5% probability.

These results are confirmed by an increase in the number of electrolytes released by pea according to the number of seeds and periods of imbibition (Figures 1A and 1B). It can be seen that the data adjusted to the increasing linear regression of 67.47 $\mu S\ cm^{-1}\ g^{-1}$ with 100 seeds, 48% higher than the electrical conductivity observed for 25 seeds (35.39 $\mu S\ cm^{-1}\ g^{-1}$) (Figure 1A). The data also adjusted to the increasing linear regression for the period of imbibition of the seeds (Figure 1B), of 61.12 $\mu S\ cm^{-1}\ g^{-1}$

in 24 h, being 32.22% higher than the electrical conductivity registered for 4 h (41.43 µS cm^{-1} g^{-1}).

These results are similar to the studies developed by Machado et al. (2011), Araújo et al. (2011), and Sponchiado et al. (2014). They also found an increase in electrical conductivity with imbibition time for forage pea, jatropha, and white oat seeds. Silva et al. (2013) observed a linear relationship between the number of electrolytes because of the time of immersion of the bean seeds in water, a fact that has little influence on the separation of treatments. Pinto et al. (2016) found an increase in conductivity with the time of imbibition, where, after a certain period, the cells are reorganized, and repair of damage to the membranes of the seeds occurs, with stabilization of the release of exudates.

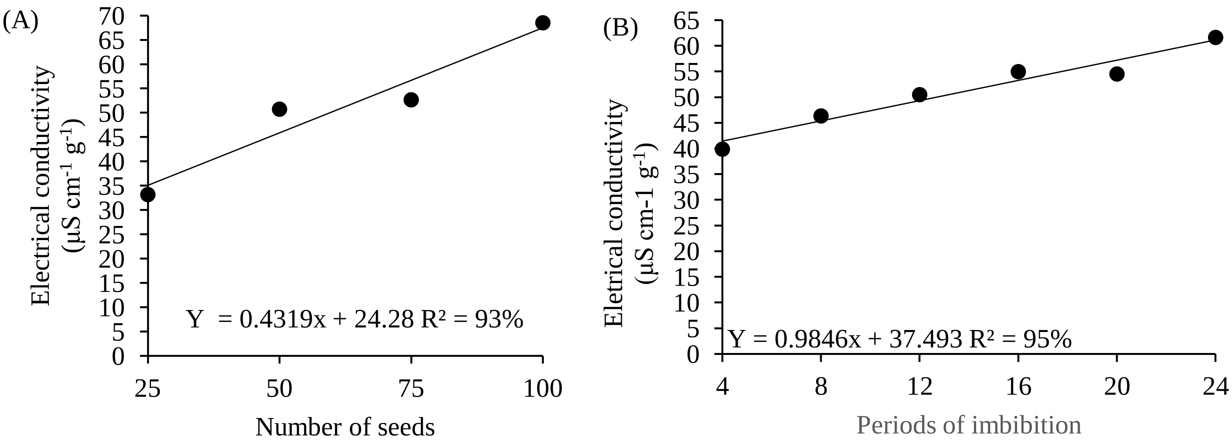

FIGURE 1 - Electrical conductivity according to the number of seeds (1A) and the period of imbibition (1B).

Torres et al. (2009), aimed to establish a methodology for the test of electrical conductivity in sesame seeds, using two periods of seed imbibition (8 and 24 h). They concluded that the period of 8 h of seed imbibition was enough to differentiate the physiological potential of the seed lots, which reduces the period of seed imbibition that usually is recommended (24 h).

The use of larger amounts of seeds allows better differentiation between lots, as it presents lower values of variation coefficient (ATAÍDE et al., 2012). However, a balance must be found between the number of seeds that allow a lower variation coefficient and lower demand for them. Machado et al. (2011) recommend 250 mL of water at a temperature of 25ºC for 24 h, to differentiate lots of pea seeds (*Pisum sativum* subsp. Arvense), whereas, Ataíde et al. (2012) recommend 50 seeds soaked in 50 mL of distilled water during the 24 h period.

Results evidenced the electrical conductivity test for pea seeds can be carried out using less amount of distilled water (75 mL), a fact of paramount importance for seed quality programs, where information about seed vigor is essential and aims to avoid waste of materials of the test.

CONCLUSION

It is recommended to use 100 seeds of Maria pea, and 24 h for the electrical conductivity test.

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