Journal of Seed Science, v.41, n.2, p.187-195, 2019 http://dx.doi.org/10.1590/2317-1545v41n2211401

Assessing the physiological quality of common bean seeds using the Vigor-S® system and its relation to the accelerated aging test¹

André Dantas de Medeiros^{2*}, Laércio Junio da Silva², Nayara Pereira Capobiango², Camila Andrade Fialho², Denise Cunha Fernandes dos Santos Dias²

ABSTRACT – Automated analysis of seed vigor stands out by allowing greater accuracy, standardization, objectivity, and speed in evaluation of the physiological potential of seed lots. The objective of this study was to evaluate the efficiency of the Vigor-S® system in assessing the physiological quality of common bean seeds compared to the information provided by the traditional vigor tests recommended for this species. Four genotypes of common bean were used, each one represented by four seed lots. Characterization of the physiological potential of the lots was carried out by the following tests: germination, first count of germination, seedling emergence, accelerated aging, and electrical conductivity. The results of these tests were compared with the data obtained from the image analysis technique, specifically the Vigor-S® system, which was used to evaluate seedling growth at two, three, and four days after the beginning of the germination test. Shoot length, primary root length, and seedling length were measured, as well as the growth index, uniformity index, and vigor index were calculated. Computerized analysis of seedling images using the Vigor-S® software is a reliable alternative for evaluation the physiological potential of bean seeds, and it produces information similar to evaluations traditionally used for that purpose.

Index terms: Phaseolus vulgaris L., seed vigor, software, seed technology, image analysis.

Avaliação da qualidade fisiológica de sementes de feijão por meio do sistema Vigor- $S^{\text{\tiny ®}}$ e sua relação com o teste de envelhecimento acelerado

RESUMO – A análise automatizada do vigor de sementes tem se destacado por permitir maior precisão, padronização, objetividade e velocidade na avaliação do potencial fisiológico de lotes de sementes. O presente trabalho teve como objetivo avaliar a eficiência do sistema Vigor-S® na determinação do potencial fisiológico de sementes de feijão comum, em comparação às informações fornecidas por testes de vigor recomendados para essa espécie. Foram utilizados quatro genótipos de feijão comum, cada um representado por quatro lotes. A caracterização do potencial fisiológico dos lotes foi realizada por meio dos testes de germinação, primeira contagem de germinação, emergência de plântulas, envelhecimento acelerado e condutividade elétrica. Os resultados desses testes foram comparados com os dados obtidos com o uso da técnica de análise de imagem, por meio do software Vigor-S®, que foi utilizado para avaliar o crescimento das plântulas aos dois, três e quatro dias após o início do teste de germinação, mediante a obtenção de valores de comprimento da parte aérea, raiz primária, plântula, e os índices de crescimento, uniformidade e vigor. A análise computadorizada de imagens de plântulas utilizando o software Vigor-S® é eficiente para a avaliar o potencial fisiológico de sementes de feijão, de forma similar às avaliações tradicionalmente utilizadas para esta finalidade.

Termos para indexação: Phaseolus vulgaris L., vigor de sementes, software, tecnologia de sementes, análise de imagens.

Introduction

Over the years, research in seed physiology and technology has contributed to the development of valuable tools for production of high-quality seeds. Thus, an important instrument to provide innovative alternatives and solutions in modern agriculture is the one that evaluates the seed vigor precisely.

Vigor tests are efficient to identify less advanced stages

¹Submitted on 11/08/2018. Accepted for publication on 12/17/2018.

²Departamento de Fitotecnia, Universidade Federal de Viçosa, 36570-900 – Viçosa, MG, Brasil.

^{*}Corresponding author < medeiros.seeds@gmail.com>

of seed deterioration and contribute to facilitate decisionmaking in seed production companies (Wendt et al., 2017), especially in establishing storage, commercialization, and quality control policies (Marcos-Filho et al., 2009).

Recently, the advent of computerized image processing has allowed quantification of various characteristics linked to seed physiological quality (Marques et al., 2015). In this respect, the use of this technology has advanced in the procedures used in assessment of seed vigor, mainly based on seedling performance, mitigating errors arising from subjective analyses.

Seedling image analysis is generally made by spatial processing, where the position and the color of the pixels are analyzed, using extraction algorithms for this purpose (Brunes et al., 2016). The technique stands out through its greater accuracy, standardization, objectivity, and speed in obtaining consistent results (Castan et al., 2018).

The need for optimizing these procedures through the use of new computational resources to refine analysis lead the development of specific systems for vigor evaluation of the seeds of different species (Marcos-Filho, 2015). Among the systems available for this purpose is Vigor-S[®], developed by the Universidade de São Paulo (USP/ESALQ) in partnership with EMBRAPA/Instrumentação Agropecuária [Crop and Livestock Instrumentation] (CNPDIA). Vigor-S® is based on evaluation of seed vigor from seedling performance through acquisition of the primary root length, hypocotyl length, and total seedling length data, as well as the root/ hypocotyl ratio and parameters based on growth rate and uniformity. An additional important advantage is that the Vigor-S® system is free for general use by private seed companies, by seed laboratories, and for academic purposes (Castan et al., 2018).

The use of software that employs the seedling image analysis technique to make inferences regarding seed quality has been studied and recommended for various species, such as squash (Silva et al., 2017), onion (Gonçalves et al., 2017), carrot (Marchi and Cicero, 2017), crambe (Leão-Araújo et al., 2017), soybean (Medeiros and Pereira, 2018; Wendt et al., 2017), common bean (Gomes-Junior et al., 2014), and maize (Castan et al., 2018), among others. However, there is not yet information that shows the effectiveness of Vigor-S® for evaluating common bean seed vigor (*Phaseolus vulgaris* L.).

Thus, the aim of the present study was to evaluate the effectiveness of the Vigor-S® system for assessment of the physiological quality of common bean seed, compared to the information provided by traditional vigor tests recommended for this species.

Material and Methods

The study was conducted at the Seed Analysis Laboratory of the Plant Science Department of the *Universidade Federal de Viçosa* in Viçosa, MG, Brazil. Three cultivars (BRS Esteio, BRS Madre Pérola, and Manteigão Fosco 11) and one elite line (VC 25) of common bean were used. The seeds were produced in Viçosa. Each genotype was initially represented by one seed lot, which first passed through aging (method described in the accelerated aging test with adjustment of the exposure time) to obtain three more lots. Therefore, four lots of each genotype were evaluated: unaged lot (L1, L5, L9, and L13), lot aged for 12 hours (L2, L6, L10, and L14), lot aged for 24 hours (L3, L7, L11, and L15), and lot aged for 36 hours (L4, L8, L12, and L16). After aging they were dried in a laboratory oven for 48 hours. The lots were characterized by the tests described as follows.

Seed moisture content: determined by the laboratory oven method at 105 °C (±3 °C) for 24 hours. Two 50 g seed samples were used for each lot. The results were expressed in mean percentage (wet basis), according to Brasil (2009).

Germination: conducted with four replications of 50 seeds per lot in rolls of paper toweling moistened with 2.5 times the dry weight of the paper and kept in a germinator at constant temperature of 25 °C. Final count was made on the ninth day after sowing (Brasil, 2009) and the results were expressed in mean percentage of normal seedlings per lot.

First count of germination: conducted jointly with the germination test. The percentage of normal seedlings obtained on the fifth day after sowing was computed (Brasil, 2009).

Seedling emergence: performed with four 50 seed replications, which were sown in multiple cell expanded-polystyrene trays. One seed was sown per cell. The trays were filled with washed sand, irrigated daily, and kept in a greenhouse. The number of emerged seedlings were evaluated daily until there was no more increase in the number of normal seedlings. The result was expressed in percentage of emerged seedlings.

Accelerated aging: conducted with four replications of 50 seeds per lot, with 220 seeds placed on a stainless steel screen in the upper third of a gerbox plastic box (11 x 11 x 3.5 cm) containing 40 mL of water. After the lid was closed, the boxes were kept in a B.O.D. chamber, regulated at 41 °C for 48 hours. After that, the germination test was performed and the percentage of normal seedlings was determined on the fifth day after setting up the test.

Electrical conductivity: four replications of 50 seeds per lot, weighed on an analytic balance with resolution of 0.001 g, were used. They were placed in plastic cups containing 75 mL of distilled water and then in germination chambers at 25 °C

for 24 hours. After this period, electrical conductivity of the solution was determined by an electrical conductivity meter (model TEC-4MP), and the mean values were calculated and expressed in μS .cm⁻¹ .g⁻¹ of seed (Vieira and Krzyzanowski, 1999).

Image analysis in Vigor-S[®]: seedlings were obtained from four replications of 16 seeds per lot, distributed in two rows on the upper third of two sheets of paper toweling and covered with a third sheet. All sheets were previously moistened with water in the amount of 2.5 times the weight of the dry paper. The 16 seeds were distributed with the hilum turned to the lower part of the paper. After covering with a third sheet of moistened paper, rolls were created, which were then placed in plastic bags and arranged vertically in B.O.D. for periods of two, three, and four days at 25 °C. At the end of each period, the seedlings were transferred from the roll of paper toweling to a sheet of blue paperboard with an area of 30 cm x 20 cm. The secondary seminal roots were removed from the seedlings evaluated at three to four days to facilitate reading by the software. Seedling images were acquired through a scanner (HP, Scanjet 200) fastened in an inverted position within an aluminum box ($60 \times 50 \times 12$ cm), adjusted to 300 dpi resolution and linked to a DELL Inspiron computer with Windows 10 operating system.

The images were individually processed by the Vigor-S® software, which provided the following variables: hypocotyl length, primary root length, total seedling length, growth index, uniformity index, and vigor index. Hypocotyl weight and primary root weight were adjusted at 10% and 90%, respectively, for calculation of the growth index. The contribution of growth and uniformity parameters used to calculate the vigor index was 70% and 30%, respectively.

Experimental design and statistical analysis: the experiment was conducted in a completely randomized experimental design with four replications. Analysis of variance (ANOVA) was performed on the data. After confirming normal distribution of errors by the Shapiro-Wilk test and homogeneity of variances by the Bartlett test, the mean values were compared by the Tukey test ($p \le 0.05$). After that, the Pearson correlation coefficients (r) were calculated for all the combinations between the seed quality evaluation tests, in which the significance of the r values was determined by the t test ($p \le 0.05$). The software used in statistical analyses was R 3.5.1 (R Core Team, 2018).

Results and Discussion

The quality tests applied for physiological characterization of the seed lots showed that the values obtained in determination of the moisture content of the four seed lots from each genotype were similar, with a variation of 1.4%, 1.1%, 0.9%, and 1.7% among the seed lots of the genotypes BRS Esteio, BRS Madre Pérola, VC 25, and Manteigão Fosco 11, respectively (Table 1). According to Marcos-Filho (2016), the uniformity of this parameter among seed lots is of utmost importance because differences greater than 2 percentage points can affect the reliability of the results of vigor tests. Moisture content is a characteristic that affects the rate of water absorption by seeds.

For the cultivars BRS Esteio and BRS Madre Pérola, the germination test allowed differentiation of the seed lots aged for 36 hours (Lots 4 and 8, respectively) from the others, in which they exhibited lower percentages of germination (86%). Nevertheless, all the seed lots exhibited values greater than 80% in the germination test, which is the minimum standard for commercialization of common bean seeds in Brazil (Brasil, 2013).

Seed lots of the line VC 25 were classified in three levels of physiological potential regarding the germination test. Lots 9 and 10 exhibited higher percentages of germination (≥92%); lot 11 had an intermediate value (85%) that did not differ from lot 10. Lot 12 had lower germination (65.5%). This lot was the only one with germination that did not meet commercialization standards.

For the cultivar Manteigão Fosco 11, analyses of variance of the germination data did not detect significant differences that could be attributed to the effects of seed lots (Table 1).

Vigor tests, on the other hand, showed greater sensitivity in stratification of the physiological potential of the seed lots than the germination test showed. For the cultivar BRS Esteio, lots 1 and 2 stood out as having greater vigor, except for the first count of germination test, which showed no difference among the lots (Table 1). The accelerated aging and electrical conductivity tests best categorized the lots, ranking them in three and four vigor levels, respectively. Lots 1, 2, 3, and 4 were classified in decreasing order regarding vigor level, which coincides with increasing order of exposure to accelerated aging, what is due the deterioration caused by exposure to high moisture and temperature.

For the other genotypes, BRS Madre Pérola, VC 25, and Manteigão Fosco 11, a similar response is observed in relation to the efficiency of vigor tests; electrical conductivity is the best test for evaluating physiological potential of common bean seeds. The accelerated aging test also proved to be efficient in characterization of the lots, except for the cultivar Manteigão Fosco 11. For that cultivar, the electrical conductivity test was the only one that showed significant differences in the vigor of the lots, indicating that the quantity of leachates of the seeds

in the test is affected by the increase in the period of exposure of common bean seeds to accelerated aging.

Since vigor tests are more sensitive, they allow less advanced stages of seed deterioration to be identified, which can facilitate decision-making regarding the destination of seed lots (Wendt et al., 2017). For Marcos-Filho (2015), the accelerated aging test provides valuable information regarding storage and field emergence potentials of seedlings. Concerning the electrical conductivity test, the greater the conductivity value, the lower seed germination and vigor is, due to loss of cell membrane integrity. Thus, the results reflect the physiological potential of the seeds, and inferences can be made regarding the storage potential of the lots (Binotti et al., 2008). Silva et al. (2014) reported that the electrical conductivity test is interesting because it quickly evaluates the capacity for cell membrane restructuring, and

this characteristic is important in vigor evaluation. It is known that degeneration of cell membranes is the first event in the deterioration process. Thus, the use of tests based on different principles can contribute to obtain more reliable responses in respect to the physiological potential of seed lots (Rocha et al., 2015).

The main objective of vigor tests is to identify possible differences in the physiological potential of seed lots that have similar germination percentages (Marcos-Filho, 2015). However, the aim of this study was to use seed lots with different levels of physiological quality induced by the artificial aging at different periods of seed exposure. Lots that contrast regarding germination allow broader verification of the efficiency of the methods for evaluation of physiological potential through automated analysis of seedling images.

Table 1. Moisture content (MC), germination (G), first count germination (FCG), moisture content after aging (MCaa), accelerated aging (AA), seedling emergence (SE), and electrical conductivity (EC) of sixteen common bean seed lots.

Lot	MC	G	FCG	MCaa	AA	SE	EC	
Lot				%		 	(μS. cm ⁻¹ . g ⁻¹)	
				BRS ESTEIO				
1	13.2	94.5 a	90	20.5	94.0 a	96.0 a	71.2 a	
2	13.9	93.5 a	90.5	20.7	94.5 a	95.5 a	86.2 b	
3	13.6	91.0 a	87	21.5	86.5 b	91.2 a	95.1 с	
4	14.6	86.0 b	82.5	22.2	78.0 с	84.2 b	122.6 d	
CV (%)	-	2.0*	4.6 ns	-	3.3*	3.5*	2.8*	
			BRS	S MADRE PÉROL	A			
5	13.1	95.0 a	95.0 a	20.3	96.0 a	90.0 a	68.13 a	
6	13.9	96.0 a	94.5 a	20.7	94.0 ab	86.5 a	75.24 b	
7	13.5	94.5 a	94.5 a	20.9	87.0 b	88.0 a	78.77 bc	
8	14.2	86.0 b	84.5 b	21.6	63.5 c	67.0 b	99.05 с	
CV (%)		2.7*	3.2*		4.2*	4.7*	4.5*	
				VC 25				
9	13.4	94.0 a	87.0 a	21.3	90.5 a	88.5 a	99.92 a	
10	14.2	92.0 ab	87.5 a	20.9	87.0 ab 81. 0 a		108.98 a	
11	13.5	85.0 b	79.5 a	20.5	79.5 b	85.0 a	108.79 a	
12	14.3	65.5 c	59.5 b	21.7	48.5 c	47.0 b	149.25 b	
CV (%)		4.5*	5.0*		6.2*	7.8*	5.1*	
			MAN	NTEIGÃO FOSCO	11			
13	13.1	94.0	86.5	20.5	96.5	97.0	53.85 a	
14	13.2	93.5	87.0	21.5	97.0	97.5	69.05 c	
15	14.8	91.0	84.5	20.6	93.5	96.0	59.29 b	
16	13.3	89.5	84.5	21.8	93.0	96.5	59.87 b	
CV (%)	-	3.3 ns	3.5 ns	-	2.2 ns	2.4 ns	3.7*	

The mean values followed by the same letter in the column, for each genotype, do not differ by the Tukey test ($p \le 0.05$). * = Significant; ns = not significant; CV = coefficient of variation.

Analyses of variance of the data obtained in the evaluations performed in Vigor-S[®] in all the periods of evaluation showed differences among the seed lots for most of the cultivars evaluated, except for the cultivar Manteigão Fosco 11 (Table 2).

For the cultivar BRS Esteio, ranking was observed on two levels of vigor for most of the variables analyzed on the second and third day of germination, with lower physiological potential for lot 4; however, it did not differ statistically from lot 3. In the evaluations performed on the fourth day after sowing, greater stratification of vigor levels can be observed; however, the ranking did not follow the same tendency of the previous periods of evaluation, in which the most vigorous lots corresponded to the least aged lots. In this case, the age of the seedlings affects the analysis made by Vigor-S*. The system proves to be more

consistent and sensitive in detecting differences in the vigor of the seed lots of this cultivar when analysis is made in younger seedlings, which gives rise to results comparable to the tests traditionally recommended for vigor evaluation. Definition of the correct time for evaluation is fundamental for generating accurate results and, according to Gomes-Junior et al. (2014), the earlier this evaluation is, the more advantageous from the practical point of view aiming at generating a faster diagnosis of the physiological quality of the lots evaluated.

In evaluation of the cultivar BRS Madre Pérola, lot 8 obtained lower seedling performance, with lower values for the variables analyzed in relation to the other lots (Table 2), which shows the sensitivity of the cultivar to deterioration due the artificial aging of at least 36 hours. The same stratification in vigor levels was also observed in the first two periods of

Table 2. Seedling lenght (SL), uniformity index (UI), growth index (GI), and vigor index (VI) of sixteen common bean seed lots at two, three, and four days after sowing.

	Second day				Third day				Fourth day			
Lot	SL	UI	GI	VI	SL	UI	GI	VI	SL	UI	GI	VI
	cm	cmindex		cm	cmindex			cmindex				
BRS ESTEIO												
1	2.29	798 a	166	355 a	6.09	810 a	419 a	536 a	8.47 ab	821	601 ab	667 ab
2	2.54	790 a	164	352 a	6.29	804 a	420 a	535 a	7.67 b	773	489 bc	574 bc
3	2.46	760 ab	156	337 ab	5.78	773 ab	369 ab	490 ab	9.19 a	826	631 a	689 a
4	2.13	703 b	125	298 b	5.18	718 b	317 b	437 b	7.60 b	770	464 c	556 с
CV (%)	10.4 ns	5.01*	13.6 ns	6.7*	10.1 ns	5.5*	10.8*	7.8*	7.1*	5.3 ns	11.7*	7.5*
BRS MADRE PÉROLA												
5	2.08	765	151 a	335 a	5.38 a	768 a	418 a	523 a	8.60 a	779 a	716 a	735 a
6	2.37	771	158 a	342 a	5.51 a	802 a	376 ab	519 a	8.84 a	797 a	701 a	730 a
7	2.08	762	153 a	336 a	5.15 a	756 a	393 a	502 a	7.01 b	712 a	520 b	578 b
8	1.73	650	108 b	271 b	3.70 b	656 b	269 b	385 b	5.31 c	569 b	382 c	438 с
CV (%)	13.7 ns	8.3 ns	13.2*	8.6*	11.1*	5.6*	15.1*	10.1*	9.6*	6.5*	10*	8.1*
					7	VC 25						
9	2.53 a	747 a	189 a	357 a	5.84 a	733 a	440 a	528 a	9.20 a	778 a	738 a	750 a
10	1.68 b	617 a	104 b	257 b	4.32 ab	624 a	316 b	408 b	5.94 b	626 bc	458 b	508 b
11	1.55 b	597 a	107 b	254 b	3.78 b	700 a	290 b	413 ab	5.54 b	668 b	437 b	506 b
12	0.72 c	301 b	48 c	124 c	2.11 c	432 b	154 с	237 с	3.63 c	524 c	271 с	347 с
CV (%)	16.3*	14.1*	16.1*	13.4*	18.1*	10.1*	18.8*	13.9*	13.6*	7.6*	14.3*	11.4*
MANTEIGÃO FOSCO 11												
13	3.98	745	308	439	7.63	805	645	693	10.79	787	964	911
14	3.95	783	292	439	7.29	775	620	666	10.23	716	873	826
15	4.05	763	307	444	7.58	784	655	694	10.89	717	942	875
16	3.38	739	249	396	7.035	753	596	643	9.28	749	824	802
CV (%)	9.8 ns	3.0^{ns}	11.5 ns	5.7 ns	9.0 ns	5.1 ns	12.4 ns	9.5 ns	8.3 ns	10.0 ns	7.9 ns	7.7 ns

Mean values followed by the same letter in the column do not differ by the Tukey test ($p \le 0.05$). *= Significant; ns = not significant; CV = coefficient of variation.

evaluation (second and third day) and a ranking in more vigor levels on the fourth day of analysis of the seedlings carried out through the Vigor-S® software. Therefore, it is probable that for a more detailed examination of the seed samples with high and uniform physiological potential, it is necessary to extend the time to evaluation of the seedlings for an additional period of 24 or 48 h, with a view toward an increase in the differences in seedling lengths in response to different vigor levels.

Speed in obtaining results is an important aspect when using computerized image analysis of seedlings (Abud et al., 2017), which, in the case of the common bean seeds used in this study, can be performed on seedlings obtained at two, three, and four days after setting up the germination test, depending on the cultivar used. Nevertheless, seedling images analyzed at an earlier time (two and three days) can provide promising results, due to the shorter time (Gomes-Junior et al., 2014) and because of the lack of need to remove secondary seminal roots, common in vigorous common bean seedlings; this increases the agility of evaluation.

Recent studies with the use of seedling image analysis have indicated evaluations at two days after sowing for maize (Mondo et al., 2011) and millet (Javorski et al., 2018) seeds; three to four days for common bean (Gomes-Junior et al., 2014), soybean (Marcos-Filho et al., 2009; Medeiros and Pereira, 2018), crambe (Leão-Araújo et al., 2017), and sunflower (Rocha et al., 2015) seeds; and five to six days for onion (Gonçalves et al., 2017) and carrot (Marchi and Cicero, 2017) seeds.

The line VC 25 was most sensitive to prior aging, according to the classification made by Vigor-S®, which was also observed in the traditional vigor evaluation tests, especially in the accelerated aging test (Table 1). The variables obtained from the software were sensitive in detecting differences in seed vigor in all the periods evaluated, highlighting the uniformity index and the vigor index. According to Silva et al. (2017), the vigor and uniformity indexes generated from the data obtained in evaluation of squash seedling growth are effective parameters for composing programs for seed quality control.

The uniformity index generated by Vigor-S®, calculated from the uniformity coefficient of Christiansen (Christiansen, 1942), also showed promising results in studies carried out by Castan et al. (2018). This index is relevant since irregular seedling emergence can lead to delays in development and variations in growth of plants in various phenological stages, which also affects harvest results (Marcos-Filho, 2016).

Leão-Araújo et al. (2017) indicated that the indexes generated using the Seed Vigor Imaging System (SVIS®), including uniformity of seedling development, were able to separate crambe seed lots in an effective manner. Results

obtained by Silva et al. (2012) showed that uniformity of seedling development can provide useful information regarding the degree of seed deterioration, initial growth potential, and uniformity of seedling emergence of crotalaria. In contrast, in studies on seeds of cucumber (Chiquito et al., 2012), sunflower (Caldeira et al., 2014), carrot (Pinto et al., 2015), maize (Marchi and Cicero, 2017), and soybean (Medeiros and Pereira, 2018), the seedling uniformity index did not correlate with the vigor of the seed lots evaluated. However, in those studies, the index proposed by Sako et al. (2001) was used.

Another parameter that proved to be efficient in detecting vigor differences among lots for most cultivars was seedling length. Marcos-Filho et al. (2009) used automated analyses in SVIS® in three-day old soybean seedlings and found that seedling length was the most sensitive parameter for indicating vigor differences among lots, exceeding the uniformity index and the vigor index.

In contrast, seed lots of the cultivar Manteigão Fosco 11 did not differ from each other for evaluations made by seedling image analysis (Table 2), just as observed in the traditional vigor tests (Table 1), except for electrical conductivity. Although the data of seedling length obtained by Vigor-S® were not sensitive enough to identify differences among the lots of this cultivar, this result is important because one of the basic requisites for introduction of new procedures of seed vigor evaluation is their equivalence with other laboratory tests that are routinely used (Gomes-Junior et al., 2014), such as accelerated aging and seedling emergence tests.

The simple correlation coefficients (Figure 1) show that the results generated by traditional tests and by the seedling analysis system exhibited significant correlations, which is an indication that the indexes generated by Vigor-S® accurately determined quality of the seed lots because there proved to be a strong dependence between them.

The accelerated aging and electrical conductivity tests, considered the most sensitive in categorization of the seed lots among the vigor tests (Table 1), showed significant positive and negative correlations, respectively, with the variables obtained by image analysis for all the specific periods of evaluation (2, 3, and 4 days) (Figure 1). These results confirm the efficiency of the Vigor-S® system for evaluating the physiological potential of common bean seeds in comparison with the tests traditionally used.

According to Gonçalves et al. (2017), the correlation between the traditional tests and image analysis is valuable for adjusting the method, especially for determining the best age of the seedlings to carry out analysis. In this sense, the strong correlation with widely used vigor tests shows the possibility of using information from the indexes generated from development of seedlings to evaluate seed vigor (Leão-Araújo et al., 2017).

The use of Vigor-S® for computerized analysis of seedling images allowed the physiological potential of seeds to be evaluated through more than one parameter, which shows the possibility of taking better advantage of the results obtained from the same evaluation. An example of analysis of seedling length carried out by Vigor-S® can be observed in Figure 2, in which the primary root of each seedling is demarcated in red and the hypocotyl in blue. Moreover, the system allows manual adjustments of the line that represents the hypocotyl and the root so that corrections can be made when the demarcation is not satisfactory. The results of analyses are presented on the processing screen and can also be exported to extensions of the Excel software, which, according to Castan et al. (2018), allow better organization of the information obtained and generate greater ease in later interpretation of the results by seed analysts.

According to Gomes-Junior et al. (2014), one of the difficulties in determining common bean seedling length in a

manual way is the tendency toward curvature of the hypocotyl of the seedlings. Thus, automation of these determinations significantly contributes to reducing the errors committed by seed laboratory analysts, who work in a routine way in evaluation of a high number of seed lots (Medeiros and Pereira, 2018).

In recent years, research in seed technology has prioritized computerized procedures for evaluation of seed vigor (Wendt et al., 2017). The results of this study show that although Vigor-S® is programmed to analyze seedling images of maize and soybean, the sensitivity observed in the variables offered by the software to detect variations in the physiological potential of common bean seed lots with different vigor levels represents an important initiative for efficiently complementing evaluation of the physiological potential of the seeds in quality control programs. As Medeiros and Pereira (2018) observed in working with the *Sistema de Análise de Plântulas* (SAPL®) [Seedling Analysis System] software, seedling image analysis by Vigor-S® in this study likewise allowed the time spent in evaluations to be reduced, generating greater agility in obtaining results.

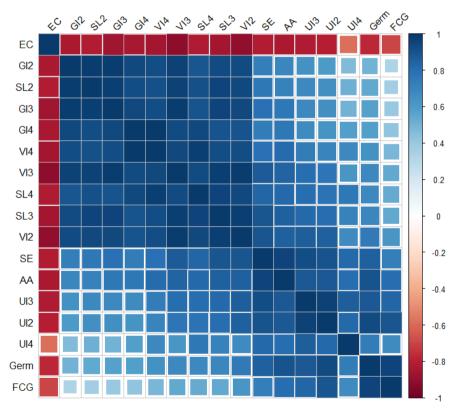


Figure 1. Pearson correlation (r) between the variables generated from traditional seed analysis and from image analysis by Vigor-S® of sixteen common bean seed lots. Germ = germination, FCG = first count of germination, AA = accelerated aging, SE = seedling emergence, EC = electrical conductivity, SL = seedling length, GI = growth index, UI = uniformity index, VI = vigor index.



Figure 2. Window of the Vigor-S® Analysis System with a sample of common bean seedlings (Cultivar Manteigão Fosco 11) at two days of germination test (A); detail of seedling 9 showing the primary root (red) and hypocotyl (blue) (B).

Conclusions

Computerized image analysis of seedlings using Vigor-S* is a consistent alternative for evaluation of common bean seed vigor and produces results compatible with those obtained by the traditional tests of seed quality evaluation.

References

ABUD, H.F.; CICERO, S.M.; GOMES-JUNIOR, F.G. Computerized image analysis of seedlings to evaluate broccoli seed vigor. *Journal of Seed Science*, v.39, n.3, p.303-310, 2017. https://doi.org/10.1590/2317-1545v39n3174582

BINOTTI, F.F.; HAGA, K.I.; CARDOSO, E.D.; ALVES, C.Z.; SÁ, M.E.; ARF, O. Efeito do período de envelhecimento acelerado no teste de condutividade elétrica e na qualidade fisiológica de sementes de feijão. *Acta Scientiarum*, v.30, n.2, p.247-254, 2008. https://doi.org/10.4025/actasciagron.v30i2.1736

BRASIL. Ministério da Agricultura, Pecuária e Abastecimento. Regras para análise de sementes. Ministério da Agricultura, Pecuária e Abastecimento. Secretaria de Defesa Agropecuária. Brasília: MAPA/ACS, 2009. 395p.

BRASIL. Ministério da Agricultura, Pecuária e Abastecimento. Instrução Normativa 45/2013, de 17 de setembro de 2013, Ministério da Agricultura, Pecuária e do Abastecimento (Anexo XI - Padrões para produção e comercialização de sementes de feijão). Brasília, DF: Diário Oficial da República Federativa do Brasil, 2013, p. 6, Seção 1.

BRUNES, A.P.; SOUZA ARAÚJO, Á.; DIAS, L.W.; VILLELA, F.A.; AUMONDE, T.Z. Seedling length in wheat determined by image processing using mathematical tools. *Revista Ciência Agronômica*, v.47, n.2, p.374-379, 2016. https://doi.org/10.5935/1806-6690.20160044

CALDEIRA, C.M.; CARVALHO, M.L.M.; OLIVEIRA, J.A.; COELHO, S.V.B.; KATAOKA, V.Y. Sunflower seed vigor determined by computerized seedling analysis. *Cientifica*, v.42, p.346-353, 2014. https://doi.org/http://dx.doi.org/10.1590/0103-8478cr20131455

CASTAN, D.O.C.; GOMES-JUNIOR, F.G.; MARCOS-FILHO, J. Vigor-S, a new system for evaluating the physiological potential of maize seeds. *Scientia Agricola*, v.75, n.2, p.167-172, 2018. https://doi.org/10.1590/1678-992x-2016-0401

CHIQUITO, A.A.; GOMES-JUNIOR, F.G.; MARCOS-FILHO, J. Assessment of physiological potential of cucumber seeds using the software Seedling Vigor Imaging System® (SVIS®). *Revista Brasileira de Sementes*, v.34, n.2, p.255–263, 2012. https://doi.org/10.1590/S0101-31222012000200010

CHRISTIANSEN, J.E. *Irrigation by sprinkling*. University of California, Berkeley, California, USA (California Agricultural Experimental Station Bulletin 670), 1942. http://www.sidalc.net/cgi-bin/wxis.exe/?IsisScript=COLPOS.xis&method=post&formato=2&cantidad=1&expresion=mfn=032293

GOMES-JUNIOR, F.G.; CHAMMA, H.M.C.P.; CICERO, S.M. Automated image analysis of seedlings for vigor evaluation of common bean seeds. *Acta Scientiarum. Agronomy*, v.36, n.2, p.195-200, 2014. https://doi.org/10.4025/actasciagron.v36i2.21957

GONÇALVES, N.R.; CICERO, S.M.; ABUD, H.F.; GONÇALVES, N.R.; CICERO, S.M.; ABUD, H.F. Seedling image analysis and traditional tests to evaluate onion seed vigor. *Journal of Seed Science*, v.39, n.3, p.216-223, 2017. https://doi.org/10.1590/2317-1545v39n3160444

JAVORSKI, M.; OTTE, D.; CASTAN, C.; SANTANNA, S.; GOMES-JUNIOR, F.G.; CICERO, S.M. Image analysis to evaluate the physiological potential and morphology of pearl millet seeds. *Journal of Seed Science*, v.40, n.2, p.127-134, 2018. https://doi.org/10.1590/2317-1545v40n2176904

LEÃO-ARAÚJO, E.F.; FARIA, J.; BARBOZA, C.; MARCOS-FILHO, J.; VIEIRA, R.D. Controlled deterioration test and use of the Seed Vigor Imaging System (SVIS®) to evaluate the physiological potential of crambe seeds. *Journal of Seed Science*, v.39, n.4, p.393-400, 2017. https://doi.org/http://dx.doi.org/10.1590/2317-1545v39n4177911

MARCHI, J.L.; CICERO, S.M. Use of the software Seed Vigor Imaging System (SVIS®) for assessing vigor of carrot seeds. *Scientia Agricola*, v.74, n.6, p.469-473, 2017. https://doi.org/10.1590/1678-992x-2016-0220

MARCOS-FILHO, J.; KIKUTI, A.L.P.; LIMA, L.B. Métodos para avaliação do vigor de sementes de soja, incluindo a análise computadorizada de imagens. *Revista Brasileira de Sementes*, v.31, n.1, p.102-112, 2009. https://doi.org/10.1590/S0101-31222009000100012

MARCOS-FILHO, J. Seed vigor testing: an overview of the past, present and future perspective. *Scientia Agricola*, v.72, n.4, p.363-374, 2015. https://doi.org/10.1590/0103-9016-2015-0007

MARCOS-FILHO, J. Seed Physiology of Cultivated Plants. Londrina: ABRATES, 2016. 659p.

MARQUES, F.R.F.; MEIADO, M.V.; CASTRO, N.M.C.R.; CAMPOS, M.L.O.; MENDES, K.R.; SANTOS, O.O.; POMPELLI, M.F. GerminaQuant: a new tool for germination measurements. *Journal of Seed Science*, v.37, n.3, p.248-255, 2015. https://doi.org/10.1590/2317-1545v37n3145605

MEDEIROS, A.D.; PEREIRA, M.D. SAPL ®: a free software for determining the physiological potential in soybean seeds. *Pesquisa Agropecuária Tropical*, v.48, n.3, p.222-228, 2018. https://doi.org/10.1590/1983-40632018v4852340

MONDO, V.H.V.; DIAS, M.A.N.; McDONALD, M.B. Seed vigor imaging system for two-day-old corn seedling evaluation. *Seed Technology*, v.33, n.2, p.191-196, 2011. https://www.jstor.org/stable/23433428

PINTO, C.A.G.; CARVALHO, M.L.M.; ANDRADE, D.B.; LEITE, E.R.; CHALFOUN, I. Image analysis in the evaluation of the physiological potential of maize seeds. *Revista Ciência Agronômica*, v.46, n.2, p.319-328, 2015. https://doi.org/10.5935/1806-6690.20150011

R CORE TEAM. R Development Core Team. R: A Language and Environment for Statistical Computing, 2018. https://doi.org/http://www.R-project.org

ROCHA, C.R.M.; SILVA, V.N.; CICERO, S.M. Avaliação do vigor de sementes de girassol por meio de análise de imagens de plântulas. *Ciência Rural*, v.45, n.6, p.970-976, 2015. https://doi.org/10.1590/0103-8478cr20131455

SAKO, Y.; MCDONALD, M.B.; FUJIMURA, K.; EVANS, A.F.; BENNETT, M.A. A system for automated seed vigour assessment. *Seed Science and Technology*, v.29, p.625-636, 2001. https://www.eurofinsus.com/media/162083/seed-vigor-imaging-system.pdf

SILVA, C.B.; LOPES, M.M.; MARCOS-FILHO, J.; VIEIRA, R.D. Automated system of seedling image analysis (SVIS) and electrical conductivity to assess sun hemp seed vigor. *Revista Brasileira de Sementes*, v.34, n.1, p.55-60, 2012. https://doi.org/10.1590/S0101-31222012000100007

SILVA, V.N.; ZAMBIASI, C.A.; TILLMANN, M.A.A.; MENEZES, N.L.; VILLELA, F.A. Condução do teste de condutividade elétrica utilizando partes de sementes de feijão. *Revista de Ciências Agrárias*, v.37, n.2, p.206-213, 2014. http://www.scielo.mec.pt/scielo.php?script=sci arttext&pid=S0871-018X2014000200011

SILVA, P.P.; BARROS, A.C.S.A.; MARCOS-FILHO, J.; GOMES-JUNIOR, F.G.; NASCIMENTO, W.M. Assessment of squash seed vigor using computerized image analysis. *Journal of Seed Science*, v.39, n.2, p.159-165, 2017. https://doi.org/http://dx.doi.org/10.1590/2317-1545v39n2171177

VIEIRA, R.D.; KRZYZANOWSKI, F.C. Teste de condutividade elétrica. In: KRZYZANOWSKI, F.C.; VIEIRA, R.D.; FRANÇANETO, J.B. (Eds.). *Vigor de sementes: conceitos e testes.* Londrina: ABRATES,1999. p.4.1-4.26.

WENDT, L.; MALAVASI, M.M.; DRANSKI, J.A.L.; MALAVASI, U.C.; GOMES-JUNIOR, F.G. Relação entre testes de vigor com a emergência a campo em sementes de soja. *Brazilian Journal of Agricultural Sciences*, v.12, p.166-171, 2017. https://doi.org/10.5039/agraria.v12i2a5435



This is an Open Access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.