

Image analysis during processing as an aid to soybean quality assessment

Análise de imagem no beneficiamento como auxiliar na avaliação da qualidade de sementes de soja

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ABSTRACT - Processing is an important step in high-quality seed production. To improve this process, image analysis enables the assessment of parameters associated with physiological quality. This study aimed to determine whether X-ray image analysis can be used as a tool in the processing process to relate the physiological quality of soya beans. The seeds were collected at the dryer exit and after bagging. The treatments included four varieties (HO Aporé IPRO, DM 73175 RSF IPRO, Brasmax Olimpo IPRO, and Brasmax Bônus IPRO). The seeds from each sampling point underwent water content, germination, and tetrazolium tests. To obtain images, the soya beans were placed in an acrylic dish containing 50 units, with four replicates per treatment, and exposed to X-rays for 10 s at an intensity of 31 kV and a distance of 55.4 cm from the radiation source. The images were processed using ImageJ software to obtain measurements of the area, perimeter, width, length, median gray, integrated density, aspect ratio, and solidity. Data were subjected to principal component and correlation analyses. The results of the present study indicate that seeds with greater sphericity tend to have higher physiological quality. Analysis of X-ray images at the end of processing is a promising tool for identifying batches of spherical seeds with a view towards higher physiological quality.

RESUMO - O processo de beneficiamento é uma etapa importante na produção de sementes de alta qualidade. No intuito de aprimorar esse processo, a análise de imagens permite avaliar parâmetros que podem ser associados a qualidade fisiológica. O objetivo do trabalho foi verificar se a análise de imagens de raios X pode ser uma ferramenta a ser implementada no processo de beneficiamento, a fim de relacioná-lo com a qualidade fisiológica das sementes de soja. As sementes foram coletadas na saída do secador e após o ensaque. Os tratamentos foram compostos por quatro cultivares (HO Aporé IPRO, DM 73175 RSF IPRO, Brasmax Olimpo IPRO e Brasmax Bônus IPRO). Nas sementes de cada ponto amostrado foi determinado o teor de água e realizados os testes de germinação e tetrazólio. Para obtenção das imagens, as sementes de soja foram dispostas em placa de acrílico contendo 50 unidades com quatro repetições por tratamento e submetidas aos raios X por dez segundos, na intensidade de 31 kV e a distância de 55,4 cm da fonte de radiação. As imagens foram processadas no software ImageJ, obtendo-se as medidas de área, perímetro, largura, comprimento, cinza mediano, densidade integrada, aspect ratio e solidez. Os dados foram estudados por meio da análise de componentes principais e análise de correlação. Os resultados dessa pesquisa indicam que sementes com maior esfericidade tendem a apresentar maior qualidade fisiológica. A análise de imagens de raios X no final do beneficiamento é uma ferramenta promissora para identificar lotes de sementes mais esféricas visando maior qualidade fisiológica.

Keywords: *Glycine max* (L) Merrill. Image J. Sphericity. Physiological quality.

Palavras-chave: *Glycine max* (L) Merrill. Image J. Esfericidade. Qualidade fisiológica.

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INTRODUCTION

Image analysis has shown potential to be used in conjunction with traditional tests to assess seed quality, improving methods and providing reliable results (SILVA et al., 2020). Among the resources available for obtaining images, the use of X-rays is recommended by the International Seed Testing Association (ISTA, 2020) and the Seed Analysis Rules (BRASIL, 2009) and is a promising for the evaluation of physical characteristics related to physiological potential (ACHA; VIEIRA, 2020; PINHEIRO et al., 2020), being a rapid and non-destructive procedure.

Studies have used X-ray images to analyze internal seed morphology, parameters related to physiological quality, and the relationship between physical characteristics and germination and vigor (ABUD; CICERO; GOMES-JUNIOR, 2018; MEDEIROS et al., 2020) in different species, such as soybean (*Glycine max* (L.) Merrill) (WENDT et al., 2014), corn (*Zea mays* L.) (CHEN et al., 2024), broccoli (*B. oleracea* L. var. *italica* Plenck) (ABUD; CICERO; GOMES-JUNIOR, 2018), okra (*Abelmoschus esculentus* (L.) (SANTOS; GOMES-JUNIOR; MARCOS-FILHO, 2020), pepper (*Capsicum* (L.)) (AHMED et al., 2020; HONG et al., 2023), papaya (*Carica papaya* (L.)) (NOGUEIRA et al.,

2024), *Brachiaria* (MEDEIROS et al., 2020), and crambe (*Crambe abyssinica* Hochst) (RIBEIRO et al., 2021; MEDEIROS et al., 2021).

When X-ray images are linked to a software, information is generated that makes it possible to analyze the size and shape characteristics of the seeds, such as the area, perimeter, length, width, and solidity (SCHNEIDER; RASBAND; ELICEIRI, 2012). Based on these physical characteristics, soya beans were mechanically processed at various stages to form lots with similar characteristics.

Processing is an important stage in the production of high-quality seeds and comprises a set of operations to which the seed is subjected, from reception to packaging and distribution. The aim of processing is to improve the characteristics of the seed lot by promoting uniformity and improving the physical and physiological quality.

Therefore, the aim of the research was to determine if X-ray image analysis could be used as a tool in the processing process to relate physical characteristics to the physiological quality of soya beans.

MATERIAL AND METHODS

This study was conducted between February and September 2023. Soybean seed samples were collected at the Unigigel seed processing unit, located in the municipality of Chapadão do Céu, GO, at the beginning (outlet of the dryer) and end (in the bag) of the process. Four varieties were used: HO Aporé IPRO, DM 73I75 RSF IPRO, Brasmax Olimpo IPRO, and Brasmax Bônus IPRO.

After collection, the samples were sent to the Seed Technology Laboratory of the Federal University of Mato Grosso do Sul, Chapadão do Sul Campus to assess the initial quality of the seeds from each sampling point. The water content was determined using the oven method at 105 °C (± 3 °C) for 24 h, with two replicates of approximately 10 g for each treatment, according to the recommendations of BRASIL (2009), expressing the results as a percentage (dry basis).

The seeds were subjected to a germination test, which was conducted on germitest paper moistened with a quantity of water equal to 2.5 times the mass of the unmoistened paper, with four replicates of 50 seeds each. The rolls were then stored in the germinator at 25 °C (BRASIL, 2009), and the seedlings were assessed five days after the test.

For the tetrazolium test, 100 seeds per treatment, with two replicates of 50 seeds, were used from each sampling point, pre-soaked in germination paper with distilled water, and stored in a germinator at 25 °C for 16 h. The seeds were then placed in plastic containers and completely immersed in a solution of 2,3,5-triphenyltetrazolium chloride at 40 °C in the dark for 3 h. The seeds were then washed under running water and individually scored as described by França-Neto and Krzyzanowski (2018).

For the X-ray test, 200 seeds of each variety were placed in four transparent acrylic plates on double-sided adhesive tape and transported to the Seed Laboratory of the

Instituto Federal Goiano, Campus Rio Verde. To obtain the images, a 'Faxitron HP' X-ray machine (model 43855A) was used, in which the seeds were exposed to radiation for 10 s at an intensity of 31 kV and a distance of 55.4 cm from the radiation source.

The images produced by the X-ray test were analyzed using ImageJ (Image Processing and Analysis in Java) software to obtain the following variables: Area: amount of two-dimensional space, i.e., the surface of the seed, expressed in cm^2 ; Perimeter: measurement of the two-dimensional contour of the seed, expressed in mm; Mode: median gray value; Width (cm); Length (cm); IntDen (integrated density): is the density based on the gray level, i.e., the sum of the image pixel values, expressed in $\text{gray cm}^2 \text{ pixel}^{-1}$; Aspect ratio (AR): obtained from the ratio between the longest and shortest axis of the ellipse circumscribing the seed (the higher the ratio, the more elongated the seed); Solidity: obtained from the ratio between the projected area and the area of the convex envelope. An increase in this value indicates that the seeds are solid.

The experimental design used was fully randomized, with four treatments (cultivars) and four replications per treatment. The data were subjected to principal component analysis (PCA) and correlation analysis using R software (R CORE TEAM, 2024) with the ggplot2 and ExpDes.pt packages.

RESULTS AND DISCUSSION

Outlet of the dryer

Samples of the four soya varieties collected at the outlet of the dryer had a water content of 9–11%. According to Simak (1991), the water content of seeds affects their optical density. The lower the moisture content of the seeds, the higher the optical density, which makes it possible to observe the internal structures of the seeds in the X-ray image.

In studies using X-ray image analysis of soybean seeds (WENDT et al., 2014), *Leucaena leucocephala* (MEDEIROS et al., 2018), millet (*Panicum miliaceum* (L.)) (JAVORSKI et al., 2018), and *Brachiaria ruziziensis* (MEDEIROS et al., 2020), it was possible to obtain images with good visualization, with water content varying between 8 and 12%, which is close to the values observed in this study.

The vectors shown in Figure 1 represent the variables obtained from the initial assessment of the physiological quality of the seeds and those obtained from the analysis of the radiographs. Multivariate PCA showed that the first two components (PC1 and PC2) accounted for 66% of the variability in the data (Figure 1).

The germination vector was close to the perimeter and opposite to the solidity. Vigor is close to solidity; viability is close to the perimeter, area, length, and width of the variables and the integrated density. Parameters such as length, width, area, and perimeter are associated with the size of the seed and the number of reserves, and are related to the viability of the seeds.

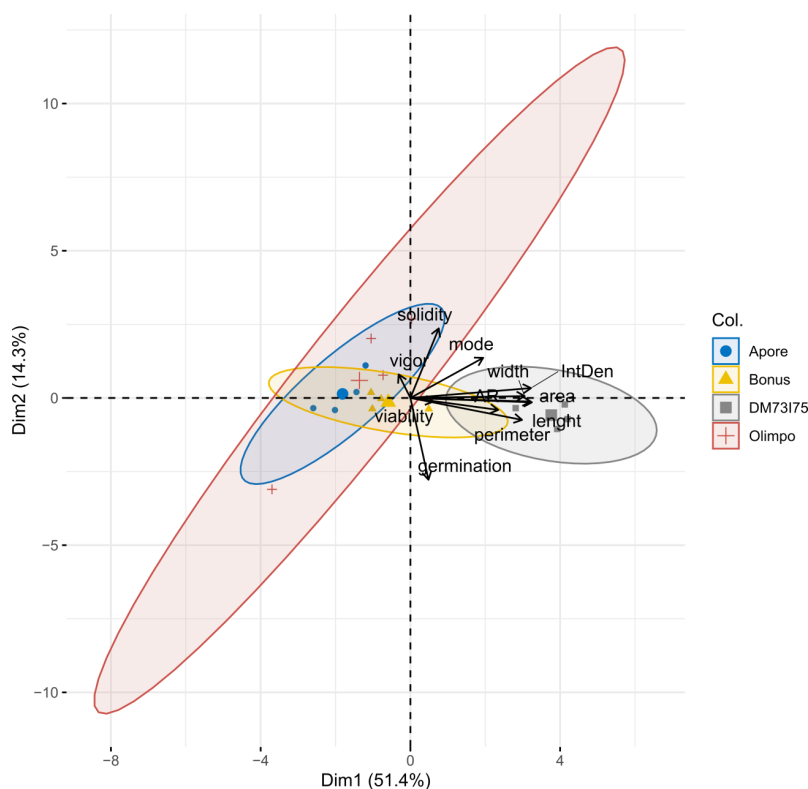


Figure 1. Biplot obtained by grouping the variables related to seed characteristics collected at the dryer outlet for four soybean varieties. AR = aspect ratio, IntDen = integrated density.

The correlation matrix (Figure 2) showed that germination had a negative correlation with median gray (mode) (-0.5; $p < 0.05$) and solidity (-0.7; $p < 0.05$), and a weak positive correlation with perimeter. Vigor showed a negative,

albeit weak, correlation with the variables that infer size: area, perimeter, width, length, and AR, and the correlation was positive only with median gray. Viability showed a weak positive correlation with all the variables.

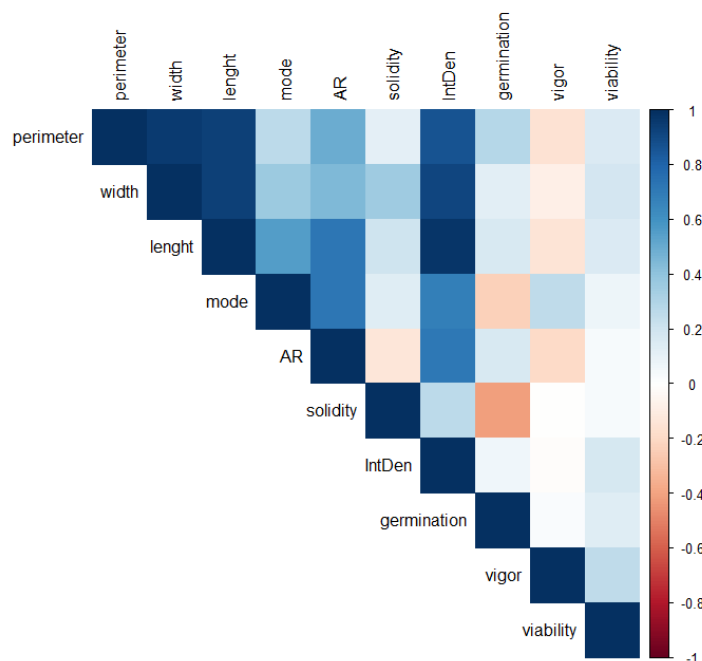


Figure 2. Correlation matrix obtained from X-ray image data and the physiological quality of seeds collected at the dryer outlet for four soybean varieties. AR = aspect ratio, IntDen = integrated density.

Therefore, it is not possible to correlate the X-ray analysis at the exit of the dryer with the physiological quality of the soya beans. Notably, the seeds leaving the dryer still go through the selection process for size, density, and shape, with non-standard seeds, such as poorly formed ones, being discarded to obtain standardized and uniform batches (MOREANO et al., 2013).

Bagging

The water content of the samples collected after bagging was 9%. Water content is an important factor in X-

ray studies because it affects the image quality. The lower the water content in the seed, the higher the optical density, which allows the internal structures of the seed to be better visualized in an X-ray image (SIMAK, 1991). Bagging is the final processing step aimed at improving the characteristics of the seed batch, achieving uniformity, and improving the physical and physiological qualities of the seed.

Multivariate PCA after bagging was performed on the data obtained for the four varieties and their characteristics were evaluated (Figure 3). The first two components (PC1 and PC2) accounted for 79% of total data variability.

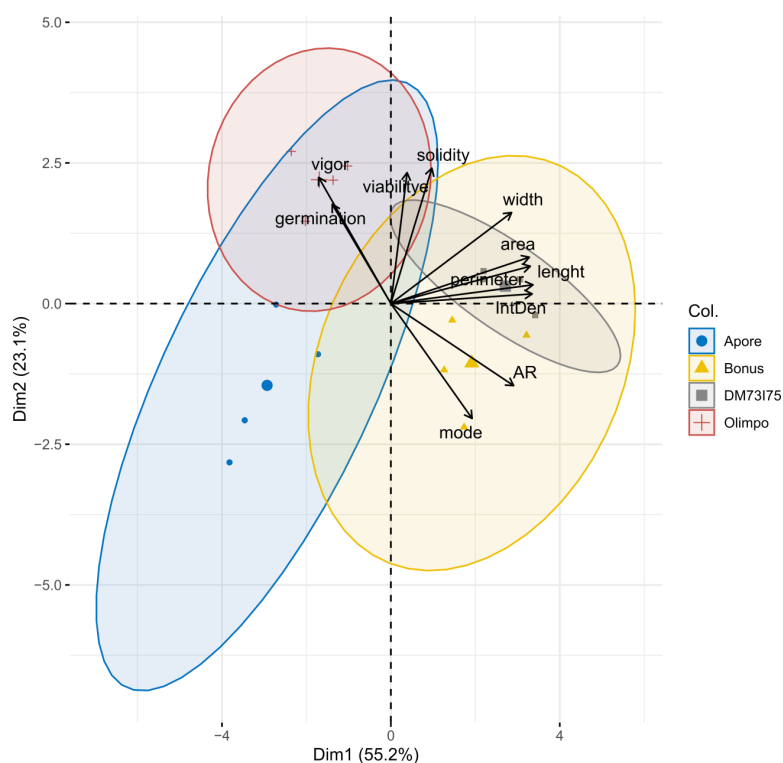


Figure 3. Biplot obtained by grouping variables related to seed characteristics collected after bagging. AR = aspect ratio, IntDen = integrated density.

According to Moreano et al. (2013), several studies have attempted to identify the influence of seed size on the physiological quality and productivity of soybeans; however, the results have been contradictory, with the authors reaching different conclusions. In broccoli (*B. oleracea* L. var. *italica* Plenck) seeds, Abud, Cicero and Gomes-Junior (2018) found that the area may reflect seed vigor, as seeds with larger areas coincide with greater reserves available for germination, a fact not demonstrated in this study because the area and germination vectors are far apart in different quadrants (Figure 3).

The vectors shown in Figure 3 represent the physiological quality, germination, vigor, viability, and variables obtained from the analysis of the radiographs. The vectors for germination, vigor, and viability were close to solidity, indicating a possible correlation. Solidity is a variable that can be used as a measure of tissue integrity, where values close to 1.0 correspond to a solid structure and lower values indicate irregular boundaries; therefore, seeds with greater

solidity may indicate that they are well formed and undamaged (RIBEIRO et al., 2021).

The vectors for germination and vigor were similar (Figure 3), and both were opposite to the median gray (mode). The variables calculated from the gray values of each pixel in the image, such as integrated and relative density and median gray, are related to the resistance of the internal tissues of the seed to the passage of X-rays, because the photons in an X-ray beam can be transmitted, scattered, or absorbed when in contact with the object.

Thus, higher gray densities indicate the presence of denser tissue, that is, a greater obstacle to the passage of X-rays, resulting in a higher level of radiopacity (light) in radiographic images (MEDEIROS et al., 2020; SILVA et al., 2020). According to Medeiros et al. (2018), images with spots indicating tissue degradation, mechanical damage, reduced embryo filling, or poor seed formation contribute to a lower level of radiopacity (dark), thereby reducing the gray values in the images.

The AR vector also proved to be the opposite of the seed germination and vigor parameters (Figure 3), and a strong negative correlation was observed between the AR and the physiological characteristics of vigor and germination (Figure 4). This implies that the more elongated the seed, the worse the physiological parameters. In soybeans, Wendt et al. (2014) observed that increasing values of the ratio between

the longest and shortest axes indicated that the seeds were more elongated, and batches with high values of this variable showed low germination, lower physiological potential, and deformed seeds. These results are in line with those obtained in the present study, from which we can conclude that round seeds have higher germination and vigor.

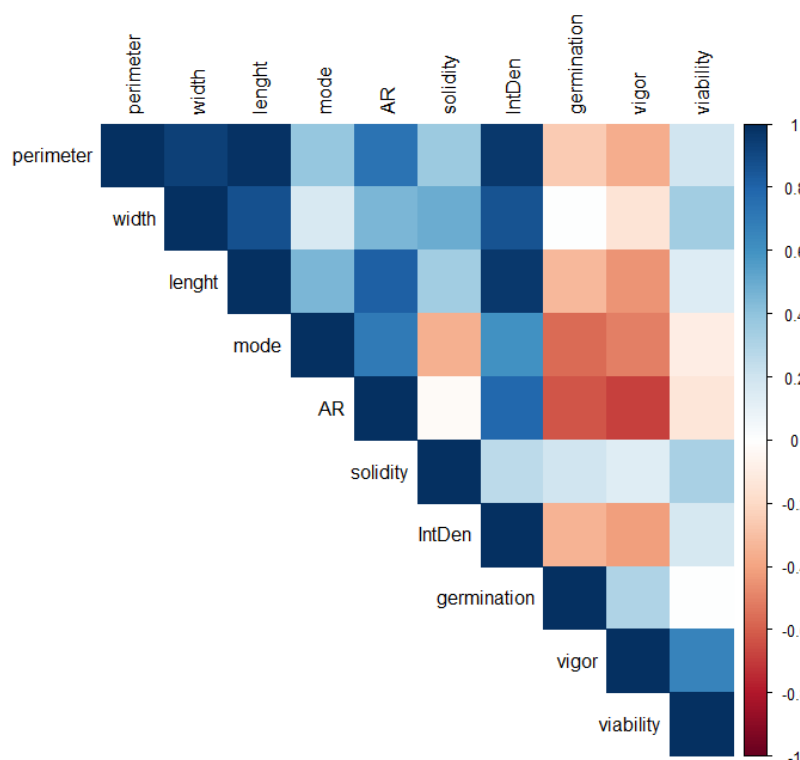


Figure 4. Correlation matrix obtained from X-ray image data and the physiological quality of seeds collected at bagging for four soybean varieties. AR = aspect ratio, IntDen = integrated density.

Germination (0.4) and vigor (0.3) showed a weak positive correlation with solidity, whereas the correlations with the other variables were negative, except for germination and width, which showed no correlation (Figure 4). Seeds arriving at the bagging plant were passed through various machines that selected the seeds, rejected damaged seeds, and formed uniform and standardized batches. The aim of soybean seed processing is to select seeds so that at the end of the process, the batches are uniform and standardized.

CONCLUSIONS

Soybean seeds with higher sphericity exhibit greater germination and vigor. Image analysis, particularly for AR and mode variables, at the final stage of processing can be used to identify lots with more spherical seeds with greater physiological potential, thus improving seed processing.

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