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# QUALITY OF BEAN SEEDS WITH DIFFERENT STORAGE UNITS

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**ABSTRACT:** Despiste having an important weight in the national GDP, the production of beans suffers considerable losses 'inside and outside the gate' and one of the bottlenecks of these losses is in the storage of seeds. Because of this, the objective of this work was to evaluate some of the main indicators that can increase the commercial productivity of grains. Relative humidity, percentage germination, electrical conductivity, temperature, and the relationship between pests and diseases and the viability of normal seeds were evaluated. The seeds used were from the BRS cultivar. The evaluation was carried out in a specific seed laboratory at PUC, at intervals of 50 days from the first evaluation to the second, this experiment being done in triplicate. For electrical conductivity and temperature, there were significant differences between the evaluations in the samples. As for the germination index, there were no significant differences at 5% probability applied in the Tukey test.
**KEYWORDS:** electrical conductivity, relative humidity, *Phaseolus vulgaris*, germination.

# QUALIDADE DE SEMENTES DE FEIJÃO COM DIFERENTES UMIDADES DE ARMAZENAMENTO

**RESUMO:** Mesmo tendo um peso importante no PIB nacional, a produção de feijão sofre perdas consideráveis ‘dentro e fora da porteira’ e um dos gargalos destas perdas está no armazenamento das sementes. Em razão disto objetivou-se, com este trabalho, avaliar alguns dos principais indicadores que possam aumentar a produtividade comercial dos grãos. Foram avaliados a umidade relativa, a germinação em porcentagem, a condutividade elétrica, a temperatura, a relação entre pragas e doenças e a viabilidade das sementes normais. As sementes utilizadas foram do cultivar BRS. A avaliação foi realizada em um laboratório especifico de sementes da PUC, em intervalos de 50 dias da primeira avaliação com a segunda, sendo este experimento feito em triplicata. Para a condutividade elétrica e a temperatura, houve diferenças significativas entre as avaliações nas amostras. Já no índice de germinação, não se mostraram diferenças significativas a 5% de probabilidade aplicados no teste de Tukey.

**PALAVRAS-CHAVE:** condutividade elétrica, umidade relativa, *Phaseolus vulgaris*, germinação.

## INTRODUCTION

Agribusiness has a great prominence in the indicators of the Gross Domestic Product (GDP) IBGE (2019), CONAB (2020), reported in its monthly-report that the national production is of 638.5 thousand tons for the common beans, 271.8 thousand tons of black common beans and 418.1 thousand tons of cowpea (CONAB 2020). Paraná is the largest producer of beans in Brazil with an area of 120 thousand hectares, with a production of 223 thousand tons per year, being produced from cowpea to black beans (CONAB 2020).

The southwestern region of Paraná accounts for more than 50% of the planted area, with the municipality of Ponta Grossa (PR) standing out in comparison to other producing regions, according to a report by Diário dos Campos (2020) reported that the cost of production per hectare revolves around R $ 3,122.28, with a productivity of 2,000 kg ha$^{-1}$, obtaining a gross revenue of R $ 9,966.67. These values are different for each producer, as some have other costs, which can reduce the value of the final profitability.

Figure 1 shows an evolution in bean prices over the months of August 2018 to April 2020, approximately 2 years.

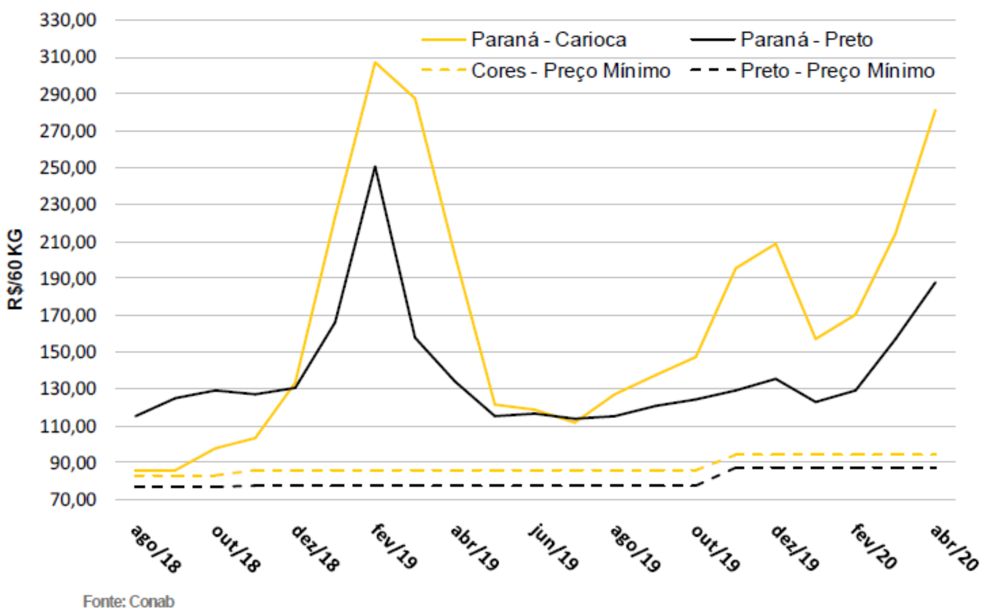



**Figure 1**. Prices received by producers in Paraná.

Beans (*Phaseolus vulgaris* L.) actively participate in these numbers (Diário dos Campos, 2020), as they are consumed by the Brazilian population from salads to special foods (Soratto et al. 2015). It is a great source of essential nutrients for human development; it has proteins, iron, calcium, magnesium, zinc, vitamins (mainly of the B complex), carbohydrates and fibers (Ferreira, 2015).

When it comes to feeding a section of the population without much income, beans stand out even more (Faroni et al., 2005). In this way, its amount of protein in its structure is high, so its consumption is high (Durigan et al., 1978).

CNABRASIL (2019), report that the daily bean consumption of beans is between 50 to 100 g per day / person, which generates 18% of proteins and 12% of calories ingested. This is because it is able to replace animal protein, as its cost is lower in the face of population inflation.

All bean production processes have their economic importance, but one stands out before other cultural treatments, such as seed production (Brackmann et al., 2002). As it is a culture extremely susceptible to diseases Tavares et al., (2015), so the post-harvest cultural treatment process is essential (Cardoso et al., 2012), in general, its storage model is not comparable with that of soybeans, corn etc., (Mota, 2016). In these crops, warehouses have large structures that monitor temperature, humidity, etc., this is no longer the case with beans (Silva et al. 2014).

Zucareli et al. (2015), demonstrate that the use of good quality seeds is essential for the proper establishment of a crop. For a more precise analysis of the quality of the seeds, it is necessary to complement the information that is provided by the 'seeders', such as the germination test, of vigor, which allows selecting the best lots for commercialization and planting, among these tests, highlights electrical conductivity (Vieira et al., 1996).

Two important tests are applied when storing cultures, such as the vigor test and the electrical conductivity test. These, in conjunction with the tetrazolium test, are considered models of biochemical processes, as both analyze the integrity of the cell membrane system (Brackmann et al., 2002). The germination analysis process is usually carried out in conjunction with other similar tests, with the objective of obtaining important information to determine seed indicators for planting (geographic location, percentage of protein, etc.) and thereby separating seeds by lots (Filho, 2005).

In conjunction with the tests described above, there is a quick and effective moisture test, which is applied by the Expedited Method. This method assesses the electrical properties of the material, and is directly dependent on moisture (Marquez, 2002).

In view of the above, the present study aims to study the methodology of germination tests, electrical conductivity and seed moisture, aiming to make them more efficient to differentiate bean seed lots in terms of physiological potential.

## MATERIALS AND METHODS

The present work was carried out in two stages, at the Seed Laboratory of the Pontifical Catholic University (PUC), campus of Toledo / PR. In the first stage, procedures for conducting electrical conductivity tests, seed moisture determination tests by the greenhouse method and by the Expedited Method and the germination test were studied.

In a second stage, also carried out in the seed laboratory of the PUC campus of Toledo, the evaluations were carried out in the same way as was done in the first stage and then the most efficient procedures were compared for the execution of each test, selected based on the data obtained in the first stage.

The methodology of this work was divided into topics, which are shown below.

A grain sample (1.5 kg) was taken, divided into 3 parts and three levels of moisture were established in the grains: 10, 13 and 16%; the samples were composed of 500 grams, then the initial moisture content of the grains was determined and 50 seeds were separated to test the germination and vigor (CE) together with this analysis, a pest/fungus survey was carried out.

The seeds that were not used were stored to perform the second evaluation in specific storage paper packages and at room temperature. The seeds went through stages, both to increase and to decrease humidity. BRS beans seeds, germ test paper, gerbox boxes, 8-digit digital scale, universal humidity meter, germination chamber, continuous flow oven, conductivity meter (Salinometer AT 255 with graphite electrode) and deionized water were used.

*Electrical conductivity methodology*

Fifty bean seeds were weighed on a four-digit precision scale (Bel Analytical Balance 0.0001g, 210g with automatic M214AIH calibration) and placed in plastic cups containing 75 mL of deionized water, placed in a germination chamber at 25 ° C, for 24 hours so that the seeds could germinate. After this period, the electrical conductivity of the seeds was read with a conductivity meter (DIGIMED DM 31), with units measured in $\mu S\ cm^{-1}\ g^{-1}$ of seeds.

*Greenhouse method at 105ºC*

Grain moisture was determined by the greenhouse method at a high temperature that remained constant, according to the Rules for Seed Analysis (Brasil, 2009). Two sub-samples of 5g wet weight were used, which were placed in greenhouses at a constant temperature of 105ºC, for a period of 24 hours. The sub-samples were re-weighed after drying.

The moisture content of the seed was calculated using the following formula:

$$\%U = \frac{WW - DW}{WW - T} x100 \qquad (1)$$

where:

WW = wet weight + container;

DW = dry weight + container;

T = tare (container).

*Expedite Method: use of appliances - electrical method*

Measurement of the electrical properties of the material depending on the humidity where the product, that is, bean seed is compressed between two electrodes and the measured electrical resistance is translated into a moisture reading, measures humidity accurately between 8 and 22%.

*Germination test methodology*

In this test, the methodology was used according to the rules for Seed Analysis (Brazil, 1992). 50 seeds were separated per lot, with the three replicates 150 seeds were used. The substrate used was the paper towel roll (germitest paper). Three sheets of paper were placed, two on the base and one on the cover. The paper was previously moistened with distilled water in an amount equivalent to three times the weight of the substrate. Germinators were used at a temperature of 25ºC and RH close to 100%. The evaluations were made on the fifth day after sowing in the laboratory and the result expressed and average percentage of normal seedlings, according to (Vieira e Krzyzanowski, 1999).

*Statistical analysis*

The Tukey test at 5% probability was used on the samples, with ANOVA analysis and applying second order polynomial regression analyzes.

## RESULTS AND DISCUSSION

Table 1 lists the evaluated parameters of a bean cultivar, subjected to conductivity tests (24 hours of soaking) and humidity of the 1st evaluation performed on August 14th and the 2nd evaluation performed 45 days later.

**Table 1**. Evaluation of the physiological potential of seeds by determining different degrees of humidity, electrical conductivity test observed CEo and corrected CE (µS $cm^{-1}$ $g^{-1}$) of beans from 2012/13 crop and germination potential.

| | RH[1] (%) | G[2] (%) | EC[3] (µS.$cm^{-1}$ $g^{-1}$) | T[4] (°C) | P/D[5] | Dead[6] | AA[7] |
|---|---|---|---|---|---|---|---|
| **1° Evaluation** | 10 | 48 | 67,06 | 22,40 | 0,0 | 1,0 | 1,0 |
| **1° Evaluation** | 13 | 40 | 81,62 | 24,10 | 3,0 | 2,0 | 7,0 |
| **1° Evaluation** | 16 | 38 | 82,06 | 24,00 | 2,0 | 7,0 | 5,0 |
| **Average 1° ev.** | - | 42[a] | 76,91[a] | 23,50[b] | 1,7 | 3,3 | 4,3[a] |
| **2° Evaluation** | 10 | 40 | 90,58 | 25,60 | 6,0 | 4,0 | 6,0 |
| **2° Evaluation** | 13 | 38 | 100,05 | 25,30 | 4,0 | 4,0 | 8,0 |
| **2° Evaluation** | 16 | 30 | 99,48 | 25,50 | 10,0 | 6,0 | 4,0 |
| **Média 2° Av.** | - | 36[a] | 96,70[b] | 25,47[a] | 6,7 | 4,7 | 6,0[b] |
| **Average 2° ev.** | - | 39 | 86,80 | 24,48 | 4,2 | 6,1 | 5,2 |
| **CV (%)** | - | 8,7 | 9,3 | 6,2 | - | - | 13,4 |
| ***p-value*** | - | 0,2372 | 0,02703 | 0,02431 | - | - | 0,4734 |

[1]**RH:** relative humidity; [2]**G:** germination; [3]**EC:** corrected electrical conductivity; [4]**T:** temperature; [5]**P/D:** pests and diseases; [7]**AA:** abnormals;

In view of this Table, we can observe that, when the humidity was at 10%, the respiratory process remained low, thus prolonging the quality of the seed. However, when it increased to 13 and 16% humidity, the breathing process accelerated, where there was a great deterioration of the evaluated product. Another factor that may have affected these indicators are injuries caused by the harvest and in the post-harvest period, as it is considered an extremely sensitive seed.

When the values of incidence of pests/diseases were analyzed, it resulted in attacks directly proportional to the moisture applied to the beans, this can be explained by the improvement of the growth environment of insects and mainly of diseases.

Faroni et al. (2005), in a bean harvest, in which the humidity was above 18%, presented the best classification by type, while the harvested grains, with 20.6 and 18.7%, indicated higher physiological quality than the harvested grains dry. When taking these grains to air storage, the result showed no significant differences in classification or color, but there was a significant reduction in their physiological potential.

Figures 2a and 2b show the polynomial regression curves in which it was possible to evaluate the direct response of moisture in relation to germination.

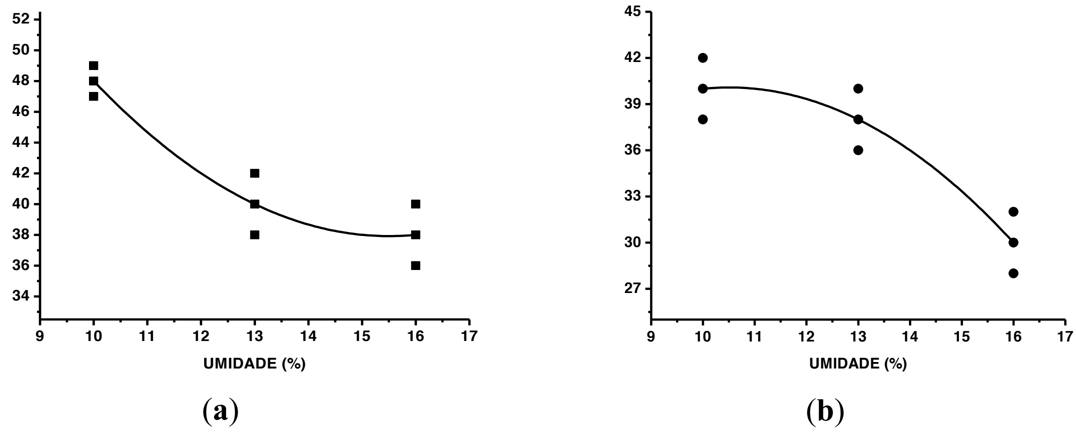



**Figure 2.** Second order polynomial regression graphs of the bean seed germination index in relation to the moisture rates of 10, 13 and 16%. (a) Graph of bean germination index in the 1st evaluation; (b) Graph of bean germination index in the 2nd evaluation.

Table 1 presented the results of germination in which there was no statistical difference between treatments, that is, the 1st and 2nd evaluations showed different values among themselves in the average of the three evaluations. Kappes et al. (2012), evaluating the germination of beans with the application of desiccants, obtained non-significant responses before the witness.

From the 1st to the 2nd evaluation, there was a 14.5% reduction in seed germination in the three moisture variables (10; 13 and 16%) (Table 1). Working with beans in positive pressure storage, Barbosa et al. (2010) observed a great loss of vigor of soybean seeds in the first six months of storage with a relative humidity of 60%.

Now, storing crambe seeds, Cardoso et al. (2012) found a reduction in germination values during the first nine months of storage. It can be compared to the results found in this research. In the second evaluation, the germination results were lower than in the first.

For the same bean cultivar analyzed, within a period of 10 to 15 days, the germination power decreased by approximately 14.5% during storage. What may have occurred was a loss in the integrity of the cell membranes of these seeds. Dias et al. (2012) and Lima et al. (2011) also pointed out this reduction in the germinative power in bean seeds with the increase in humidity, when stored in a specific location.

In addition to analysis of variance, regression analysis is an important tool for obtaining statistically effective responses, as it is formed by a set of algorithm methodologies that analyze and interpret the relationship between variables with good approximation, increasing or decreasing the relationship between the independent variables and the dependent variable (Abbad e Torres 2002).

Figures 2a and 2b, represent second order polynomial regression curves. The equations for each curve are represented on them.

Equation 2, mathematically models the germination index regression curve of the first evaluation in beans, together with its regression coefficient ($R^2$). In Equation 3, the mathematical model of the germination index regression curve of the second evaluation in beans is presented, with its regression coefficient ($R^2$).

$$G(\%) = 0{,}13608RH^2 + 3{,}54599RH + 23{,}39957 \; R^2 = 0{,}87 \quad (2)$$

Where:

**G(%):** germination rate in percentage (dependent variable);

**RH (%):** percentage relative humidity (independent variable).

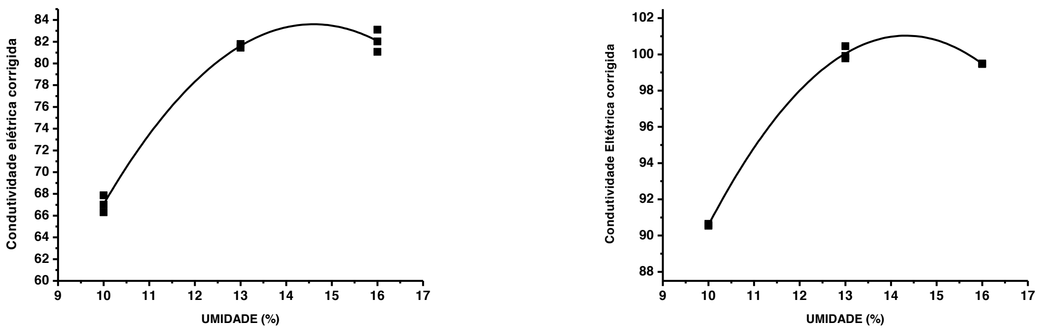
$$G(\%) = 0{,}15713UM^2 + 4{,}09458RH + 25{,}8648 \; R^2 = 0{,}83 \quad (3)$$

Where:

**G(%):** germination rate in percentage (dependent variable);

**RH (%):** percentage relative humidity (independent variable).

Figures 3a and 3b show the polynomial regression curves, in which it was possible to evaluate the direct response of humidity in relation to electrical conductivity.




**(a)** **(b)**

**Figure 3.** Second order polynomial regression graphs of the electrical conductivity of the bean seed in relation to the moisture rates of 10, 13 and 16%. (a) Graph of the conductivity of beans in the 1st evaluation; (b) Graph of the conductivity of beans in the 2nd evaluation.

The electrical conductivity was significantly influenced by the interaction with moisture in the seed. In (Figure 3.a), the increase was significant until the humidity of 13%, with a maximum peak of 83 in electrical conductivity. In (Figure 3.b), the growth was also exponential, but with a maximum peak of electrical conductivity at 13% humidity under a significant percentage difference (26.0%).

Zuchi et al. (2013), evaluating the electrical conductivity of soybean seeds, they were able to measure an increase in the release of electrolytes from the seeds, this generates a reduction in the physical and physiological characteristics of the seeds, the same occurred in the beans seeds at a humidity above 10%.

Vieira e Krzyzanowsky (1999), mention that the conductivity test has a great role in the functional analysis of the seed, evaluating its vigor and its physiological quality. According to Table 1, the conductivity values increased in the first evaluation, with an increase in humidity. In the second evaluation, the results were close to the maximum required.

On the other hand, Santos et al. (2005), report that this test (the electrical conductivity test), can be considered a negative point for the physiological state of the seed. This is because it can lose the internal structures of the membranes, which can generate a weakness in the defense of pathogens.

The increase in conductivity values in the first assessment can be corroborated with the results of Costa et al. (2010) who, evaluated in several grains, that the conductivity is directly proportional in relation to the humidity, that is, the higher the humidity the greater the conductivity.

Figures 3a and 3b show the second order polynomial regression curves, in which the equations for each curve are presented.

Equation 4, mathematical model of the electrical conductivity regression curve of the first evaluation on beans, together with its regression coefficient ($R^2$). In Equation 5, it represents the mathematical model of the electrical conductivity regression curve of the second evaluation in beans, with its regression coefficient ($R^2$).

$$CE = 0{,}774RH^2 - 20{,}95RH - 71{,}26 \quad R^2 = 0{,}98 \qquad (4)$$

Where:

**EC:** electrical conductivity (dependent variable);

**RH** (%): percentage relative humidity (independent variable).

$$EC(\%) = 0{,}5659RH^2 - 15{,}375RH + 15{,}743 \quad R^2 = 0{,}99 \qquad (5)$$

Where:

**EC:** electrical conductivity (dependent variable);

**RH** (%): percentage relative humidity (independent variable).

Ferreira (2018) mentions that the determination coefficient R2 has important characteristics to evaluate a statistical result. In this evaluation, he determined that the values of $R^2$ must be between 0 and 1. If the value of $R^2$ is 1, the regression equation is 100% related to the dependent variable as a function of the independent variable. Table 2 presents the moisture data verified in two stages, one at the beginning of the work and the other at the end, together with the other assessments.

**Table 2.** Determination of humidity by the Expedito Method (electrical method).

| [1]$MA_{me}$ (%) | 1° Evaluation | 2° Evaluation |
|---|---|---|
| **10%** | 10,5 | 11,1 |
| **13%** | 13,4 | 12,8 |
| **16%** | 15,5 | 12,8 |
| **Average** | 13,3[a] | 12,2[a] |
| **CV (%)** | 12,3 | |
| ***p-value*** | 0,5940 | |

[1]**$MA_{me}$ (%):** moisture analyzed by the expedited method; **CV (%):** coefficient of variation.

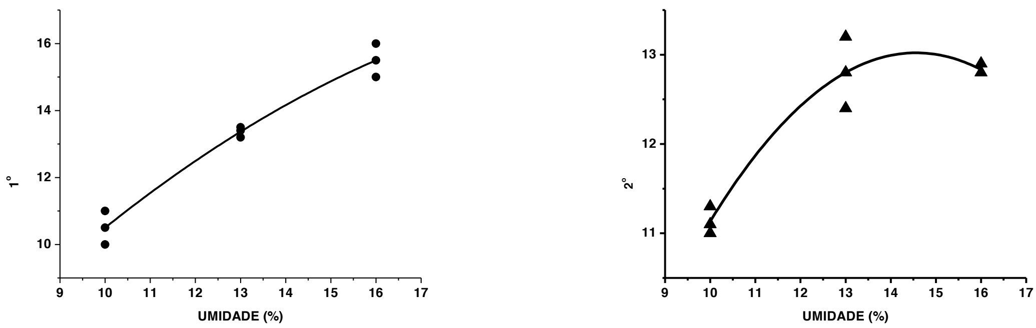
One of the factors that has caused the increase in the degree of humidity would be the effect of hygroscopicity (property that certain materials have to absorb water), of seed with the environment, as well as the attack of woodworms. According to Lazzari (2005), after a period of 30 days of storage, bean seeds infested with woodworms, had 1% in the degree of increased humidity.

The degree of moisture in the seeds is a very important factor in the conservation of the physiological state of the grain (grains and seeds) (Pinto et al., 2007). Amarante et al. (2007), when the humidity is low, the respiratory rate is decreased and the metabolism is also reduced, but if there is high humidity the process is reversed, causing serious damage, such as grain deterioration, decreased vigor and even the presence of pests.

Thus, it is noted the great importance of keeping seeds in controlled environments, because, in addition to maintaining their physical integrity for several years, it does not lose its viability and is still protected against stored grain pests (Catão et al., 2010).

Grain moisture (Table 2), determined by the difference in initial moisture, was not statistically different between the 10, 13 and 16% samples, by the expedite method, between the first and the second evaluation. The first evaluation showed better results with values above 2 percentage points in relation to the second evaluation.

Figures 4a and 4b show the polynomial regression curves, in which the direct response of humidity in relation to humidity can be evaluated by the expedite method.




(a) (b)

**Figure 4.** Polynomial regression graphs of second order of moisture by the expeditious method of bean seed in relation to humidity rates of 10, 13 and 16%. (a) Graph of humidity by the expeditious method of beans in the 1st evaluation; (b) Graph of humidity by the expedite method of beans in the 2nd evaluation.

Equation 6, mathematical model of the moisture regression curve calculated by the Expedito method of the first evaluation on beans, together with its regression coefficient ($R^2$). Equation 7 presents the mathematical model of the moisture regression curve calculated by the expeditious method in the second evaluation on beans, with its regression coefficient ($R^2$).

$$EC = 9{,}7468RH^2 - 1{,}0246RH + 1{,}1023 \quad R^2 = 0{,}96 \tag{4}$$

Where:

**EC:** electrical conductivity (dependent variable);

**RH** (%): percentage relative humidity (independent variable).

$$EC = 0{,}11034RH^2 - 1{,}116RH + 9{,}435 \quad R^2 = 0{,}91 \tag{5}$$

Where:

**EC:** electrical conductivity (dependent variable);

**RH** (%): percentage relative humidity (independent variable).

## CONCLUSIONS

With this work, we can conclude that the initial water content of the bean seeds can influence the interpretation of the results of the electrical conductivity test and germination tests.

The correct storage of seeds can maintain both their physical integrity and genetic characteristics important for the maintenance of species and prevention against attack by pests and insects.

Therefore, attention must be paid to the correct storage of seeds, maintaining an ideal temperature, controlled humidity and control of pests and diseases.

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