

## Original Research Article

# Physiological quality of rice seeds stored in different environments and packages

### ABSTRACT

**Aims:** Evaluate the physiological quality of rice seeds during storage in different packages and environments, identifying the best condition to conserve the seeds.

**Study design:** Completely randomized with four replicates.

**Place and Duration of Study:** Plant Science Laboratory and Genetics Laboratory of the Federal University of Piauí, Bom Jesus-PI, from April to October 2016.

**Methodology:** Seeds of rice were stored for 180 days in plastic bottles, kraft paper packaging, trifoliate paper packaging and polyethylene bags in cold and humid chamber (10 °C and 85% RH), in a temperature controlled environment (25 °C) and in an uncontrolled environment (21-36.4°C and 30-67% RH). The water content and vigor characteristics (first counting germination and germination speed index and electrical conductivity) were evaluated before (control), and at 90 and 180 days of storage, additional tests were also evaluated during these periods the Absolute growth rate of the root (AGRR), Absolute growth rate of the aerial part (AGRAP), Relative growth rate of the root (RGRR) and Relative growth rate of the aerial part (RGRAP).

**Results:** The values of AGRAP and RGRAP were reduced, differing from the others, for the trifoliate paper packages (0.168 cm day<sup>-1</sup>) and polyethylene bag (0.168 cm dia<sup>-1</sup>) in environment 1 at 180 days of storage. The plastic bottles presented the highest values for the characters 1<sup>st</sup> count (%) and germination (%) in all environments and storage periods. At the end of the storage (180 days) the environment with temperature control was the one that presented the best results of 1<sup>st</sup> counting of germination (%) and germination (%). At 90 and 180 days, the plastic bottles presented higher Germination speed index. The environment with temperature control presented better results for electrical conductivity (μS cm<sup>-1</sup> g<sup>-1</sup>).

**Conclusion:** The environment with temperature control is efficient in storing rice seeds for 180 days using a plastic bottle package. During storage, the greatest changes in the physiological quality of rice seeds are verified in the cold and humid chamber environment and in the polyethylene bag package.

**Keywords:** deterioration; viability; vigor; *Oryza sativa* L.

## 1. INTRODUCTION

Rice, *Oryza sativa* (L.), is the third largest cereal crop in the world, being surpassed by corn and wheat (Storck 2004). Rice is considered to be the most socioeconomically important product in many developing countries, serving as a staple food for about 2.4 billion people, especially for low-income population (Kenndy & Burligame 2003).

Rice grains harvest occurs seasonally during the year, which makes post-harvest stages important and determinant to maintain seed quality. During storage, there are several factors that interfere with seed quality, starting with the control of physical-chemical, biochemical and metabolic reactions (Aguar et al., 2012, Garcia & Benito 2006).

Temperature and relative humidity are the main factors that affect seed quality during storage (Smaniotto et al., 2014), having a direct relationship in the process of seed viability loss during storage and changes in the product quality, in contrast, by-products (Kong et al., 2008, Malaker et al., 2008). According to Cardoso et al. (2012), the deterioration process is inevitable but can be delayed depending on the storage conditions and the seed characteristics.

In addition to the environment, the packaging used in storage should help to reduce the rate of the deterioration process, keeping the initial seed moisture content stored in order to reduce

respiration (Tonin & Perez 2006). Seed deterioration is related to the characteristics of the packaging containers, depending on the greater or lesser ease of water vapor exchange between the seeds and the atmosphere (Marcos Filho 2015).

Therefore, the objective of this study was to evaluate the physiological quality of rice seeds during storage in different packages and environments, identifying the best condition to conserve the seeds.

## 2. MATERIAL AND METHODS

The experiment was conducted at the Plant Science Laboratory and Genetics Laboratory of the Federal University of Piauí, Bom Jesus-PI, from April to October 2016. Rice seeds, cultivar BRS Sertaneja were used.

The seeds were packed in plastic bottles, kraft paper packaging, trifoliate paper packaging and polyethylene bags, and then stored for a period of six months in a cold and humid chamber (10 °C and 85% RH), in a temperature controlled environment (25 °C) and in an uncontrolled environment (21-36.4 °C and 30-67% RH). Each package had, in average 0.2 kg of seeds. All packaging was properly sealed.

The water content and vigor characteristics (first counting, germination and germination speed index and electrical conductivity) were evaluated before (control) at 0 day, and at 90 and 180 days of storage. Seed samples from each treatment were submitted subjected to the evaluations, and the electrical conductivity and humidity test were not tested at 90 days. The following evaluations were performed:

Determination of the degree of relative humidity - the greenhouse method was used at 105 ± 3 °C for 24 h, using two replicates for each sample, according to the Rules for Seed Analysis (Brasil 2009).

Germination test - 200 seeds of each treatment using "Germitest" paper, moistened with distilled water, in a ratio of 2.5 times the weight of the paper. The paper with 50 seeds each were placed in a BOD-type germination chamber at 25 °C and photoperiod programmed for 16 hours, until the end of the test. The evaluation was performed 10 days after, and the normal seedlings were evaluated according to Brasil (2009).

First germination count - Performed together with the germination test, recording the number of normal seedlings 5 days after the test implementation according to Brasil (2009).

Germination speed index (GSI) - Carried out together with the germination test. The evaluations of the normal seedlings were performed daily, at the same time, from the 1<sup>st</sup> day until the 10th day. After obtaining the number of normal seedlings, the germination speed was calculated, using the formula indicated by Edmond & Drapala (1958).

Electrical conductivity (EC): Four subsamples of 50 seeds were weighed (0.001g precision) and placed in plastic cups containing 75 ml of deionized water at 25 °C for 24 hours. After this period, the electrical conductivity of the soaking solution was determined and the results expressed in  $\mu\text{S cm}^{-1} \text{ g}^{-1}$  of seed (AOSA, 1983).

Absolute growth rate of the root (AGRR) and Absolute growth rate of the aerial part (AGRAP) - was held with the germination test, measuring the root and shoot length of the seedlings on the 1st and 10th day, obtained from the equation:  $\text{TCA} = (\text{W2}-\text{W1}) / (\text{T2}-\text{T1})$ . Where, M2 = final length; M1 = initial length; T2 - T1 = time interval, with results expressed in  $\text{day}^{-1} \text{ cm}$ .

Relative growth rate of the root (RGRR) and Relative growth rate of the aerial part (RGRAP) - Performed together with the germination test, measuring the root and shoot length of the seedlings on the 1st and 10th day, obtained from the equation:  $\text{TCR} = (\ln \text{W2} - \ln \text{W1}) / (\text{T2} - \text{T1})$ , where:  $\ln$  is the logarithm neperian; Wl final length; M1 = initial length; T2 - T1 = time interval; with results expressed in  $\text{cm cm}^{-1} \text{ day}^{-1}$ .

The experimental design was completely randomized with four replicates, the experiment had different factorial schemes, and the characteristics AGRR, AGRAP, RGRR and RGRAP had 2x4x3 scheme being two storage periods, four packages and three environments following the mathematical model:

$$y_{izkj} = \mu + p_i + a_z + r_k + p_i a_z + p_i r_k + a_z r_k + p_i a_z r_k + e_{izkj} \text{ being:}$$

$y_{izkj}$  = observed in the plot corresponding to  $j$ -ésimo storage period of  $z$ -ésimo environment

inside of  $k$ -ésimo storage container  $j$ -ésima repetition;  $\mu$  = mean effect of the experiment  $p_i$  = effect

**Comment [R1]:** Mention the seed source. Selection of this rice cultivar for the study should be justified.

**Comment [R2]:** Indicate the thickness of the polythene bags.

**Comment [R3]:** Please provide the size of all these containers.

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**Comment [R5]:** Please explain clearly. Were the evaluations done at 0, 90 and 180 days of storage except for electrical conductivity and humidity test at 90 days???

**Comment [R6]:** What is this? Is it the protocol for seed analysis?? This is missing in the reference list.

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**Comment [R7]:** Provide the formula as the reader cannot refer the original paper for each calculation.

**Comment [R8]:** Provide details how did you maintain the control for determining EC.

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**Comment [R11]:** Are these replicates for each packages? How many packages were there in total for the whole study?

**Comment [R12]:** Include the 0th day (control) in the factor for storage period.

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87 of *i-ésimo* storage period;  $a_z$  = effect of *z-ésimo* environment;  $r_k$  = effect of *k-ésimo* storage  
 88 container;  $p_i a_z$  = effect of the interaction between *i-ésimo* storage period with the *z-ésimo*  
 89 environment;  $p_i r_k$  = effect of the interaction between *i-ésimo* storage period with *k-ésimo* storage  
 90 container ;  $a_z r_k$  = effect of the interaction between *z-ésimo* environment with *k-ésimo* storage  
 91 container;  $p_i a_z r_k$  = effect of the triple interaction between *i-ésimo* storage period with *z-ésimo*  
 92 environment and *k-ésimo* storage container;  $e_{izkj}$  = effect of the error associated with observation of  
 93 the plot containing *i-ésimo* storage period of *z-ésimo* environment inside of *k-ésimo* storage container  
 94 of *j-ésima* repetition.

95 The germination test, first germination count and GIS had a 2x4x3 + 1 factorial experimental  
 96 design, two storage periods, four storage containers and three environments with one control  
 97 (characteristics evaluation before the application of the treatments) following the mathematical model :

98  $y_{izk(t)j} = \mu + T_t + p_{i(t)} + a_{z(t)} + r_{k(t)} + p_i a_{z(t)} + p_i r_{k(t)} + a_z r_{k(t)} + p_i a_z r_{k(t)} + e_{izk(t)j}$  being:

99  $y_{izk(t)j}$  = observed in the plot corresponding to *i-ésimo* storage period of *z-ésimo*  
 100 environment inside of *k-ésimo* storage container inside of *t-ésimo* type ( control or treatment) of *j-ésima*  
 101 repetition;  $\mu$  = mean effect of the experiment;  $p_{i(t)}$  = effect of *i-ésimo* storage period within  
 102 the type;  $a_{z(t)}$  = effect of *z-ésimo* environment inside the type;  $r_{k(t)}$  = effect of *k-ésimo* storage  
 103 container within the type;  $p_i a_{z(t)}$  = effect of the interaction between *i-ésimo* storage period with *z-ésimo*  
 104 environment;  $p_i r_k$  = effect of the interaction between *i-ésimo* storage period with *k-ésimo*  
 105 storage container within type;  $a_z r_{k(T)}$  = effect of the interaction between *z-ésimo* environment with *k-ésimo*  
 106 storage container within the type;  $p_i a_z r_{k(t)}$  = effect of triple interaction between *i-ésimo*  
 107 storage period with *z-ésimo* environment and *k-ésimo* storage container within the type;  $e_{izk(T)j}$  =  
 108 effect of the error associated with observation of the plot containing *i-ésimo* storage period in *z-ésimo*  
 109 environment inside of *k-ésimo* storage container of *j-ésima* repetition inside of the type.

110 The electrical conductivity character followed the experimental design of factorial 4x3 + 1,  
 111 with four storage containers and three environments with a control (characteristics evaluation before  
 112 the application of the treatments) following the mathematical model:

113  $y_{zk(t)j} = \mu + T_t + a_{z(t)} + r_{k(t)} + a_z r_{k(t)} + e_{zk(t)j}$  being:

114  $y_{izk(t)j}$  = observed in the plot corresponding to *z-ésimo* environment inside of *k-ésimo*  
 115 storage container inside of *t-ésimo* type (control or treatment) of *j-ésima* repetition;  $\mu$  = mean effect of  
 116 the experiment;  $a_{z(t)}$  = effect of *z-ésimo* environment inside of the type;  $r_{k(t)}$  = effect of *k-ésimo*  
 117 storage container inside of the type;  $a_z r_{k(T)}$  = interaction effect inside of *z-ésimo* environment with *k-ésimo*  
 118 storage container within type;  $e_{izk(T)j}$  = effect of the error associated with the observation of  
 119 the plot *z-ésimo* environment inside of *k-ésimo* storage container of *j-ésima* repetition within type.

120 After the preliminary analyzes, when the significance of the interaction was observed, the  
 121 necessary unfolding was performed and from these results the comparison between the adjusted  
 122 means of the treatments by the Tukey test at 5% of probability was obtained. All statistical analyzes  
 123 were performed with SAS® software (1999).

### 124 3. RESULTS AND DISCUSSION

125 The AGRR, AGRAP characteristics of the rice were highly significant for all the studied  
 126 factors, with triple interaction significance. Proving the effect on these characters of the period, the  
 127 container and storage environment of rice seeds (Table 2). The interaction between the factors  
 128 causes different behaviors of the periods according to the container and the environment; therefore,  
 129 there is no pattern that causes better systematic conditions.

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134 **Table 1. Moisture content (%) of rice seeds before and after storage of 180 days in different**  
 135 **packages and environments**

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| Packing / Storage Environment                 | Moisture content (%) |
|-----------------------------------------------|----------------------|
| <b>Witness Moisture</b> (Before Storage)      | 8,0                  |
| Plastic bottles / Cold and humid chamber      | 9,5                  |
| Plastic bottles / Temperature controlled      | 8,6                  |
| Plastic bottles / Uncontrolled environment    | 8,1                  |
| Kraft paper / Cold and humid chamber          | 9,5                  |
| Kraft paper / Temperature controlled          | 8,9                  |
| Kraft paper / Uncontrolled environment        | 8,9                  |
| Trifoliolate paper / Cold and humid chamber   | 9,8                  |
| Trifoliolate paper / Temperature controlled   | 8,6                  |
| Trifoliolate paper / Uncontrolled environment | 8,5                  |
| Polyethylene bags / Cold and humid chamber    | 10,1                 |
| Polyethylene bags / Temperature controlled    | 9,6                  |
| Polyethylene bags / Uncontrolled environment  | 9,1                  |

136  
 137 The values of AGRR and AGRAP decreased, differing from the others, for the trifoliolate and  
 138 polyethylene bags in cold and humid chamber at 180 days of storage (Table 4). These results agree  
 139 with statement that the longevity of seeds that are stored in different types of packaging may vary due  
 140 to water vapor exchange (Alves & Lin 2003).

141 Trifoliolate paper and polyethylene bag have characteristics that provide little or no resistance  
 142 to the water vapor exchanges of the seeds with the medium (Alves & Lin 2003), and because **they**  
 143 **seeds have a are** hygroscopic, **characteristic, that is, the ability to they exchange either absorb or**  
 144 **loose moisture with depending upon the environment air moisture within the container in which they**  
 145 **are stored age container are, the seed absorb or loose water** until they **are reach a stable with the**  
 146 **environment air moisture content with the air enclosed in the container** (Schauren 2011). This factor  
 147 causes an increase in deterioration, which is proven in the seed moisture content values stored in  
 148 these two types of packaging (Table 1), considering that Smaniotto et al. (2014) observed decreases**ed**  
 149 **in** seed vigor related to deterioration with increasing humidity, and according to Corte et al. (2010) this  
 150 deterioration **provides** slow**ser** seedling growth. It was also possible to observe that in the other  
 151 packages and environments, the means did not differ.

152 Depending on the storage conditions of the seeds, deterioration can be delayed or  
 153 accelerated. The seeds are **submitted subjected** to the **greatest respiratory activity**, increase the  
 154 wear and tear on the seeds' reserves, thus accelerating the deterioration process (Peske et al., 2006).

**Comment [R20]:** You should clearly mention when this happens. Is this during increased moisture levels??

155 It is likely that the temperature and RH of the storage environment contributed to the seed  
 156 deterioration when associated to these types of packages, since the cold and humid chamber  
 157 environment presented lower mean values of AGRR and AGRAP (Table 4), as Smaniotto (2014)  
 158 states that although the most important factors in seed conservation are not considered, temperature  
 159 and RH contribute significantly to their deterioration, affecting the speed of the biological processes.  
 160 The conditions of relative humidity and temperature during storage in which the seeds will obtain their  
 161 specific hygroscopic equilibrium, will determine the maintenance of their physiological quality, for a  
 162 greater or lesser time (Borges et al. 2009).

163 At 180 days of storage, AGRR values were higher than those observed at 90 days (Table 4),  
 164 indicating that the storage period influenced the growth of rice seedlings.

165 The AGRR character was not affected by any of the factors tested in rice seeds (Table 2)  
 166 showing that storage period, container and environment do not directly interfere with this character.  
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**Table 2. Summary of variance analysis of Absolute growth rate of the root (AGRR) and Absolute growth rate of the aerial part (AGRAP), Relative growth rate of the root (RGRR) and Relative growth rate of the aerial part (RGRAP) in different forms of seed storage rice**

| VF                  | DF | Mean square         |         |                      |          |
|---------------------|----|---------------------|---------|----------------------|----------|
|                     |    | AGRR                | AGRAP   | RGRR                 | RGRAP    |
| Storage period (SP) | 1  | 0.241**             | 0.084** | 0.001 <sup>ns</sup>  | 0.0095** |
| Packaging (P)       | 3  | 0.031 <sup>ns</sup> | 0.019** | 0.002 <sup>ns</sup>  | 0.0025** |
| Environment (E)     | 2  | 0.034 <sup>ns</sup> | 0.013*  | 0.004 <sup>ns</sup>  | 0.0037** |
| SP x P (T)          | 3  | 0.003 <sup>ns</sup> | 0.012*  | 0.0005 <sup>ns</sup> | 0.0020** |
| SP x E (T)          | 2  | 0.047 <sup>ns</sup> | 0.063** | 0.004 <sup>ns</sup>  | 0.0078** |
| P x E (T)           | 6  | 0.040 <sup>ns</sup> | 0.045** | 0.002 <sup>ns</sup>  | 0.0042** |
| SP x P x E (T)      | 6  | 0.045 <sup>ns</sup> | 0.016** | 0.003 <sup>ns</sup>  | 0.0023** |
| Error               | 95 | 0.028               | 0.004   | 0.002                | 0.0005   |
| <b>CV%</b>          |    | 26.22               | 16.52   | 18.74                | 13.63    |
| <b>Mean</b>         |    | 0.642               | 0.384   | 0.207                | 0.162    |

*N<sup>ns</sup>, \*, \*\* not significant, significant at 5 and 1%, respectively, of probability by the test F.*

In general, the plastic bottle package presented the highest values for the 1<sup>st</sup> germination count (%) and germination (%) characters in all environments and storage periods. On the other hand, trifoliate paper and polyethylene bag stored in the cold and humid chamber at 180 days had the lowest values (Table 7), as shown in the AGRAP and RGRAP variables. Da Silva et al. (2010) working with the storage of rice, maize and beans seeds, with different packages, concluded that, regardless of the species, the plastic bottle package presented better values of 1st count (%) and germination (%), this was due to the seed moisture content that were stored in the permeable packages, in which they suffered a greater influence of the atmospheric conditions of the storage site, since this type of packaging does not offer any resistance to the water vapor exchanges of the seeds with the medium in which they are stored different than the impermeable, and this is shown in the values of moisture degree of the seeds in Table 1.

Therefore, the use of packaging that allows or not a certain water vapor exchange between the seed and the outside environment is important to avoid fluctuations in the degree of seed moisture. The reduction of metabolic activity, with a reduced deterioration rate, allows the maintenance of the physiological quality for a longer period (Cardoso et al., 2012).

The 1<sup>st</sup> germination count (%) at 180 days of storage, the lowest averages were observed in the uncontrolled environment (Table 4), proving the increase in the respiratory rate in the seeds, and according to Marini et al. (2013), this increase allows degenerative transformations, which can lead to the rapid consumption of their reserves, at the beginning of the germination process, reducing respiratory efficiency and, consequently, leading to less use in the synthesis processes, causing a decrease in germination and seed vigor. According to Mano (2012) the seed deterioration under high temperature conditions occurs from the interference of the metabolic activities of the cell, due to the oxidation of lipids, proteins and nucleic acids.

At the end of the storage (180 days), the temperature control environment showed the best results for the 1<sup>st</sup> germination count (%) and germination (%) (Table 4), due to the uniformity of the values, regardless of the type of packaging, different than the control, but absolute values presented a smaller decrease when compared to the other environments. A similar result was verified by Almeida et al. (2010), who emphasized that the physiological quality of stored seeds is better when kept in an environment where the temperature is controlled.

Seed packaging influenced the results of the germination speed index (Table 4). At 90 and 180 days, the plastic bottle package presented higher values, since this type of packaging can preserve or alter the minimum physiological characteristics of the seeds during storage, reducing the deterioration and according to Marini et al. (2013) provides a slower growth in the seedlings.

These results show that the type of packaging used while storing the seeds is important to preserve the physiological quality of the seeds, which agrees with Bessa (2015) and Carvalho et al. (2016).

Considering that the solute leakage from the seeds and, consequently, a higher electrical conductivity value, are associated with lower seed vigor (Machado et al., 2011, Marcos-Filho 2015),

makes it possible to confirm that the environment that provided the best results was with temperature control, with no significant difference between the packages used, including the control (Table 4). This result confirms the results obtained in the previous test.

**Table 3. Summary of analysis of variance of First germination count, Germination, Germination speed index (GSI) and Electrical conductivity in different forms of storage of rice seeds.**

| VF                  | DF      | Mean square                |             |         |                         |
|---------------------|---------|----------------------------|-------------|---------|-------------------------|
|                     |         | 1 <sup>st</sup> ger. Count | Germination | GSI     | Electrical conductivity |
| Treatment           | 24 (12) | 213.9**                    | 2097.4**    | 223.5** | 17,65*                  |
| Type (T)            | 1       | 1120.7**                   | 3432.0**    | 847.5** | 2,158 <sup>ns</sup>     |
| Storage period (SP) | 1       | 253.5**                    | 1998.4**    | 248.4** | -                       |
| Packaging (P)       | 3       | 232.6**                    | 3466.7**    | 385.7** | 12,15 <sup>ns</sup>     |
| Environment (E)     | 2       | 20.04 <sup>ns</sup>        | 2923.0**    | 141.7** | 5,355 <sup>ns</sup>     |
| SP x P (T)          | 3       | 163.4**                    | 1522.0**    | 138.0** | -                       |
| SP x E (T)          | 2       | 195.9**                    | 1577.6**    | 232.6** | -                       |
| P x E (T)           | 6       | 227.8**                    | 1685.0**    | 168.9** | 27,09**                 |
| SP x P x E (T)      | 6       | 128.9**                    | 1805.0**    | 156.0** | -                       |
| Error               | 75 (39) | 21.15                      | 70.16       | 5.474   | 6,967                   |
| CV%                 |         | 19.64                      | 13.78       | 14.88   | 12,20                   |
| Mean                |         | 11.60                      | 60.80       | 15.73   | 21,62                   |

*ns*, \*, \*\* not significant, significant at 5 and 1%, respectively, of probability by the test F.

**Table 4. Deployment of means of Absolute growth rate of the root, Absolute growth rate of the aerial part, Relative growth rate of the root, 1<sup>st</sup> germination count, Germination, Germination speed index and Electrical conductivity of rice seeds stored for a period of six months in a cold and humid chamber (1), in a temperature controlled environment (2) and in an uncontrolled environment (3)**

| Environment/<br>Packaging                                                 | 90 days               |                       |                       | 180 days              |                       |                       |
|---------------------------------------------------------------------------|-----------------------|-----------------------|-----------------------|-----------------------|-----------------------|-----------------------|
|                                                                           | 1                     | 2                     | 3                     | 1                     | 2                     | 3                     |
| Absolute growth rate of the root (cm day <sup>-1</sup> )                  |                       |                       |                       |                       |                       |                       |
| Plastic bottles                                                           | 0.487 Aa <sup>a</sup> | 0.401 Aa <sup>a</sup> | 0.401 Aa <sup>a</sup> | 0.435 Aa <sup>a</sup> | 0.369 Aa <sup>a</sup> | 0.380Aa <sup>a</sup>  |
| Kraft paper                                                               | 0.465 Aa <sup>a</sup> | 0.378 Aa <sup>a</sup> | 0.379 Aa <sup>a</sup> | 0.438 Aa <sup>a</sup> | 0.378 Aa <sup>a</sup> | 0.379Aa <sup>a</sup>  |
| Trifoliolate paper                                                        | 0.414 Aa <sup>a</sup> | 0.394 Aa <sup>a</sup> | 0.417 Aa              | 0.168 Bb <sup>b</sup> | 0.377 Aa <sup>a</sup> | 0.370Aa <sup>a</sup>  |
| Polyethylene bags                                                         | 0.399 Aa <sup>a</sup> | 0.420 Aa <sup>a</sup> | 0.405 Aa <sup>a</sup> | 0.081 Bb <sup>b</sup> | 0.453 Aa <sup>a</sup> | 0.424Aa <sup>a</sup>  |
| Absolute growth rate of the aerial part (cm day <sup>-1</sup> )           |                       |                       |                       |                       |                       |                       |
| Plastic bottles                                                           | 0.186 Aa <sup>a</sup> | 0.170 Aa <sup>a</sup> | 0.170 Aa <sup>a</sup> | 0.177 Aa <sup>a</sup> | 0.162 Aa <sup>a</sup> | 0.165 Aa <sup>a</sup> |
| Kraft paper                                                               | 0.182 Aa <sup>a</sup> | 0.165 Aa <sup>a</sup> | 0.165 Aa <sup>a</sup> | 0.177 Aa <sup>a</sup> | 0.165 Aa <sup>a</sup> | 0.165 Aa <sup>a</sup> |
| Trifoliolate paper                                                        | 0.172 Aa <sup>a</sup> | 0.167 Aa <sup>a</sup> | 0.173 Aa <sup>a</sup> | 0.076 Bb <sup>b</sup> | 0.163 Aa <sup>a</sup> | 0.163 Aa <sup>a</sup> |
| Polyethylene bags                                                         | 0.169 Aa <sup>a</sup> | 0.174 Aa <sup>a</sup> | 0.170 Aa <sup>a</sup> | 0.056 Bb <sup>b</sup> | 0.180 Aa <sup>a</sup> | 0.175 Aa <sup>a</sup> |
| Relative growth rate of the root (cm cm <sup>-1</sup> day <sup>-1</sup> ) |                       |                       |                       |                       |                       |                       |
|                                                                           | 0.592 <sup>b</sup>    |                       |                       | 0.692 <sup>a</sup>    |                       |                       |
| 1 <sup>st</sup> germination count (%)                                     |                       |                       |                       |                       |                       |                       |
| Witness                                                                   | 28.0 A                | 28.0 A                | 28.0 A                | 28.0 A                | 28.0 A                | 28.0 A                |
| Plastic bottles                                                           | 17.5 Ba <sup>a</sup>  | 10.5 Bb <sup>a</sup>  | 17.5 Ba <sup>a</sup>  | 23.0 Aa <sup>a</sup>  | 13.5 Bb <sup>a</sup>  | 9.5 Bb <sup>b</sup>   |
| Kraft paper                                                               | 4.0C Da <sup>b</sup>  | 6.5B Ca <sup>b</sup>  | 6.5 Da <sup>a</sup>   | 24.0 Aa <sup>a</sup>  | 13.0 Bb <sup>a</sup>  | 8.0 Bb <sup>a</sup>   |
| Trifoliolate paper                                                        | 10.0 Ca <sup>a</sup>  | 10.0 Ba <sup>b</sup>  | 14.0B Ca <sup>a</sup> | 0.5 Bc <sup>b</sup>   | 17.0 Ba <sup>a</sup>  | 10.0 Bb <sup>a</sup>  |
| Polyethylene bags                                                         | 1.5 Db <sup>a</sup>   | 3.5 Cb <sup>b</sup>   | 10.0 CDa <sup>a</sup> | 0.0 Bb <sup>a</sup>   | 19.0 Ba <sup>a</sup>  | 13.0 Ba <sup>a</sup>  |
| Germination (%)                                                           |                       |                       |                       |                       |                       |                       |

**Comment [R21]:** What is this??? Is this the control?? Change.

|                    |                       |                       |                       |                      |                      |                       |
|--------------------|-----------------------|-----------------------|-----------------------|----------------------|----------------------|-----------------------|
| Witness            | 89.5 A                | 89.5 A                | 89.5 A                | 89.5 A               | 89.5 A               | 89.5 A                |
| Plastic bottles    | 86.0 ABa <sup>a</sup> | 76.0 Ba <sup>a</sup>  | 86.5 Aa <sup>a</sup>  | 68.0 Ba <sup>b</sup> | 69.0 Ba <sup>a</sup> | 71.0 Ba <sup>b</sup>  |
| Kraft paper        | 43.5 Cb <sup>b</sup>  | 73.0 BCa <sup>a</sup> | 44.5 Cb <sup>a</sup>  | 75.5 Ba <sup>a</sup> | 59.5 Bb <sup>b</sup> | 55.5 Cb <sup>a</sup>  |
| Trifoliolate paper | 75.0 Ba <sup>a</sup>  | 62.0 Cb <sup>a</sup>  | 74.5 Ba <sup>a</sup>  | 3.0 Cc <sup>b</sup>  | 69.5 Ba <sup>a</sup> | 54.5 Cb <sup>b</sup>  |
| Polyethylene bags  | 33.5 Cb <sup>a</sup>  | 33.5 Db <sup>b</sup>  | 82.0 ABa <sup>a</sup> | 4.5 Cb <sup>b</sup>  | 70.5 Ba <sup>a</sup> | 60.0B Ca <sup>b</sup> |

#### Germination speed index (seeds day<sup>-1</sup>)

|                    |                      |                      |                      |                      |                       |                       |
|--------------------|----------------------|----------------------|----------------------|----------------------|-----------------------|-----------------------|
| Witness            | 29.9 A               | 29.9 A               | 29.9 A               | 29.9 A               | 29.9 A                | 29.9 A                |
| Plastic bottles    | 21.9 Ba <sup>a</sup> | 15.7 Bb <sup>b</sup> | 21.6 Ba <sup>a</sup> | 23.8 Ba <sup>b</sup> | 21.8 Bab <sup>a</sup> | 19.3 Bb <sup>a</sup>  |
| Kraft paper        | 7.4 Db <sup>b</sup>  | 13.2 Ba <sup>b</sup> | 9.6 Db <sup>b</sup>  | 24.5 Ba <sup>b</sup> | 18.2 Cb <sup>a</sup>  | 13.6 Dc <sup>a</sup>  |
| Trifoliolate paper | 16.5 Ca <sup>a</sup> | 12.5 Bb <sup>b</sup> | 17.4 Ca <sup>a</sup> | 1.01 Cc <sup>b</sup> | 22.03 Ba <sup>a</sup> | 15.5 CDb <sup>a</sup> |
| Polyethylene bags  | 5.3 Db <sup>a</sup>  | 5.7 Cb <sup>b</sup>  | 15.0 Ca <sup>a</sup> | 0.9 Cc <sup>b</sup>  | 22.8 Ba <sup>a</sup>  | 17.1 BCb <sup>a</sup> |

#### Electrical conductivity ( $\mu\text{S cm}^{-1} \text{ g}^{-1}$ ) at 180 days

|                    |           |          |            |
|--------------------|-----------|----------|------------|
| Witness            | 20,91 B   | 20,91 A  | 20,91 AB   |
| Plastic bottles    | 25,17 Aa  | 20,07 Ab | 23,24 ABab |
| Kraft paper        | 21,66 Aba | 21,51 Aa | 20,03 Ba   |
| Trifoliolate paper | 16,59 Cb  | 21,61 Aa | 23,70 Aa   |
| Polyethylene bags  | 21,24 Ba  | 23,08 Aa | 22,26 Aba  |

Means followed by the same uppercase letters in the columns, lowercase in the lines within the storage period and overwritten in the rows between periods within the same environment do not differ by the Tukey test at 5% probability; Means followed by the same uppercase letters in the columns, lowercase in the rows within the storage period and underwritten in rows between periods within the same environment do not differ among themselves by the Tukey test at 5% probability.

## 4. CONCLUSION

1. The environment with temperature control is efficient in storing rice seeds for 180 days using a plastic bottle package.
2. During storage, the greatest changes in the physiological quality of rice seeds are verified in the cold and humid chamber environment and in the polyethylene bag package.

## REFERENCES

- Aguiar, R. W. S. ; Brito, D.R. ; Ootani, M.A. ; Fidelis, R.R. ; Peluzio, J. N. Effect of carbon dioxide, temperature and storage on soybean seeds and associated mycoflora. Journal of Agronomy, vol. 43, n. 3, p. 554-560. 2012.
- Almeida, F.A.C. ; Jerônimo, E.S. ; Alves, N.M.C. ; Gomes, J.P. ; Silva, A.S. Study of techniques for the storage of five oilseeds under environmental and cryogenic conditions. Brazilian Journal of Agroindustrial Products, v.12, n.2, p.189-202. 2010
- Alves, A.C., and Lin, H.S. Type of packaging, initial moisture and storage period in bean seeds. Scientia Agraria, v.4, n.1-2, p.21-26. 2003.
- ASSOCIATION OF OFFICIAL SEED ANALYSTS. Seed vigor testing handbook. East Lansing. 88p. (AOSA, Contribution, 32). 1983.
- Bessa, J. F.V. ; Donadon, J.R. ; Resende, O. ; Alves, R.M.V. ; Sales, J.D.F. ; Costa, L.M. Storage of crambe in different packages and environments: Part I - Physiological quality. Brazilian Journal of Agricultural and Environmental Engineering, v.19, n.3, p.224-230. 2015.
- Borges, S. ; Borges, E.E. L. ; Corrêa, P. C. ; Brune, A. Hygroscopic equilibrium and viability of angico-red seeds (*Anadenanthera peregrina* (L.) Speng) under different environmental storage conditions. Scientia Forestalis, v.37, p.475-481.
- Cardoso, R.B. ; Binotti, F.F.S. ; Cardoso, E.D. Physiological potential of crambe seeds as a function of packaging and storage. Tropical Agriculture Research, v. 42, n.3, p.272-278. 2012.
- Carvalho, E. R. ; Oliveira, J.A. ; Mavaieie, D.P.R. ; da Silva, H. W. ; Lopes, C. G. M. Pre-packing cooling and types of packages in maintaining physiological quality of soybean seeds during storage. Journal of Seed Science, v.38, n.2, p.129-139. 2016.

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270 Corlett, F. M. F. ; Barros, A. C. S. A. ; Villela, F. A. Physiological quality of urucum seeds stored in  
 271 different environments and packaging. Revista Brasileira de Seentes, vol. 29, no. 2, p.148-158. 2007.  
 272 Silva, F. S. ; Porto, A. G. ; Pascuali, L.C. ; Da Silva, F. C. Feasibility in storing seeds in different  
 273 packages for small rural properties. Revista de Ciências Agro-Ambientais, Alta Floresta, v.8, n.1,  
 274 p.45- 56. 2010.  
 275 Edmond, J.B.; Drapala, W.J. The effects of temperature, sand and soil, and acetone on germination  
 276 of okra seed. Proceedings of the American Society for Horticultural Science, v.71, p.428-434. 1958.  
 277 Garcia, F. G.; Benito, M. E. G. Seed germination of five Helianthemum species: effect of temperature  
 278 and presowing treatments. Journal of Arid Environments, v. 65, n. 1, p. 688-693. 2006.  
 279 Kenndy, G.; Burligame, B. Analysis of food composition data on rice from a plant genetic resources  
 280 perspective. Food Chemistry, v.80, n.4, p.589 - 596. 2003.  
 281 Kong, F.; Chang, S. K. C.; Liu, Z.; Wilson, L. A. Changes of soybean quality during storage as related  
 282 to soymilk and tofu making. Journal of Food Science, v.73, n.3, p.134-144. 2008.  
 283 Machado, C.G.; Martins, C.C.; Santana, D.G.; Cruz, S.C.S.; Oliveira, S.S.C. Adequação do teste de  
 284 condutividade elétrica para sementes de *Pisum sativum* subsp *Arvense*. Ciência Rural, v.41, n.6,  
 285 p.988-995. 2011.  
 286 Malaker, P. K.; Mian, I. H.; Bhuiyan, K. A.; Akanda, A. M.; Reza, M. M. A. Effect of storage containers  
 287 and time on seed quality of wheat. Bangladesh Journal of Agricultural Research, v.33, n.3, p.469-477.  
 288 2008.  
 289 Mano J. Reactive carbonyl species: Their production from lipid peroxides, action in environmental  
 290 stress, and the detoxification mechanism. Scientia Resaerch Center, v 45, p 1–8. 2012.  
 291 Marcos-Filho, J. Physiology of seeds of cultivated plants. Londrina: ABRATES, 660 p. 2015.  
 292 Marini, P. ; Moraes, C. L. ; Larré, C. F. ; Lima, M. C. ; De Moraes, D. M. ; From Amarante, L.  
 293 Indicative of quality loss of rice seeds under different temperatures through enzymatic and respiratory  
 294 activity. Interciencia, vol. 38, no. 1, p. 54-59. 2013.  
 295 Peske, S.T. ; Lucca Filho, O.A. ; Barros, A.C.S.A. Seeds: Scientific and technological foundations.  
 296 2.ed. rev. e ampl. Pelotas: Ed. University  
 297 Smaniotto, T.A.S. ; Resende, O. ; Marçal, K.A. F. ; de Oliveira, D. E. C. ; Simon, G. A. Physiological  
 298 quality of soybean seeds stored under different conditions. Brazilian Journal of Agricultural and  
 299 Environmental Engineering, v.18, n.4, p.446-453. 2014.  
 300 Storck, C.R. Variation in chemical composition of rice grains submitted to different treatments.  
 301 Dissertation (Master in Science and Food Technology), Federal University of Santa Maria. 108 p.,  
 302 2004.  
 303 Tonin, G.A. ; Perez, S. C. J. G. A. Physiological quality of porous *Ocotea* porosa seeds (Nees et  
 304 Martius ex. Nees) seeds after different storage and seeding sowing conditions. Brazilian Journal of  
 305 Seeds, v. 28, n. 2, p. 26-33, 2006.  
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