

*Full Length Research Paper*

## Storage of crambe fruit subjected to different drying conditions

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**This study aimed to determine the effect of drying conditions and storage time on crambe seed quality. Crambe fruits with a moisture content of  $27.0 \pm 1.0\%$  (wet basis; w.b.) were used. These fruits were dried in a forced-air oven and were kept under controlled temperatures of 35, 45, 60, 75 and 90°C at relative humidities of 20.9, 8.7, 6.8, 4.8 and 2.3%, respectively. During the drying process, trays containing 0.4 kg of fruit were periodically weighed until the final drying point of  $7.0 \pm 1.3\%$  (w.b.). The fruits were packed in glass jars covered with permeable fabric. Each container had 400 g of fruit, and the containers were kept under ambient conditions (temperature of  $26 \pm 3^\circ\text{C}$  and relative humidity of  $55 \pm 12\%$ ). The temperature and relative humidity of the environment were recorded by a digital datalogger. The samples were evaluated at 0, 3, 6, 9 and 12 months for moisture content, electrical conductivity (EC), germination percentage and germination speed index (GSI). Drying the crambe fruit at high temperatures caused a decrease in the physiological quality of the seeds. There was an increase in the amount of electrolytes released by the seeds over time. Seeds stored for 12 months at room temperature had superior germination regardless of drying temperature.**

**Key words:** Physiological quality, *Crambe abyssinica*, biodiesel.

### INTRODUCTION

Biodiesel has proven to be a genuine alternative to replace fossil diesel oil. In Brazil, a proposal to replace fossil fuel for fuels obtained from biomass has existed since 1920. However, it was the oil crisis in the 1970s that led the federal government to create the National Alcohol Program (Programa Nacional do Álcool, Proálcool), which has made the replacement of gasoline with ethanol a reality. Tests performed with different mixing ratios of biodiesel in diesel fuel have shown viable

technical results. However, the decrease in oil prices and the high production cost of biodiesel compared to standard diesel have paralyzed the progress of the commercial use of biodiesel in Brazil and worldwide, but current problems related to the availability of affordable oil and its impacts on the environment have revived research into biodiesel around the world (Osaki and Batalha, 2008). Basic guidelines of the Brazilian Biodiesel Production Program aim to highlight Brazil in the production of renewable fuels, because the country has competitive advantages over other countries, such as land availability for the expansion of agriculture as well as enough water and energy to practice agriculture for energy production in a competitive manner (Benedetti et

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al., 2006).

Several oleaginous species are being tested or used for the production of biodiesel, such as soy, castor, palm, sunflower, peanut, crambe, oilseed radish, rapeseed and babassu (Mello et al., 2007).

Although, Brazil has a great diversity of agricultural products for the production of vegetable oils and biodiesel, many crops still have an extractive nature, with no commercial plantations to assess their real potential. Thus, crambe (*Crambe abyssinica*) is an option for oil extraction in the Brazilian Midwest.

In Brazil, studies related to culture and crambe oil production have been recently undertaken, because domestic producers and researchers have access to the FMS Brilhante cultivar, which is commercialized by the Mato Grosso do Sul (MS) Foundation. When evaluating the production potential of crambe as well as the physicochemical characteristics of the oil and biodiesel obtained from these grains, Jasper (2009) concluded that biodiesel from *C. abyssinica* Hochst complies with the standards established by Resolution No. 7 of the National Petroleum, Natural Gas and Biofuels Agency (Agência Nacional de Petróleo, Gás Natural e Biocombustíveis, ANP).

Crambe is a plant of the Brassicaceae family and is a potential source material for biofuel. The crambe fruit is a silique that is initially green, but has a yellowish color upon maturation, and each fruit contains a single spherical seed that is green or greenish brown (DESAI et al., 1997).

This crop has the advantages of high yield, tolerance to drought, tolerance to frost and cycle precocity, which lasts approximately 90 days. Due to these characteristics, crambe can be grown in the winter season if sown immediately after soybean harvest as an alternative to planting the second crop. However, this oleaginous crop will only be successful in Brazilian soil if the productivity levels are satisfactory. To this end, the use of superior quality seeds can significantly contribute to the achievement of production goals (Neves et al., 2007), and the proper drying and storage of the seeds can ensure the quality and performance of the crop in the field.

Crambe should be dried to no more than moisture content of 10% (wet basis, w.b.) before storage (Oplinger et al., 1991). The drying process partially removes water from the seed by the simultaneous transfer of heat from the air to the seed and by the transfer of mass through water vapor flow from the seed to the air. The drying process is a dynamic process and depends on the relative humidity of the air (Villela and Peres, 2004). In drying, air is used to transport water from the seeds out of the system. Forced hot air is used to heat the seeds, causing the internal water to migrate to the outer surface to be evaporated, thus reducing the moisture content of the mass of the seeds being dried (Peske and Villela, 2003).

In artificial drying, air absorbs water from the product in

the form of vapor at the same time that air supplies heat to the system. The seeds, which are hygroscopic, undergo changes in their moisture content according to environmental conditions. When the seeds come in contact with air, they perform exchanges until their vapor pressure and temperature have similar values reaching levels of energy, water and thermal balance. However, the temperature of the drying air must be controlled within certain limits, thus avoiding possible physicochemical and biological damage to the seeds (Elias, 2002).

Crambe seeds must be stored and commercialized with a moisture content of up to 10% (w.b.) (Knights, 2002) and cleaned to a maximum of 2% of impurities (Springdale, 2005). If the seeds are harvested with high moisture content, natural or forced ventilation can be used for drying. It is not recommended to dry the seeds with air without heat if the moisture content exceeds 20% (w.b.). To maintain the quality of the seed, the maximum recommended drying temperature is 43°C (Golz, 1993; Knights, 2002).

Crambe seeds are considered orthodox due to their physiological behavior during storage. The ability of the orthodox seed cells to survive desiccation involves the synthesis of proteins known as late embryogenesis abundant (LEA) proteins, which are produced in late embryogenesis, as well as the accumulation of soluble sugars and presence of antioxidants, which allow the cytoplasm to reach the so-called vitreous state (Berjak, 2006). Seeds with this behavior, as characterized by Roberts (1973), are tolerant to desiccation and can be stored with low moisture content in environments with low temperature and relative humidity.

Seed survival in storage essentially depends on the moisture content. This reliance can be attributed to the fact that physiological reactions increase quantitatively when the moisture content rises. The identification of adequate moisture content of the seed increases the storage capacity and decreases the risk of deterioration. The metabolic activity and damage caused by insects and microorganisms are influenced by the moisture content in the seed. Deteriorative reactions occur at high moisture contents and can be reduced with low water levels. However, studies have shown that important reactions occur more readily in a dry state than in a wet state and that the nature of these reactions varies with moisture content (Marcos Filho, 2008). Thus, this study aimed to analyze the effect of drying conditions and storage time on the quality of crambe fruit for up to 12 months.

## MATERIALS AND METHODS

Fruits of the FMS Brilhante cultivar were used, which was developed by the MS Foundation. The fruits were produced in the 2010 crop at the Experimental Field of the Federal Institute of Education, Science and Technology of Goiás, Rio Verde Campus (Instituto Federal de Educação, Ciência e Tecnologia Goiano - IF Goiano - Câmpus Rio Verde) located in Rio Verde-GO at 17° 47'

53° latitude (S) and 51° 55' 53" longitude (W). The experiment was conducted in the Post-Harvest Laboratory of Plant Products and Seed Laboratory (IF Goiano- Câmpus Rio Verde). The fruit harvesting was done manually when the moisture content was  $27.0 \pm 1.0\%$  (w.b.), which was determined by gravimetry using an oven at  $105 \pm 3^\circ\text{C}$  for 24 h (Brasil, 2009).

Crambe drying was performed in an oven with forced ventilation kept at the controlled temperatures of 35, 45, 60, 75 and  $90^\circ\text{C}$  and average relative humidities of 20.9, 8.7, 6.8, 4.8 and 2.3%, respectively. During the drying process, trays containing 0.4 kg of fruit were periodically weighed until it reached the final drying point of  $7.0 \pm 1.3\%$  (w.b.), which has been established for the safe storage of this product.

The temperature and relative humidity of the drying air were monitored by means of a psychrometer installed inside the experimental dryer. The fruits were packed in glass jars (400 g of fruit per jar) covered by voile fabric on October 10, 2010, and the jars were stored for 12 months. During this period, the temperature and relative humidity of the ambient air were recorded by a digital datalogger.

The samples (four replicates) were evaluated at 0, 3, 6, 9 and 12 months. The following parameters were measured: moisture content, electrical conductivity (EC), germination percentage and germination speed index (GSI).

The EC test was performed according to the methodology described by Vieira and Krzyzanowski (1999). A total of 4 subsamples of 50 fruits were used from each treatment, and the subsamples were weighed on a scale with a resolution of 0.01 g. The samples were placed in plastic cups to soak with 75 ml of deionized water and were kept in a biochemical oxygen demand (BOD) incubator with controlled temperature of  $25^\circ\text{C}$  for 24 h. The solutions containing the fruit were slightly shaken for leachate uniformity, which was immediately proceeded by a reading on a portable digital conductivity meter (Instrutherm model CD-850), and the results were divided by the mass of the 50 fruits and the result was expressed in  $\mu\text{S cm}^{-1} \text{g}^{-1}$  of seeds.

Germination test was conducted with four subsamples of 25 fruits from each treatment. The fruits were packed in Gerbox boxes on blotting paper moistened with distilled water, which was equivalent to 2.5 times the dry substrate mass, to achieve adequate moisture and uniformity of the test. The samples were kept in a Mangelsdorf germinator set at a constant temperature of  $25 \pm 2^\circ\text{C}$ . The evaluations were performed every two days from the second day after sowing until 32 days were completed according to the criteria established in the Rules for Seed Analysis (Brasil, 2009). The average germination percentage was calculated, and the GSI was calculated as follows:  $\text{GSI} = n_1 \cdot d_1^{-1} + n_2 \cdot d_2^{-1} + n_3 \cdot d_3^{-1} \dots n \cdot d_n^{-1}$ ; where  $n_1$  is the number of seeds germinated on the first day of counting;  $n_2$  is the number of seeds germinated on the second day of counting;  $n_3$  is the number of seeds germinated on the third day of counting;  $n_n$  is the number of seeds germinated on the  $n^{\text{th}}$  day of counting;  $d_1$  is the first day;  $d_2$  is the second day;  $d_3$  is the third day; and  $d_n$  is the  $n^{\text{th}}$  day (Maguiri, 1962).

The design was completely randomized according to a split plot design with five controlled drying temperatures (35, 45, 60, 75 and  $90^\circ\text{C}$ ) in the plots and five evaluation months (0, 3, 6, 9 and 12 months) in the subplots. The averages were compared by Tukey's test at a 5% significance level.

## RESULTS AND DISCUSSION

The drying times of the crambe fruit when considering the reduction of moisture content from  $27.0 \pm 1.0$  to  $7.0 \pm 1.3\%$  (w.b.) and with drying temperatures of 35, 45, 60, 75 and  $90^\circ\text{C}$  were 13.75, 7.75, 5, 3.75 and 3.26 h,

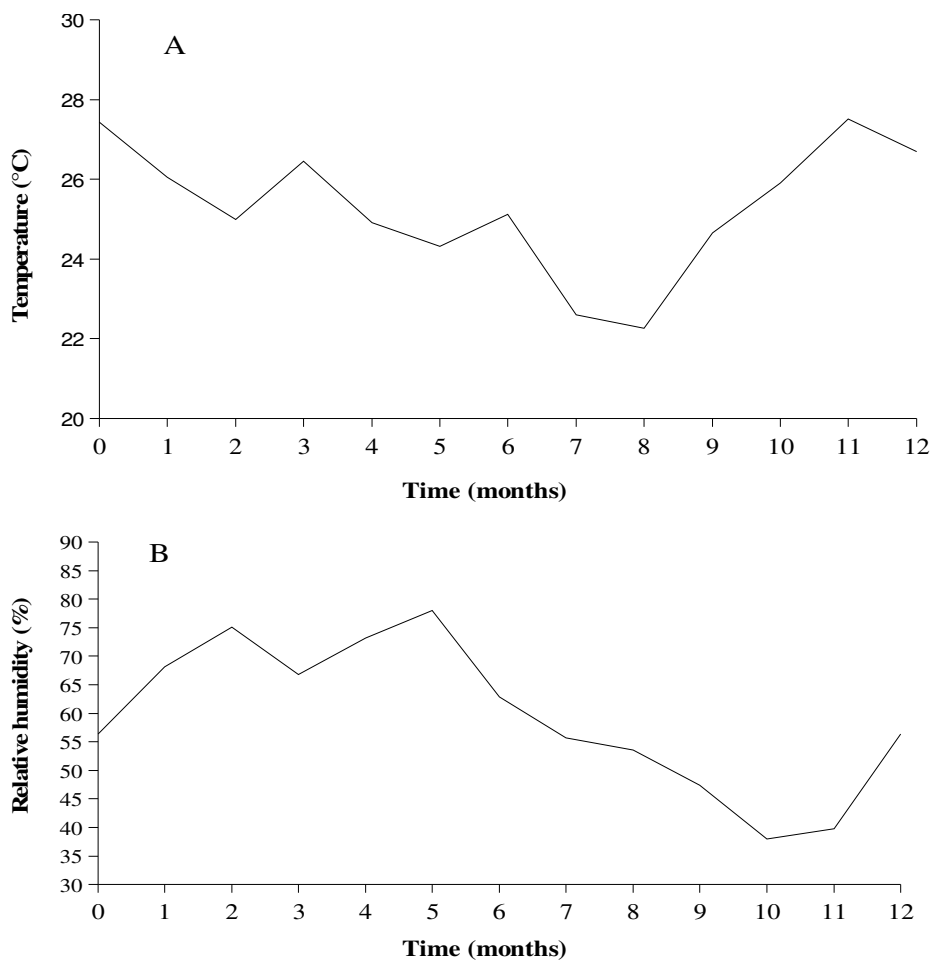
respectively. These values were similar to the *Jatropha* seed for drying temperature conditions of 30, 40, 50, 60 and  $70^\circ\text{C}$  (Ullmann et al., 2010). When drying crambe fruit at temperatures of 30, 40, 50, 60 and  $70^\circ\text{C}$ , Costa et al. (2011, 2012a) found that increasing the drying temperature increases the drying rate and decreases the time required for drying when considering a moisture content reduction from  $21.0 \pm 1.3$  to  $7.0 \pm 1.4\%$  (w.b.).

Figure 1 shows the average monthly values of temperature and relative humidity of the air where the crambe fruits were stored. The average temperature in storage was  $24.89 \pm 2.63^\circ\text{C}$  with fluctuations over time (Figure 1A). The relative humidity of the air during the first six months of storage from November to April was higher with an average of 70.69% (Figure 1B), and there was a decreasing trend in the relative humidity in the last months of storage from July to October with an average of 39.82%, due to the variability of the temperature along the year. The variation in the relative humidity and temperature caused changes in the moisture content of seeds, which can affect the physiological quality of the seeds.

Table 1 shows the analysis of variance of the evaluated characteristics. The drying temperature and time of storage influenced the physiological quality of the crambe fruit. Table 2 shows the values of moisture content in the crambe fruit under different drying temperatures during storage. The storage of the crambe fruit in glass containers covered with permeable fabric under ambient conditions at a temperature of  $26 \pm 3^\circ\text{C}$  and relative humidity of  $55 \pm 12\%$  allowed for variations in moisture content during storage. Changes in air conditions cause constant changes in this variable. At the end of the storage period, there was a decrease in moisture content for all drying temperatures due to the temperature and relative humidity of the air being moderately lower at this time of the year (July to October) (Figure 1B), which made the fruit reach equilibrium with the ambient conditions in which they were stored.

As indicated by the EC values shown in Table 3, there was an increase in the amount of electrolytes released by the seeds over the course of the storage period. The amount of electrolytes released tended to increase more substantially with the fruit that were dried at a temperature of  $90^\circ\text{C}$ , thereby confirming the influence of time and drying temperature on the amount of solutes leached into the solution.

At the beginning of storage, the crambe fruit showed low values of EC for the five drying conditions with values between 0.516 and  $0.610 \mu\text{S cm}^{-1} \text{g}^{-1}$ . At 12 months, however, the fruit that were dried at  $45^\circ\text{C}$  showed the lowest EC value, which was equivalent to the initial sample. These results indicated better preservation of the crambe fruit when dried at a temperature of  $45^\circ\text{C}$ , which probably kept the cell membranes better organized, thus controlling the release of solute during soaking. This coincides with the study of Knights (2002) in which it was recommended that the seed quality should maintain a



**Figure 1.** (A) Average temperature during storage at ambient conditions ( $24.89 \pm 2.63^\circ\text{C}$ ). (B) Average relative humidity during storage at ambient conditions ( $55 \pm 12\%$ ).

**Table 1.** Summary of the analysis of variance for moisture content, electrical conductivity (EC), germination and germination speed index (GSI) during storage of the crambe fruit.

Analyzed variable	Variation source	Average square	CV (%)
Moisture content (% w.b.)	Drying	0.0002**	8.04
	Months	0.0054**	8.05
	Drying $\times$ Months	0.0001**	
EC ( $\mu\text{S cm}^{-1} \text{g}^{-1}$ )	Drying	0.0257*	6.35
	Months	0.0922**	4.93
	Drying $\times$ Months	0.0483**	
Germination (%)	Drying	3354.9650**	25.51
	Months	2592.0650**	19.68
	Drying $\times$ Months	331.1775**	
GSI (dimensionless)	Drying	12.9207**	30.32
	Months	15.4527**	21.62
	Drying $\times$ Months	1.7461**	

\*\*Significant at 1% according to the F test. \*Significant at 5% according to the F test. <sup>NS</sup>, Not significant.

**Table 2.** Moisture content (% w.b.) of the crambe fruit subjected to different drying temperatures and during storage under ambient conditions during 12 months.

Drying (°C)	Storage time (months)				
	0	3	6	9	12
35	9.12 <sup>aB</sup>	8.35 <sup>aB</sup>	10.55 <sup>aA</sup>	7.03 <sup>aC</sup>	5.77 <sup>aD</sup>
45	9.23 <sup>aAB</sup>	8.37 <sup>aB</sup>	10.09 <sup>aA</sup>	6.97 <sup>aC</sup>	6.17 <sup>aC</sup>
60	7.43 <sup>bBC</sup>	8.26 <sup>aB</sup>	10.44 <sup>aA</sup>	6.85 <sup>aC</sup>	6.37 <sup>aC</sup>
75	7.63 <sup>bB</sup>	7.85 <sup>aB</sup>	10.48 <sup>aA</sup>	6.45 <sup>bC</sup>	5.76 <sup>aC</sup>
90	7.28 <sup>bB</sup>	8.15 <sup>aB</sup>	9.52 <sup>aA</sup>	6.97 <sup>aB</sup>	5.52 <sup>aC</sup>

The averages followed by the same lowercase letters in the column and uppercase letters in the rows do not differ according to Tukey's test at 5% significance.

**Table 3.** Electrical conductivity (EC;  $\mu\text{S cm}^{-1}\text{g}^{-1}$ ) of the crambe fruit subjected to different drying conditions during 12 months.

Drying (°C)	Storage time (months)				
	0	3	6	9	12
35	0.57 <sup>abcC</sup>	0.59 <sup>bBC</sup>	0.59 <sup>abBC</sup>	0.64 <sup>abAB</sup>	0.66 <sup>abA</sup>
45	0.61 <sup>aA</sup>	0.58 <sup>bAB</sup>	0.55 <sup>bB</sup>	0.60 <sup>bAB</sup>	0.60 <sup>bAB</sup>
60	0.58 <sup>abAB</sup>	0.59 <sup>bAB</sup>	0.53 <sup>bB</sup>	0.63 <sup>abA</sup>	0.63 <sup>abA</sup>
75	0.52 <sup>cC</sup>	0.61 <sup>abAB</sup>	0.57 <sup>abB</sup>	0.60 <sup>abAB</sup>	0.64 <sup>abA</sup>
90	0.55 <sup>bcB</sup>	0.66 <sup>aA</sup>	0.61 <sup>aA</sup>	0.67 <sup>aA</sup>	0.66 <sup>aA</sup>

The averages followed by the same lowercase letters in the column and uppercase letters in the rows do not differ according to Tukey's test at 5% significance.

maximum drying temperature of 43.33°C.

Tables 4 and 5 show the germination percentage and GSI values of the crambe fruit with different drying temperatures and storage for 12 months in a natural environment. The germination potential was higher in fruit dried at temperatures of 35, 45 and 60°C during storage.

The crambe seeds that were not stored had low germination percentages in all drying temperatures, thereby indicating that all recently harvested seeds exhibited primary dormancy. This mechanism is common in recently harvested seeds of several species (Brasil, 2009). The study of dormancy in crambe seeds is necessary when considering that the germination capacity may increase with storage time. When studying the viability of recently harvested crambe seeds subjected to different drying conditions and moisture contents, Costa et al. (2012a, b) and Faria (2010) observed a low germination percentage. The same behavior was also observed by Oliva (2010) when drying crambe fruit and storing the fruit for eight months in paper bags. In papaya seeds, the germination of seeds derived from recently harvested fruit is low with germination percentages of 40 and 14.75% for fruit harvested in January and September, respectively (Aroucha et al., 2005). Castor bean seeds also exhibit dormancy when recently harvested, but the dormancy breaks after nine months of storage (Lago et al., 1979).

According to Marcos Filho (2005), dormancy has a

depth inversely proportional to its age; that is, it is more intense in recently harvested seeds. Thus, the gradual overcoming of dormancy was achieved with storage in which the crambe aged resulting in a higher germination percentage and greater GSI.

During storage, the germination percentage increased regardless of drying temperature up to the third month after which the values varied (Table 4), and a lower germination percentage was observed at high drying temperatures.

Moreover, there was a higher germination index at a temperature of 35°C when compared to other temperatures, and a temperature of 90°C resulted in the lowest germination index (Table 4). Martins et al. (2005) studied papaya seeds from the Formosa group, and they also observed an increase in the germination power between zero and three months of storage and a decrease between three and six months of storage. However, it should be emphasized that the drying temperature and time of exposure to high temperature may have also influenced these variables (Marcos Filho, 2005; Kohama et al., 2006). In seeds, there are basically two types of water as follows: free (absorbed) and adsorbed water. The free water moves and is rapidly lost when the seeds are being dried. The adsorbed or retained water is attached to macromolecules, which participate in metabolic reactions, and when the adsorbed or retained water

**Table 4.** Germination (%) of crambe seeds stored under ambient conditions for up to 12 months.

Drying (°C)	Storage time (months)				
	0	3	6	9	12
35	27.25 <sup>ab</sup>	65.50 <sup>aA</sup>	55.25 <sup>aA</sup>	51.75 <sup>abA</sup>	58.50 <sup>abA</sup>
45	23.00 <sup>aC</sup>	53.50 <sup>abAB</sup>	51.00 <sup>ab</sup>	61.25 <sup>aAb</sup>	68.00 <sup>aA</sup>
60	22.50 <sup>aC</sup>	49.50 <sup>abAB</sup>	46.75 <sup>ab</sup>	62.25 <sup>aAB</sup>	64.00 <sup>abA</sup>
75	23.00 <sup>aC</sup>	46.50 <sup>bAB</sup>	60.75 <sup>aA</sup>	38.50 <sup>bBC</sup>	47.75 <sup>bAB</sup>
90	20.50 <sup>aA</sup>	21.50 <sup>cA</sup>	22.00 <sup>bA</sup>	18.50 <sup>cA</sup>	21.75 <sup>cA</sup>

The averages followed by the same lowercase letters in the column and uppercase letters in the rows do not differ according to Tukey's test at 5% significance.

**Table 5.** Germination speed index (GSI) of the crambe fruit subjected to storage for 12 months.

Drying (°C)	Storage time (months)				
	0	3	6	9	12
35	1.32 <sup>aC</sup>	3.75 <sup>aA</sup>	2.29 <sup>aBC</sup>	2.97 <sup>abAB</sup>	3.93 <sup>abA</sup>
45	1.08 <sup>aD</sup>	3.00 <sup>abBC</sup>	2.09 <sup>aCD</sup>	3.47 <sup>aB</sup>	4.75 <sup>aA</sup>
60	1.14 <sup>aD</sup>	2.68 <sup>bBC</sup>	1.91 <sup>aCD</sup>	3.54 <sup>aB</sup>	4.86 <sup>aA</sup>
75	1.27 <sup>aC</sup>	2.45 <sup>bBC</sup>	2.50 <sup>aAB</sup>	2.00 <sup>bBC</sup>	3.02 <sup>bA</sup>
90	1.03 <sup>aA</sup>	1.03 <sup>cA</sup>	0.88 <sup>bA</sup>	0.91 <sup>cA</sup>	1.16 <sup>cA</sup>

The averages followed by the same lowercase letters in the column and uppercase letters in the rows do not differ according to Tukey's test at 5% significance.

is lost by drying, it can cause a decrease in seed germination (Marcos Filho, 2005).

According to Silva and Vieira (2006), the GSI is included among the best-known vigor tests. The GSI test is easily performed, because data collection is performed during the germination test. The germination speed test considers that lots with faster germinating seeds are more vigorous, and therefore, there is a direct relationship between germination speed and seed vigor. Thus, as with germination percentage, the GSI was higher with these drying temperatures (between 35 and 90°C). This index increased until the third month of storage after which the values decreased and fluctuated (Table 5). Moreover, at a temperature of 90°C, there was a lower GSI as compared to other temperatures.

When comparing these results with the moisture content (Table 2), a direct relationship was observed between these parameters (Tables 2 to 5), that is, both the germination percentage and GSI varied over time for the analyzed treatments. Thus, when the moisture content in seeds was higher, there was a decrease in germination resulting in a lower GSI.

## Conclusions

Drying at high temperatures (90°C) resulted in lower physiological quality of the crambe fruit over the storage period. Generally, there was an increase in the amount of electrolytes released by the fruit during the storage period.

The storage of the crambe fruit for 12 months at room temperature promoted the break of seed dormancy, thereby increasing the germination percentage.

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