PHYSIOLOGICAL AND SANITARY QUALITIES OF MAIZE LANDRACE SEEDS STORED UNDER TWO CONDITIONS

Qualidade fisiológica e sanitária de sementes de variedades crioulas de milho armazenadas em duas condições

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ABSTRACT

The preservation of seed quality during the storage period depends not only on the conditions during production and harvesting but also on the storage and maintenance of appropriate storage product conditions. Thus, the aim of this study was to evaluate the physiological and sanitary qualities of maize landrace seeds stored under two conditions. The maize seed batch varieties Oito carreiras, Cabo roxo and Lombo baio were used. Tests included germination, first count, cold test, accelerated aging and sanity. Based on the results it was concluded that the physiological quality of these seed varieties decreased with the storage period. The major fungi identified in the maize seeds during storage were from the genera *Aspergillus*, *Fusarium* and *Penicillium*, which caused deterioration and reduction of the physiological quality. Storage using a paper bag at a temperature of 10 °C did not prevent the deterioration of maize seeds but was more effective at preserving the quality of the seed compared with a plastic bag at room temperature.

Index terms: *Zea mays*; storage; sanity; germination.

RESUMO

A preservação da qualidade das sementes, durante o período de armazenagem, não depende somente das condições de produção e de colheita, mas das condições de armazenamento e de manutenção adequada de estocagem do produto. Dessa forma, objetivou-se, neste trabalho, avaliar a qualidade fisiológica e sanitária de sementes de variedades crioulas de milho, quando armazenadas em duas condições. Foram utilizados lotes de sementes de milho pertencentes às variedades Oito carreiras, Cabo roxo e Lombo baio. Os testes realizados foram testes de germinação, primeira contagem de germinação, teste de frio, envelhecimento acelerado e sanidade. Por meio dos resultados, foi possível concluir que a qualidade fisiológica das sementes de variedades crioulas diminuiu com o período de armazenamento. Os principais fungos encontrados nas sementes de milho, durante o armazenamento, foram os dos gêneros *Aspergillus*, *Fusarium* e *Penicillium*, os quais provocaram deterioração e redução da qualidade fisiológica. O armazenamento em saco de papel, à temperatura de 10 °C não evitou a deterioração das sementes de milho, mas foi mais eficiente na preservação da qualidade da semente em relação à embalagem plástica, à temperatura ambiente.

Termos para indexação: *Zea mays*; armazenamento; sanidade; germinação.

INTRODUCTION

Seed quality is influenced by storage conditions between harvest and sowing. During storage, temperature and relative humidity are the main factors influencing seed quality, especially vigor. The relative humidity is related to the water content of seeds and controls the metabolic processes. The temperature influences the speed of the biochemical processes and indirectly affects the water content of seeds (Carvalho; Nakagawa, 2012).

Storage is crucial to preserve the physical, physiological and sanitary seed qualities and also provides seed for scientific research and agriculture. Seeds age naturally during storage. This deterioration continues until the seeds are no longer viable. If the storage conditions are not adequate, the physiological quality of seed batches may deteriorate (Baudet, 2012).

The packaging used and the environment in which the seeds stored are important considering that deterioration is also associated with the characteristics of the packaging that contains the seeds (this determines the amount of water vapor exchange between the seed and the atmosphere) and the environmental conditions under which seeds are stored. There are many different

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types of packaging type depends on the species the water content of the seeds the conditions and the storage period (Freitas, 2009; Baudet, 2012; Carvalho; Nakagawa, 2012).

The detection of seed deterioration using vigor tests is an important component in assessing the physiological quality and contributes to solving problems of the seed industry such as storage. Despite the considerable increase in knowledge regarding the analysis of seeds, many varieties of landrace seeds require basic information concerning the ideal conditions for germination and vigor. These varieties have considerable genetic variability and require studies that assess the physiological and sanitary qualities. Inn addition, analysis of the chemical composition of seeds especially during the storage period, is needed. The objective of this study was to evaluate the physiological and sanitary qualities of maize landrace seeds stored under two conditions.

MATERIAL AND METHODS

The experiment was conducted in the Didatic Laboratory of the Research Seed and Plant Pathology Laboratory in the Federal University of Santa Maria, Brazil.

The seeds of landrace maize Oito carreiras, Lombo baio and Cabo roxo (2011/2012 season) from the Association of Landrace Seed Keepers of Ibarama, Brazil (29°25'10"S; 53°08'05"W, altitude: 317 m) were used. These seed batches were produced in the same area and under the same temperature and environmental conditions (humid subtropical climate).

The maize seeds were stored for nine months under two conditions, i.e., condition 1 (C_1): in paper bags at a temperature of 10 °C and condition 2 (C_2): in plastic packaging at room temperature, in Santa Maria / Brazil (29º42'24"S; 53º48'42"W, altitude: 116 m) in a humid subtropical climate.

The physiological quality of the seeds of all varieties was assessed every three months the seeds were subjected to the two storage conditions for a total of nine months. The sanitary quality was assessed every three months for a total of nine months for the varieties Oito carreiras and Lombo baio, and a total of six months for the variety Cabo roxo (this test could not be completed in the latter variety because there was an insufficient number of seeds). The following tests were conducted:

Water content: determined using the oven method at 105 ± 3 °C for 24 hours using two samples of seeds; the results are expressed as a percentage (Brasil, 2009).

Germination test: conducted at 25 °C using eight replications of 50 seeds distributed on paper rolls moistened with an amount of water equivalent to 2.5 times the weight of the substrate. Seed counting was performed seven days after sowing, and the results are expressed as a percentage of normal seedlings (Brasil, 2009).

First germination count: performed with the germination test, where the percentage of normal seedlings was determined on the fourth day after the beginning of the test (Brasil, 2009), except for the variety Oito carreiras (fifth day) because preliminary tests have not published.

Accelerated aging: conducted using plastic boxes containing 40 ml of water maintained in an incubator chamber set at a temperature of 45 °C (Bittencourt et al., 2012) for 96 hours. At the end of each period eight replicates of 50 seeds were set to germinate. Seed counting was performed on the fourth day after sowing, except for Oito carreiras (fifth day) because preliminary tests have not yet been published. The results are expressed as a percentage of normal seedlings.

Cold test: conducted using eight replicates of 50 seeds distributed on paper rolls moistened with distilled water at a ratio of 2.5 times the weight of the paper. The rolls were placed inside plastic bags and maintained at a controlled temperature (10 °C) inside a chamber for seven days. After this period, the seeds were transferred to a germination chamber at a temperature of 25 °C where they remained for five days. The results are expressed as a percentage of normal seedlings (Barros et al., 1999).

Sanity test: conducted using eight replicates of 50 seeds that were distributed in plastic boxes lined with two sheets of sterilized filter paper moistened using sterile distilled water. Incubation was performed for 7 days in a chamber set at a temperature of 22 ± 3 °C with a 12 hour photoperiod. During this period, the assessment and identification of fungi were performed with the aid of an optical stereoscopic microscope and specialized bibliography (Barnett; Hunter, 1999).

Statistical analysis: a completely randomized design was used and the treatments were arranged in a 2 x 4 (condition x storage period factorial). Comparisons between treatment means were performed using the Tukey test at a 5% probability, and when significant effects were observed, regression analysis was performed. The variables expressed as percentages were converted to sine $\sqrt{x/100}$.

RESULTS AND DISCUSSION

Evaluation of physical and physiological seed qualities

During the nine months under the two storage conditions, the water content of the seeds did not exceed a value of 13%. According to Baudet (2012), the most important factor affecting the conservation of seeds is water content, which should not exceed 13% in stored seeds.

Regarding the physiological quality of the maize seed, it was observed that the germination and vigor of the lots of the three maize landrace seeds were significantly reduced during the nine months under both storage conditions (Figures 1, 2, 3).

In the Oito carreiras batch, it was observed that the seed quality decreased with the storage period (Figure 1). In the accelerated aging and cold tests, storage under condition 1 (paper bag at a temperature of 10 °C) showed the highest values of normal seedlings compared with the storage condition 2 (plastic container, room temperature). The vigor expression of variety Oito carreiras was more affected by high temperature and humidity conditions (accelerated aging) than by exposure to low temperatures (cold test), and this is probably related to genetic factors.

The seed quality of the variety Cabo roxo decreased with the storage period (Figure 2). In all tests, significant differences were observed between the two storage conditions. Storage under condition 1 showed the highest values of normal seedlings in relation to storage under condition 2, except for the results of the first germination test. The reduction in the number of normal seedlings was more pronounced in the accelerated aging test compared with the cold test.

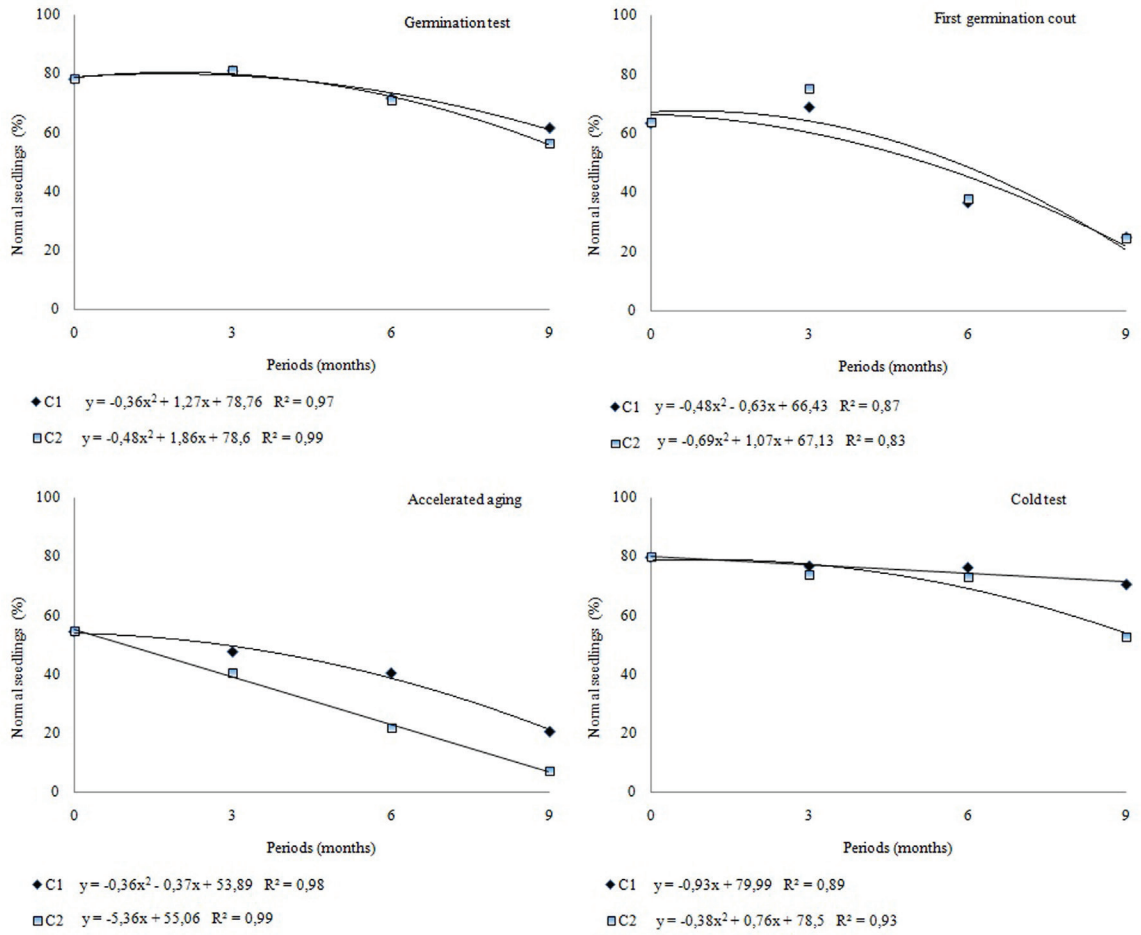

Figure 1: Evaluation of the physiological quality of maize seed variety Oito carreiras submitted to different periods and storage conditions (C1: paper bag, temperature of 10 °C and C2: plastic packaging at room temperature).

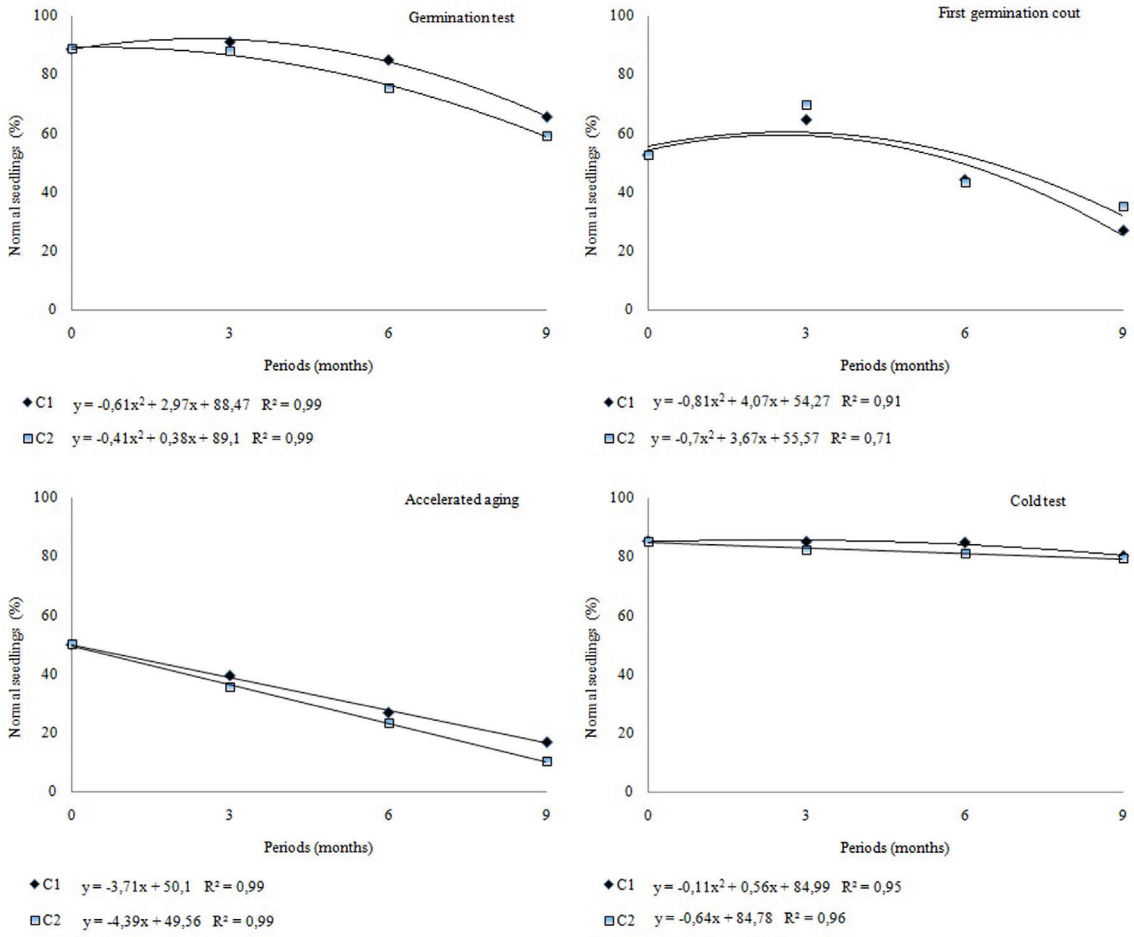

Figure 2: Evaluation of the physiological quality of maize seed variety Cabo roxo subjected to different storage periods and storage conditions (C1: paper bag, temperature of 10 °C and C2: plastic packaging at room temperature).

It was observed that with an increase in the storage period, there was a decrease in the percentage of normal seedlings in the seed batch of the variety Lombo baio (Figure 3). Similarly, in the accelerated aging test, we observed the largest decrease in the percentage of normal seedlings under both storage conditions, and the highest values were found in the seeds stored under condition 2. Storage under condition 1 (paper bag, temperature of 10 °C) resulted in the highest values of normal seedlings compared with storage under condition 2 (plastic packaging at room temperature).

The differences observed in vigor (first counting, accelerated aging and the cold test) and initial seed germination among the varieties Oito carreiras, Cabo roxo and Lombo baio may be related to genetic factors. According to Baudet (2012), not all varieties or individual seeds within the same genetic group survive the same period of time under a wide range of storage conditions. Longevity is a genetically determined trait that varies among species and varieties of the same species.

On the other hand, according to the same author, a factor that should be taken into account in a significant decrease in the germination of stored seeds is the natural deterioration of seeds. This is an irreversible process and consequence of the genetic constitution of the seed, the environment during its development and its chemical composition.

In this work, storage under condition 1 (paper bag, temperature of 10 °C) did not prevent the deterioration of the maize seed but was significantly more efficient in preserving seed quality compared with condition 2 (plastic packaging at room temperature). In accordance

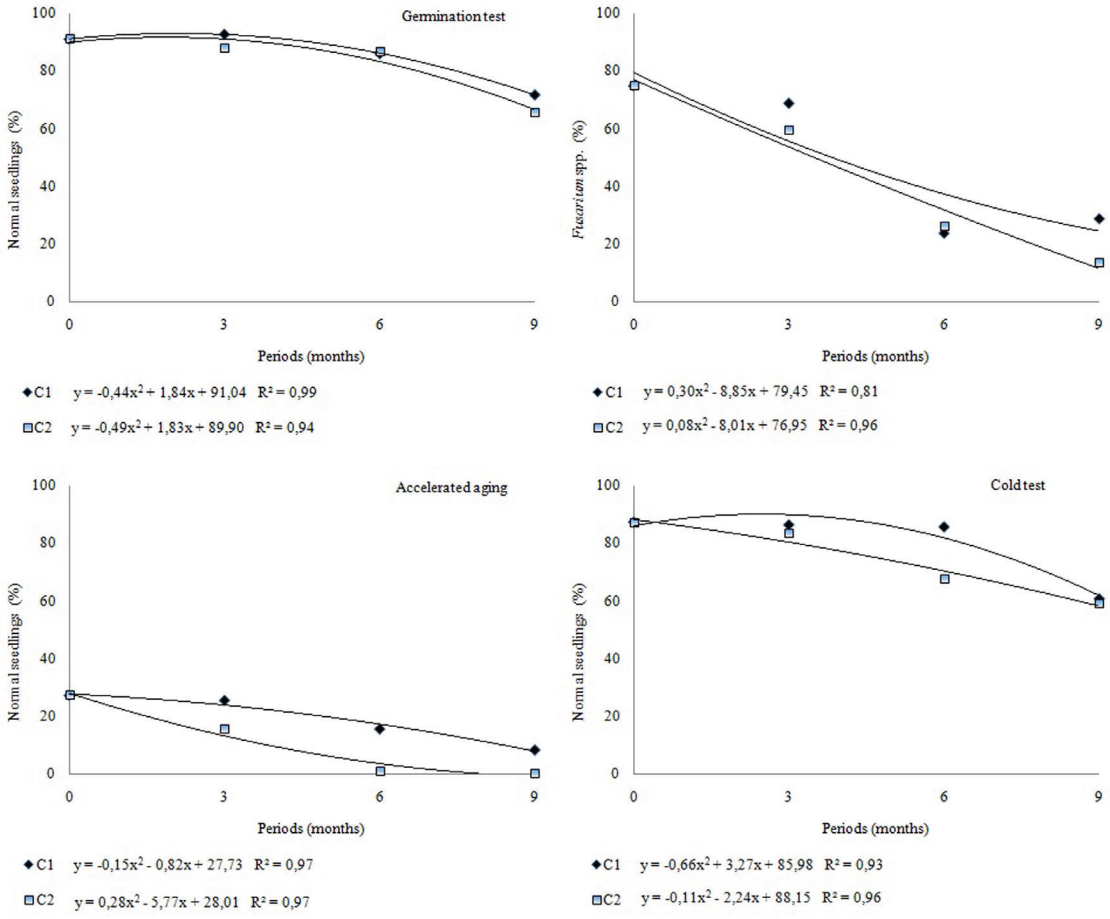

Figure 3: Evaluation of the physiological quality of maize seed variety Lombo baio submitted to different storage periods and storage conditions (C1: paper bag, temperature of 10 °C and C2: plastic packaging at room temperature).

with Baudet (2012), seed storage under controlled environmental conditions (temperature and relative humidity) allows the maintenance of seeds for long periods of time. However, the reduction in vigor depends on the period of time that the seed will be stored, the type of seed and the conditions of storage.

Silva et al. (2010) concluded that for small farms storing corn seed is viable when the seeds are stored in waterproof packaging. These authors reported that the physiological quality of seeds analyzed decreased throughout the storage period, regardless of the type of packaging used. Moreover, the seeds stored in permeable containers had the lowest quality (germination and vigor) at the end of the storage period when compared to semi-permeable and impermeable packaging.

Moreover, Antonello et al. (2009) stated that the behavior of the seeds of maize landraces during storage for a period of six months depended on the seed variety and the packaging used. Catão et al. (2013) reported that the storage of maize seed in plastic containers was generally effective for maintaining the physiological quality and to a lesser extent, the sanitary quality of the seeds.

Temperature is a very important factor in storage and may have influenced the results obtained. Temperature variations in the environment during seed storage in plastic packaging although they were not quantified in this study may have been too high and accelerating breathing directly affects the rate of chemical reactions as well as the activity of micro-organisms according to Marcos Filho (2005).

The increase in respiration triggers other processes such as increases in the enzymatic activity and free

fatty acid content. The temperature increases the rate of metabolic and enzymatic reactions, causing an acceleration in the rate of deterioration. The result is a rapid decline in germination and seed vigor (Baudet, 2012).

Evaluation of the sanitary quality of seeds

Among the fungi detected in the seeds of the maize landraces in the sanitary test (Figures 4, 5 and Table 1), there was a high incidence of field fungi such as *Fusarium* spp., and *Aspergillus* spp. and *Penicillium* spp., which are considered storage fungi. These results are similar to those observed by Catão et al. (2013) for maize landrace seeds naturally infested with fungi under pre- and post-storage conditions. Other fungi such as *Rhizopus* spp., *Rhizoctonia* spp., *Trichoderma* spp., *Verticillium* spp., *Cladosporium* spp. and *Mucor* spp. were also identified, but their occurrence was only observed in a few samples and at a low incidence.

In the batch variety Oito carreiras, the incidence of *Aspergillus* spp. tended toward equilibrium after six months of storage. *Fusarium* spp. increased until the third month under condition 1 and condition 2 and then decreased (Figure 4). The seeds stored under condition 1 (paper bag, temperature of 10 °C) had a lower incidence of *Aspergillus* spp. and a higher incidence of *Fusarium* spp. and *Penicillium* spp. compared with condition 2 (plastic packaging, room temperature). In the seeds stored in the plastic packaging, the initial incidence of *Aspergillus* spp. was low (13%) but then increased until the ninth month (100%). This tendency may explain the reduction in the incidence of *Penicillium* spp. due to the competition between the two fungal genera for the nutritional substrate.

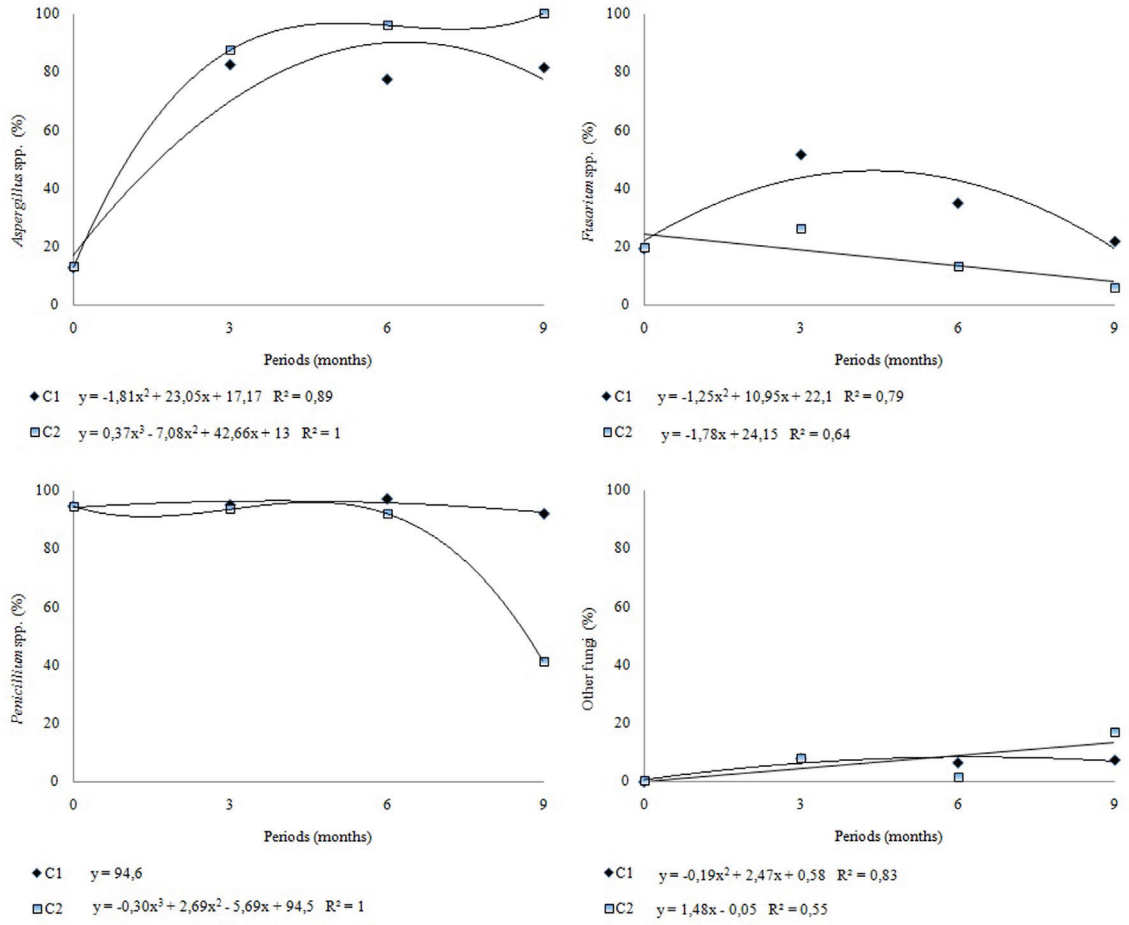

Figure 4: Incidence of fungi in seed of the maize variety Oito carreiras under different storage periods and storage conditions (C1: paper bag, temperature of 10 °C and C2: plastic packaging at room temperature).

A plot of the maize seed variety Lombo baio and *Fusarium* spp. indicated that the fungal incidence was initially 80% but then decreased during the storage period for both studied conditions. Moreover, the incidence of *Penicillium* spp. was high during the nine months of storage under both conditions (1 and 2) (Figure 5). The high incidence of *Penicillium* spp. during storage may also be related to the low percentage of *Aspergillus* spp. (1-28%) due to competition between the two fungal genera for the nutritional substrate.

In the batch variety Cabo roxo, significant differences were observed between storage periods and storage conditions. Over the six months of storage, there was a high incidence of fungi of the genus *Penicillium* (Table 1). The incidence of *Fusarium* spp. was initially high but then decreased significantly during storage. There was a higher incidence of fungi of the genus *Aspergillus* after three months and *Penicillium* spp. after six months of storage in plastic packaging at room temperature (C2).

According to Coutinho et al. (2007), many factors affect the quality of maize seeds. Fungi are considered the most important because they are related to low germination rates and low vigor, and they accelerate the process of deterioration during storage.

Moreover, storage fungi such as *Aspergillus* spp. and *Penicillium* spp. are present on freshly harvested seed, usually in very low percentages. These fungi are able to survive in an environment with low humidity and proliferate in succession to field fungi, causing deterioration of the seed and the production of toxins that can poison humans and animals (Carvalho; Nakagawa, 2012).

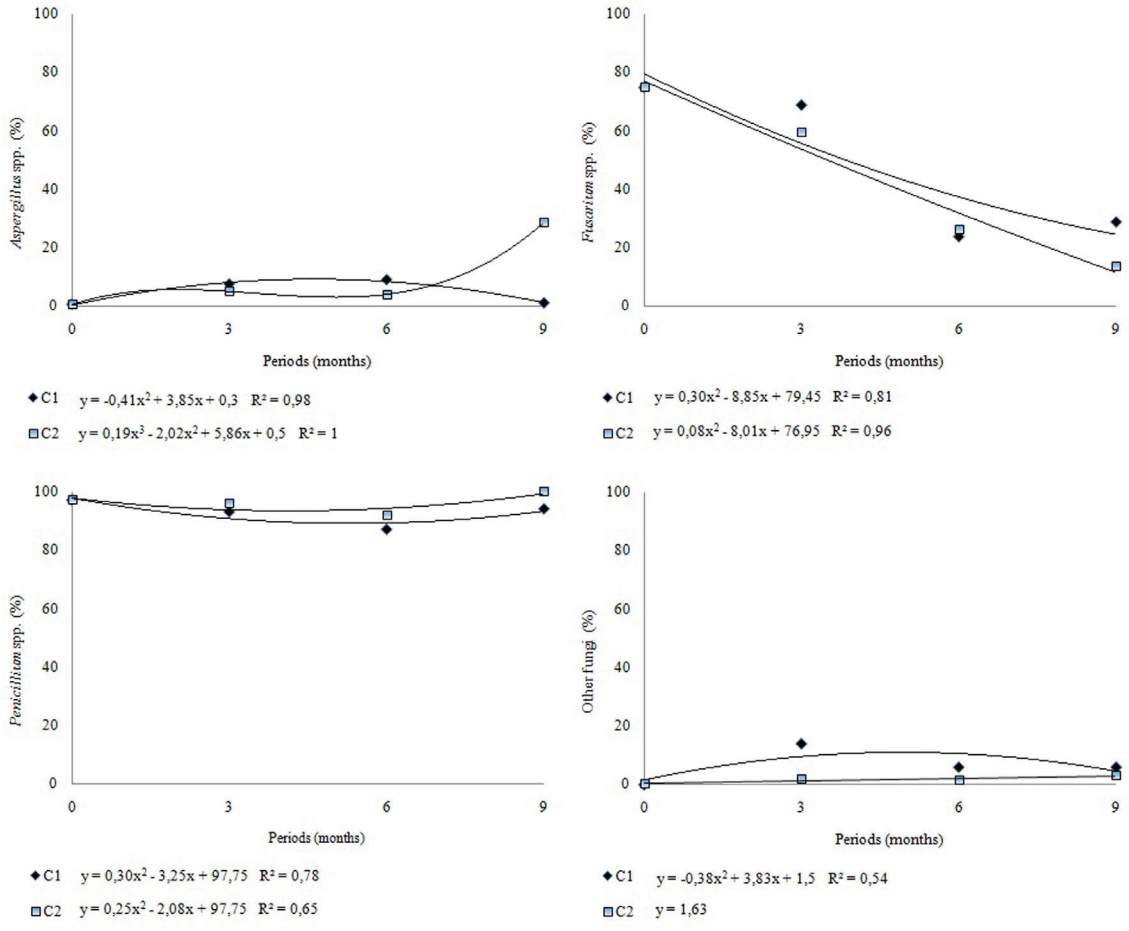

Figure 5: Incidence of fungi in seeds of the maize variety Lombo baio under different storage periods and storage conditions (C1: paper bag, temperature of 10 °C and C2: plastic packaging at room temperature).

Table 1: Incidence of fungi (%) in the maize seed variety Cabo roxo subjected to different storage periods and storage conditions.

Fungi	Months	Condition 1	Condition 2	CV (%)
Aspergillus spp.	0	0bA*	0cA	20.22
	3	83aA	88aA	
	6	0bB	17bA	
Fusarium spp.	0	77aA	77aA	25.22
	3	52bA	26bB	
	6	37bA	40bA	
Penicillium spp.	0	95aA	95aA	11.51
	3	61bB	94aA	
	6	98aA	92aA	

* Averages followed by the same lowercase letters within a column and the same capital letter within a row are not significantly different at the 5% probability level based on Tukey's test.

During the storage of maize seeds, a high temperature accelerates respiration, which directly affects the rate of chemical reactions as well as the activity of micro-organisms (Marcos Filho, 2005). These micro-organisms attack the seeds and in combination with metabolic processes, accelerate the deterioration of the seeds and can produce toxins that damage membranes and inhibit seed germination (Freitas, 2009; Baudet, 2012).

CONCLUSIONS

The physiological quality of the maize landrace seeds decreased with the storage period.

The major fungi found in the maize seeds during storage were from the genera *Aspergillus*, *Penicillium* and *Fusarium*, and they caused deterioration and reduced the physiological quality.

Paper bag storage at a temperature of 10 °C did not prevent deterioration of the maize seeds but was more effective at preserving the quality of the seed compared with the plastic packaging at room temperature.

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