Original Research Article

Germination at the Constant Temperature of Amaranth Seeds BRS Alegria Storaged with Different Conditions of Sealing of Packaging and Water Content

ABSTRACT

The objective of this work was to evaluate the physiological quality of amaranth seeds, cultivar BRS Alegria, stored in open and closed polyethylene packages and in two water contents during the 13 - month period. The experimental design was completely randomized in a factorial scheme with two initial water contents (10% and 8%), and two packing conditions (sealed and unsealed), with four replications, totaling 16 plots. The germination was done with photoperiod of 12 hours and constant temperature of 25 ° C, for 7 days. The variables analyzed were percentage of germination (PG), first germination count (FGC), non-germinated seeds (NGS), germination speed index (GSI) and mean germination time (MGT). Seeds with packing sealing and higher water content presented better results for PG, NGS and GSI (77.5%, 22.5% and 24.88, respectively) and for FGC, the sealing was better than the non-sealing of the packages , regardless of the water content (61.25%). It was concluded that packaging sealing and water content of 10% are recommended for storage of amaranth seeds.

Keywords: Amaranthus cruentus, first germination count, germination speed index, mean germination time.

1. INTRODUCTION

The Amaranth (*Amaranthus cruentus* L.) is one of the species that has good grain production and has rapid growth [1]. It is rich in proteins, compared to most cereals, and of high biological value, with a high content of lysine, methionine and sulfur amino acids, limiting in other cereals and legumes [2,3,4,5,6]. Due to its agronomic characteristics and easy adaptation, the potential of amaranth to the Brazilian savanna, increasing the income in the agricultural productive chain is great [1,7]. The BRS Alegria cultivar is a national variety selected for its adaptation to savanna climate [1].

On a small scale, amaranth grains are naturally dried through exposure to ambient air [8]. The importance of grain drying increases as production increases [9]. When amaranth is grown on a large scale, artificial drying is necessary to reduce the moisture level and ensure good conservation [10]. The main advantage of drying is the possibility of anticipating the harvest, allowing the producers to obtain stock and supply high quality products, in the case of grains by organoleptic and nutritional characteristics and, in the case of seeds, by the high percentage of germination and vigor [9,11,12,13].

The ability of a seed to maintain its quality during storage depends on the longevity inherent to the species, its initial quality and environmental storage conditions [14]. Thus, the seed can be grown under a rigorous system, have adequate harvest, and processed to the highest purity, but can be lost if stored under inadequate conditions.

UNDER PEER REVIEW

The physiological quality of stored seeds may be related to the type and conditions of packaging used [15]. When the seeds are stored in permeable containers, their water content varies according to the variations of the humidity of the air and in semipermeable packages there is some resistance to the exchanges, but nothing that completely prevents the passage of moisture [16]. The permeability of the packages can also relate to their own fence. Polyethylene packages, when violated, now have characteristics of permeable packaging, while when properly sealed, they are semipermeable [15].

BRS Alegria cultivar with 10% water content can be safely stored for ten months of cold storage. At 11.7% water content, stored at higher temperatures, its physiological quality is impaired [17].

Despite advances in amaranth research, little information related to the seed storage process is found [17]. In this context, the present study had the objective of evaluating the physiological quality of amaranth seeds, BRS Alegria cultivar, stored in open and closed polyethylene packages and in two water contents during the period of 13 months (10 months in ambient conditions and 3 months in cold chamber).

2. MATERIAL AND METHODS

The work was carried out in the Laboratory of Seeds of the Faculty of Agronomy and Animal Science of the Federal University of Mato Grosso. Seeds of *Amaranthus cruentus* L. BRS Alegria cultivar were used from plants grown between March and June 2017, at the Rio Verde Technological Research and Development Foundation, MT 449, Km 8, in the Lucas do Rio Verde city, Mato Grosso state, geographic coordinates 13°00'27 "S and 55°58'07"W, with an average elevation of 387 meters. The climate is tropical (Aw), according to the classification of Köppen [18], and the predominant soil is dystrophic Red-Yellow Latosol of clay texture, and flat relief.

The harvest was done manually on 06/06/2017, with a maximum water content of 22%, the panicles being carefully removed to avoid grain dropping, and then they were trodden by means of mechanical track and the seeds dried in the sun under until they reach 12% moisture, as recommended by Spehar et al. [1]. Due to the small size of the seeds there was still a large amount of straw, and to minimize such impurities the seeds were passed in forced-blow dryer blower with counterflow flow at the mean velocity of 4.5 ms-1 and air temperature of 30 °C. The process of removing the impurities through the blow dryer at 30 °C also allowed a slight drying of the seeds, decreasing the water content to approximately 10% (Table 1).

Part of the seeds were subjected to drying in a forced circulation oven at 45 °C, and the seeds were distributed in steel screens with a mesh of less than 1.0 mm (smaller than seeds) until reaching a mean water content of 8%, forming two batches of different water contents, as shown in Table 1.

The water content of the seeds was determined by the oven method at 105 ± 3 °C for 24 hours, with 5 g samples of seeds packed in aluminum containers, in triplicate, according to the Manual of Rules for seed analysis [19]. The results were expressed as percentage (wet basis) for each treatment.

The lots were stored in closed (semipermeable) polyethylene packages. Seeds were stored in a laboratory environment for 10 months, with maximum and minimum temperatures of 33.8 °C and 22.1 °C, respectively, and maximum relative humidity of 76% and minimum of 22%, measured with a thermo- hygrometer, model 303C. After this period the batch characterization was done, with the means of four replications, which is found in Table 1.

Table 1. Pre-test results of BRS Alegria amaranth seeds germination stored for 10 months.

Water content	Variables

UNDER PEER REVIEW

	PG	FGC	NGS	GSI	MGT
9.82%	69.33	69.33	30.67	17.33	2.05
7.84%	62.00	60.00	38.00	15.42	2.07
Mean	65.67	64.67	34.33	16.38	2.06

PG: percentage of germination; FGC: first germination count; NGS: non-germinated seeds; GSI: germination speed index; and MGT: mean germination time.

After this characterization, where part of the packages were violated, different lots were allowed between closed and open packages. These seed lots were stored in a cold room, as recommended by Nobre et al. [17], with averages of 18 °C of temperature and 60% of relative humidity, in the Laboratory of Seeds of the Faculty of Agronomy and Animal Science of the Federal University of Mato Grosso, for another three months in order to verify the effect of the conditions of packaging.

The germination test was carried out in transparent plastic boxes (gerboxes), size 11 cm x 11 cm x 3 cm. The seeds were treated with 0.5% sodium hypochlorite solution (NaClO) for one minute to eliminate possible contaminations. The substrate used was paper, two blots were previously sterilized at 105 °C for 4 hours, moistened in the amount equivalent to 2,5 times its dry mass. The treated seeds were placed to germinate in the blotches moistened with distilled water, evenly distributed in the quantity of 50 seeds. Germination occurred at a constant temperature of 25 °C and photoperiod of 12 hours, as recommended by Crivelari Costa and Bianchini [20] for amaranth seeds BRS Alegria. BOD (Biochemical Oxygen Demand) incubator was used.

The variables analyzed for the pre-test and germination test were: percentage of germination (PG), first germination count (FGC), non-germinated seeds (NGS), germination speed index (GSI) and mean germination time (MGT).

The number of germinated seeds was evaluated daily, using the initial protrusion of the primary root with approximately 1.0 mm in length, also called physiological germination, until the seventh day after sowing. stabilization), when PG and NGS were calculated using the criteria established in the Rules for seed analysis [19]. The FGC was performed together with the germination test, and evaluated on the third day after sowing.

The germination speed index was calculated with the germination test and calculated according to Maguire [21], using the formula presented in Equation 1:

$$GSI = \sum_{i=1}^{N} \left(\frac{G_i}{N_i}\right) \qquad \text{(Eq. 1)}$$

Where: GSI is the germination speed index, dimensionless; G_1, G_2 and G_i is the number of germinated seeds in the first count, second count, ... i-th counting, respectively, dimensionless; and N_1, N_2 and N_i is the number of sowing days at the first count, second count, ... i-th counting, respectively, dimensionless.

The mean germination time was calculated using the expression proposed by Labouriau [22] presented in Equation 2:

$$MGT = \frac{\sum_{i=1}^{k} n_i t_i}{\sum_{i=1}^{k} n_i} \qquad \text{(Eq. 2)}$$

In which: MGT is the mean germination time, in days; k is the last germination time of the seeds, in days; n_i is the number of seeds germinated at time t_i (not the cumulative number, but that referred to the i-th observation), dimensionless; t_i is the time between the beginning of the experiment and the i-th observation, in days.

UNDER PEER REVIEW

The experiment of seeds stored for 13 months (10 + 3) was arranged in a completely randomized design, in a factorial scheme with two water contents (10% and 8%), and two packaging conditions (sealed and unsealed), with four replications, totaling 16 plots. The data were submitted to the Shapiro-Wilk normality test [23], and to the analysis of variance, and the means were compared by the Tukey test at a 5% probability level. The analysis was performed through the computer program System for Analysis of Variance - SISVAR [24].

3. RESULTS AND DISCUSSION

As can be seen in Table 2, where the results of the Shapiro-Wilk test are presented, data from the BRS Alegria amaranth germination test stored for 13 months were presented in an approximately normal distribution and could be submitted to analysis of variance and to the mean test, except for the variables Water Content (WC) and MGT, which were made descriptive analyzes.

Table 2. Shapiro-Wilk test values for BRS Alegria amaranth seed germination test data stored for 13 months.

Test	n	Variables					
		WC	PG	FGC	NGS	GSI	MGT
Wc	16	0.776	0.955	0.975	0.955	0.975	0.863
Pr<Wc	16	0.001	0.585	0.913	0.585	0.916	0.021
Wt	16	0.887					

Wc: calculated Shapiro-Wilk value; Wt: tabulated Shapiro-Wilk value, according to number of plots (n) and significance level (5%); WC: water content; PG: percentage of germination; FGC: first germination count; NGS: non-germinated seeds; GSI: germination speed index; and MGT: mean germination time.

It can be verified, according to the data (Table 3), that: for sealed packs, the lot with water content of 10% lost water in relation to the initial water content, while that of 8% remained close to initial. In the unsealed packages, the seeds absorbed water in both batches. The increase of the water content for unsealed packages was expected for experimental conditions [15].

For the lot with initial water content of 10%, the sealing caused the seeds to decrease their water content, and to the unsealed, the opposite occurred. For the lot with initial water content of 8%, the water content in the seeds was maintained when the packages were sealed, but increased when they were open. The relative humidity of 60% of the cold chamber may have influenced the results found.

This shows that the packaging sealing kept the water content of the seeds lower than the unsealed packages for the two water contents studied, while open packages increased the water content of the seeds. This is because when the packages are violated, their water content varies according to the variations of the humidity of the air, but when sealed, the polyethylene packages, which are semipermeable, create resistance to this exchange, keeping the initial water content for more time [16].

Alves and Lin [15] found similar results in the variations of the initial water contents of 11% and 15% of bean seeds stored for 21 months for polyethylene packaging. These seeds enter into hygroscopic equilibrium with the environment, remaining between above 11% and below 15% for the conditions studied. For them, the polyethylene packaging was the one that maintained the water content of the seeds better.

UNDER PEER REVIEW

Also, these authors still observed that polyethylene packages, when violated, now have characteristics of permeable packages, while when properly sealed, they retain their original characteristics [15].

Table 3. Variation of the initial water content, in %, of BRS Alegria amaranth seeds stored for 13 months.

Sealing of packaging	Water content		
	10%	8%	Mean
Sealed	8,58	8,31	8,45
Unsealed	11,07	9,02	10,05
Mean	9,83	8,67	

For germination percentage (Table 4), non-germinated seeds (Table 6) and germination speed index (Table 7), the interaction between packing factors and seed water contents was statistically significant. For the first germination count variable (Table 5) there was no difference in the interaction, only the packaging condition for FGC, which presented a statistical difference (Tukey, 0.5%).

In Table 4 is showed the mean values of germination percentage (PG) for BRS Alegria amaranth seeds stored for 13 months. For sealed packages, the water content of 10% was better than 8%. In unsealed packages, the water contents presented similar results. It was observed that, for water content of 10%, the seal allows a better maintenance of the physiological quality of the seeds, while for the water content of 8%, there was no difference between the packaging conditions.

This decrease in the percentage of germination may be due to the increase in the temperature of the drying air combined with the increase of the initial water content and the time of storage. This same result was found by Afonso Júnior and Corrêa [25], evaluating the drying effects of bean seeds (*Phaseolus vulgaris* L.). They observed that germination decreased with increasing drying air temperature, initial water content and storage time.

Table 4. Variation of the initial water content, in %, of BRS Alegria amaranth seeds stored for 13 months.

Sealing of packaging	Water content		
	10%	8%	Mean
Sealed	77.50 Aa	51.50 Bb	64.50
Unsealed	35.00 Bb	47.00 Bb	41.00
Mean	56.25	49.25	

Means followed by the same letter, uppercase in line and lowercase in the column, do not differ statistically from each other by the Tukey test at a 5% probability level.

Only the germination of the seeds stored with initial water content of 10% in sealed packaging was higher than the pre-test data. These values of PG fell within the marketing standard [26].

Another possible cause for loss of germination to 8% water content was explained by Lin [27,28], who reported that the water absorption by the seeds to reach their hygroscopic equilibrium, causes the deterioration of the plasma membrane of seeds and decreases their physiological quality.

A similar result was obtained in the first germination count (Table 5). There was no difference between water contents (p-value of 6.18%), which means that only the packaging conditions were meaning, the best result was obtained for sealed packages, regardless of the water content. Values between 50% and 69% of first count were also observed by Nobre et al. [17], for BRS Alegria amaranth seeds.

Tabela 5. Averages of the first germination count (FGC), in %, of the germination test of BRS Alegria amaranth seeds stored for 13 months.

Sealing of packaging	Water content		
	10%	8%	Mean
Sealed	71.50	51.00	61.25 a
Unsealed	33.50	47.00	40.25 b
Mean	52.50	49.00	

Means followed by the same letter do not differ statistically from each other by the Tukey test at a 5% probability level.

Table 6 shows the values of non-germinated seeds (NGS) for BRS Alegria amaranth seeds stored for 13 months. According to the results, for sealed packages, the lot with initial water content of 10% obtained less quantity of non-germinated seeds than the lot with 8%. For unsealed packages, high amounts of non-germinated seeds were found, regardless of the initial contents of the seeds. For the lot with initial water content of 10%, the sealing decreased the quantity of non-germinated seeds, whereas for 8%, there was no difference, being greater the quantity of seeds not germinated, independent of the condition of packaging sealing.

Among the non-germinated seeds are likely hard and deteriorated seeds. Similar values were found by Nobre et al. [17], for BRS Alegria amaranth seeds stored at 50% relative humidity and 26 °C temperature, with water content of 10%. They found 17% of hard seeds under these conditions.

Tabela 6. Non-germinated seeds (NGS), in%, of the germination test of BRS Alegria amaranth seeds stored for 13 months.

Sealing of packaging	Water content		
	10%	8%	Mean
Sealed	22.50 Bb	48.50 Aa	35.50
Unsealed	65.00 Aa	53.00 Aa	59.00
Mean	43.75	50.75	

Means followed by the same letter, uppercase in line and lowercase in the column, do not differ statistically from each other by the Tukey test at a 5% probability level.

Similar behavior is found for the rate of germination (Table 7), however, for sealed packages, the lot with initial water content of 10% presented a statistically similar result in relation to the 8% batch. Unsealed packages presented different results, being higher for the lot with initial water content of 8%. For the lot with initial water content of 10%, the seal increased the GSI while for the 8% water content, there was no difference between the packaging conditions.

It is still possible to observe that the lot with initial water content of 8% maintained the GSI of the pre-test, while the lot of 10% had the GSI improved when the packages were kept sealed, but was impaired when the packages were violated.

Tabela 7. Germination speed index (GSI) means of germination test of BRS Alegria amaranth seeds stored for 13 months.

Sealing of packaging	Water content		
	10%	8%	Mean
Sealed	24.88 Aa	19.78 Ab	22.33
Unsealed	8.91 Bb	15.37 Ab	12.14
Mean	16.89	17.58	

Means followed by the same letter, uppercase in line and lowercase in the column, do not differ statistically from each other by the Tukey test at a 5% probability level.

The mean germination time values of BRS Alegria amaranth seeds stored for 13 months are shown in Table 7. According to the data presented, the water content of 10% took longer to germinate than the water content of 8%, regardless of the packaging conditions. This is because the lower the water content, the faster the water absorption, which leads to rapid germination and may be detrimental to the good development of the seedling. The organization of the cells is greater at slower germination.

Tabela 8. Mean germination time (MGT), in days, of the germination test of BRS Alegria amaranth seeds stored for 13 months.

Sealing of packaging	Water content		
	10%	8%	Mean
Sealed	2,08	1,50	1,79
Unsealed	2,20	1,69	1,94
Mean	2,14	1,60	

Also, through the TMG data it is possible to notice that the first germination count has values close to the final percentage of germination, because the amaranth germinates relatively fast, between 1.52 and 2.23 days (considering the emission of the primary root of 2.0 mm). Similar results were found by Costa and Dantas (2009), who obtained emergence of up to three days after sowing, reinforcing the rapid germination of amaranth.

4. CONCLUSION

It was concluded that sealed packages and water content of 10% is recommended for BRS Alegria amaranth seed storage, since it maintains its physiological quality for a longer time.

Polyethylene packages, when violated, have the characteristics of permeable packaging, while when properly sealed, they retain their original characteristics.

UNDER PEER REVIEW

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UNDER PEER REVIEW

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UNDER PEER REVIEW