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Qualidade fisiológica e avaliação econômica de sementes tratadas de feijão-caupi armazenadas em diferentes embalagens

Physiological quality and economic evaluation of treated cowpea seeds stored in different packaging

Calidad fisiológica y evaluación económica de semillas tratadas de frijol caupí almacenadas en diferentes envases

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Resumo

O objetivo deste trabalho foi avaliar a qualidade fisiológica de sementes de feijão-caupi, e a viabilidade econômica do armazenamento em embalagens e produtos químicos. As sementes foram tratadas quimicamente com os ingredientes ativos: fipronil; fipronil + piraclostrobina + metil tiofanato; fludioxonil + metalaxyl. Sementes sem tratamento químico constituíram a testemunha. Em seguida, foram acondicionadas em sacos de algodão (permeável); sacos de ráfia (permeável); sacos de papel kraft (semipermeável); e garrafas do tipo PET (impermeável) e armazenadas por 90 dias. As seguintes avaliações foram realizadas: grau de umidade de sementes; teste de germinação; primeira contagem de germinação; índice de velocidade de germinação; comprimentos de hipocótilo e radícula e pesos de massa seca de hipocótilo e radícula. Por fim, realizou-se a análise de custos. A qualidade fisiológica das sementes de feijão-caupi é reduzida de forma mais acentuada sob condições de armazenamento em embalagens permeáveis e tratadas com fludioxonil + metalaxyl. A embalagem impermeável sem adição de tratamento químico, diminui a deterioração da qualidade fisiológica em sementes de feijão-caupi e é viável economicamente no período de armazenamento de até 90 dias.

Palavras-chave: *Vigna unguiculata*; Potencial fisiológico; Sustentabilidade; Viabilidade econômica.

Abstract

The objective of this work was to evaluate the physiological quality of cowpea seeds and the economic viability of storage in different types of packaging and with different chemical treatments. Seeds were submitted to one of the following chemical treatments of active ingredients: fipronil; fipronil + pyraclostrobin + thiophanate methyl; and fludioxonil + metalaxyl. Seeds without chemical treatment constituted the control. Seeds were placed in permeable cotton bags, permeable raffia bags, semipermeable kraft paper bags, and impermeable PET bottles. The following were evaluated: seed moisture content; germination test; first germination count; germination speed index; hypocotyl length; root length and hypocotyl dry mass and root dry mass. Cost analysis of treatments was also performed. The physiological quality of cowpea seeds was more markedly reduced under storage conditions in permeable packages with fludioxonil + metalaxyl treatment. Impermeable packaging without chemical treatment decreased deterioration of physiological quality in cowpea seeds and is economically viable for a storage period of up to 90 days.

Keywords: *Vigna unguiculata*; Physiological potential; Sustainability; Economic viability.

Resumen

El objetivo de este trabajo fue evaluar la calidad fisiológica de las semillas de frijol caupí, así como la viabilidad económica del almacenamiento en diferentes paquetes y productos químicos. Las semillas fueron tratadas químicamente con los ingredientes activos: fipronil; fipronil + piraclostrobina + metil tiofanato; fludioxonil + metalaxil. Las semillas sin tratamiento químico constituyeron el control. Fueron empacados en bolsas de algodón (permeables); bolsas de rafia (permeables); bolsas de papel kraft (semipermeables); y botellas tipo mascota (impermeables) y almacenadas durante 90 días. Se llevaron a cabo las siguientes evaluaciones: grado de humedad de la semilla; prueba de germinación; primer recuento de germinación; índice de velocidad de germinación; longitudes de hipocotilo y radícula y pesos de materia seca de hipocotilo y radícula. Realizó el análisis de costos. La calidad fisiológica de las semillas de frijol caupí se reduce más notablemente bajo condiciones de almacenamiento en envases permeables y se trata con fludioxonil + metalaxil. El empaque a prueba de agua del tipo de botella para mascotas, sin la adición de tratamiento químico, disminuye el deterioro de la calidad fisiológica en las semillas de caupí y es económicamente viable en el período de almacenamiento de hasta 90 días.

Palabras clave: *V'igna unguiculata*; Potencial fisiológico; Sostenibilidad; Viabilidad económica.

1. Introduction

Cowpea [*Vigna unguiculata* (L.) Walp.], is an economically important legume in Brazil, being the second most consumed type of bean in the country. It is an excellent source of energy and protein and its cultivation generates jobs and income (Rocha et al., 2017).

Brazil stands out as one of the world's largest bean producers, accounting for about 13% of global production (Brasil, 2016), however, it is the third largest importer of legumes (Conab, 2018). The need to import beans reveals that the country still does not have production capable of supplying the domestic market, and thus there is a need to develop technologies capable of increasing crop productivity to meet the demands of national consumption.

Despite exhibiting good adaptability in the Northeast region of Brazil, cowpea has low yields due to factors such as climatic adversity, especially irregular rainfall (Freire Filho et al., 2011). This, in turn, impacts plant development (Martins et al., 2018), even though cowpea is more tolerant to low rainfall than other species (Boukar et al., 2018). Therefore, avoiding the storage of seeds in conditions that are inadequate for the preservation of seed quality (Resende et al., 2008) is of high importance for crop planning in order to tolerate the low amount of available water.

Considering the importance that good seed quality has on the development of crops, the maintenance of physiological potential is necessary. This can be achieved through alternatives such as seed treatment and storage in appropriate packaging (Marcos Filho, 2015), as well as the adoption of new cultivars that enable farmers to market a better-quality product (Alves et al., 2009).

Seed storage under controlled or uncontrolled conditions at a low or high technological level can be effective in maintaining physical performance in terms of germination and vigor. In addition, seed treatment stands out as being used over the years as a protective control measure (Reddy, 2013), being practically a component of integrated pest management. Nonetheless, studies that validate the action of insecticides and fungicides on cowpea are important, given the high number of synthetic products developed and released on the market today.

Products chosen for treatment should be those that efficiently eradicate pathogens or insects yet do not significantly debilitate seeds (Ludwig et al., 2011). These products must not be toxic to plants, humans or the environment, and be non-corrosive, easy to use, inexpensive, and provide full conditions for the maintenance of seed viability (Peske & Baudet, 2012). Such products would then contribute to the preservation of the natural environment and the health of farmers (Hoi et al., 2009).

Thus, the objective of the present study was to evaluate the physiological quality and economic viability of cowpea seeds stored under different packaging and chemical treatments.

2. Materials and Methods

The research has an exploratory character, as considered by Pereira et al. (2018) and, it was performed in a laboratory simulating conditions found in the field and quantitative. The experiment was carried out at Phytotechnics Laboraty of Federal University of Piauí, Bom

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Jesus, state of Piauí, Brazil, using cv. BR 17 Gurguéia cowpea seeds without previous treatment.

The studied cultivar is recommended for cultivation in the North and Northeast regions of Brazil. One-hundred cowpea grains weigh 12.5 g, and the species has a 70-80-day cycle after emergence. It is semi-prostrate and belongs to the sempre-verde commercial group (Embrapa, 2008).

The seed lot was initially analyzed for germination and vigor by evaluating moisture content and performing germination, first germination count and hypocotyl and root length tests, all prior to storage.

The experimental design was completely randomized with four replications in a 4x4 + 1 factorial arrangement (seed treatments, types of packaging and storage period) for a total of 17 treatments. The seed treatments were fipronil; fipronil + pyraclostrobin + thiophanate methyl; and fludioxonil + metalaxyl; and the control (treatment without chemical addition). The seeds were subsequently packed in four different types of packages containing on average 0.4 kg of seeds in each treatment: permeable cotton cloth bags; permeable raffia bags; semipermeable kraft paper bags; and impermeable PET bottles. The seeds were stored for 90 days (December 2016 to March 2017) in a large farm shed located in Uruçuí, state of Piauí, Brazil (Lat. 9º05’20.4’’S; Long. 44º20’55.1’’W, and Alt. 283 m), with temperatures between 22 °C and 30.3 °C and relative humidity of 42% to 68%.

Seeds were analyzed for physiological quality and economic viability at the end of the three months of storage as follows:

Degree of humidity (H%): Two replicates were performed for each experimental unit in a greenhouse at 105 ± 3 ° C for 24 hours, according to the methodology described in the rules for seed analysis (Brasil, 2009).

Germination test (G%): Performed using four replicates of 50 seeds for each treatment. Seeds were sown on a roll of Germitest® paper moistened with water at 2.5 times the weight of dry paper and maintained in B.O.D. at a temperature of 25 ºC and a photoperiod of 16 hours for seven days, at which time the evaluation was performed. Results are expressed as percentage of normal seedlings.

First germination count (FGC): Conducted together with the germination test. Normal seedlings were counted on the fifth day after sowing. Results are expressed as percentage of normal seedlings. *Germination speed index* (GSI): Done as part of the germination test. Normal germinated seedlings were counted daily (1st to 8th day). The index was calculated using the formula proposed by Maguire (1962).

Hypocotyl length (HL) *and root length* (RL): Performed using ten repetitions of 20 seeds on Germitest® paper pre-moistened with distilled water and sealed in bags within B.O.D. at 25 ºC for seven days. Normal emerged seedlings (primary root and hypocotyl) were measured using a graduate (cm) ruler (Nakagawa, 1999).

Hypocotyl dry mass (HDM) *and root dry mass* (RDM): Evaluated for normal seedlings obtained from the germination test. Cotyledons were removed, deposited in paper bags and placed in an oven at 80 ºC for 24 hours (Nakagawa, 1999). Hypocotyl and root were then weighed using a precision scale (mg).

Treatment costs were analyzed at the end of the experiment taking into consideration local values of the chemicals and packaging used, based on December 2016, and converted into American dollars (US$). The volume of each package was standardized at 930g of seeds so that the price of each package for each kg of seed, as well as the costs of applying the chemicals, could be calculated.

The following was then calculated: (1) PLS = [(G% x P%)/100], where PLS = pure live seeds; G% = percentage of germinated seeds in each treatment; and P% = percentage physical purity of seeds.

With PLS, the following was calculated (2): EP = (NP/ PLS), where EP = emergence of field plants; NP = estimated number of plants per hectare; and PLS = pure live seeds.

Next, the following was calculated: (3) NS = [(EP x WTS)/1000], where NS = number of seeds required per hectare; EP = emergence of field plants; and WTS = weight of one thousand seeds (g).

Finally, cost per treatment was calculated by summing all costs of each treatment from the equation (4): TC = ((SC kg^{-1} x NS) + PC + CTC)), where: TC = total cost (US$); SC kg^{-1} = seed cost per kilogram (US$); NS = estimated number of seeds per hectare; PC = packaging cost (US$); and CTC = chemical treatment cost (US$).

Analysis of variance was performed with the effects of the treatments being evaluated by F-test for the response variables (H%), (G%), (FGC), (GSI), (HL), (RL), (HDM) and (RDM), considering each combination of factors as a treatment and the initial measurement. A factorial analysis was then done with the additional of the control according to the mathematical model (5): $y_{ij(z)} = \mu + C_{i(z)} + S_{j(z)} + C_{i(z)}\, S_{j(z)} + e_{ij(z)}$ where, $y_{ij(z)}$ = Observation associated with the *i*-th chemical treatment in combination with the *j*-th form of storage within the *z*-th type (initial or treatment); μ = average of the experiment; $C_{i(z)}$ = effect of the *i*-th chemical treatment within type; $S_{j(z)}$ = effect of *j*-th storage packaging within type; $C_{i(z)}\, S_{j(z)}$

= effect of interaction of *i*-th chemical treatment with *j*-th packaging within type; and $e_{ij(z)}$ = error associated with observing *i*-th chemical treatment associated with *j*-th packaging within type. The usual model for a completely randomized design in a factorial scheme was followed for the other characteristics. After the analysis of variances and detection of the significance of interactions and the means were compared by Tukey test (P<0.05) using SAS® statistical program.

3. Results and Discussion

All analyzed variables presented a significant interactive effect (P<0.01) by the F test, which indicates the existence of variability among the studied treatments (treated seeds x packaging).

Analyzing seed moisture (H%) after the storage period revealed that there was an increase in seed water content in almost all packaging associated with seed treatments, except when stored in kraft paper and treated with fipronil + pyraclostrobin + thiophanate methyl or with fludioxonil + metalaxyl, in which seed water content decreased by 4.22% and 7.04%, respectively, in relation to the initial content, but not differing from the control (Table 1).

Table 1. Average values of technological characters of seeds of cowpea cv. BR 17 Gurgéia submitted to four different chemical treatments, four types of storage and initial treatment. Bom Jesus, Piauí, Brazil.

	--------------- Humidity (%) ---------------				-------------- Germination (%) --------------			
Treat[a]	Fipronil	(Fip+Pyr+Met)[c]	(Flu+Met)[d]	(Cont)[e]	Fipronil	(Fip+Pyr+Met)[c]	(Flu+Met)[d]	(Cont)[e]
Initial[b]	14.2 C	14.2 BC	14.2 C	14.2 B	82.0 A	82.0 A	82.0 A	82.0 ABC
Coton	20.6 Ab	15.4 BCc	23.4 Aa	20.2 Ab	97.0 Aa	94.6 Aa	81.0 Aa	87.6 ABa
Raffia	20.2 Aa	19.0 Aa	20.8 Ba	19.5 Aa	97.6 Aa	88.0 Aa	33.0 Bb	81.6 BCa
Kraft	17.8 Ba	13.6 Cb	13.2 Cb	15.2 Bb	93.6 Aa	96.0 Aa	28.6 Bc	65.6 Cb
PET	15.4 Ca	16.1 Ba	15.2 Ca	15.6 Ba	96.0 Aa	97.0 Aa	95.0 Aa	98.6 Aa
Average	17.38				83.06			
CV[f] (%)	1.03				7.90			
	--- First germination count (FGC) ---				--- Germination speed index (GSI) ---			
Initial	23.8 B	23.8 B	23.8 B	23.8 B	-	-	-	-
Cotton	79.0 Aa	94.6 Aa	81.6 Aa	81.0 Aa	31.50 Aa	33.98 Aa	29.03 Aa	28.85 Ba
Raffia	94.6 Aa	92.0 Aa	31.0 Bc	58.6 Bb	32.05 Aa	33.43 Aa	10.98 Bc	23.78 Cb
Kraft	91.0 Aa	87.0 Aa	28.6 Bc	56.0 Bb	31.95 Aa	32.08 Aa	11.43 Bc	22.00 Cb
PET	96.0 Aa	91.6 Aa	94.6 Aa	91.6 Aa	36.15 Aa	37.15 Aa	34.35 Aa	35.83 Aa
Average	74.8				29.03			
CV[f] (%)	10.70				8.35			

Means followed by the same uppercase letter in columns and lowercase letter in rows do not differ from each other by the Tukey test (P<0.05); [a]treatments; [b]initial measurement; [c]fipronil +

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pyraclostrobin + thiophanate methyl; [d]fludioxonil + metalaxyl; [e]control, without chemical treatment; [f]coefficient of variation. Source: Data from research result. Source: Authors.

The loss of water from seeds treated with these products, in addition to physiological changes, may indicate fungal activity, which may lead to reduced germination performance and seedling survival. Possibly another factor must be related to the characteristic resistance of cowpea to water vapor exchange with the relative humidity of the environment in semipermeable packaging, such as kraft paper. Eliud et al. (2010) found that temperature and relative humidity of 30.8 ºC and 80.1%, respectively, showed their greatest reduction in common bean (*Phaseolus vulgaris*) virility after 30 days of storage under controlled conditions, which is only a third of the storage time of the present experiment.

The highest water content after storage occurred for seeds in cotton bags treated with fludioxonil + metalaxyl, which had a 64.78% increase in relation to initial seed content. This demonstrates that seeds stored in packaging with higher permeability are subject to variation in air humidity, and that the type of packaging directly influences the maintenance of seed moisture during storage (Neves et al., 2014). It is possible that stored seeds sought hygroscopic equilibrium according to the relative humidity of the storage environment (Baudet & Villela, 2012).

Reducing water content leads to reduced seed viability and vigor, as seeds have a complex antioxidant defense system involving enzymatic and non-enzymatic components (Mittler et al., 2002; Kibinza et al., 2011; Sofo et al., 2015).

For germination percentage, all treatments with fipronil and fipronil + pyraclostrobin + thiophanate methyl, regardless of the packaging, did not differ from the initial value, as well as the control treatment in cotton bags and PET bottles. However, seeds treated with fludioxonil + metalaxyl stored in raffia and kraft bags had significant reductions of 59.75% and 65.12%, respectively, in relation to initial germination. The germination percentage for seeds stored in these types of packaging was also significantly affected in thte treatment without application.

Reduction in seed germination percentage using fludioxonil + metalaxyl can be explained by the fact that the concentration of the product caused a reduction in the decomposition rate of starch stored in the germinating seeds. This effect was observed with the use of cypermethrin by Obidola et al. (2019) when studying the phytotoxicity of this insecticide in cowpea germination, and who found that low concentrations of insecticide increase a-amylase enzyme activity in the first days of storage, which subsequently reduces;

nonetheless, high concentrations affect germination power with a reduction in cowpea starch levels.

Likewise, seeds that were not submitted to any kind of chemical treatment experienced a reduction in germination, also when packed in raffia and kraft bags. When stored in raffia bags, there was a 20% reduction in germination compared to initial germination, demonstrating that greater sensitivity of cowpea may occur during the initial development of seedlings after sowing. Similar results were obtained by Carvalho et al. (2016), who studied prepackage cooling in different types of soybean (*Glicine max* L.).

This negative effect on germination makes seed storage unfeasible since it is well below 80%, which is the stipulated standard for the commercialization of bean seeds (Brasil, 2013). The reduction in germination in these cases may be associated with several mechanisms involved in the deteriorative process of seeds. One such mechanism is lipid peroxidation, which is linked to a series of degenerative changes initiated from the formation of reactive oxygen species (ROS) (Halliwell, 2006; Ahmad et al., 2009; Izumi et al., 2010), with the production and accumulation of ROS resulting in metabolic disorders that may lead to oxidative destruction of the cell (Caverzan et al., 2019).

The longevity of seeds stored in different types of packaging may vary due to moisture exchange (Eliud et al., 2010; Guedes et al., 2012). However, no significant differences in germination were observed in treatments with fipronil + pyraclostrobin + thiophanate methyl and only fipronil among the different packaging used, which may indicate no changes in enzymatic activities that contribute to cowpea seed germination, in the respective chemical treatments and during the storage period evaluated. Therefore, this performance is important since it indicates no phytotoxic effect of these active ingredients on cowpea seeds.

Regarding the influence of treatments on first germination count (FGC), satisfactory results were observed with the use of fipronil + pyraclostrobin + thiophanate methyl, which had a positive effect for all packaging, with averages of 90.15 and 91.3% normal seedlings, respectively (Table 1), showing stimulation of the germination process since FGC was initially 23.8%. Similar results for cowpea were obtained by Oliveira et al. (2015), however, it is noteworthy that these authors observed an intense reduction in seed vigor after 90 days of storage with the use of fipronil + pyraclostrobin + thiophanate methyl under controlled conditions of temperature and humidity, indicating a probable phytotoxic effect of the active ingredients, impairing physiological quality of seeds after longer storage.

The control treatments (without chemical treatment) and fludioxonil + metalaxyl provided higher numbers of normal seedlings when packaged in PET bottles and cotton bags,

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which differed statistically from seeds packaged in raffia bags and kraft paper (Table 1). This demonstrseas that the associated us of fludioxonil + metalaxil in semipermeable (kraft) and permeable (raffia) packaging can contribute to the reduction of germination characteristics of cowpea.

Seeds packed in PET were observed to be superior with regard to germination speed (GSI), reflecting the higher germination performance of these seeds (Table 1). This behavior is accepted to be a result of this type of packaging being able to preserve or minimally alter the physiological characteristics of seeds during storage, and thus reduce deterioration.

Even so, it was observed that the control treatment (without chemical treatment) was statistically inferior in the majority of the used packaging, except for the use of PET bottles in relation to treated seeds. Therefore, it is feasible to treat seeds for storage and subsequent cultivation. Similarly, as the other characteristics evaluated in the germination process, seeds treated with fipronil and fipronil + pyraclostrobin + thiophanate methyl promoted the highest values of GSI and did not differ depending on the packaging used, indicating that somehow these compounds promote the germination process independently of the packaging used.

The treatments of fipronil and fipronil + pyraclostrobin + thiophanate methyl expressed the highest results for growth according to hypocotyl length, but both did not differ when compared to initial length and the different types of packaging, similar to what was found for the germination variables (Table 2).

Table 2. Average values of technological characters of seeds of cowpea cv. BR 17 Gurgéia submitted to four different chemical treatments, four types of storage and initial treatment. Bom Jesus, Piauí, Brazil.

	--------- Hypocotyl length (cm) ---------				------------- Root length (cm) -------------			
Treat[a]	Fipronil	(Fip+Pyr+Met)[c]	(Flu+Met)[d]	(Cont)[e]	Fipronil	(Fip+Pyr+Met)[c]	(Flu+Met)[d]	(Cont)[e]
Initial[b]	10.8 A	10.8 A	10.8 A	10.8 AB	7.4 B	7.4 B	7.4 A	7.4 B
Cotton	16.3 Aa	15.8 Aa	11.0 Aa	12.6 Aba	13.4 Aa	11.2 ABab	7.7 Ab	8.5 ABab
Raffia	17.1 Aa	14.1 Aa	0.0 Bc	6.7 Bb	12.0 Aa	6.9 Bab	0.0 Bc	4.2 Bbc
Kraft	13.2 Aa	13.0 Aa	0.0 Bb	9.6 ABa	10.6 Aa	10.6 Aba	0.0 Bb	5.6 Bab
PET	15.5 Aa	15.5 Aa	12.2 Aa	16.3 Aa	13.1 Aa	13.1 Aa	10.0 Aa	13.8 Aa
Average	13.07				8.70			
CV[f] (%)	3.32				25.86			
	------------ Hypocotyl dry mass (mg) ------------				----------------- Root dry mass (mg) -----------------			
Cotton	0.36 Aa	0.39 Aa	0.44 Aa	0.40 Aa	0.11 Ba	0.04 Aa	0.02 Aa	0.04 Aa
Raffia	0.42 Aa	0.21 Aab	0.00 Bb	0.10 ABb	0.11 Ba	0.04 Aa	0.00 Aa	0.10 Aa
Kraft	0.33 Aa	0.10 Aa	0.00 Bb	0.08 Ba	0.61 Aa	0.18 Ab	0.00 Ab	0.05 Ab
PET	0.49 Aa	0.34 Aa	0.16 ABa	0.09 ABb	0.12 Ba	0.13 Aa	0.34 Aa	0.14 Aa
Average	0.86				0.78			
CV[f] (%)	9.13				9.77			

Means followed by the same uppercase letter in columns and lowercase letter in rows do not differ from each other by the Tukey test (P<0.05); [a]treatments; [b]initial measurement; [c]fipronil + pyraclostrobin + thiophanate methyl; [d]fludioxonil + metalaxyl; [e]control, without chemical treatment; [f]coefficient of variation. Source: Data from research result. Source: Authors.

These results can be explained by the active ingredients used as cell bioactivators increasing germination, emergence and seedling development (Avelar et al., 2011). This work corroborates that of Ferreira et al. (2016), who studied the effects of fungicide and insecticide treatment on soybean seed quality before and after storage and found an improvement in vigor of seeds treated with fipronil and fipronil + pyraclostrobin + thiophanate methyl after two months of storage.

Hypocotyl length was also significantly affected, and the combined effect of fludioxonil + metalaxyl on seeds stored in PET bottles and raffia bags did not differ from values for the initial treatment (Table 2). Hypocotyl length is important because it can contribute to a larger or smaller lodging of plants in the field which can influence harvest (Rocha et al., 2009) since more upright plants contribute to mechanical harvesting (Matos Filho et al., 2009). Growth variables were not measured in kraft paper and raffia bag packaging using fludioxonil + metalaxyl due to the attack of pathogens (fungi) on seeds, which completely destroyed the structures to be measured. Fungi promote endocarp disruption and, as a consequence, can potentially reduce seed germination (Delgado-Sánchez et al., 2013).

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The behavior observed in the seeds with the occurrence of fungi may be associated with the fungicide not having promoted protection against pathogenic attacks and environmental stress. Nevertheless, these results may help in the importance of using fungicides in seed treatment, since this protection should contribute to further plant development.

According to Castro et al. (2006), fungicides aim to control the fungal population and thus reduce their deleterious effects. However, there they can vary in their results, and are capable of modifying metabolism and even plant morphology. In the control treatment, although not statistically different, the seeds packed in PET bottles had higher HL in relation to seeds of the other types of packaging, and even in relation to the initial treatment. This may have been due to the reduced capacity for gas exchange with the external of the impermeable packaging environment, which would avoid increases in humidity (Table 2), in addition to providing greater protection against the entry of organisms harmful to seed health.

The fipronil treatment differed positively in root length for all packaging types and in relation to chemical treatments, with significantly higher values (Table 2). For the treatment with fipronil + pyraclostrobin + thiophanate methyl only the seeds in raffia bag packaging had values lower than the initial values. The action of the active ingredients in the seeds may have induced changes in cellular proteins, which, according to Hakeem et al. (2012), undergo changes in their expression levels when under stressful environmental conditions.

The treatments fludioxonil + metalaxyl and control with packaging of cotton bags and PET bottles showed significant results in relation to the initial treatment, probably because these treatments favored root growth. With kraft paper and raffia packaging, seeds had values equal to zero, due to the presence of fungi in the seedlings, which may be associated with the environmental conditions of storage.

The undesirable results regarding the fludioxonil + metalaxyl treatment can be explained by the residual effect of the chemical not lasting the entire storage period. This does not imply that the active ingredients are ineffective, but possibly that they did not remain viable over time, with storage fungi, such as the genera *Aspergillus* and *Penicillium*, developing later and causing the cited undesirable effects. Similar results were found by Pereira et al. (2007) with the use of fludioxonil + metalaxyl and imidacloprid + thiodicarb in the control of *Penicillium*. These authors explain that only when the seeds were stored and treated did the chemicals protect them, because the tendency is that the population of microorganisms increases after storage.

The overall results for hypocotyl dry mass (HDM) were similar to those for hypocotyl length (HL) because HDM results from HL; that is, hypocotyl size may reflect, more or less, dry mass.

The results for root dry mass (RDM) differed from those for root length (RL) in relation to packaging and chemical treatments (Table 2). In the treatment with fipronil alone, kraft paper and cotton bags did not different in relation to root length.

The seeds treated with fipronil + pyraclostrobin + thiophanate methyl and conditioned in raffia bags differed in relation to RL, but positively (Table 2), because although length was less than in the other treatments, the roots may have been thicker, as implied by greater dry mass. For the control, kraft paper and raffia bag packaging had higher values of RL, demonstrating that longer roots can have greater weight, but less thickness.

Analysis of storage costs for cowpea seeds in this experiment revealed that the use of PET bottles without chemical treatment had the lowest cost (Table 3), being 70.39% lower than the higher cost treatment of kraft paper packaging with fludioxonil + metalaxil. This information is important considering the good germination results without the use of chemical pesticides for the treatment of PET bottles and control.

Table 3. Costs for storing seeds of cowpea cv. BR 17 Gurgéia for 90 days in different packaging and chemical treatments. Bom Jesus, Piauí, Brazil.

Packaging	(Fipronil)	(Fip+Pyr+Met)[b]	(Flu+ Met)[c]	(Control)[d]
Cotton	US$[a] 199.07	US$ 198.70	US$ 225.19	US$ 206.70
Raffia	US$ 197.64	US$ 212.73	US$ 525.65	US$ 221.55
Kraft	US$ 205.55	US$ 195.61	US$ 633.76	US$ 275.58
PET	US$ 205.10	US$ 198.00	US$ 196.38	US$ 187.62

[a]American dolar; [b]fipronil + pyraclostrobin + thiophanate methyl; [c]fludioxonil + metalaxyl; [d]control, without chemical treatment. Source: Data from research result.

Furthermore, this result is also of great importance, in practical terms, from the productive and economic point of view, as well as the social and environmental standpoint. The capacity of PET bottles to prevent water vapor exchange with the environment makes the conservation of seeds possible without the use of chemical pesticides. Thus, there is no need for costs related to purchasing chemicals, which makes this a suitable method, especially for rural families on small farms.

It is noteworthy that eliminating the practice of pre-treatment of seeds in the production process would provide gains for the health of workers involved in the operation of chemical application, as well as promote the preservation of natural resources by not disposing of contaminants into the agroecosystem. In addition, safe pesticide handling and use practices are often overlooked by farmers, indicating the need for public policies that implement reductions in the use of harmful pesticides (Macharia et al., 2013; Mengistie et al., 2017) and improve technical assistance to small farms (Stadlinger et al., 2011).

However, the results obtained by not treating cowpea seeds do not imply recommending no preventive application of the tested products. There remains the possibility that residual effects present in seeds when disposed in the soil provide greater protection regarding pests and diseases compared to those without previous treatment.

The current model used by large properties involves seed storage and transport using raffia bag packaging. The present study found that seeds treated with fipronil and packaged in raffia bags experienced an increase of 5.34%, compared to lower cost treatments (Table 3). Furthermore, relating these data to those obtained for germination percentage (Table 1), demonstrates the viability of deciding the type of treatment and storage most appropriate for conserving physiological quality of cowpea seeds, taking into consideration the management options of larger enterprises.

Overall, this study presents alternatives, aimed at being effective and sustainable, to the various storage methods that protect agricultural products from mold and insects, since aerobic respiration is more restricted (Navarro et al., 2010). For cowpea, this may be related to increasing free fatty acid content, decreasing oxygen concentration and increasing carbon dioxide (CO_2) concentration due to seed respiration. Although seed deterioration is inevitable, the rate can be reduced by factors such as temperature and humidity (Cardoso et al., 2012).

The alternative of greater economic viability for the treatment with cotton bags was when it was associated with the fipronil + pyraclostrobin + thiophanate methyl treatment, which had a reduction of 3.87% compared to the treatment without application. The treatment using fludioxonil + metalaxyl proved to be the least economically viable option for storage in cotton bags because it increases cost by 8.95% compared to the control.

The least economically viable treatment among those tested was kraft paper packaging combined with fludioxonil + metalaxyl, which resulted in low germination after 90 days of seed storage.

4. Final Considerations

Storage in PET (impermeable) bottles without chemical treatment promotes the maintenance of cowpea seed physiological quality and is economically viable for up to 90 days.

The research provides important information about cowpea seeds under storage conditions with or without chemical treatment and the effects on the physiological attributes of the seeds. This information can help seed producers, farmers and for future scientific work.

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