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Effects of packaging material and seed treatment on Weevil (*Callosobruchus maculatus* (F) Coleoptera: Bruchidae) infestation and quality of cowpea seeds

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This study was conducted to determine the effects of seed treatments using Neem leaf powder, pepper fruit powder, camphor, groundnut oil and wood ash in combination with five different packaging materials (cloth bag, paper bag, glass bottle, polythene bags and plastic container) on the viability and germination vigor of cowpea seeds. The experiment was laid out in a completely randomized block design in a 5 by 6 factorial with three replications. 1 kg of dried cowpea seeds with a uniform moisture content of 11% were stored for 6 months in different packaging materials in combination with six different seed treatments: No treatment (control), Camphor (Chemical treatment) -4 g/kg of seeds, Neem (*Azadirachta indica* L.) leaf powder (Botanical) -10 g/kg of seeds, Groundnut Oil / Palm oil - 5 ml/kg seeds, Powdered Dry pepper (Capsicum spp) -10 g/kg of seeds and wood ash 1 kg/kg of seeds. Indoor temperatures were between 25.3 and 30.7°C, outdoor temperatures between 26.1 and 31.1°C and relative humidity between 61.0 and 82%. Results obtained from the study showed that packaging material and seed treatment had high significant effect on the vigor and germination percentage of cowpea seeds. Germination of seeds treated with powdered pepper (73%) and neem leaf powder (72.5%) were significantly higher than other treatments. Seeds stored in plastic containers had the highest vigor and germination percentages (61.1 and 77.1% respectively) followed by glass bottle (60.3 and 72.2% respectively) after 6 months storage period. It seems that for better storage of cowpea seeds for a period not exceeding 6 months it is preferable to use plastic containers and glass bottles in combination with dry powdered pepper or neem leaf powder to maintain seed vigor and viability.

Key words: Cowpea, germination, packaging material, seed treatment, vigor.

INTRODUCTION

Cowpea (*Vigna unguiculata* (L) Walp) is the second most important grain legume grown in Sierra Leone after groundnut and is used mainly as a food grain. It contains 23% protein by weight and plays an important role in

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providing the protein requirements of Sierra Leoneans especially those living in the rural areas (Crop Production Guidelines for Sierra Leone, 2005). Cowpea also supplements the income of many small-scale farmers and contributes to the maintenance of soil fertility by fixing nitrogen in the soil. The use of quality seeds in cultivation is one of the most important factors that can increase farm level yield. Although seed quality is governed by genetic makeup, seed storage and retention of viability are important for seed vigor (Deepa et al., 2011). As seed is considered as one of the important basic agricultural input for obtaining higher yields. Therefore production of quality seeds of high germination is of utmost importance in a seed program. Seed security is a key to the attainment of household food security among resource poor farmers in developing countries (Gari, 2004). Among the causes of seed insecurity in Africa is inadequate facilities and the use of inappropriate methods for seed storage among rural farmers. This impairs the maintenance of sufficient and safe seed sources. Successful seed storage is key to farmers' seed security and may also enable communities to generate income through collecting, storing and selling seeds. Seed storage problems are partly responsible for farmer's failure to save seeds of traditional crops (Delouche, 1982).

Several factors affect the longevity of seeds in storage and these include the variety of seed, initial seed quality, storage conditions, moisture content, insects pests, bacteria and fungi. The poor storage of cowpea seeds is a problem because of cowpea weevil (*Callosobruchus maculatus*) infestation. The infestation starts in the mature standing crop, and then at the threshing floor and eventually carried into store house, leading to seed deterioration under ambient storage conditions. There is the growing awareness of the deleterious consequences resulting from the widespread use of many chemical pesticides. The safe and feasible approach is the treatment of seeds with botanicals, which are safe, economical, cheap, eco-friendly in nature and non-toxic to man and animals. Among the botanicals, using castor, neem powder, neem oils, lemon grass are known to be effective protectants against storage insects; they can reduce infestation and maintain the quality of the seed in terms of viability and vigour for longer period in storage. Study by Basavegowda et al. (2013) found that Chickpea (*Cicer arietinum*L.) had higher seed germination and vigour index but lower insect damage and seed infection when commercial cold storage structures are used for maintaining seed quality. Duruigbo (2010) studied the treatments of seeds using edible, non edible oils, fungicides, insecticides and plant products and its influence on seed viability and insect infestation. Maintenance of seed quality during the storage period is important not only for successful crop production but also for maintaining the quality and integrity of the seeds that are in constant threat of genetic erosion (Barua et al., 2009). To maintain the quality of seeds during storage the standardization of suitable seed treatment and packing material is most important because seed treatment is the basic measure to assure adequately healthy crops at emergence and during further growth of plants (Wani et al., 2014).

The unavailability of quality seeds is a major problem affecting cowpea production in Sierra Leone. Information on prolonging the shelf life of cowpea seeds under ambient storage conditions is very limited in Sierra Leone. The aim of this research was to collect data on techniques for storage of cowpea seeds. The specific objectives are i) to identify a suitable packaging material for better storage of cowpea seed, ii) to determine the effect of seed treatment with chemicals and plant products on viability and vigour during storage and iii) to determine the effect of packing material on insect pest damage.

Cowpea (*V. unguiculata* (L) Walp) is the second most important grain legume after groundnut that is grown in Sierra Leone and is used mainly as a source of protein food and also supplements the income of many small-scale farmers. Although seed quality is governed by genetic makeup, seed storage and retention of viability are important for seed vigor (Deepa et al., 2011). Failure of farmers to save seeds of traditional crops could be due to inadequate facilities and the use of inappropriate methods for seed storage. The poor storage of cowpea seeds is a recipe for the cowpea weevil (*C. maculatus*) to infest and destroy seeds. Infestation may start in the mature standing crop, at the threshing floor, and eventually carried into the store house leading to seed deterioration under ambient storage conditions. Factors such as the variety of seed, initial seed quality, storage conditions, moisture content, insect pests, bacteria and fungi also affect the longevity of seeds in storage. Attempts have been made by several workers with many crops to develop methods for maintaining the viability and vigour of seeds for longer storage. Wani et al. (2014) found that seeds of maize treated with Captan (3 g/kg) and packed in Cloth bags and stored for 9 months under ambient conditions maintained maximum seed quality compared to the use of Castor oil or Vitavex in cloth bags and or polythene bags. Study by Basavegowda et al. (2013) found that Chickpea (*C. arietinum* L.) had higher seed germination and vigour index but lower insect damage and seed infection when commercial cold storage structures are used for maintaining seed quality. Duruigbo (2010) studied the treatments of seeds using edible, non edible oils, fungicides, insecticides and plant products and its influence on seed viability and insect infestation. Maintenance of seed quality during the storage period is important not only for successful crop production but also for maintaining the quality and integrity of the seeds that are in constant threat of genetic erosion (Barua et al., 2009). To maintain the quality of seeds during storage the standardization of suitable seed

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MATERIALS AND METHODS

The experiment was conducted in 2012 in the Seed laboratory of the Njala Agricultural Research Centre (NARC) in Sierra Leone. The experiment was laid out in a completely randomized block design in a 5 by 6 factorial with three replications. 1 kg of dried cowpea seeds with a uniform moisture content of 11% before packaging was put in five different packaging materials (cloth bag, paper bag, glass bottle, polythene bags and plastic container) in combination with six different seed treatments: No treatment (control), Camphor (Chemical treatment) – 4 g/kg of seeds, Neem *Azadirachta indica* L.) leaf powder (Botanical) – 10 g/kg of seeds, Groundnut Oil / Palm oil – 5 ml/kg seeds, powdered dry Pepper (Capsicum spp) -10 g/kg of seeds and wood ash 1 kg/kg of seeds. Sufficient quantities of Neem leaves and dry pepper fruits were obtained, sun-dried for 5 days and ground into powder. The ground material was sieved using 1.0 mm wire mesh sieve. The treated seeds were stored for 6 months with indoor temperatures ranging from 25.3 to 30.7°C, outdoor temperatures from 26.1 to 31.1°C and relative humidity from 61.0 to 82%.

Data was collected on seed samples drawn from each treatment on the following parameters after storage for 6 months: moisture content, indoor and outdoor temperatures, relative humidity, 100 seed weight, percentage damaged seeds, percentage germination and percentage vigor. The indoor temperatures, outdoor temperatures and relative humidity were measured for 6 months using a Thermohygrometer at 9:00 am and 3:00 pm each day and monthly means recorded. The percentage of damaged seeds in each treatment were determined by dividing the numbers of seeds from 100 randomly selected seeds with holes/perforations by beetles by the total randomly collected seeds and expressed as a percentage. Percentage germination was calculated by dividing the number of germinated seeds by the total number of seeds sown after 8 days and expressed as a percentage. Percentage vigor was calculated by dividing the number of seeds that germinated at first count after 5 days by the total number of seeds sown. Data were subjected to analysis of variance (ANOVA) using Genstat Release version 7.2DE. Duncan's multiple range test (DMRT) was used for mean separation. Regression and correlation analysis were explored to examine relationships among various parameters.

RESULTS AND DISCUSSION

Results showed August had the highest indoor and outdoor relative humidity (79.2 and 82% respectively) whilst the lowest relative humidity was recorded in the month of April (61.0%) (Table 1). The high indoor temperature during the initial storage period and the high relative humidity (RH) especially at the 3 month storage period must have provided favourable conditions for the growth and multiplication of cowpea storage pests. McCormack (2004) also found that increasing the temperature during storage will increase insect activity resulting to increase seed damage. There was also significant ($P < 0.05$) difference in mean 100 seed weight of the cowpea seeds among the different packaging materials. Cowpea seeds stored in the plastic container had the highest mean 100 seed weight (14.9 g) after 6 months of storage, followed by seeds stored in the Glass bottle (14.8 g). However, there was no significant difference between the seed stored in the plastic container and glass bottle (Table 2). The lowest mean 100 seed weight (10. g) was recorded for seeds stored in cloth and paper bags. Similarly, seeds stored in the glass bottle and Plastic containers had the least seed damage and weevil severity score (Table 2). Packaging material significantly ($P < 0.001$) affected the percentage of damaged seeds and weevil severity score. The lowest percent damage in cowpea seeds was obtained by seeds stored in plastic containers (4.4%) whilst the highest was recorded by seeds stored in the cloth bag (39.7%). The seeds that were stored in the glass bottle and plastic container which had the lowest percentages of damage seeds also had the lowest weevil severity score (1.4 and 1.3 respectively). A strong positive a correlation ($r = 0.96$) was also observed between the percentage of damaged seeds and weevil severity score (Table 3). However, there was a negative correlation ($r = - 0.8$) between percent damage seeds and germination percentage.

It was also observed that the type of packaging material used to store cowpea seeds had significant effect on vigor and germination capacity. Highly significant ($P < 0.001$) difference was observed in mean vigor and germination percentages among cowpea seeds stored in the different packaging materials. The seeds stored in plastic containers which had the highest vigor (61.6%) also had the highest germination percentage (77.1%), followed by seeds stored in glass bottles which had vigor percentage of 60.3% and mean germination percentage of 72.2% (Figure 1). Seeds stored in cloth bags which recorded the least vigor percentage (43.4%) also had the lowest germination percentage (53.3%). There was also significant difference in mean germination percentage between seeds stored in the paper bag, polythene, glass bottle and plastic container and those stored in cloth bags (Figure 1). The results also indicate that there was strong positive correlation ($r = 0.94$) between percentage vigor and germination percentage (Table 3). Similar findings were also reported by Gurmit and Hari (1992) who found that storing seeds in vapour proof containers maintain the desired quality of seeds for

Table 1. Mean Monthly indoor and outdoor Temperature and Relative Humidity of storage environment during storage period.

Month	Temperature (°C)		Relative humidity (%)	
	Indoor temperature	Outdoor temperature	Indoor relative humidity	Outdoor relative humidity
April	30.7	30.3	62.2	61.0
May	30.2	31.1	65.6	66.4
June	29.0	29.6	70.1	72.0
July	27.5	28.5	74.0	74.2
August	25.3	26.1	79.2	82.0
September	28.3	29.2	75.8	77.8

Table 2. Effect of packaging material on mean 100 seed weight seed damage and weevil infestation of cowpea seeds.

Packaging material	Mean 100 seed weight (g)	Percent damage seed	Weevil severity score
Glass bottle	14.6 [a]	7.1 [a]	1..4 [a]
Paper bag	10.8 [c]	35.6 [c]	2.7 [c]
Cloth bag	10.8 [c]	39.7 [c]	2.8 [c]
Polythene bag	11.7 [b]	25.8 [b]	2.2 [b]
Plastic container	14.9 [a]	4.4 [a]	1.3 [a]
LSD (0.05)	0.56	4.2	0.2
CV (%)	9.6	40	22.6

Means followed by the letter in the same column are significantly difference ($p < O.05$) (DMRT).

Table 3. Correlation coefficients (r) of physiological and physical traits of cowpea seeds after 6 months storage.

Percent- D	-0.8243				
Vigor	0.9428	-0.7732			
MC	-0.4079	0.1826	-0.4742		
Seed wt	0.6041	-0.7898	0.6054	- 0.0089	
Weevil severity	-0.7921	0.9650	-0.7578	0.2002	-0.7821
	Germ	Percent- D	Vigor	MC	Seed wt

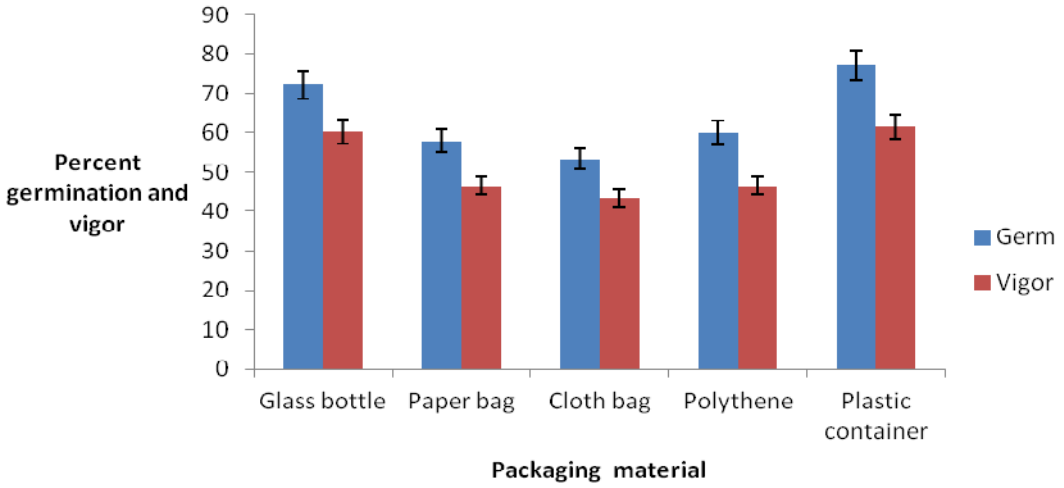

Figure 1. Effect of packaging material on vigor and germination of cowpea seeds.

Table 4. Interaction between period of storage and seed treatment on germination of cowpea seeds.

Storage period	Seed treatment					
	No treatment	Camphor	Neem leaf powder	Groundnut oil	Powdered dry pepper	Wood Ash
0	92 [a]	92 [a]	92 [a]	92 [a]	92 [a]	92 [a]
3	61.5 [d]	65.4[dc]	72.5[b]	67.8[b]	73.0 [b]	67.3[bc]
6	22.0[g]	45.5[e]	62.0[d]	29.0[f]	58.0[d]	47.5 [e]
LSD (0.05)	Storage period x Seed treatment 5.2					
CV (%)	9.2					

longer period than moisture pervious containers like cloth bag and gunny bag. This could be due to the fact that the impervious containers encouraged the activity of weevil resulting to higher percent seed damage and weevil severity and consequently low vigor and germination. Impervious containers also provided suitable environment and offered protection against contamination and serve as barriers against the escape of seed treatment chemicals than moisture pervious containers. The insects suffocate as soon as the oxygen in the container is used up. The percent seed damage caused by weevils significantly affected seed weight, vigor and germination of the seeds. The low germination could also be due to decrease in the seed weight caused by weevil damage resulting in less food reserve available in the seed to facilitate the growth of the seed.

There was a significant interaction between storage period and seed treatment of cowpea seeds. Generally, germination percentage of cowpea seeds decreased with months after storage. At 3 months after storage cowpea seeds treated with powdered pepper had the highest germination (73.0 %) followed by seeds treated with Neem leaf powder (72.5.5%) whilst the untreated cowpea seeds recorded lowest germination percentage (61.5%). This resulted in 33% reduction in germination of untreated seeds compared to 20.6% and 21.2% reduction in germination of seeds stored in powdered pepper and Neem leaf powder respectively.

At both 3 and 6 months after storage, significant ($P < 0.05$) differences were observed between the treated cowpea seeds and the untreated. Similar trend was also observed . after 6 months storage The cowpea seeds treated with neem leaf powder and dry pepper powder had the highest germination percentage (62 and 58% respectively) followed by the seeds treated with wood ash (47.5%) and camphor (45.5%) respectively. The lowest germination percent (22.0%) was again recorded by the untreated cowpea seeds (Table 4).

The results of the study clearly show that neem leaf powder, powdered pepper, wood ash and groundnut oil were effective in protecting cowpea seeds up to 3 months in storage. However, seed quality deteriorated rapidly resulting in low vigor and germination after 6 months. These results do not agree with the findings of Duruigbo (2010) who reported that seeds treated with neem leaf powder gave higher mean germination percentage for cowpea than seeds treated with black peeper, pepper fruit and soya bean oil and palm oil. This could be attributed to ovicidal and insecticidal action of azadiractin contained in neem leaf. It also has the ability to penetrate the cuticle of the insects, kills insects by flooding their spiracles thus causing asphyxiation. Similar results were also reported by Maraddi (2002) who observed that cowpea seeds treated with neem leaf powder at 5 g per kg of seeds recorded higher germination (39.5%) and vigour index (10.72) compared to control (34.2 and 8.64%) at the end of 10 months of storage period. The results of the current study contradict earlier report by Osekre and Ayertey (2002) who suggested the use of palm oil and coconut oils to control cowpea beetles. Although the groundnut oil reduced seed damage due to weevils, it adversely affected germination after 3 months of storage. The low germination percentage of seeds treated with groundnut oil could be attributed to impairment of respiration by the seed at the time of the test.

Conclusion

Results obtained from the present study suggest that plastic containers and glass bottles are better packaging materials for cowpea storage than polythene bags, paper bag and cloth bag under ambient storage. Application of neem leaf powder, powdered pepper and wood ash were most effective in protecting seeds from weevil damage and maintaining good seed quality and viability up to six month. The combination of neem leaf powder, powdered pepper wood ash and airtight containers in plastic containers significantly reduced seed deterioration. For short term storage it is best to package cowpea seeds in moisture proof containers than cloth or paper bags to prolong shelf life and maintain good seed quality. The use of this technology by resource poor farmers is low cost and will allow them to save enough seeds of high quality for future planting. However, farmers should not store their cowpea seeds in cloth bags or paper bags.

Declaration of conflict of interest

Ernest G. Kamara hereby declare that there is no financial or any other interest behind the development

and submission of this manuscript and that it is purely based on self aspiration and desire to build up my academic career as a researcher.

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