Effect of Season and Storage Method on Seed Quality of Slender Leaf (*Crotalaria Sp.*) in Western Kenya

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Abstract

Slender leaf (Crotalaria sp.) is an important source of nutrients, income and traditional medicines in Kenya. However, its production is constrained by lack of quality seeds. The aim of the study was to investigate the effect of various storage methods on seed quality of slender leaf and recommend the best method(s) of seed storage to the farmers in Western Kenya. Seeds from various morphotypes of slender leaf that had high germination percentage (90% and above) were stored in Kakamega and Siaya Counties using pots, jars, brown paper bags and polythene bags. Seeds were also stored in the freezer at (at temperature of -2^0 C) at University of Eldoret. Storage period was 3 months. Seed viability and vigour, were determined for each storage method. Data obtained from storage experiments was subjected to ANOVA and T-tests using Statistical Analysis Software (SAS). Season of growth and storage methods significantly influenced seed quality in Kakamega and Siaya. Seeds stored in pots, brown paper bags, jars and freezer had higher seed quality than those stored in polythene bags. It was concluded that in order to obtain high quality seeds farmers should store slender leaf seeds in pots or brown paper bags or plastic jars or freezer. Farmers should avoid storing seeds in polythene bags as it results in low quality seeds.

Key words: slender leaf, seed quality, storage

INTRODUCTION

Slender leaf is an important African Indigenous Vegetable (AIVs) which is a source of nutrients as well as revenue in Kenya (Abukutsa, 2011; Ebert, 2014; Mwaura, 2014; Simon, 2016). Slender leaf can also be used as green manure; livestock fodder, nematode suppression and a trap crop for witch weed (Nyalemegbea et al., 2011; Sikuku and Okello, 2013). It also has medicinal uses. Slender leaf is used to cure malaria, stomach pains and general swellings (Opande, et al, 2017; Uusiko et al., 2010). Enhanced production of these vegetables can help farmers to tap into all these uses.

Most farmers use farm-saved seed for the production of slender leaf vegetables. However farmers' traditional vegetable seed production is erratic and the quality of the seed is variable due to the differences in agronomic, harvest and post harvest practices (Abukutsa, 2011). Limited research has been undertaken on the production of quality AIV's seed on farmers' fields and seed storage methods (Abukutsa, 2014; Afari-Sefa *et al.,* 2011).

Seed storage has been shown to affect seed quality of crops hence influence germination potential. Factors that affect seed quality in storage include duration of storage, temperature in storage structure, seed moisture and the relative humidity in the storage structure, oxygen pressure, storage pests and diseases (Walters et al., 2010, Mbofung *et al.*, 2013, Wei *et al.*, 2011). All these factors interact with each other to bring about the aging of the seed. Aging is composed of deteriorative events superimposed on the seed capacity to detoxify and repair. The ability to repair membranes and detoxify toxins produced by respiration varies from species to species and from cultivar to cultivar (Zheng and Huang-cheng, 2014). Since the process of seed deterioration in storage varies from species to species this study set out to investigate the response of slender leaf seeds to different storage conditions.

MATERIALS AND METHODS

This study was part of a research that investigated agronomic and post harvest practices that influence seed quality in slender leaf vegetables. The research was done in 3 stages. The first stage was seed collection. Slender leaf seed samples were collected from farmers in Kakamega and Siaya Counties using scientific germplasm collection techniques (Maina, et al., 2012). The second stage was morphological characterization where the seeds were sown in boxes placed in a green house at the University of Eldoret. After the growth of plants 3 different varieties were identified using morphological descriptors. These were *C. brevidens intermedia*, *C. ochroleuca* and *C. brevidens parviflora*. Seeds from each variety were harvested separately and cleaned. They were then sown in boxes placed in the green house for purification. The 3rd stage involved field experiments conducted in Kakamega and Siaya Counties. The seeds from the green house experiments were used to establish plants in the field. The effect of agronomic and post harvest practices (variety, leaf defoliation, stage of seed harvest, drying and threshing methods) on seed quality was investigated. Percent germination (PG) and speed of germination index (SGI) were determined for various combinations of these factors. The procedures and results of these 3 stages have been reported in greater detail elsewhere (Maina, et al, 2013).

In the 4th stage (which is the subject of this paper) seeds obtained from field experiments in Siaya and Kakamega counties for different seasons (long and short rain seasons) were stored. Seeds selected for storage were those with high PG for each slender leaf varieties. These were *C. brevidens intermedia* seeds harvested at black pod stage from non defoliated plants, shelled by hand and dried in the sun (PG - 91%); *C. ochroleuca* seeds harvested at brown pod stage from plants that were non defoliated, hand shelled and dried in the sun (PG - 90%) and *C. brevidens parviflora* seeds harvested at black pod stage from non defoliated plants, hand shelled and dried in the sun (PG - 94%). Storage experiments were conducted between May and July 2006 for the 1st season and January to March 2006 for the 2nd season in Kakamega and Siaya counties. Average room temperature was 23^{0}C and relative humidity 85% between May and July 2006 in Kakamega County. In Siaya County, the average room temperature was 25^{0}C and relative humidity 80% during the same period. Between December and February 2006 the average room temperature was 26^{0}C while average humidity was 80% in Kakamega County. In Siaya County the average temperatures was 28^{0}C and relative humidity 65% for the same period.

Moisture content of the seeds before storage was determined by drying five replications of 50 seeds for each treatment in a well ventilated oven at 103^{0}C for 17 hours according to ISTA, 2015. After removing the seeds from the oven, seeds in the dishes were allowed to cool for about 30 to 45 minutes inside a dessicator before their weights were taken. Moisture content was determined by the following formula

% Seed moisture content = Initial seed weight before drying (g) – Seed weight after drying (g) ---X 100
Initial seed weight before drying (g)

Seeds were stored in 5 different ways, in pots, brown paper bags, plastic jars, polythene bags and freezer (at -2 ^{0}C). Amount of seed stored in each container or foil paper (for the freezer method) was 5 grams which was measured using a weighing balance. There were 3 replicates for each storage method. The seeds were stored for 90 days. Germination tests were then conducted in the laboratory to investigate seed viability and vigour for the different storage methods. Percent germination was used to determine seed viability. Seed vigour was determined by speed of germination test which was incorporated in the standard germination test described above.
The speed of germination index was determined by the formula below.

$$\text{Speed of germination index (SGI)} = \sum N/D$$

SGI – speed of germination index; N- Number of normal seedlings that germinated per day; D - Day after sowing
The higher the SGI the higher the seed vigour.
Data collected from the laboratory experiments were subjected to Analysis of variance (ANOVA) in the SAS software. T-tests were used to compare seasons and sites as far as percent germination and speed of germination indices were concerned. Means for the different storage methods were separated using LSD procedure.

RESULTS

Seed Moisture

Average seed moisture before storage for all the seeds was from 12% in Kakamega. In Siaya, average moisture range for the seeds was 11%.

Effect of Season on Seed Quality of Slender Leaf Varieties

Crotalaria brevidens intermedia seeds harvested from the long rains significantly differed from those harvested in the short rains in PG but not in SGI in the Kakamega and Siaya Counties. Seeds harvested in the long rains season had significantly ($P \leq 0.05$) higher percent germination than those from the short rains season (Figure 1(i)). Speed of germination index of the seeds did not significantly ($P \geq 0.05$) differ with season (Figure 1 (ii).

i) ii)

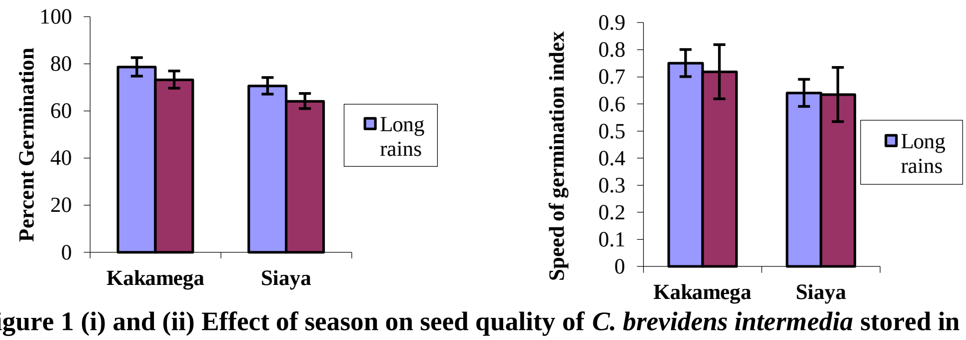


Figure 1 (i) and (ii) Effect of season on seed quality of *C. brevidens intermedia* stored in Kakamega and Siaya counties.

In Kakamega, season significantly ($P \leq 0.05$) affected SGI but not PG for the *C. ochroleuca* seeds. Seeds from the long rain seasons had significantly ($P \leq 0.05$) higher PG and SGI compared to those from the short rain season (Table 2).

Table 2: Percent germination and speed of germination index for the long and short rain seasons in Kakamega

Season	Seed quality parameter Percent germination	Speed of germination index
Long rain	83.40±0.34a	0.80±0.01a
Short rain	81.20± 0.36b	0.76±0.01b

Means with the same letters within each seed quality parameter are not significantly different at $P \leq 0.05$

Percent germination (PG) and SGI index of *C. brevidens parviflora* did not significantly ($P \geq 0.05$) differ for the two seasons in Siaya. Percent germination and SGI are shown in Table 3.

Table 3: Percent germination and speed of germination index for the long and short rain seasons in Siaya

Season	Seed quality parameter Percent germination	Speed of germination index
Long rain	68.0±0.45a	0.64±0.01a
Short rain	67.5±0.46a	0.63±0.01a

Means with the same letters within each seed quality parameter are not significantly different at $P \leq 0.05$

Effect of Storage Method on Seed Quality of Slender Leaf Varieties

Method of storage significantly ($P \leq 0.05$) affected all the quality parameters of *C. brevidens intermedia seeds*. Seeds stored in the pot, brown paper bag and freezer had significantly ($P \leq 0.05$) higher PG and SGI than those stored in plastic jars and polythene bags in both Kakamega and Siaya [Figure 2 (i) and (ii)].

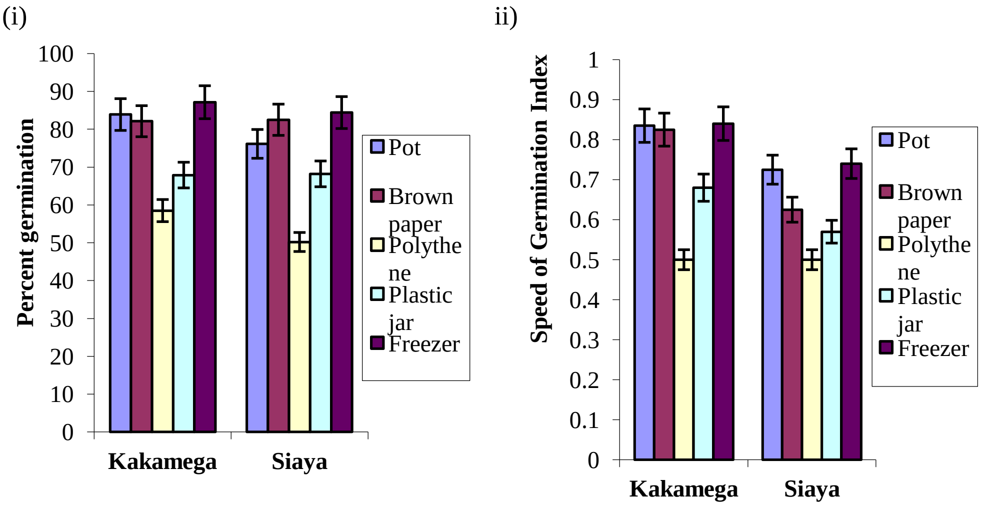


Figure 2 (i) and (ii) Effect of storage method on seed quality of *C. brevidens intermedia* in Kakamega and Siaya Counties.

Season and storage methods did not significantly interact for both PG and SGI in the *C. brevidens intermedia*.

Storage methods significantly affected PG and SGI of *C. ochroleuca* in Kakamega. Seeds stored in the pot, brown paper bag and plastic jars had significantly ($P \leq 0.05$) higher PG than those stored in polythene bags and freezer. Seeds stored in brown paper bag and plastic jars had significantly ($P \leq 0.05$) higher SGI than the other methods (Figure 3 and 4).

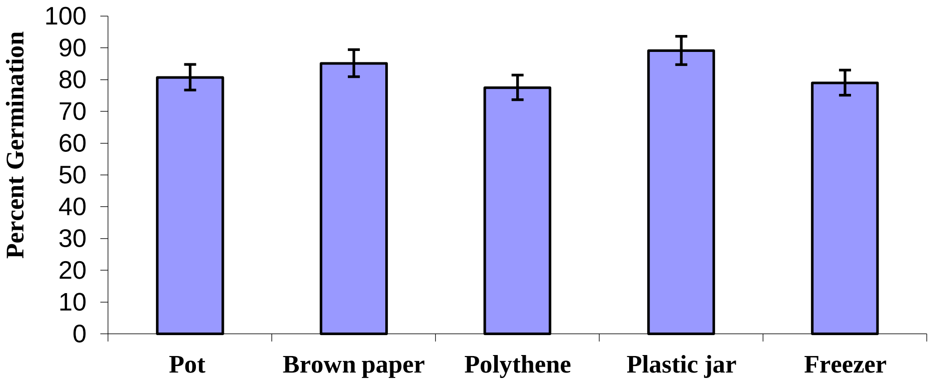


Figure 3: Effect of storage method on percent germination of *C. ochroleuca* seeds in Kakamega County

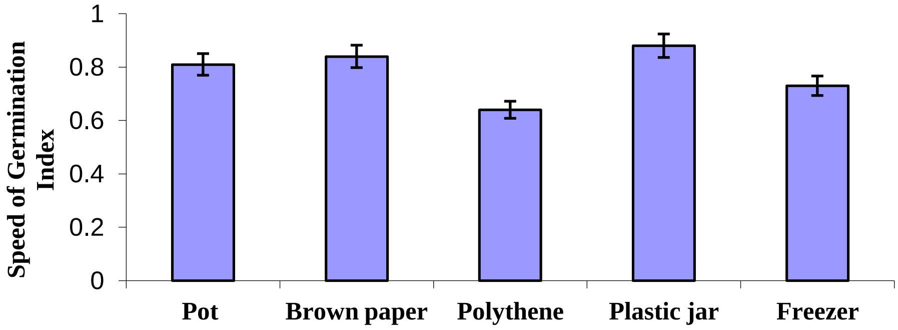


Figure 4: Effect of storage methods on speed of germination indices of *C. ochroleuca* seeds in Kakamega County

There was significant interaction between season of growth and storage methods for SGI in Siaya. Seeds of *C. ochroleuca* harvested after the long rain season and stored in brown paper bags had significantly ($P \leq 0.05$) higher SGI than other storage methods from the long and short rain seasons. *C. ochroleuca* seeds harvested in the long rains and stored in the freezer did not differ much from those harvested in the short rains as far as SGI was concerned (Figure 5). Generally the long rains season gave better quality seed than the short rains season for all the storage methods (Figure 5).

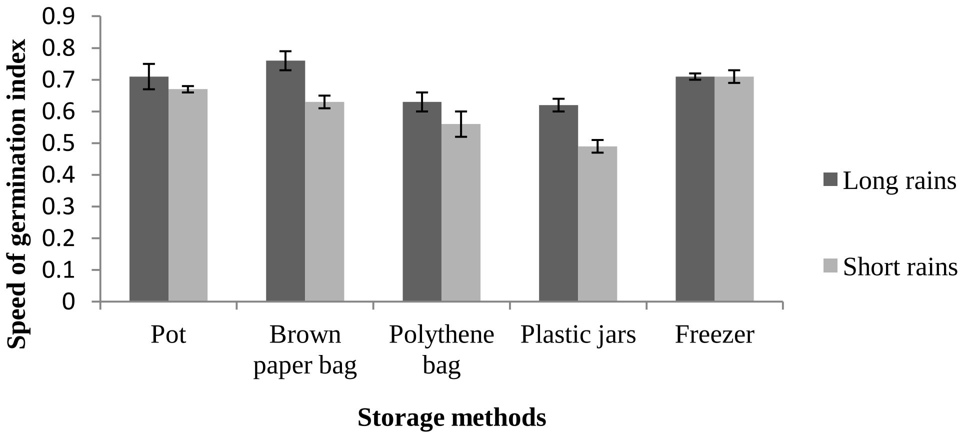


Figure 5: Speed of germination indices of significant interaction between season and storage methods for *C. ochroleuca* seeds in Siaya County

Method of storage significantly affected PG and SGI index of *C. brevidens parviflora* seeds stored in Siaya. Seeds of this variety which were stored in pot and freezer had significantly higher PG and SGI than other methods in Siaya (Figure 6 and 7).

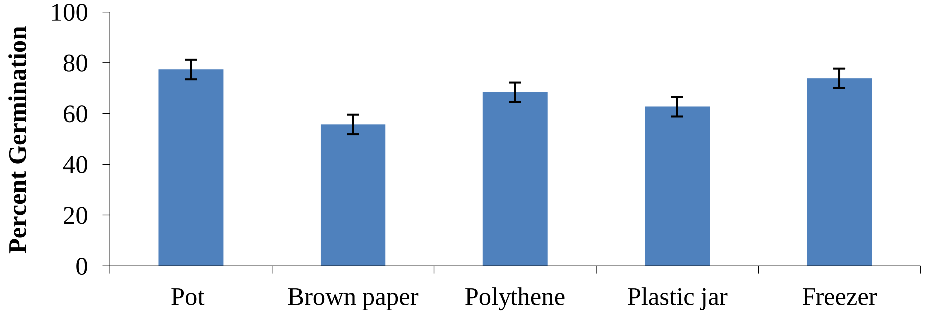


Figure 6: Effect of storage methods on percent germination of *C. brevidens parviflora* seeds in Siaya County.

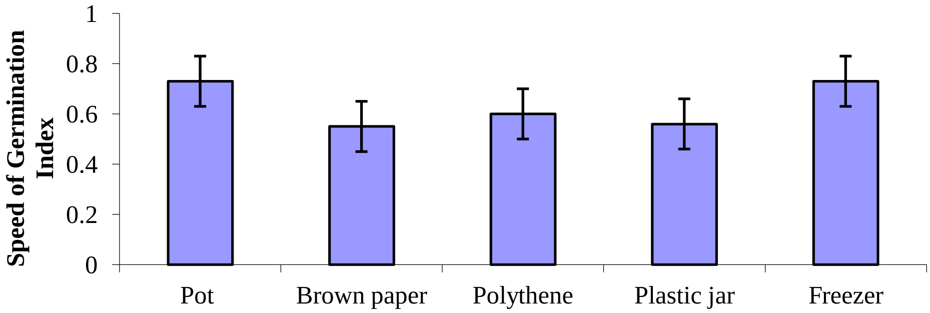
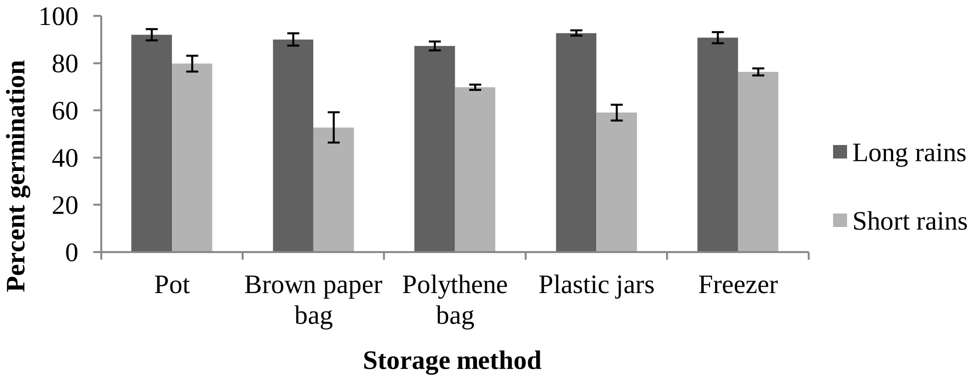


Figure 7: Effect of storage methods on speed of germination indices of C. *brevidens parviflora* seeds in Siaya County.

For C. *brevidens parviflora* seeds there were significant interaction (P≤0.05) between season of growth and storage methods for the 2 seed quality parameters in Kakamega. Seeds harvested after the long rain seasons and stored in pots had significantly higher (at P ≤ 0.05) PG and SGI than others in this interaction (Figure 5.10 (i) and (ii).

(i)



(ii)

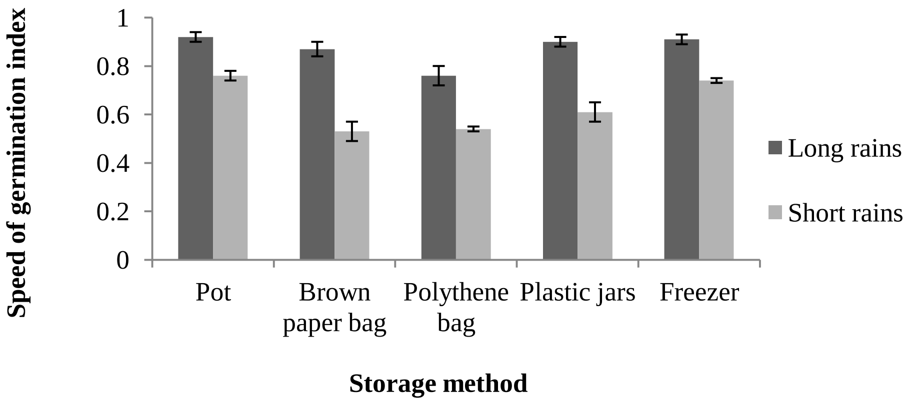


Figure 8 (i) and (ii) Significant two way interaction between season and storage methods for percentage germination and speed of germination index for *C. brevidens parviflora* seeds in Kakamega County

DISCUSSION

C. brevidens intermedia seeds obtained from the long rains season had higher percent germination (PG) and speed of germination index (SGI) [though not significant at $P \leq 0.05$ for the latter] than seeds from the short rains in Kakamega and Siaya Counties. Even though PG significantly differed in the two seasons, SGI is more important as it has to do with the field performance of the crop (Finch-Savage and Bassel, 2016). These results indicated that farmers could store seeds from both seasons and still get a mean value of above 0.70 for SGI.

Season from which *C. ochroleuca* seeds were obtained significantly affected SGI in Kakamega. Percent germination (PG) did not significantly differ for the two seasons. *C. ochroleuca* seeds from the long rains season had significantly higher SGI than those from the short rains; mean values were above 0.8 indicating that farmers could still store seeds from both seasons. This may have been due to suitable ecological conditions found in Kakamega during both the long and short rain seasons that ensured the high quality seeds are obtained for storage (GOK, 2006). In Siaya, *C. ochroleuca* seeds from the long rains had significantly higher PG and SGI than seeds from the short rains. Farmers can still store seeds from both seasons and still get good quality because mean germination was in the 70s and mean SGI was above 0.7.

C. brevidens parviflora seeds obtained from the long rains in Kakamega had significantly higher PG and SGI than those from the short rains. The difference between the two seasons was large indicating that for this morphotype farmers need to harvest and store seed from the long rains to get good crops. This slender leaf morphotype seems to be more vulnerable to moisture stress. Reason(s) to this observation need to be investigated further.

Effect of Storage Method on Seed Quality of Slender Leaf Varieties

Method of storage significantly affected PG and SGI in both Kakamega and Siaya for all the slender leaf varieties. For all the slender leaf varieties the most suitable methods of storage were the pot, brown paper bag or freezer as they had significantly higher PG and SGI than polythene bags and plastic jars. Similar results were obtained for *Corchorus olitorius* where seeds stored in tin cans had higher seed quality than those stored in polythene bags (Razzak, et al., 2013). Another study also reported that *Corchorus capusularis* seeds stored in earthen pots had higher seed quality than those stored in tin cans (Mazed, 2014).

Temperature, seed moisture, relative humidity, oxygen pressure and biotic factors during storage have been shown to affect seed quality (Walters, 2010). Temperatures in the pot and freezer in the present study were lower than the ambient temperatures. This ensured that the seeds do not respire too fast producing toxic compounds that kill the embryos of the seeds. This is in line with a study on *Sesbania* seeds that indicated seeds stored in a freezer had higher germination and vigour index than other those stored at room temperature (Karim et al., 2016; Mbofung et al., 2013). Due to numerous pores of the pots and brown paper bags the humidity in the storage containers is the same as that of the room. This may explain where the PG were not in the 90s but ranged between 50- 80% in the present study. High moisture levels in the storage container encourage the seed to respire and fungi growth which deteriorates the seed (Razzak *et al.*, 2013; Alhamdan *et al.*, 2011).

Pot and brown paper bags also had numerous pores that ensured gaseous exchange between the storage container and the atmosphere. This ensured a good supply of oxygen which is needed by the seeds which respire slowly during storage. Carbon (IV) oxide was also removed further improving storage conditions of the seed. The plastic jars enclosed enough air to ensure oxygen availability and non accumulation of carbon (IV) oxide to toxic levels. Polythene bags enclosed less air and hence oxygen was not available to the respiring seeds and there was accumulation of carbon (IV) oxide. This is in line with reports that indicated that adequate gaseous exchange is important (Walters, 2010).

Storing seed in the pots and brown paper bags also provided dark conditions which are associated with preserving seed quality. Sunlight has been associated with reducing the life of seeds due to the fact that photoreceptors found in seeds are stimulated and this leads to increased metabolic reactions that breakdown stored food (Wei, 2011).

Storing seeds in pots, brown paper bags and plastic jars was comparable to storing seeds in the freezer. At farm level, farmers may not afford to use freezers due to high cost and sometimes lack of electricity. However they could embrace the use of pots, brown paper bags and plastic jars which are readily available and do not cost much. They can re-use the brown paper bags used for packaging wheat or maize flour to store seeds instead of buying them. Recycled plastic jars can also be easily obtained from the home or market.

There were differences between the percent germination and speed of germination indices for most the storage methods. This is in line with other studies that indicate seed speed of germination index is more sensitive to storage than percent germination depending on the crop species (Finch- Savage and Bassel, 2016).

Generally, season and methods of storage interacted significantly more often for the SGI (which was found in *C. ochroleuca* and C. *brevidens parviflora*,) than for PG (which occurred for only *C. brevidens parviflora*). This is understandable since speed of germination index, is a measure of seed germination under field conditions which are normally not optimal (Finch- Savage and Bassel, 2016). When season and method of storage interacted, *C. brevidens parviflora* seeds stored in pots in Kakamega had high SGI. *C.*

ochroleuca seeds stored in plastic jars in Siaya also had high SGI. This implies that these methods provided the appropriate conditions for storage as discussed above. It is also important to note that when 2 or more methods were superior to others and when the interactions between seasons and storage methods were considered one of the methods emerged better than the rest. For example in Siaya *C. ochroleuca* seeds stored in pots, brown paper bag and freezer had higher speed of germination index than those stored in polythene bags and plastic jars. When interaction between season and storage method is considered, brown paper bags were superior to pots and the freezer as far as SGI was concerned.

CONCLUSIONS

Season significantly affected percent germination and speed of germination index. Methods storage also significant affected percent germination and speed of germination index of slender leaf seeds. Seeds of slender leaf stored in pots, brown paper bags, plastic jars and freezer generally had higher seed quality than those stored in the polythene bags in both sites and seasons. Farmers need to avoid using polythene bags for storage of seeds.

REFERENCES

Abukutsa-Onyango, M.O. 2011. Keynote Paper. Strategic Repositioning of Agrobiodiversity in the Horticultural Sector for Sustainable Development in Africa. In Agricultural Innovations for Sustainable development. Contributions for the Finalists of 2009/2010 African-wide Women and Young Professionals in Science and Competitions.Accra, Ghana 2011, 8-16.

Abukutsa-Onyango, M.O. 2014. Researching African Indigenous Fruits and Vegetables – Why? Accessed 6/5/2018. Available at: Knowledge.cta.int/Dossiers/Commodities/Vegetables/feature articles/Researching-African-Indigenous-Fruits-Vegetables-Why.

Afari-Sefa, et al., 2011.Vegetable breeding in Africa: constraints, complexity and contributions towards achieving food and nutritional security. Food security 4(1): 115-127.

Alhamdan, A.M., Alsadon, A.A. and Khalil, S.O, 2011. Influence of storage conditions on seed quality and longevity of four vegetable crops. American-Eurasian J. Agric. & Environ. Sci. 11(3): 353-359.

Ebert A.W. 2014. Potential of underutilized traditional vegetables to contribute to food and nutritional safety, income and more sustainable production systems. Sustainability 6: 319-355.

Finch-Savage W.E. and Bassel, G.W., 2016. Seed vigour and crop establishment: extending performance beyond adaptation. Journal of Experimental Botany Vol. 67(3): 567-59.

GOK, 2006. Ministry Of Agriculture Report, 2006 – 2008. Government Printer. Nairobi, Kenya.

ISTA, 2015. International rules for seed testing. Basserdorf, Switerland. International seed testing association.

Karim, M.T., Fazla, H., Darwar A.K.M.G, Ali, M.A., 2016. Effect of storage conditions on seed quality of Sesbania spp. Accessions. Agriculturists 14 (1):83-91.

Maina, F.N. W., L. S., Gohole and R. M. Muasya, 2013. Effect of agronomic and post harvest practices on seed quality of slender leaf vegetables. In the 13th Workshop on Sustainable Horticultural Production in the Tropics, 3rd and 6th December 2013 at Meru University of Science and Technology, Meru.

Maina, F.N. W., R. M. Muasya and L. S., Gohole, 2012. Morphological characterization of jute mallow, *Corchorus sp*. to assess its genetic diversity in Western Kenya. Baraton Interdisplinary Research Journal Vol. 2 No. 2, 2012: 21-9.

Mazed H.E.M., 2014. Effect of storage container on seed quality attributes of Deshi Jute (*Corchorus capasularis* L.) Bangladesh Agron. J. 16 (11):70-5

Mwaura S.N. et al., 2014. African leafy vegetables and household well being in Kenya. a disaggregation by gender. Current Research Journals of Social Sciences 6(4): 82-94.

Mbofung, G.C.Y., Goggi, A.S., Leandro, L.F.S. and Mullen, R.E., 2013. Effects of storage temperature and relative humidity on viability and vigour of treated soybean seeds. Crop Sci. 53: 1086-1095.

Nyalemegbe, K.K., Osakpa, T.Y. and Danson S.K.A, 2011. Ecological considerations in the selection of leguminous plants as cover crops on the vertisols of Accra plains in Ghana. Journal of Ghana science association 13:1-4

Opande, G.T., Musyimi, D.M. and Kaberia, M.J., 2017. Phytochemical screening and antimicrobial effect of leaf and root extracts of *C. brevidens* on *Canida albicanus*, *Staphylococus amerous*, *Escherichia coli* in Maseno Kenya. International journal of science invention Vol. 6 (6): 33-40.

Razzack, M.A. and Azad, A.K., 2013. Effect of storage container on seed quality attributes of Tossa Jute (*Corchorus Olitorius* L.). Bangladesh Agron. J. 16 (11):61-6

Sikuku, P.A., Musyimi, D.M., Kariuki, S. and Okello, S.V., 2013. Response of slenderbox (*Crotalaria ochroleuca*) to water deficient. Journal of biodiversity and environmental Sciences, Vol. 3 (12): 245-252.

Simon J. et al., 2016. Improving income and nutrition of small holder farmers in Eastern Africa using a market driven approach to enhance value driven production of African Indegenious Vegetables. African Journal of Biotechnology.15 (7): 199-206.

Uusiko, N.P., Oleofse, A., Doudu, K.G., Bester, M.J., Feber,M., 2010. Nutritional value of leafy vegetable of Sub Saharan Africa and their potential contribution to human health. Journal of food composition and analysis 23:499-509.

Walters, C., Ballesteros, D. and Vertucci, V.A., 2010. Structural mechanism of seed deterioration: standing the test of time. Plant Science179: 565-573.

Wei, X., Liao, J., Mingxi, J., 2011. Effect of pericarp, storage conditions, seed weight, substrate moisture content, light, GA3 and KNO_3 on germination of Eupelea pleiospermum. Seed Science and technology 38(1): 1-13

Zheng, Y. and Huan-Cheng, M., 2014. Effects of seed aging on seed germination and seed reserve utilization in Mumian Hort. Technology Vol. 24 (4): 471-4

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