



Assessment of seed quality of pea (*Pisum sativum* L.) influenced by different storage containers at different storage periods

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Abstract

Three types of containers and four storage periods were used as experimental treatments. The experiment was laid out by Completely Randomized Design (CRD) with four replications. Seed quality factors *viz.*, moisture content, germination percentage, vigor index, percentage of abnormal seedling, normal seedlings and dead seed were recorded every 15 days interval. The initial moisture content of seed in plastic container, poly bag, and gunny bag were 9.25%, 9.22% and 10.33%, respectively, but it was increased with increasing storage time after 60 days (10.00%, 10.6% and 14.00%). The germination percentage was higher at 15 days after storage (DAS) for different containers like plastic containers (85%), poly bag (79%) and gunny bag (78%) than after 60 DAS (69%, 61% and 56%), respectively. The percentage of abnormal seedling was increased from 10 to 24%, 14 to 31%, and 15 to 33% in plastic container, poly bag, and gunny bag, respectively, from 15 DAS to 60 DAS. The percentage of dead seed was increased from 4 to 6%, 7 to 8% and 8 to 12% in plastic container, poly bag and gunny bag respectively, from 15 DAS to 60 DAS. The vigor indexes of seedling were 14.58, 12.85 and 10.92 at 15 DAS in plastic container, poly bag and gunny bag, respectively, which attained at 10.39, 10.26 and 10.08 at 60 DAS, respectively. Several fungal infection and lesion was found during seed health tests. The results revealed that the germination percentage decreased with the increasing the storage period. So this study opined that initial moisture and storage container has significant effect on the quality of pea seeds and plastic container is the best one for storing the seeds with high benefits.

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Introduction

Pea (*Pisum sativum* L.) is one of the important pulse crops as well as vegetables belong to (Leguminosae) Fabaceae family. The Mediterranean region, western and central Asia and Ethiopia have been indicated as centers of origin. The area under this crop was 19,000 hectares and the production was 14,000 MT during the year 2010-11 (BBS, 2012). It was exported to Middle East and United Kingdom by Hortex Foundation. Bangladesh earns Tk. 1456.33 million (US\$ 24.70 million) in the year 2003-04 by exporting vegetables, which constitutes 60.08 percent of the earnings from agricultural products (Karim *et al.*, 2005). Gregor Johann Mendel who is known as the "father of modern genetics", worked with seven characteristics of pea plants in his experiments.

In Bangladesh presently pea is grown at a very small area of 12477 ha of land and annual production is about 3410 metric tons (BBS, 2005). The average dry seed yield is only 0.75 t/ha which is lower than other countries such as USA 3.94 t/ha and France 3.23 t/ha. The low yield is mainly due to lack of good quality seeds and lack of high yielding variety (Anonymous, 1990).

Lack of quality seed is one of the main problems for pea production. Seeds are the basic input for production. 10-15% production was found reduced due to use of poor quality seed (Huda, 2001). Major part of seed quality deteriorates at the time of storage in our country. Most of the people of Bangladesh store their seeds as they store their food grain. The most important biotic and abiotic factors that affect seed quality are relative humidity, temperature, insect and fungus. The relative humidity and temperature act constantly on seed. Due to high relative humidity seed sets equilibrium at high percentage of moisture. Higher temperature and higher moisture percentage increase respiration. As a result the quality of the seed becomes bad and the seed is unusable for crop production.

High relative humidity and high temperature cause high moisture content in pea seed and results in low germination with low vigor seedlings at the end of storage.

Safe condition was defined as those, which maintain seed quality without loss of vigor for three years (Harrington, 1960). Seed equilibrium moisture is lower than 14% for cereal seeds and 11% in oil seeds (Harrington, 1972) or, more properly, ranges from 12% to 13% and from 8% to 9.5%, respectively and from 7% to 8% for most of the vegetables seeds (Lovato, 1976). This means a seed moisture in "equilibrium" with 50% RH at 30°C or 60% RH at 20°C (Delouche *et al.*, 1973), or with 65% RH but with a temperature higher than 33°C for only a few hours (Harrington, 1972). Therefore, proper control of seed moisture content and seed storage conditions becomes essential for the maintenance of seed viability and vigor, especially in humid tropical regions.

Seed moisture and storage temperature are two of the most important factors affecting seed longevity during storage. Storage experiments have shown that reduction in moisture content extends the time of Douglas fir and Western Hemlock seeds. Seed health has also been found to be influenced by the seed quality of different crops as has been reported by Fakir (1989).

Maintenance of seed quality during storage period is important not only for crop production in the following year but also for the maintenance integrity of the seeds because of their constant threat and of genetic erosion. Little works have been found on seed storage of pea seed in Bangladesh. Information relating pea seed storage and effects of biotic and abiotic factors on quality of pea seed storage are crucial.

Thus the present research work was conducted to identify the best container for storing pea seed, to determine the abiotic factors on quality of pea seeds, to determine the biotic factors on the storing performance on the pea seed and to assess the interaction between storage container and storage periods the storage.

Materials and methods

Location

The study was conducted at the plant pathology laboratory of the Hajee Mohammad Danesh Science & Technology University, Dinajpur (HSTU) during 20 March to 20 May, 2016 for around two months to observe the effects of variation in Pea seed quality preserved in different seed containers. The experiment was conducted in the room condition with three seed storage containers.

Seed collection

Pea seeds were supplied by the Course Coordinator of the course and used in this experiment. Seeds were kept in three supplied storage containers and stored at room temperature and RH for 2 months.

Experimental treatments

The following two sets of treatments were included in the experiment:

A. Storage material: 3

i) Plastic container ii) Poly bag iii) Gunny bag

B. Storage period: 4

i) 15 days ii) 30 days iii) 45 days iv) 60 days

Experimental design

The experiment was laid out in Completely Randomized Design (CRD) with four replications.

Sampling

Sampling was carried out at 15 days intervals up to 60 days i.e. 4 times. At each sampling, samples were taken randomly from each storage container.

Regular Testing of Seed Kept in Three Containers

During the storage period seeds samples were taken every fortnightly from the containers for determination of change of moisture content (Wet basis), germination percentage, presence of insect and others activities.

Moisture Test

Moisture content was determined by using high constant temperature oven method following international rules for seed testing in the seed technology laboratory of HSTU.

5g of seeds from each 3 containers were taken and poured in a small container with cover and kept in an oven maintained at a temperature of 130°C to 133°C for a period of 1 hour. The moisture content of seeds (wet basis) was determined by the following formula (Anonymous, 1999).

$$MC (\%) = \frac{M_2 - M_3}{M_2 - M_1} \times 100$$

M_1 = Weight of container + cover

M_2 = Weight of container + cover + pea seed before drying

M_3 = Weight of container + cover + pea seed drying

Germination Test

Germination test was conducted using sand as substratum. The plastic trays with seed were incubated at room temperature and irrigated at every odd day. After 8 days, germination percentage was recorded. The normal seedlings and abnormal seedlings and ungerminated seed were classified according to the prescribed rules given by ISTA.

$$\text{Germination (\%)} = \frac{\text{Number of seeds germinated}}{\text{Number of seeds tested}} \times 100$$

Seeds were stored at room condition in different containers and germination test was performed every 8 days of interval beginning from collection of seeds up to 40 days to observe the rate of natural aging of Maize seeds stored in different containers.

Seedlings obtained from standard germination test were used for seedling evaluation. Normal or abnormal seedlings were classified according to the rules of ISTA (1993). Seedling shoot and root length was measured on day I of the germination test. Ten plant samples from each plastic pot were harvested and shoot and root length of individual plant was recorded.

Vigor Index (VI)

Seedlings obtained from standard germination test were used for seedling evaluation. Normal or abnormal seedlings were classified according to the rules of ISTA (1993). Seedling shoot and root length was measured on day 8 of the germination test. Ten plant samples from each plastic container were harvested and shoot and root length of individual plant was recorded.

Vigor index (VI) was calculated from a daily count of germinated seeds until it reached a constant value, using the following formula:

$$\text{Vigor index VI} = \frac{\text{Number of normal seedlings}}{\text{Days to first count}} + \dots + \frac{\text{Number of normal seedlings}}{\text{Days to final count}}$$

Insects and Diseases

In every fortnightly presence and number of insects and diseased seeds were also observed in the stored Pea seeds. Numbers of insects were counted from 25 g of seeds and the infestation of pathogen and insect was observed.

Record Keeping of Dry Bulb and Weight Bulb Temperature and Relative Humidity

Reading of dry bulb and weight bulb temperature and relative humidity of inside the room and outside the room were recorded in 10.00 am and 4.00 pm every day. Amount of rainfall and maximum and minimum temperature were collected from Wheat Research Center, Nosipur, Dinajpur.

Statistical analysis

Statistical analysis of the data was done using MSTAT-C and MS Excel software. Correlation studies were also computed following the standard procedure as described by Gomez and Gomez (1984).

Results

The results obtained on the moisture content of seed, germination capacity and seedling evaluation and seed health test of pea seed at various storage container and storage period have been presented below.

Effect of abiotic factors

Effects of storage container on moisture content of pea seed at different Days after Storage (DAS)

Effect of storage container on moisture content of pea seeds in different storage period is given in Table1.

Table 1. Effect of storage container on moisture content of pea seeds at different DAS.

Storage container	Moisture content (%) in different storage period			
	15 DAS	30 DAS	45 DAS	60 DAS
Plastic container	9.25 b	9.89 c	9.95 c	10.0 b
Poly bag	9.22 b	10.0 b	10.5 b	10.6b
Gunny bag	10.33 a	12.2 a	13.3 a	14.0 a
CV (%)	5.22	7.12	4.80	8.40

Mean bearing same letter(s) within same column do not differ significantly at 5% level of significance. DAS= Days after Storage.

Result revealed that the moisture content of seeds significantly influenced by different storage container in various observation period. The seed of gunny bag was retained highest moisture content 10.33, 12.2, 13.3, and 14% at 15, 30, 45 and 60 days after storing respectively. The plastic container gained the lowest moisture content 9.25, 9.89, 9.95 and 10.0% during 15, 30, 45 and 60 days after storing respectively. Results also revealed that the moisture content of seed also increased proportionally with increased number of days at different storage container. The lowest moisture content (9.25%) found in seed of plastic container at 15 days; whereas, the highest moisture continent (14%) found in seed of gunny bag at 60 days. As seed is highly hygroscopic living material;

it absorbs moisture from air if it is stored in an environment where relative humidity is higher than seed moisture content. For this reason, seeds absorbed moisture from the ambient air and tended to equilibrium with relative humidity. The rate of absorbance was higher in gunny bag because of gunny bag is not air tight container but tin and plastic container are moisture proof so, increasing rate was lower in air tight container.

Effects of storage container on germination of pea seed at different DAS

Seed germination test provides to the ability of seeds to germinate and produce a seedling that will emerge from the soil and develop onto a healthy vigorous plant.

The seed of plastic container showed the highest germination 69-85%, whereas, the lowest germination found in gunny bag (56-78%) during 15, 30, 45 and 60 days after observation respectively.

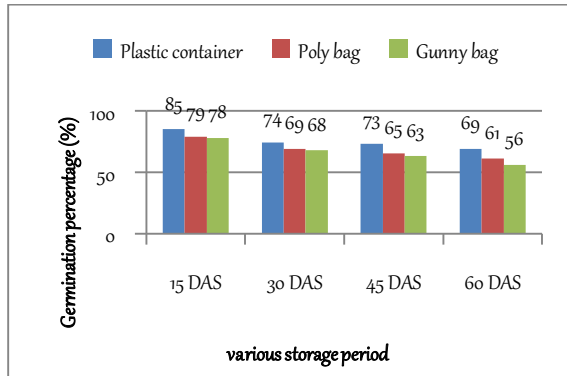


Fig. 1. Effect of storage container on germination percentage of pea seeds at different DAS.

Effects of storage container on normal seedling at different DAS

Effect of storage container on number of normal seedling of pea seeds in different storage period is given in Table 2.

Results showed that the number of normal seedling was significantly influenced by different storage container at various observation periods.

The seed of gunny bag always produced the lowest number of normal seedling 78, 70, 63 and 56 at 15, 30, 45 and 60 days after storing respectively.

The plastic container produced the highest number of normal seedling 83, 77, 71 and 67 at 15, 30, 45 and 60 days after storing respectively. The intermediate number of normal seedling was produced by poly bag seeds.

Table 2. Effect of storage container on number of normal seedling in different DAS.

Storage container	Number of normal seedling during storage period			
	15 DAS	30 DAS	45 DAS	60 DAS
Plastic container	83 a	77 a	71 a	67 a
Poly bag	79 b	71 b	65 b	61 b
Gunny bag	78 b	70 b	63 c	56 c
CV (%)	5.0	4.3	3.2	6.0

Mean bearing same letter(s) within same column do not differ significantly at 5% level of significance.

Effects of storage container on abnormal seedling at different DAS

Effect of storage container on number of abnormal seedling of pea seeds in different storage period is given in Fig. 2.

Result revealed that the number of abnormal seedling was significantly influenced by different storage container in various observation periods. The seed of gunny bag always produced the highest number of abnormal seedling 15, 21, 27 and 33 at 15, 30, 45 and 60 days after storing respectively. The plastic container produced the lowest number of abnormal seedling 10, 15, 21 and 24 at 15, 30, 45 and 60 days after storing respectively. The intermediate number of abnormal seedling was produced by poly bag seeds.

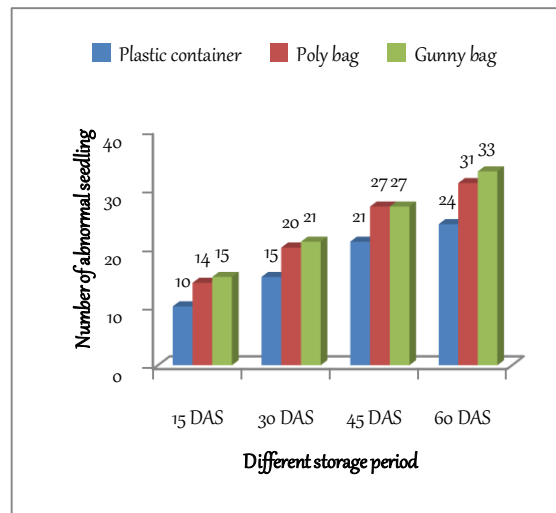


Fig. 2. Effect of storage container on number of abnormal seedling at different DAS.

Effects of storage container on dead seedling at different DAS

Effect of storage container on number of dead seed of pea in different storage period is given in Table 3.

Table 3. Effect of storage container on number of dead seeds at different DAS.

Storage container	Number of dead seed during different storage period			
	15 DAS	30 DAS	45 DAS	60 DAS
Plastic container	4 c	5 c	5 c	6 c
Poly bag	7 b	9 b	8 b	8 b
Gunny bag	8 a	10 a	11 a	12 a
CV (%)	6.5	2.1	6.0	4.5

Mean bearing same letter(s) within same column do not differ significantly at 5% level of significance.

Result showed that the number of dead seed was significantly influenced by different storage container in various observation periods. The seed of gunny bag obtained the highest number of dead seed 8,10,11 and 12 at 15,30,45 and 60 days after storing respectively,; whereas the lowest number of dead seed found in plastic container 4, 5,5 and 6 at different storing periods respectively.

Effects of storage container on fresh ungerminated pea seed at different DAS

Effect of storage material on number of fresh ungerminated pea seed in different storage period is given in Table 4.

Table 4. Effects of storage container on fresh ungerminated pea seed at different DAS.

Storage container	Number of fresh ungerminated seed during storage period			
	15 DAS	30 DAS	45 DAS	60 DAS
Plastic container	2 b	1 c	1 c	2 c
Poly bag	4 a	4 b	4 b	3 b
Gunny bag	4 a	5 a	5 a	5 a
CV%	3.2	3.7	3.7	3.6

Means followed by same letters are not significant at 5% level of significance.

Result revealed that the number of fresh ungerminated was significantly influenced by different storage material in various storage periods.

The seed of gunny bag obtained the highest number of dead seed 4, 5, 5 and 5 at 15, 30, 45 and 60 days after storing respectively; whereas the lowest number of fresh ungerminated seed was found in plastic container 2, 1, 1 and 2 at different storing periods respectively.

Effects of storage container on seedlings root length, shoot length and root- shoot ratio of pea seeds at different DAS

In case of shoot length, highest length was observed in poly bag (27.08 cm) at 15 DAS and the lowest length was 24.3 cm in gunny bag at 60 DAS.

On the other hand in case of root length, highest length was observed in poly bag (25.18 cm) at 15 DAS and the lowest length was 11.01 cm in gunny bag at 60 DAS. But there is no significant difference in shoot length among the containers.

Anyway, in the long run, plastic container showed slightly better performance (25.85 cm). In case of root-shoot ratio, it is clear that in each container, higher length of roots was noticed than shoots. The highest ratio was found in plastic container at 15 DAS (0.95) and the lowest ratio at 60 DAS in gunny bag was 0.70 (Table 5). This result confirmed that the root length of pea seedling is higher than the length of shoot.

Table 5. Effects of storage container on seedlings root length shoot length and root- shoot ratio at different DAS.

Storage containers	Shoot length (cm)				Root length (cm)				Root-Shoot ratio			
	15 DAS	30 DAS	45 DAS	60 DAS	15 DAS	30 DAS	45 DAS	60 DAS	15 DAS	30 DAS	45 DAS	60 DAS
Plastic container	26.24 b	26.23 a	25.9 a	25.85 a	24.92 b	22.29 a	21.75 a	18.87 b	.95 a	.85 a	.84 a	.73 b
Poly bag	27.08 a	26.10 a	25.6 a	25.15 a	25.18 a	22.44 a	20.48 ab	19.11 a	.93 a	.86 a	.80 b	.76 a
Gunny bag	26.25 b	26.02 b	25.45 a	24.3 b	22.57 c	20.03 b	18.57 c	17.01 c	.86 c	.77 b	.73 c	.70 c
CV%	3.2	3.4	3.7	4.2	3.6	3.7	3.9	3.5	3.7	3.6	4.2	3.9

Means followed by same letters are not significant at 5% level of significance.

Effects of storage container on thousand seed weight at different DAS 1000 seed weight was significantly different due to various storage container and different evaluation period (Table 6).

Results showed that the 1000-seed weight was increased in gunny bag compared to poly bag and

plastic container due to gaseous exchange between seed and environment. The highest seed weight was found 60 days (55, 53 and 52g) at gunny bag, ploy bag and plastic container respectively; whereas the lowest seed weight found in 15 days at different storage containers.

Table 6. Effect of storage container on number of 1000-seed weight at different DAS.

Storage container	1000-seed weight during different storage period			
	15 DAS	30 DAS	45 DAS	60 DAS
Plastic container	50 c	51 c	51 c	52 c
Poly bag	51c	52 b	52 b	53 b
Gunny bag	52b	53 a	54 a	55 a
CV%	3.7	3.2	3.4	3.7

Mean bearing same letter(s) within same column do not differ significantly at 5% level of significance. DAS= Days after Storage.

Result also showed that the seed weight of gunny bag is higher compared with poly bag and plastic containers.

Effects of storage container on vigor index at different DAS

In case of vigor index, the highest vigority (14.58) was found in plastic

container at 15 DAS (Table 7.) and the lowest were observed in gunny bag at 60 DAS (10.08). The tendency of vigority loss was slow which reflected in the germination percentage.

Table 7. Effects of storage container vigor indexat different DAS.

Storage container	Vigor index			
	15 DAS	30 DAS	45 DAS	60 DAS
Plastic container	14.58 a	14.02 a	11.39 a	10.39 a
Poly bag	12.85 b	11.20 b	11.02 a	10.26 a
Gunny bag	10.92 c	10.88 c	10.24 b	10.08 b
CV%	3.5	4.0	4.5	5.1

Means followed by same letters are not significant at 5% level of significance.

Effect of biotic factors

Seed health test

Effects of storage container on fungal infection at different DAS

During seed health test several fungal infections were observed in pea seeds which have been presented in Fig. 3.

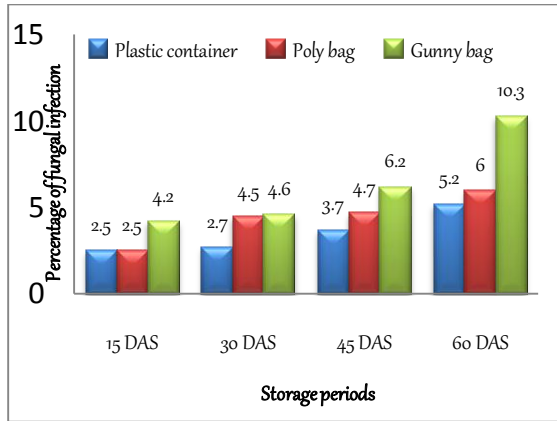


Fig. 3. Effect of storage container on fungal infection at different DAS.

Results revealed that 2.5, 2.7, 3.7 and 5.2 % fungal infection were observed in pea seeds at 15, 30, 45 and 60 DAS respectively at plastic container while 2.5, 4.5, 4.7 and 6% fungal infection at 15, 30, 45 and 60 DAS respectively at poly bag and 4.2, 4.6, 6.2

and 10.3% fungal infection at 15, 30, 45 and 60 DAS respectively at gunny bag. The highest fungal infections were observed in gunny bag.

During seed health tests several fungal infections were observed. Among the observed fungus *Fusarium*, *Ascochyta* and *Colletotrichum* are notable. The highest observed fungal growth was *Fusarium* followed by *Ascochyta* and *Colletotrichum* respectively in pea seeds. It was clearly observed that different levels of susceptibility to fungal infections by pea seeds.

Effects of storage container on lesion of seeds at different DAS

Several lesions were observed in pea seeds. The highest lesion (3) was found in gunny bag at 60 DAS (Table 8) and the lowest were observed in plastic container at 15 DAS (1).

Table 8. Effects of storage container on lesion of seeds at different DAS.

Storage container	Number of lesion of seeds			
	15 DAS	30 DAS	45 DAS	60 DAS
Plastic container	1a	1 a	2 a	2 a
Poly bag	1a	2 a	2 a	2 a
Gunny bag	1a	2 a	3 b	3 b
CV%	3.5	4.0	4.5	5.1

Means followed by same letters are not significant at 5% level of significance.

Effects of storage container on insects of pea seeds at different DAS

Effect of storage container of insects of pea seeds in different storage period is given in Fig. 4.

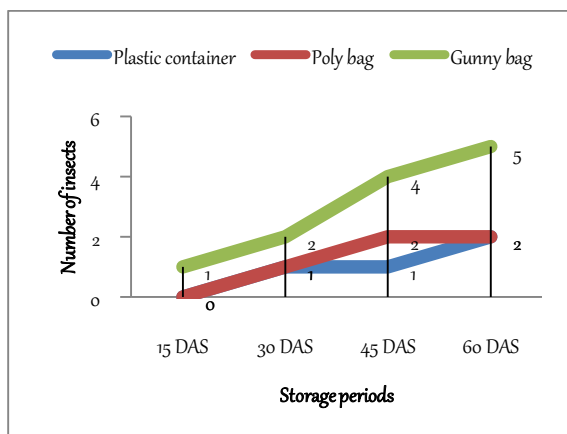


Fig. 4. Effect of storage container on insect infestation at different DAS.

Results revealed that the number of insects significantly influenced by different storage container in various observation period. In the seed of gunny bag found the highest number of insects (1, 2, 4 and 5) at 15, 30, 45 and 60 DAS respectively while lowest number of insects was observed in plastic container such as 0, 1, 1 and 2 at 15, 30, 45 and 60 DAS respectively.

Discussion

Result revealed that the moisture content of seeds significantly influenced by different storage container in various observation period. The seed of gunny bag always retained the highest moisture content and the plastic container gained the lowest moisture content. Brandenbing *et al.* (1961) found that moisture content of 5-6% was favorable for such a prolong storage condition. Higher moisture content during pea seed storage is one of the chief reasons that it losses viability sooner.

The germ inability of pea seeds was found decreased with time of storage. This decrease was closely related with the high moisture contents of the seeds. Seeds of gunny bag and poly bag with high moisture content during storage period lost their germination capacity at a similar rate than plastic container. The seed of plastic container showed the highest germination, whereas the lowest germination found in gunny bag. Similar result was showed by Doijode (1999).

Result showed that the number of normal seedling was significantly influenced but different storage container in various observation periods. The seed of gunny bag always produced the lowest number of normal seedling than plastic container. Result showed that the number of abnormal seedling was significantly influenced by different storage container in various observation periods. The seed of gunny bag always produced the highest number of abnormal seedling than plastic container.

Result revealed that the number of dead seed was significantly influenced by different storage container in various observation periods. The seed of gunny bag obtained the highest number of dead seed, whereas the lowest number of dead seed found in plastic container at different storing periods respectively. Amin *et al.* (2008) also reported similar results in his study.

Result showed that the number of fresh ungerminated seed was significantly influenced by different storage container in various observation periods. The seed of gunny bag obtained the highest number of fresh ungerminated seed, whereas the lowest number of dead seed found in plastic container at different storing periods respectively. The results showed similarity with the results of Amin *et al.* (2008).

Result revealed that root and shoot ratio of seed was significantly influenced by different storage container in various observation periods. The seed of gunny bag represents less difference in root and shoot ratio while plastic container showed the highest. Barton (1943) also reported similar results in his study.

1000 seed weight was significantly different due to various storage container and different evaluation period. Result showed that the 1000 seed weight was increased in gunny bag compared to poly bag and plastic container due to gaseous exchange between seed and environment. The highest seed weight was found in 60 days at gunny bag, poly bag and plastic container respectively; whereas the lowest seed weight found in 15 days at different storage containers. Islam (2006) also observed similar results in his study.

In case of vigor index, the highest vigority was found in plastic container and the lowest was observed in gunny bag. Islam (2006) also observed similar results in his study.

Result showed that during seed health tests several fungal infections were observed. The fungus may cause deleterious effect on seed health. Khanna (1977) conducted a study on feeding potential of insect pests of stored of stored grain. He also observed similar results in his study.

Result revealed that the number of insects significantly influenced by different storage container in various observation period. In the seed of gunny bag found the highest number of insects than other storage container. Afzal *et al.* (1999) stated that storage of pea seed should be dried to a moisture content of 8-10%, have foreign container removed, be stored in a cool dry place, and be protected from insect and rodents.

Conclusion

From the overall results it may be concluded that moisture content, storage container affected seed quality of pea during storage period. Moisture content, 1000-seed weight, abnormal seedlings, dead seed were increased and germination, normal seedlings were decreased with the increase of storage period. The research also revealed that the plastic container was the best and gunny bag was the worst storage container up to 60 days after storage.

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