



Seed Quality of Stored Soybean (*Glycine max* L.) as Influenced by Storage Containers and Storage Periods

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Received: 12 February 2014

Accepted: 14 June 2014

Abstract

An experiment was conducted at Bangabandhu Sheikh Mujibur Rahman Agricultural University, Gazipur during April 2013 to June 2013 to study the effect of three storage containers (tin container, polythene bag and cloth bag); and five storage periods (0, 15, 30, 45, and 60 days) on the seed quality of soybean. The experiment was laid out in Completely Randomized Design (CRD) with four replications. The soybean seeds stored in tin container showed maximum germination capacity with high germination index, highest seedling growth, seedling dry weight per plant and vigour index. The seeds stored in cloth bag had the lowest seed quality during the testing period. The highest 1000-seed weight, moisture content, electrical conductivity, abnormal seedlings and number of dead seeds were recorded in cloth bag; where the equivalent lowest values were recorded in tin container. The moisture content, 1000-seed weight, electrical conductivity, number of abnormal seedlings and dead seeds were increased with the longer period of storage. In addition, germination index, seedling growth, dry weight per plant of soybean seedling, and vigor index decreased with the increase in storage period. Among the three containers, tin container was the best and the cloth bag was the worst storage container for soybean seed storage for long period.

Keywords: Soybean, seed quality, storage container, storage period

1. Introduction

Soybean (*Glycine max* L.) is a very important protein rich oilseed crop extensively grown in the tropical, sub-tropical as well as the temperate region. Soybean oil is the largest component of the world's edible oils. The world production of edible oils consists of 30% soybean (Assefa, 2008). Soybean seed contains about 40-45% protein and 20-22% oil, 20-26% carbohydrate and a high amount of Ca, P and vitamins (Rahman *et al.*, 2011). In Bangladesh, in terms of area and production, soybean is a negligible crop concentrated in a very few locations.

During the period 2009-10 in Bangladesh, cultivation of soybean covered about 55,000 hectares of land and produced about 90,000 metric tones of seeds (Anonymous, 2011). The country's total demand for edible oil is around 1.3 million metric tons per year. Of the amount, 0.15 to 0.2 million metric tons come from mustard oil while the rest is met with imported soybean, rapeseed and palm oil (Rahman and Chowdhury, 2010).

Regardless of the high yield potential and other advantages, the average yield of soybean in Bangladesh is very low (1500-2000 kg ha⁻¹)

(BBS, 2009). However, Soybean cultivation is hindered due to lack of good quality seed (Rahman *et al.*, 2013; Amin *et al.*, 2009). Quality seed can increase the yield upto 30% keeping the other production factors the same (BARI, 1993). Quality seed that provides adequate plant stand is the basis for profitable production and expansion of soybean crop. In order to increase the production of soybean, a source of high quality, disease free seed must be established and maintained.

Storage condition is the key part of the maintenance of seed quality during storage. Generally, the quality of seed of soybean deteriorates during the storage period. In Bangladesh, seed quality deteriorates at the time of storage due to the prevailing bad weather which favours insect, and disease attack. Poor storage conditions greatly affect seed vigour (Heydecker, 1979); reduce respiration and seedling size. Exposure of seeds to moisture, temperature, relative humidity and activity of pests are related to the retention of seed viability in storage. Types of container also regulate temperature, relative humidity, and seed moisture content. Therefore, it is necessary to store seeds in suitable containers. Indeed, types of container that can provide suitable condition to maintain seed quality for longer period is very essential for seed preservation.

However, very little work has been carried out in Bangladesh in this regard. Keeping this in view, the present study was undertaken to determine the effect of different storage containers and storage periods on the seed quality of stored soybean.

2. Materials and Methods

2.1. Experimental site and design

The experiment was conducted at the Bangabandhu Sheikh Mujibur Rahman Agricultural University (BSMRAU), Gazipur during April 2013 to June 2013. BARI Soybean-5 was used in the experiment. The experiment was laid out in the Completely Randomized

Design (CRD) with four replications. Three storage containers: 1) tin container, 2) polythene bag and 3) cloth bag; and five storage periods (0, 15, 30, 45, and 60 days) were used in this experiment. Seeds were stored at room temperature (24 to 30°C) and relative humidity (70 to 82%) up to 60 days. Seed samples were taken randomly from each storage container and were tested at the end of each storage period as described above. Quality of soybean seeds was assessed by moisture content, viability, vigour index, electrical conductivity and observation of biotic factors as per the following procedures.

2.2. Data Collection and Statistical Analysis

2.2.1. Moisture content test

Moisture content was determined at every 15 days during experimental period by using low constant temperature oven method (103°C, 18 hr) following International Rules for Seed Testing (ISTA, 1999) in the seed Technology Laboratory of BSMRAU. The moisture content of seed (wet basis) was determined following the formula (Anonymous, 1999):

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

Where,

M_1 = wt. of container + cover

M_2 = wt. of container + cover + soybean seed before drying

M_3 = wt. of container + cover + soybean seed after drying

2.2.2. Viability test

Viability test was done through germination test. Four hundred seeds were tested for germination from each container for every storage period in the pots as per the rules of ISTA, 1999. In pot, germination test was conducted using sand as substratum. The plastic pots were placed in room temperature (24 to 30°C) for germination. Seedlings were counted every day up to the completion of germination at the fifteenth day. Data on abnormal seedlings, dead seeds, and shoot and root lengths were recorded from the experiment. Abnormality of the seedlings was

determined according to the prescribed rules given by ISTA (1999). Germination percentage was calculated following Krishnasamy and Seshu (1990) as:

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

2.2.3. Germination index (GI)

The germination index (GI) was calculated as described in the Association of Official Seed Analysts (AOSA, 1983).

$$\text{Germination index (GI)} = \frac{\text{Number of germinated seed}}{\text{Days to first count}} + \dots + \frac{\text{Number of germinated seed}}{\text{Days to final count}}$$

2.2.4. Seed vigour index

This was calculated by determining the germination percentage and seedling length of the same seed lot. For seed vigour index data were recorded on germination up to 15 days of sowing. Then root length and shoot length were measured from seedlings of pots for calculating seed vigour index. Seed vigour index was calculated using the following formula (Baki and Anderson, 1970):

$$\text{Seed vigour index} = \text{Germination (\%)} \times (\text{Mean shoot length} + \text{Mean root length})$$

2.2.5. Seedling vigour index

Ten plant samples from each pot were harvested on the day 15 of the germination test and dried at 70°C for 72 hours for dry matter yield. Dry weight of those samples was measured. Seedling vigor index was calculated according to the following formula (Jayraj and Karivaratharaju, 1992):

$$\text{Seedling vigour index} = \text{Mean germination (\%)} \times \text{Dry matter content per seedling}$$

2.2.6. Electrical conductivity test

For electrical conductivity test, a sample of 50 seed was taken from each treatment in a conical flask containing 50 ml de-ionized water and was

incubated at 20°C for 24 hours following Ali *et al.* (2004). After 20 hours, water of the beaker containing seeds was decanted in order to separate the seeds. The electrical conductivity of the decanted water containing seed leachate was measured with a conductivity meter (Model-CM-30ET). Four repetition measures were made for each sample of seed.

2.2.7. Observation of biotic factors

For the observation of biotic factors that influence seed quality, 50 g of seeds was taken and the number and kind of insect or pathogens present were observed.

2.2.8. Statistical analysis

Analysis of variance was conducted using MSTAT-C software (Russel and Eisensmith, 1993) and significance of differences among means was compared employing least significant difference (LSD) test at a 0.05 probability level.

3. Results and Discussion

3.1. 1000-seed weight and moisture content of soybean as influenced by storage container and storage period

Effect of containers and storage periods on 1000-seed weight and moisture content of soybean seeds are presented in Table 1. Storage container showed significant effect on 1000-seed weight and moisture content at different storage periods. At 0-day storage, 1000-seed weight of different containers' seeds was similar (74.85 g) but it increased with increasing storage time. The maximum 1000-seed weight was recorded in cloth bag, while the lowest was in tin container at 15, 30, 45 and 60 days after storage (DAS), respectively. In addition, the initial moisture content was similar (8.30%) in different containers' seeds, but it increased with the increase in storage period. Seeds in cloth bag gave the highest moisture and tin container resulted in the lowest moisture at 15, 30, 45 and 60 DAS, respectively which was significantly different from those in polythene bag, except 15 DAS.

Table 1. Effect of storage containers and storage periods on 1000- seed weight and moisture content of soybean

Containers	1000-seed weight (g) at					Moisture content (%) at				
	0-day storage	15 DAS	30 DAS	45 DAS	60 DAS	0-day storage	15 DAS	30 DAS	45 DAS	60 DAS
Tin container	74.85	75.45b	77.51c	79.97b	82.11b	8.3	8.38b	8.55c	9.78c	10.88c
Polythene bag	74.85	77.35a	80.42b	81.78ab	85.74a	8.3	8.60b	9.15b	11.61b	13.15b
Cloth bag	74.85	79.14a	82.25a	84.66a	87.20a	8.3	8.90a	10.85a	12.90a	15.01a
LSD _{0.05}		1.83	3.93	3.38	2.80		0.25	0.26	0.40	0.24
CV (%)		4.57	3.07	2.57	2.06		1.84	1.68	2.19	1.16

Means followed by different letters within same column are significantly different by the LSD test at 0.05 level; DAS = Days after storage

Table 2. Effect of storage containers and storage periods on germination percentage and germination index of soybean seed

Containers	Germination (%) at					Germination index (GI) at				
	0-day storage	15 DAS	30 DAS	45 DAS	60 DAS	0-day storage	15 DAS	30 DAS	45 DAS	60 DAS
Tin container	83.00	82.25a	81.75a	81.00a	79.75a	17.03	16.98a	16.10a	15.34a	13.89a
Polythene bag	83.00	80.0ab	78.25b	76.50b	72.50b	17.03	15.53b	14.22b	12.93b	11.88b
Cloth bag	83.00	78.75b	76.50c	71.25c	66.50c	17.03	14.74b	13.06c	12.02b	10.97c
LSD _{0.05}		2.69	1.89	6.30	4.36		0.83	0.73	1.59	0.36
CV (%)		2.10	1.49	5.17	3.73		3.30	3.17	7.40	1.83

Means followed by different letters within same column are significantly different by the LSD test at 0.05 level; DAS = Days after storage

1000-seed weight and moisture content were increased gradually up to 60 DAS and were higher in cloth bag. As seed is highly hygroscopic living material; it absorbs moisture from air. The rate of absorbance was higher in cloth bag because of cloth bag is not air tight container, whereas tin container and polythene

bag are moisture proof. The hydrophilic nature of high protein content of soybean helps in more absorption of water, increases the hydrolytic enzyme activity, enhances respiration and increases in free fatty acid content and finally, deteriorates the seed quality (Monira *et al.*, 2012; Khalequzzaman *et al.*, 2012).

3.2. Germination percentage (GP) and germination index (GI) of soybean seed as influenced by storage container and storage period

Germination is the most important function of a seed as an indicator of its viability and worth as seed. There was a significant difference in the percentage of germination due to different storage containers for all storage periods in the study (Table 2). Seeds packed in tin container retained seed viability for longer time than in cloth bags. Initial germination was 83.00% for all containers at 0-day storage. Both germination percentage (GP) and germination index (GI) decreased with the increase of storage period. The highest GP and GI were recorded for seeds in tin container, whereas the lowest values were recorded in cloth bag at 15, 30, 45 and 60 DAS, respectively, which were statistically identical to polythene bag at 15 DAS.

Germination of soybean seeds was reported to be affected by storage containers during storage period (Shelar, 2008; Singh and Dadlani, 2003). Harrington (1973) found that a package which is moisture proof or moisture resistant would be more valuable in prolonging germination ability and vigor. Germination percentage and germination index of soybean seeds were decreased gradually in cloth bag. The reduction was more accentuated as the moisture content of

the grains increased. By increasing moisture content from 8.3 to 15.01% in cloth bag (Table 1), the rate of germination percentage decreased from 83 to 66.50%; and germination index decreased from 17.03 to 10.97% at 60 DAS (Table 2). During the storage period 70-82% RH was prevailing. That's why initial moisture content of the seeds of non-air tight cloth bag was low and it increased over time by absorbing moisture from the surrounding atmosphere. The warm temperature and concomitant high relative humidity in the store reduced seed viability due to increased respiratory activity, decreased stored food materials and increased degree of invasion by seed-borne or storage fungi (Dejene, 2004).

3.3. Abnormal seedling and dead seed of soybean as affected by storage container and storage period

Abnormal seedling and number of dead seed increased with the increase of storage period (Table 3). The initial abnormal seedling was 6.00% for all containers at 0-day storage. The highest abnormal seedling percentage was recorded in cloth bag and the lowest values were recorded in tin container, which were identical to those in polythene bag at 15 and 45 DAS, respectively. The initial dead seed in tin container, polythene bag and cloth bag was 4.25%, but it increased with increasing storage time.

Table 3. The existence of abnormal seedlings and dead seeds of soybean as affected by storage containers and storage periods

Containers	Abnormal seedling (%) at					Dead seed (%) at				
	0-day storage	15 DAS	30 DAS	45 DAS	60 DAS	0-day storage	15 DAS	30 DAS	45 DAS	60 DAS
Tin container	6.00	6.75b	7.25b	7.50b	8.00c	4.25	4.50c	5.25b	5.75c	7.50b
Polythene bag	6.00	7.00b	8.25ab	9.50b	10.50b	4.25	5.50b	6.75ab	7.75b	9.75b
Cloth bag	6.00	8.50a	9.75a	12.5a	15.50a	4.25	6.50a	8.50a	11.50a	14.50a
LSD _{0.05}		1.83	1.71	2.07	2.50		0.92	3.71	3.13	2.96
CV (%)		15.41	12.68	13.13	13.80		10.50	33.97	23.49	17.47

Means followed by different letters within same column are significantly different by the LSD test at 0.05 level; DAS = Days after storage

The highest dead seed percentage was recorded in cloth bag, whereas the lowest values were recorded in tin container at 15, 30, 45 and 60 DAS, respectively, which was statistically identical to those in polythene bag at 60 DAS.

The maximum abnormal seedlings and dead seeds were recorded in cloth bag and the minimum in tin container. Increasing rate of abnormal seedling and dead seed was observed higher in cloth bag, due to high moisture and fungal activities in soybean (Monira *et al.*, 2012). Seed quality deterioration during storage was due to the damage to membrane, enzyme, proteins and nucleic acid. With the accumulation of time, such degenerative changes result in complete disorganization of membranes and cell organelles and ultimately causing death of the seed (Roberts, 1972).

3.4. Seedling growth of soybean as affected by storage container and storage period

Seed quality of soybean was evaluated by seedling characters, i.e. length and weight of seedling. Seedling growth is considered to be important tool that can be used for assessing the magnitude of deterioration. Seedling characters like shoot length, root length varied over storage containers and storage period. Storage container showed significant effect on root length and shoot length which decreased with the increase in storage period (Table 4). Initial root length was 16.48 cm and shoot length was 33.76 cm for all containers at 0-day storage. Seeds in tin container gave the highest root and shoot length and cloth bag resulted in the lowest root and shoot length at 15, 30, 45 and 60 DAS, respectively. Moisture content had the influential effect on seed storage and high moisture content in cloth bags obviously deteriorated seedling growth (Copeland, 1976).

3.5. Seedling dry matter of soybean as affected by storage container and storage period

Seedling dry weight of soybean seed was obtained for seed of three containers (tin container, polythene bag, and cloth bag) which

indicate that dry weight of cloth bags' seedling was lower than the other container's seed (Table 5). Seedling dry weight per plant was decreased with the increase of storage period. Initial soybean seedling dry weight was 33.13 mg plant⁻¹ for all containers at 0-day storage. The highest seedling dry weight per plant was obtained from tin container for all storage periods. On the contrary, the lowest dry weight per plant was recorded from cloth bag for all storage periods, which was statistically identical to that in polythene bag at 30 DAS.

The deterioration of seed due to high moisture absorption in cloth bags, which is a progressive process accompanied by accumulation of metabolites, and which progressively depresses germination and growth of seedling with increased age, ultimately reduces the dry matter and vigour of soybean seeds during storage.

3.6. Vigour index of soybean as affected by storage container and storage period

Effects of containers and storage periods on seed and seedling vigour index of soybean are presented in Table 6. Both seed and seedling vigour index were decreased with the increase of storage period. Initial seed vigour index was 4169.92 for all containers at 0-day storage. Storage containers had significant effect on seed vigour index. Seed vigour index was decreased with the increase of storage period. The highest seed vigour index was obtained from tin container, and the lowest vigour index was obtained from cloth bag for all storage periods. Initial seedling vigour index was 2749.79 for all containers. The highest seedling vigour index was obtained from tin container, and the lowest seedling vigour index was obtained from cloth bag at 45 and 60 DAS. The highest seed and seedling vigour index was obtained from tin container, which was followed by polythene bag and the lowest were obtained from cloth bag for all storage periods, which might be due to low vigour of seeds for long storage periods, high moisture and fungi infestation (Singh and Dadlani, 2003).

Table 4. Effect of storage containers and storage periods on root length and shoot length of soybean seedlings

Containers	Root length (cm/plant) at					Shoot length (cm/plant) at				
	0-day storage	15 DAS	30 DAS	45 DAS	60 DAS	0-day storage	15 DAS	30 DAS	45 DAS	60 DAS
Tin container	16.48	14.15a	13.98a	13.28a	12.07a	33.76	33.01a	32.49a	31.78a	29.90a
Polythene bag	16.48	13.27b	12.01b	10.75b	8.64b	33.76	31.34b	29.28b	27.80b	24.53b
Cloth bag	16.48	12.20c	10.42c	8.13c	6.02c	33.76	29.55c	27.06c	23.65c	20.93c
LSD _{0.05}		0.87	0.75	0.42	1.10		1.03	1.93	2.44	2.52
CV (%)		4.14	3.84	2.47	7.73		2.06	4.07	5.51	6.27

Means followed by the different letter within same column are significantly different by the LSD test at 0.05 level; DAS = Days after storage

Table 5. Effect of storage containers and storage periods on dry weight (mg) per plant of soybean seedlings

Containers	Dry weight (mg/plant) at				
	0-day storage	15 DAS	30 DAS	45 DAS	60 DAS
Tin container		32.01a	29.18a	28.00a	26.15a
Polythene bag	33.13 ^{NS}	26.48ab	23.31b	20.75b	16.60b
Cloth bag		23.60b	19.05b	15.30c	11.05c
LSD _{0.05}		6.74	5.30	5.76	1.82
CV (%)		15.40	13.90	16.87	6.35

Means followed by the different letter within same column are significantly different by the LSD test at 0.05 level; DAS = Days after storage

3.7. Electrical conductivity (EC) of soybean seed as affected by storage container and storage period

Electrical conductivity (EC) test of seed leachate provides a quick decision about seed vigour. Storage containers had significant effect on EC of soybean seed (Table 7). Initial EC of seed leachate was 89.27 $\mu\text{S cm}^{-1}$ for all containers at 0-day storage. The highest EC of seed leachate was recorded from the seeds stored in cloth bag for all storage periods. Conversely, the lowest EC was recorded from tin container for all storage periods, which was statistically identical to polythene bag at 15 DAS.

EC of seeds showed a highly negative correlation with germination percentage (Fig. 1). The negative relationship between EC and seed germination indicated that more cells leachate escaped from low quality seed and lowered the germination capacity of soybean seed, which is in agreement with the findings of Halim *et al.* (2012) in onion seeds. High EC of seed is assumed due to membrane deterioration during the imbibitions period of lower quality seeds. In tin container, the quality of seed was good and that is why they leak less leachate compared to that of low quality seeds stored in cloth bag. The vigour of cloth bag's seed was decreased over time of storage due to greater moisture absorption (Baki and Anderson, 1970).

Table 6. Effect of storage containers and storage periods on seed and seedling vigour index of soybean

Containers	Seed vigour index at					Seedling vigour index at				
	0-day storage	15 DAS	30 DAS	45 DAS	60 DAS	0-day storage	15 DAS	30 DAS	45 DAS	60 DAS
Tin container	4169.92	3880.51a	3798.97a	3653.55a	3550.72a	2749.79	2632.04a	2385.87a	2253.20a	2084.60a
Polythene bag	4169.92	3569.66b	3231.25b	2947.05b	2626.71b	2749.79	2118.75ab	1824.03b	1587.10b	1203.00b
Cloth bag	4169.92	3289.43c	2865.88c	2261.70c	1777.94c	2749.79	1855.60b	1457.25b	1086.50c	733.15c
LSD _{0.05}		264.20	121.4	322.7	376.50		524.6	418.3	364.9	118.00
CV (%)		4.61	2.30	6.83	8.88		14.89	13.84	13.89	5.51

Means followed by the different letter within same column are significantly different by the LSD test at 0.05 level; DAS = Days after storage

Table 7. Effect of storage containers and storage periods on the electrical conductivity (EC) of soybean seed

Containers	Electrical conductivity ($\mu\text{S cm}^{-1}$) at				
	0-day storage	15 DAS	30 DAS	45 DAS	60 DAS
Tin container		126.75b	195.25c	214.88c	291.50c
Polythene bag	89.27 ^{NS}	172.25b	258.75b	308.75b	427.75b
Cloth bag		220.00a	319.50a	441.25a	715.76a
LSD _{0.05}		47.72	55.97	56.93	106.00
CV (%)		17.25	13.57	11.07	13.86

Means followed by the different letter within same column are significantly different by the LSD test at 0.05 level; DAS = Days after storage

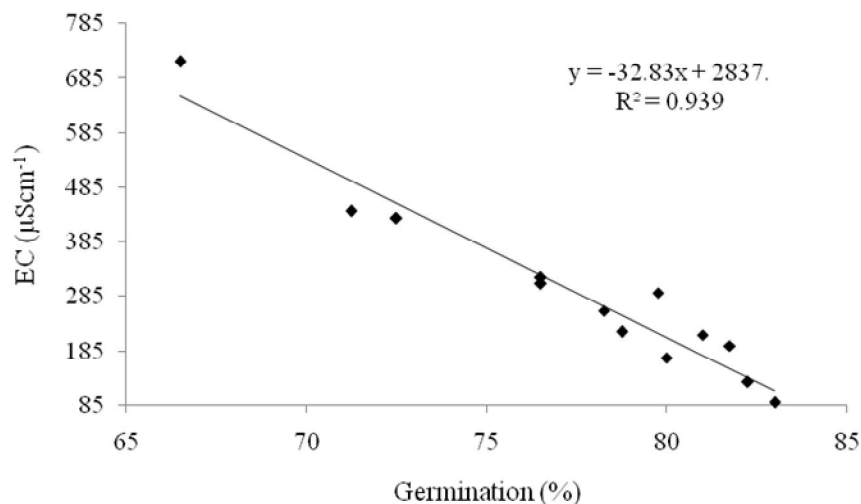


Fig. 1. Relationship between germination (%) and electrical conductivity (EC) of soybean seeds

3.8. Effect of Biotic Factors

Biotic factors like insect and fungi were investigated during the storage period in different storage containers. No insect was found in any storage container for the whole storage periods. In general, prevalence of fungi was found higher in the seeds of cloth bag and the lowest was found in seeds of tin containers. Number of dead seeds (Table 3) indicates the presence of fungi but these were not identified by any pathological test.

3.9. Seed colour of soybean as affected by storage container and storage period at 60 DAS

Initial seed colour of soybean was yellowish. After 60 days of storage, seed colour of tin container was similar as initial colour, but seeds of cloth bag changed into fade or brown colour. Fade or brown colour might be due to increased moisture and fungi infestation (Khalequzzaman, 2012) in cloth bags.

4. Conclusions

The soybean grains generally deteriorated with storage and deterioration was particularly strong for grains stored in cloth bag. High temperature, high relative humidity, and moisture in the storage environment are the principal factors involved in deterioration of soybean seed quality during storage. Loss of germination capacity was the final manifestation of seed deterioration. When seed moisture content was increasing the rate of germination percentage was decreased. The deterioration rate was also the highest in seeds of cloth bag. The electrical conductivity, abnormal seedling and dead seed were increased with increasing storage time, but the increasing rate was higher in seeds of cloth bag. The shoot and root length of seedling and seedling vigour was the lowest in cloth bag at the end of storage period. Dry weight per plant of soybean decreased in all cases but the rate of deterioration was also the highest in cloth bag.

It may be concluded that the conventional practices of soybean seed storage in cloth bags are not suitable because there is a possibility of moisture gain and pests attack, which play important role in deterioration of soybean seed quality and viability during storage. Air tight tin container will be the best means for soybean seed storage for long time.

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