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## **Soybean Seed Quality as Affected by Cultivars, Threshing Methods and Storage Periods**

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### **ABSTRACT**

Poor quality in some soybean seed during storage can be due to their physiological characteristic or mechanical causes. The soybean seeds are susceptible to mechanical injury mediated damage during post harvest handling. These injuries lead to deterioration during storage. The purpose of this investigation was to evaluate the effects of different threshing methods i.e., hand threshing, beating by stick threshing and mechanical threshing on some soybean cultivars i.e., Giza 21 (G-21), Giza 35 (G-35) and Giza 111 (G-111) yield, seed quality and its longevity during storage. A field experiment was carried out at the Experimental Farm of Agronomy Department, Faculty of Agriculture, Mansoura University, Egypt during 2010 and 2011, followed by a laboratory experiment under the laboratory conditions of Seed Technology Research Unit in Mansoura, Dakahlia Governorate, Seed Technology Research Department, Field Crops Research Institute, Agricultural Research Center. Seed quality tests were performed on each treatment, directly after threshing (0 month), three or six months storage. The results showed significant differences among soybean cultivars in all studied traits. G-21 surpassed G-35 and G-111 cultivars on seed yield and related attributes, oil and protein %, germination, germination after accelerated aging %, seed and seedling vigor. Threshing methods insignificantly affected oil and protein percentages, but exhibited significant effect on seed yield and seed quality. Threshing by hand or by stick methods were reduced seed quality less than mechanical method. Seed viability of soybean cultivars decreased gradually with increasing storage period up to six months. G-111 showed lower resistance to storage conditions, which might be classed as poor "storer" under the experimental conditions. Higher viability of G-21 seed (as indicated by germination score) than others (G-35 and G-111) might be classified as a good "storer". Assessment of some soybean cultivars seed quality during storage by monitoring germination and germination after aging percentages, in addition to seed and seedling vigor measurements may be reliable indicators for damages occurred after mechanical threshing method.

**Key words:** Soybean, seed quality, cultivars, threshing, storage periods

### **INTRODUCTION**

The major complaint from the farmers who tender their seed for processing is after that, this seed has been found to exhibit lower germination. Soybean [*Glycine max* (L.) Merrill] is an important legume crop. Wide variation in yield attributes, seed yield, nutritional value and seed

quality were reported for soybean cultivars by many investigators i.e., El-Harty *et al.* (2010), Shairef *et al.* (2010), Mostafa (2011) and El-Sayed (2012) and according to them the differences in soybean cultivars might be due to the genetic makeup factors.

Soybean crop, as all cereal crops, begins to lose quality when they are harvested, processed or stored. The losses of seed viability resultant destitution in plant stand which is the basis for appropriate production and expansion of this crop mainly in tropical and subtropical countries. Soybean seed deteriorates faster than those of most other crops (Priestley *et al.*, 1985). Besides inherent poor storability, mechanical damage is mainly responsible for seed quality deterioration becomes increasingly by more important problems such as storage deterioration, insect infestation and diseases (Wilson and McDonald, 1992). Soybean seed being inherently a weak structure is more susceptible to mechanical injury which increases its deterioration and loss on seed quality (Tekrony *et al.*, 1987; Lori *et al.*, 2001) during post harvest handling (Shelar, 2008). The thin coat of the seed is fragile and if too dry it can develop cracks while handling, leading to deterioration. These cracks further multiply during storage (Ujjinaiah and Sreedhara, 1998; Parde *et al.*, 2002). Free fall is a major cause of damage to soybeans during handling which not only rebound on quantitative losses but also seed quality (Parde *et al.*, 2002). Bartsch *et al.* (1979) reported that a more common form of readily visible physical seed damage is seed coat and radical or bruising of the cotyledons fractures under the seed coat which are difficult to detect and remove during threshing. Nave (1979) reported that a seed producer must be interested with maintaining threshing and separating efficiency while avoiding unnecessary impact damage of seed. Shrivastava and Ojha (1986) stated that soybean is susceptible to mechanical damage, hence germination declines by 10% when dropped from a height of only 1 m. Mechanical threshing causes more damage to the seed compared to manual threshing (Saini *et al.*, 1982). Threshing by hand method recorded higher laboratory germination percentage, least physical damage and electrical conductivity in soybean seed compared to beating on cement floor (Reddy *et al.*, 1995). Jha *et al.* (1996) threshed soybean seed by hand beating, machine threshing, or tractor treading and kept under ambient storage conditions and then tested for seed quality. He found that hand beating resulted in higher germination levels and less deterioration of seed than the other two techniques at all stages of storage. Since the seed quality and longevity depends upon the initial quality of seed and storage method (Shelar, 2008). So, the type and amount of mechanical damage caused during post harvest greatly influence the viability and vigor of soybean seed during storage (Wilson and McDonald, 1992). Mayeux *et al.* (1972) found that the germination of soybean seed is influenced by the percentage of cracks in stored seed. Wein and Kueneman (1981) found that a large variation in quality loss of soybean seed with cultivar during storage. Wilson and McDonald (1992) observed that soybean genotypes differ in inherent poor storability and their capacity to maintain viability and vigor during storage.

The purpose of this investigation was to evaluate the effects of damage occurred during some threshing methods and different storage periods on yield and seed quality of some soybean cultivars.

## **MATERIALS AND METHODS**

**Field experiment:** A field experiment was carried out at the Experimental Farm of Agronomy Department, Faculty of Agriculture, Mansoura University, Egypt during two successive growing

summer seasons of 2010 and 2011. The main objective of this study was to determine: (1) the performance of soybean cultivars i.e., Giza 21 (G-21), Giza 35 (G-35) and Giza 111 (G-111) on yield attributes and (2) the effect of some threshing methods i.e., hand threshing (Hand), beating by stick threshing (Stick) and mechanical threshing (Mechanical) of studied cultivars on yield, oil and protein percentages. In each season, the experiment was laid-out in completely randomized block design with three replications.

**Seed coat percentage:** The pods were air dried to approximately 12% moisture content and hand-threshed. Ten seeds were soaked in distilled water and incubated at 5°C for 15-16 h. The seed coat was separated from each seed using a razor blade. The seeds (without seed coats) and the seed coats were dried in a hot air oven at 100°C for 24 h. After drying, the seeds (without seed coats) and seed coats were weighed separately and the seed coat percentage was calculated (Kuo, 1989). The percentages of seed coat were 8.50, 8.10 and 7.60% for studied soybean cultivars Giza 21, Giza 35 and Giza 111, respectively.

The soil in the experimental field was clay in texture with EC 1.66 dS m<sup>-1</sup> and pH 7.90. Each experimental unit area included 31 ridges each of 60 cm width and 3.5 m length (1/64.52 fed.). The preceding winter crop was Egyptian clover in both seasons. The experimental field was well prepared and calcium super phosphate (15.5% P<sub>2</sub>O<sub>5</sub>) was applied during soil preparation at the rate of 150 kg/fed.

Soybean seeds were thoroughly mixed with nodulating bacteria and sown in hills, 10 cm apart at eastern side of ridges on the first week of May in both seasons. After three weeks, only two healthy plants remained in each hill. Nitrogen and potassium fertilizers were applied in the forms of urea (46.5% N) and potassium sulphate (48% K<sub>2</sub>O) at the rate of 60 kg N/fed. and 48 kg K<sub>2</sub>O/fed. in two equal doses (after thinning and three weeks later). The normal cultural practices for growing soybean crop were followed.

When plants of soybean cultivars reached harvest maturity (about 90% of the pods were yellow, but before they had turned brown), a sample of ten randomly guarded plants from one outer ridge of each experimental unit was taken and the following characteristics were recorded; number of branches/plant, number of pods/plant, pod length, number of seeds/pod, number of seeds/plant, 100-seed weight and seed yield/plant.

Seed yield (kg/fed.) was determined from the rest of the ridges (30 ridges) of each experimental unit, where pods were harvested by hand stripping and dried in open air to reduce moisture content to 10-12%. Each threshing method was carried out on pods which harvested from each 10 ridges of each experimental unit (1/200 fed.) as follows: (1) Hand threshing: Pods were hand threshed. (2) Beating by stick threshing: The dried pods contained in jute bags were threshed by beating with stick and the seeds were separated from the pod walls and another plant parts by sieving. Sieving (round hole) used to eliminate the small, immature and insect damaged seeds. (3) Mechanical threshing: The dried plant threshed by threshing machine at speed of 450 rpm/kg.

Seed oil content estimated by taking seed samples of dried seeds of each plot, cleaned and ground into very fine powder by grinder. The seed oil percentage was determined following AOAC (1990) using Soxhlet apparatus. Seed crude protein percentage was estimated as per the improved Kjeldahl method of AOAC (1990).

All data of each season were statistically analyzed according to the technique of Analysis of Variance (ANOVA) for the completely randomized block design for yield attributes and factorial completely randomized block design for seed yield/fed., oil and protein % and then combined analysis was done between both seasons as published by Gomez and Gomez (1984), by means of "MSTAT-C" Computer software package. The treatment means were compared using the Least Significant of Differences (LSD) as outlined by Waller and Duncan (1969).

**Laboratory experiment:** A laboratory experiment was carried out under the laboratory conditions of Seed Technology Research Unit in Mansoura, Dakahlia Governorate, Seed Technology Research Department, Field Crops Research Institute, Agricultural Research Center during 2010 until 2012 years. The purpose of the experiment was to assess seed quality resulted from the previous field experiment. The studied treatments were; (1) soybean cultivars (G-21, G-35, G-111), (2) threshing methods (Hand, Stick, Mechanical) and (3) storage periods (0, 3, 6 months). Draw samples were divided into three parts, the first was evaluated directly to seed quality tests (0 storage) and the others were stored in cloth bags under the laboratory condition for 3 and 6 months. After each storage period (0, 3 and 6 months), samples of seed were drawn at random from each treatment. Random sample of 400 seeds per each treatment were sown in four replications in sterilized sand culture and incubated at 25°C for 8 days and then used to evaluate every seed test done on each treatment per the rules of International Seed Testing Association (ISTA, 1985). Germination percentage was expressed by the percentage of seed germinating normally after eight days. Then the same four replications were used to evaluate seedling length (cm) from 10 normal seedlings taken at random from each replicate at the end of standard germination test, then dried in a forced air oven at 105°C for 24 h to obtain seedlings dry weight and expressed as grams. The accelerated aging test was carried out to predict the storage potential of seed. Each sample of 400 seeds were sterilized, weighted and placed in accelerate ageing chamber at 40°C and 100% relative humidity for 48 h. After this exposure period, the seed sample was submitted to the standard germination test. Electrical conductivity (Matthews and Powell, 1981) of seed leakages was determined for 50 individually weighed seeds per sample placed into 250 mL distilled water at 20°C. After 24 h the leakage electrical conductivity was measured using the CMD 830 WPA. The conductivity was expressed as  $\mu\text{mhos/g}$  of seed. Mean germination time was calculated according to Ellis and Roberts (1981):

$$\text{Mean germination time} = \frac{(N1 \times T1) + (N2 \times T2) + (N3 \times T3) + (N4 \times T4)}{N1 + N2 + N3 + N4}$$

Where, N1, N2, N3 and N4 are first, second, third and fourth count, respectively and T1, T2, T3 and T4 are time of first, second, third and fourth count, respectively. Germination rate was estimated according to Bartlett (1937):

$$\text{Germination rate} = \frac{a + (a+b) + (a+b+c) + \dots + (a+b+c+m)}{n(a+b+c+\dots+m)}$$

Where, a, b and m are number of seedlings emerged at the first count, second count and final count and n is number of counts.

Collected data were subjected to the statistical analysis according to the technique of Analysis of Variance (ANOVA) for the factorial completely randomized design and then combined analysis was done between both seasons as following Gomez and Gomez (1984), by means of "MSTAT-C" Computer software package. The treatment means were compared applying the least significant of differences (Waller and Duncan, 1969).

## RESULTS AND DISCUSSION

**Field experiment:** The obtained results show that number of branches and pods/plant, pod length, number of seeds per pod and per plant 100-seed weight, seed yield per plant (Table 1), seed yield per feddan, oil and protein percentages (Table 2) were significantly affected in the studied soybean cultivars over both seasons. Giza 21 cultivar significantly surpassed other studied cultivars (Giza 35 and Giza 111) in all studied characters. Giza 35 cultivar ranked after Giza 21 cultivar followed by Giza 111 cultivar in case of yield attributes, seed yield, oil and protein % over both seasons. This variation might be due to the genetic makeup factors of studied soybean cultivars. These results are in agreement with those stated by Shairef *et al.* (2010), Mostafa (2011) and El-Sayed (2012).

Threshing methods (hand threshing, beating by stick threshing and mechanical threshing) significantly affected seed yield per feddan, but did not show significant effect on oil and protein percentages over both seasons (Table 2). Hand threshing of soybean significantly increased seed yield/fed. compared with beating by stick threshing and mechanical threshing by 6.06 and 11.28% over both seasons.

Table 1: Quantitative analyses of branch, pod and seeds of studied cultivars over both seasons

Characters treatments	No. of branches/plant	No. of pods/plant	Pod length (cm)	No. of seeds/pod	No. of seeds/plant	100-seed weight (g)	Seed yield/plant (g)
<b>Cultivars</b>							
G-21	2.88	73.33	5.80	3.27	198.9	20.88	36.77
G-35	2.27	69.22	4.71	3.05	185.4	16.88	31.22
G-111	1.88	65.11	4.09	2.86	176.9	13.77	22.72
LSD at 5%	0.31	4.02	0.53	0.27	8.8	2.04	3.71

Table 2: Seed yield/fed., oil and protein percentages as affected by cultivars and threshing methods over both seasons

Characters treatments	Seed yield (kg/ fed)	Oil (%)	Protein (%)
<b>Cultivars</b>			
G-21	1725.2	20.26	40.78
G-35	1626.9	20.01	40.00
G-111	1467.6	19.90	39.15
LSD at 5%	40.4	0.20	0.66
<b>Threshing methods</b>			
Hand	1705.2	20.07	39.97
Stick	1601.8	20.05	40.01
Mechanical	1512.7	20.06	39.96
LSD at 5%	27.7	ns	ns

ns: Not significant

The interaction between cultivars and threshing methods exhibited significant effect on seed yield/fed. over both seasons only, while oil and protein percentages did not showed significant effect. The highest seed yield/fed was resulted from G-21 cultivar when threshed by hand method. Whereas, the lowest seed yield/fed was produced from G-111 cultivar when threshed by mechanical method (Fig. 1).

**Laboratory experiment:** Data of the standard germination, seed and seedling vigor traits as affected by the studied factors are presented in Table 3. There was a significant difference between soybean cultivars in standard germination percentages. G-21 and G-35 cultivars showed the highest germination percentages (85.5 and 85%) while, G-111 produced the lowest value, which was 83.3%. Also, there was a significant difference between soybean cultivars on seed vigor traits as measured by (germination percentage after aging, electrical conductivity, mean germination

Table 3: Germination and seed and seedling vigor quantitation, as affected by cultivars, threshing methods and storage periods over both seasons

Characters treatments	Seed vigor				Seedling vigor		
	Germination (%)	Germination after aging (%)	Electrical conductivity ( $\mu$ mhos/g)	Mean germination time (day)	Germination rate	Seedling length (cm)	Seedling dry weight (g)
<b>Cultivars</b>							
G-21	85.5	66.0	0.032	3.28	0.795	24.16	0.685
G-35	85.0	63.5	0.041	3.63	0.769	22.05	0.656
G-111	83.3	63.9	0.047	3.77	0.740	26.31	0.740
LSD at 5 %	1.6	1.2	0.007	0.17	0.005	0.48	0.016
<b>Threshing methods</b>							
Hand	85.9	67.0	0.037	3.46	0.810	24.41	0.706
Stick	84.7	64.0	0.039	3.57	0.759	24.13	0.697
Mechanical	83.1	62.5	0.044	3.65	0.735	23.98	0.679
LSD at 5 %	1.6	1.2	0.007	0.17	0.005	0.48	0.016
<b>Storage periods</b>							
0 month	90.6	76.1	0.037	3.31	0.827	24.33	0.737
3 month	83.6	66.1	0.040	3.57	0.763	24.14	0.708
6 month	79.5	51.3	0.043	3.79	0.713	24.04	0.636
LSD at 5 %	1.6	1.2	0.007	0.17	0.005	0.48	0.016

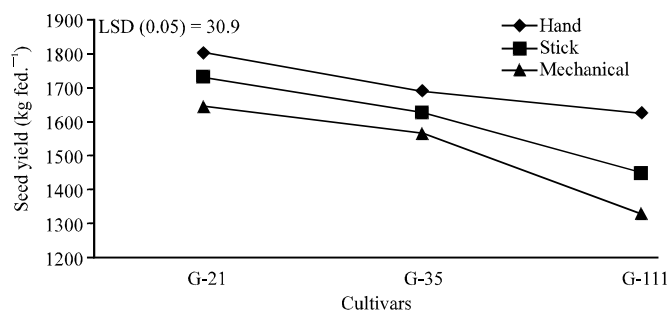


Fig. 1: Seed yield (kg/fed.) as affected by the interaction between cultivars and threshing methods over both seasons

time and germination rate). G-21 surpassed other cultivars on germination percentages after accelerated aging. The possible reason of lower germination given by G-111 and G-35 after aging test might be that tolerance of subjected seed had declined in vigor leading to slower germination.

Compared to higher electrolyte loss recorded in G-35 (0.041  $\mu$  mhos/g seed) and G-111 (0.047  $\mu$  mhos/g seed), lowest leakage loss noticed in the G-21 soybean cultivar was indicative of higher seed vigour and quality. Poor seed vigour and quality may also be ascribed to relatively thin seed coat 8.1 and 7.6% measured respectively in G-35 and G-111 cultivars than the 8.5% in G-21. Relatively thick seed coat in G-21 than the G-35 and G-111 offers higher resistance to injuries that might be occurring during threshing practice and storage condition including accelerated ageing (Marwanto, 2004).

Also the results indicated that cultivars treatment had a significant effect on seed and seedling vigor (Table 3). For example, G-21 gave the maximum germination rate. Seedling vigor traits as nominated by seedling length and its dry weight also significantly differed between the studied varieties where, G-111 surpassed G-35 and G-21 in seedling length and its dry weight.

The threshing methods treatments had statistically significant effect on standard germination percentage, germination percentage after accelerated aging, seed and seedling vigor characters. Threshing soybean by hand resulted in higher and faster germination with normal seedlings and heaviest than threshing by stick or machine (Table 3). Soybean seed threshed by machine had lost viability rapidly to the seed threshed by hand or stick beating. The threshing methods particularly by machine, produces more breaks, cracks, bruises and abrasions in seeds which in turn resulted in abnormal seedlings (Green *et al.*, 1966; Reddy *et al.*, 1995).

Increasing storage period to three and six months induced significant reduction in germination percentage, germination after aging percentage, seed and seedling vigor (Table 3). The reduction in germination percentage after threshing to three months was 7.0% which more than from three to six months period (4.1%). While, the depression values of germination percentage after aging conversed (10.0 and 14.8%).

The seed and seedling vigour declined gradually during storage and reached minimum after six months of storage. Tekrony *et al.* (1993) stated that germination of soybean seed declines more rapidly during storage than other grain crops. Delouche (1974) reported that the seed germination and seedlings vigor declined with increasing storage period.

Cultivars $\times$ threshing methods $\times$ storage periods interaction were detected in germination percentage (Fig. 2). During the first storage period (0 month), G-21 was the higher value in

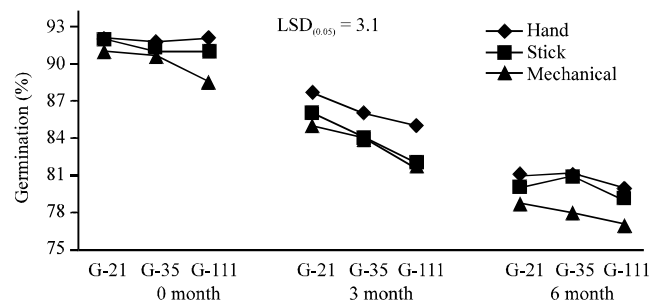


Fig. 2: Germination percentage as affected by the interaction among cultivars, threshing methods and storage periods over both seasons



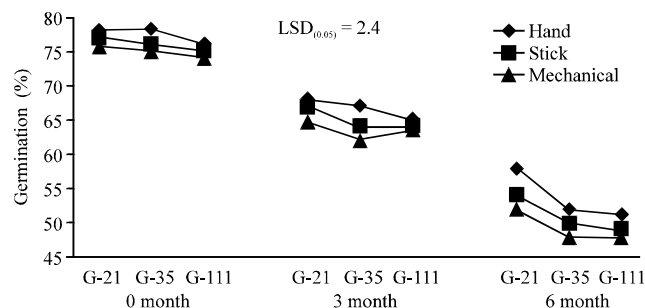


Fig. 3: Germination after aging as affected by the interaction among cultivars, threshing methods and storage periods over both seasons

germination percentage followed by G-35 then G-111 which was the lowest. Conversely, germination percentage was unaffected by threshing methods. Following three months of storage, G-21 gave maximum germination percentage when threshed by hand and strongly declined with mechanical method in the same period. Similar, trends observed after 6 months storage i.e., the decrease in germination percentage continued until reach to the minimum values when G-111 threshed by machine but did not reach less than accepted level of certified soybean seed (75%). During storage, injured or deeply bruised areas may serve as centers for infection and results in deterioration, whereas injuries close to vital parts usually bring about the most rapid losses of viability (Bewley and Black, 1984). The deleterious effects of mechanical threshing and storage on reducing soybean seed quality were also reported by Jha *et al.* (1996) and Shelar (2008).

There was significant effect among the three cultivars when threshed by three different methods during storage periods on germination percentage after accelerated aging (Fig. 3). The result showed that maximum percentage produced with G-21 cultivar, while the lowest once resulted from G-111 cultivar. The tolerance of G-111 to aging chamber condition was lower than other studied cultivars, which produced the lowest seed vigor and that was evident with mechanical threshing method. Since seed aging is associated with deteriorative changes in membranes (Parrish and Leopold, 1978) this indicates that G-21 was resistant to incubator aging, where mechanism of resistance was more affected by seed coat thickness G-111 (7.6%). Similar observation has also been reported by other investigators (Stanway, 1978; Dassou and Kueneman, 1984; Horlings *et al.*, 1991) and suggested that a seed coat characteristic plays an important role in resistance to seed deterioration. Meanwhile, seed coat relative impermeability to water absorption is the main factor involved (Kuo, 1989).

It is concluded that increasing storage periods to three and six months produced more weakness on seed vigor particularly with mechanical threshing method.

This may be related to the amount of damage on seed coat that resulted in increased seed deterioration with advance in storage period finally causing reduced germination percentage in accelerated aged seeds. These observations suggest that most cultivars resistant to the conditions of incubator aging were also resistant to deterioration in storage (Marwanto, 2004). This is consistent with result reported by Wein and Kueneman (1981) and Jha *et al.* (1996).

G-111 showed maximum seedling length and the heaviest dry weight when threshed by hand (Fig. 4, 5). Conversely, G-21 exhibited short seedling length and lighter dry weight when

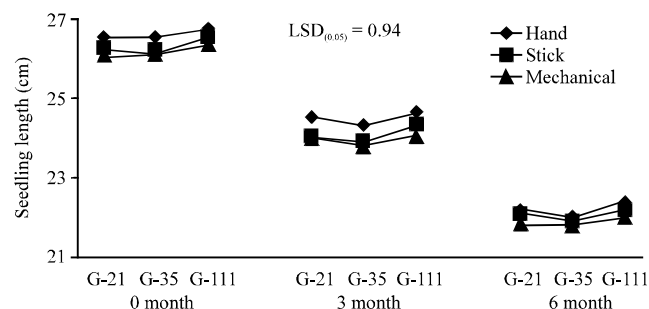


Fig. 4: Seedling length as affected by the interaction among cultivars, threshing methods and storage periods over both seasons

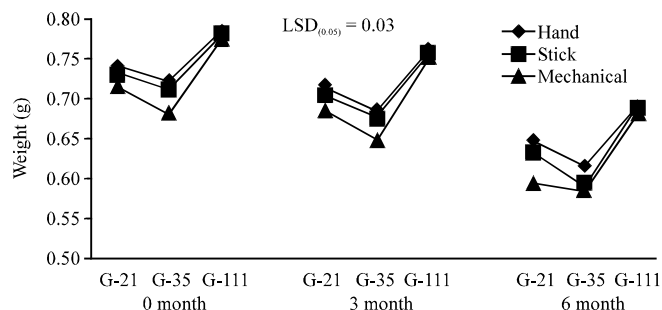


Fig. 5: Seedling dry weight as affected by the interaction among cultivars, threshing methods and storage periods over both seasons

threshed by mechanical method; these values declined gradually as the storage period increased to three months and reached to the minimum values after six months storage. Similar results were also reported by Shelar (2008) and Mayeux *et al.* (1972) when working with quality of stored soybean cultivars.

## CONCLUSIONS

Results of this study indicate that threshing some soybean cultivars and stored for six months resulted in lowering seed quality as indicated by reduction in seed germination, seed and seedling vigor. Among cultivars included in these studies, there were inherent cultivars differences in yield and seed quality after threshing and during storage. Reduced seed coat thickness played a significant role in decreasing seed quality. G-111 cultivar has thinner seed coat than G-21 and G-35 which resulted in susceptible for mechanical damage and increase in rate of deterioration further multiply after six months storage and ultimately resulted in less seed and seedling vigor. The consistently maintained higher seed viability by G-21 (as indicated by germination score) than others, might be classified as a good "storer". Irrespective inherent cultivar good storability, producer must be concerned with maintaining threshing efficiency to avoid invisible seed injuries, which reflects positively on soybean seed quality.

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