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Changes in Viability, Vigour and Chemical Composition of Soybean Seeds Stored Under the Humid Tropical Conditions

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ABSTRACT. Seeds of five soybean genotypes were stored for nine months under controlled and ambient humid tropical conditions, arranged in a completely randomized factorial design. Periodically seeds were germinated in sand at 30 C and in a field to determine the loss in seed viability and vigour. Seeds were also assessed for changes in chemical composition during storage. The seeds under ambient storage exhibited a sharp decline in viability beyond three months and the percentage viability was near zero after nine months; however, the drop in viability was insignificant under controlled storage. The loss in seed vigour was more remarkable and significantly greater under ambient than controlled storage. The genotypes differed significantly in viability and vigour at all periods of storage. A greater change in seed moisture coinciding with a larger loss in viability and vigour was evident under ambient than controlled storage conditions. The reduction in protein and ash and increase in carbohydrate, oil and free fatty acid in seed was less under controlled than ambient storage. A remarkable increase was noticeable in free fatty acid during storage under ambient conditions.

INTRODUCTION

One of the basic needs in agriculture is quality seed, characterised by high viability and vigour (Yaklich *et al.*, 1979). These two characters cannot be differentiated in storage tests (Justice and Bass, 1978) especially in seed lots of crop species like soybean (*Glycine max* (L.) Merr.) that rapidly deteriorate under ambient storage in subtropical and tropical environments, with high temperatures and humidities (Delouche, 1975; Tenne *et al.*, 1978; Arulnandhy *et al.*, 1984). Viability and vigour tend to decline as seed ages (Copeland, 1976) that inturn determine the crop stand and final production (Roberts, 1972; Singh and Gupta, 1982).

Evaluation of viability, vigour and chemical composition of any seed lot is of utmost importance to judge the quality. With this objective in view, studies on viability, vigour and chemical composition of seed of different soybean genotypes stored under ambient and controlled conditions are presented in this paper. These factors were considered and studied from the same seed lots simultaneously. Soybeans were selected because of their relative importance as a high protein food crop in Sri Lanka and their susceptibility to seed deterioration under local conditions.

MATERIALS AND METHODS

Seeds of five soybean genotypes, including three promising advanced breeding lines and two standard cultivars "Pb-1" and "Bosssier" were assessed for viability, vigour and chemical composition after storage under ambient (maximum temperatures 26-36 C, minimum temperatures 18-26 C and mean RH 73.2 $\pm$ 5.9%) and controlled (constant at 20 $\pm$ 1 C and 50 $\pm$ 1% RH) conditions for a period of nine months at the Agricultural Research Station in Maha Illuppallama. The seeds for this study were obtained from a planting on 18 October 1983 at the same site and were hand harvested at full maturity in January 1984 at the development stage R_8 (Fehr *et al.*, 1971). Seeds were sun-dried and kept in cold storage till 9 March 1984. The experiment was a factorial (5 genotypes X 2 storage conditions) arranged in a completely randomized design with four replications. Germination tests were performed initially and after three, six and nine months of storage.

Twenty-four 100 g seed samples of each genotype were placed in paper bags and stored under ambient or controlled conditions. For each testing period, four samples of each genotype were removed from each storage and seed moisture content was determined using a Dickey John Moisture Tester (Dickey John Co-operation, Illinois). Twenty-five seeds of each sample were drawn at random, treated with Captan 80% WP at 3g/kg of seeds and planted in moist sterilized sand contained in plastic boxes at a constant temperature of 30 $\pm$ 1 C. Seeds were germinated under illumination for eight hours a day, using flourescent lamps 750-1000 lux. Another 25 Captan-treated seeds of each sample were planted in the field with conditions favourable for germination; however, the soil temperature varied at different planting (Table 1). Emerged seedlings were counted after five and eight days of planting.

Table 1. Soil temperature at a depth of 5 cm during field germination in germination tests.

Storage period months	Germination test no.	Soil temperature ^{0} C mean	Soil temperature ^{0} C maximum
0	1	31.0 $\pm$ 1.1	38.4
3	2	30.4 $\pm$ 0.6	34.2
6	3	31.0 $\pm$ 1.2	38.0
9	4	24.7 $\pm$ 0.6	25.2

Table 2. Moisture percentage of soybean seeds stored under ambient and controlled storage.

Storage period months	Ambient		Controlled	
	range	mean	range	mean
0	10.0 – 10.8	10.3	10.0 – 10.8	10.3
3	11.2 – 11.8	11.8	8.6 – 9.0	8.9
6	10.1 – 10.3	10.2	8.0 – 8.3	8.2
9	12.1 – 12.4	12.2	8.4 – 9.0	8.6
Mean moisture % during storage		11.3		8.6
Standard deviation		1.1		0.4

Seedlings were carefully uprooted after eight days, examined and classified in accordance with the criteria established by the Association of Official Seed Analysts (1970). Reported germination values included only normal seedlings. Seedling length of five random seedlings from each sample was measured from the base of the cotyledons to the root tip and seedling dry weight was obtained gravimetrically after drying in an oven at 60 C for 48 hours.

The percentage composition of protein, carbohydrate, oil, free fatty acid and ash seed of each genotype was determined at the initiation and at the end of storage period. Official methods of American Oil Chemists' Society (1977-78) were adopted for determination.

The germination percentage was utilized as a measurement of seed viability. Germination percentage was transformed to arcsine scale for the purpose of statistical analysis. The seedling length and dry weight were used to measure the seed vigour, addition to emergence rate which was computed as follows

1/2 [(*number of seedlings emerged after 5 days of planting*) / 5
PLUS
(*number of seedlings emerged after 8 days of planting*) / 8]

This is a modification of the method used by Maquire (1962).

RESULTS AND DISCUSSION

The mean initial germination in sand at 30 C and in the field was 87.6 and 52.9% respectively (Tables 3 and 4) and the mean seed moisture content before storage was 10.3% (Table 2). The storage conditions had a drastic effect on seed viability (Tables 3 and 4). The genotypes under ambient storage exhibited a sharp decline in germination (by 24.3% after three months, 73.7% after six months and 84.4% after nine months in sand germination and by 4.2% after three months, 51.5% after six months and 52.9% after nine months in field germination). However, the drop in germination was much less under the controlled storage conditions and the germination was, respectively 82.9 and 77.7% after a storage period of nine months in sand and field germination tests.

Table 3. Mean germination percentage (P) of soybean genotypes in sand at 30C after three durations of storage under ambient and controlled conditions. Arcsine transformed values are in column A.

Storage condition	Genotype	Initial germination		Storage period (months) 3		6		9	
		P	A	P	A	P	A	P	A
Ambient	PM 78-25	90.5	72.1	80.0	63.5	16.6	24.1	16.0	24.3
	PM 78-13	88.7	70.3	81.1	64.2	44.9	42.1	0.0	4.1
	PB -1	88.3	70.1	61.2	51.5	7.4	15.8	0.0	4.1
	Bossier	86.2	68.2	36.3	37.1	0.0	4.1	0.0	4.1
	F 73-14	84.4	66.8	58.3	49.8	0.0	4.1	0.0	4.1
	Mean	87.6	69.5	63.4	53.2	13.9	18.0	3.2	5.7
Controlled	PM 78-25	90.5	72.1	93.4	75.1	95.8	75.2	93.4	75.4
	PM 87-13	88.7	70.3	95.8	75.2	81.0	64.2	83.4	66.0
	Pb-1	88.3	70.0	90.8	72.4	84.4	65.8	89.5	71.1
	Bossier	86.2	68.2	89.3	70.9	70.6	57.6	82.0	64.9
	F 73-14	84.4	66.8	81.4	64.5	56.0	48.5	66.0	54.3
	Mean	87.6	69.5	90.1	72.2	77.6	62.8	82.9	66.3
LSD for genotype within storage condition at P = 0.05			NS		11.7		11.4		12.0
LSD for storage condition at P = 0.05			NS		7.1		2.1		5.4
Interaction (genotype X storage condition)			NS		NS		NS		NS

Table 4. Mean germination percentage (P) of soybean genotypes in the field after three, six and nine months of storage under ambient and controlled conditions. Arcsine transformed values in column A.

Storage condition	Genotype	Initial germination		Storage period (months) 3		6		9	
		P	A	P	A	P	A	P	A
Ambient	PM 78-13	73.1	58.7	63.3	52.7	2.1	8.1	0.0	4.1
	PM 78-25	59.1	50.2	63.2	52.7	2.4	8.9	0.0	4.1
	Pb -1	56.1	48.5	47.0	43.3	2.4	8.9	0.0	4.1
	Bossier	40.2	38.9	27.2	31.4	0.0	4.1	0.0	4.1
	F 73-14	35.9	36.8	42.8	40.9	0.0	4.1	0.0	4.1
	Mean	52.9	46.6	48.7	44.2	1.4	6.8	0.0	4.1
Controlled	PM 78-13	73.1	58.7	78.1	62.1	63.2	52.6	81.7	64.7
	PM 78-25	59.1	50.2	87.1	69.0	84.9	67.2	75.6	60.4
	Pb-1	56.1	48.5	71.5	57.7	83.0	65.7	72.2	58.2
	Bossier	40.2	38.9	88.9	70.6	48.7	44.4	85.1	67.3
	F 73-14	35.9	36.8	81.3	64.8	48.8	44.4	73.7	59.2
	Mean	52.9	46.6	81.4	64.7	65.7	54.8	77.7	61.9
LSD for genotype within storage condition at P = 0.05			6.4		10.4		12.5		12.0
LSD for storage condition at P = 0.05			NS		4.7		5.6		5.4
Interaction (genotype X storage condition)			NS		NS		NS		NS

On average, the genotypes under controlled conditions always maintained significantly higher germination than under ambient conditions. Also the germination of the genotypes differed significantly at all periods under both storage environments. These results are in agreement with the observation made by Toole and Toole (1946), Singh and Gunasena (1979) and Arulnandhy *et al.*, (1984).

Emergence rate, seedling length and dry weight describe the seed vigour in this experiment. The mean emergence rates were 3.5 and 1.3 per day initially in sand and field tests (Table 5). The mean initial seedling length was 29.7 and 7.1 cm and seedling dry weight was 106.3 and 107.3 mg in sand and field germination, respectively (Tables 6 and 7). On the average, the vigour of the genotypes declined with storage time (Tables 5, 6 and 7). However, the loss in seed vigour was more remarkable and significantly greater under ambient than controlled storage. The genotypes showed significant differences in emergence rate, seedling length and dry weight during storage. Indeed the genotypes stored under controlled conditions maintained the initial seed vigour to a great extent throughout the storage period (Tables 5, 6 and 7) and similarly with seed viability (Tables 3 and 4).

A lower initial germination percentage and emergence rate was apparent in field than sand germination (Tables 3, 4 and 5). This effect could be attributed to the stress resulting from high soil temperature in the field during seed germination and emergence (Table 1). These temperatures were above optimal for soybean germination. The temperature for sand germination was however, maintained at 30 C, which is the approximate optimum for soybean germination. The same argument could apply for seeds stored for three months under controlled conditions. These seeds showed remarkably higher germination and emergence rate than the initial values when tested in the field (Tables 1, 4 and 5). Further more, a short time increase of viability and vigour of seed during the first three months of storage under controlled conditions can be expected due to a decline in seedborne micro-organisms and seed coat hardness during storage.

The seed moisture content increased under ambient and decreased under controlled storage conditions (Table 2). A greater change in seed moisture during storage period coinciding with a larger loss in viability and vigour of seed was noticed under ambient than controlled storage conditions (Tables 2-7). The greater loss in viability and vigour of seed

Table 5. Emergence rates (day^{-1}) of five soybean genotypes in sand and field germination tests after three durations of storage under two conditions.

Storage condition	Genotype	Sand germination				Field germination			
		Initial emergence rate	Storage period (months)			Initial emergence rate	Storage period (months)		
			3	6	9		3	6	9
Ambient	PM 78-25	3.7	3.0	0.8	0.4	1.8	2.2	0.2	0.2
	Pb-1	3.6	2.2	0.3	0.0	1.1	1.7	0.1	0.0
	PM 78-13	3.5	2.8	1.7	0.0	1.7	2.2	0.1	0.0
	F 73-14	3.4	2.3	0.0	0.0	0.9	1.4	0.0	0.0
	Bossier	3.4	1.4	0.0	0.0	0.9	0.9	0.0	0.0
	Mean	3.5	2.3	0.6	0.1	1.3	1.7	0.1	0.0
Controlled	PM 78-25	3.7	3.8	3.7	3.7	1.8	3.3	3.1	3.2
	Pb-1	3.6	3.5	3.3	3.1	1.1	2.6	3.0	3.0
	PM 78-13	3.5	3.8	3.2	3.3	1.7	3.1	2.3	2.9
	F 73-24	3.4	3.2	2.2	2.8	0.9	3.1	1.8	2.8
	Bossier	3.4	3.5	2.8	3.4	0.9	3.1	2.0	3.0
	Mean	3.5	3.5	3.1	3.3	1.3	3.1	2.4	3.0
LSD for genotype within each storage condition P = 0.05		NS	0.6	0.4	0.4	NS	0.6	0.6	0.5
LSD for storage condition P = 0.05		NS	0.3	0.3	0.2	NS	0.3	0.3	0.2
Interaction (genotype X storage condition)		NS	NS	NS	NS	NS	NS	NS	NS

Table 6. Seedling length (cm) of five soybean genotypes stored under two conditions in sand and field germination tests.

Storage condition	Genotype	Sand germination: Initial seedling length	Sand germination: Storage period (months) 3	6	9	Field germination: Initial seedling length	Field germination: Storage period (months) 3	6	9
Ambient	Pb-1	31.8	28.6	6.2	0.0	7.4	8.9	1.6	0.0
	PM 78-13	31.3	29.6	21.0	0.0	6.5	7.9	1.1	0.0
	PM 78-25	29.4	28.8	12.6	8.4	8.0	7.6	1.4	0.0
	F 73-14	28.1	27.0	0.0	0.0	6.6	7.9	0.0	0.0
	Bossier	27.9	23.3	0.0	0.0	7.0	7.5	0.0	0.0
	Mean	29.7	27.5	7.9	1.7	7.1	7.9	0.8	0.0
Controlled	Pb-1	31.8	32.7	31.3	25.9	7.4	8.6	10.6	12.9
	PM 78-13	31.3	24.5	32.8	29.4	6.5	8.7	9.6	11.5
	PM 78-25	29.4	30.0	28.4	27.5	8.0	9.0	9.1	11.4
	F 73-14	28.1	30.0	23.5	23.9	6.6	8.9	9.3	12.2
	Bossier	27.9	30.4	26.1	25.1	7.0	9.7	9.9	12.3
	Mean	29.7	30.3	28.4	26.3	7.1	9.0	9.7	12.1
LSD for genotype within each storage condition at P = 0.05		NS	3.8	5.7	3.7	NS	NS	2.6	1.6
LSD for storage condition at P = 0.05		NS	1.7	2.6	1.7	NS	0.6	1.2	0.7
Interaction (genotype X storage condition)		NS	NS	NS	NS	NS	NS	NS	NS

Table 7. Seedling dry weight (mg) of soybean genotypes stored under two conditions in sand and field germination tests.

Storage condition	Genotype	Sand germination: Initial seedling dry weight	Sand germination: Storage period (months) 3	6	9	Field germination: Initial seedling dry weight	Field germination: Storage period (months) 3	6	9
Ambient	F 73-14	141.6	146.1	0.0	0.0	135.0	145.3	0.0	0.0
	Bossier	129.2	114.8	0.0	0.0	138.0	105.2	0.0	0.0
	PM 78-13	98.4	89.0	100.3	0.0	92.0	113.6	15.1	0.0
	Pb-1	93.5	110.1	26.7	0.0	106.0	117.0	17.0	0.0
	PM 78-25	68.7	88.1	47.6	25.1	65.5	90.5	16.9	0.0
	Mean	106.3	109.6	34.9	5.0	107.3	114.3	9.8	0.0
Controlled	F 73-14	141.6	159.8	176.8	181.4	135.0	166.4	162.7	155.7
	Bossier	129.2	146.6	195.2	190.5	138.0	137.0	184.0	126.1
	PM 78-13	98.4	142.8	149.9	142.7	92.0	137.1	136.1	108.0
	Pb-1	93.5	144.9	142.4	136.0	106.0	134.7	155.4	103.0
	PM 78-25	68.7	79.3	93.1	90.9	65.5	110.7	94.0	86.3
	Mean	106.3	134.7	151.5	148.3	107.3	137.2	146.4	115.8
LSD for genotype within each storage condition at P = 0.05		14.2	32.8	35.4	24.8	32.5	31.4	35.0	11.1
LSD for storage condition at P = 0.05		NS	14.7	15.8	11.1	NS	13.0	15.7	7.4
Interaction (genotype X storage condition)		NS	NS	NS	NS	NS	NS	NS	NS

under ambient storage may also be due to high seed moisture combined with high storage temperature (maximum temperatures 26 – 36 C) which prevailed during ambient storage. These results support the conclusion of other researchers that high seed moisture and high storage temperature related to rapid seed deterioration (Justice and Bass, 1978; Tenne *et al.*, 1978; Gregg, 1982; Arulnandhy *et al.*, 1984).

Germination values were significantly and positively correlated with emergence rate, seedling length and seedling dry weight measured at different periods during storage (Table 8). The correlation coefficient observed indicate a close association of viability with vigour of soybean seed.

The percentage chemical composition of seed of five soybean genotypes at the beginning and at the end of storage under ambient and controlled conditions are presented in Table 9. Protein and ash decreased and carbohydrate, oil and free fatty acid increased in seed under both conditions of storage. However, the reduction in protein and ash and the increase in carbohydrate, oil and free fatty acid in seeds was less under controlled than ambient storage. The seeds stored under ambient conditions showed a remarkable increase in free fatty acid after nine months (Table 9) and the viability of these seeds dropped to almost zero during this period (Table 3).

Indications from these tests are that the loss in viability and vigour of soybean seed is rapid under the humid tropical conditions beyond three months of storage. The ability of controlled storage to minimize seed moisture fluctuation and maintain low seed moisture helps in preserving seed viability and vigour in soybean under these conditions. Hence, controlled storage facilities at constant temperatures and humidities, 20 C and 50% relative humidity in this case, should be considered if prolonged storage of soybean seed under the humid tropical conditions is necessary. The loss in viability and vigour of soybean seed is accompanied by a decrease in protein and ash and an increase in carbohydrate, oil and free fatty acids in seed. However, the remarkable change is noticeable in free atty acid in deteriorated soybean seeds. The data also indicate that there is genetic variability in maintenance of seed viability and vigour during storage under the humid tropical conditions, a trait that should be considered in cultivar selection and development for such areas.

Table 8. Correlation coefficient between the measurements of viability and vigour of soybean seed obtained from sand (SG) and field (FG) germination tests.

	Seedling dry weight		Seedling length		Emergence rate	
	SG	FG	SG	FG	SG	FG
Germination	0.99**	0.92**	0.98**	0.96**	0.99**	0.93**
Emergence rate	0.91**	0.84**	0.97**	0.94**		
Seedling length	0.92**	0.88**				

** Significant at $P = 0.01$

Table 9. Percentage chemical composition of seed of five soybean genotypes at the beginning and end of storage under ambient and controlled conditions.

Condition	Months in storage	Chemical composition	PM 78-25	PM 78-13	F 73-14	Pb-1	Bossier	Mean	Percent change
Before storage	0	Protein	48.8	49.0	38.5	40.3	38.5	43.0	-
		Oil	18.2	17.6	21.3	21.0	20.5	19.7	-
		Free fatty acid	0.5	0.5	1.0	0.5	0.6	0.6	-
		Carbohydrate	15.9	16.8	25.5	22.6	22.3	20.6	-
		Ash	7.4	5.1	4.5	5.8	6.9	5.9	-
Ambient storage	9	Protein	41.1	41.1	33.4	38.5	33.5	37.5	-12.1
		Oil	21.6	19.0	24.8	23.9	22.8	22.4	+13.8
		Free fatty acid	1.0	2.0	1.5	1.6	1.1	1.4	+113.3
		Carbohydrate	22.8	27.1	30.1	24.6	29.9	26.9	+30.6
		Ash	6.5	4.8	4.3	4.3	4.5	4.9	-16.9
Controlled storage	9	Protein	47.2	41.1	34.6	40.3	42.0	41.1	-4.4
		Oil	19.6	18.5	24.4	21.0	21.2	20.9	+6.3
		Free fatty acid	0.7	1.0	1.0	1.1	0.7	0.9	+50.0
		Carbohydrate	18.7	28.5	28.7	25.5	23.0	24.9	+20.9
		Ash	6.5	4.8	4.8	5.0	4.8	5.1	-13.6

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