ISSN 0972-5210



INFLUENCE OF SEED TREATMENTS AND PACKAGING MATERIALS ON SEED LONGEVITY OF ALFALFA (*MEDICAGO SATIVA* L.) CV. RL-88

P. R. Arvind Kumar*, B. C. Channakeshava and R. Siddaraju

Department of Seed Science and Technology, University of Agriculture Sciences, College of Agriculture, G.K.V.K., Bengaluru - 560 065 (Karnataka), India.

Abstract

A Laboratory experiment was conducted to study the seed longevity of alfalfa Cv. RL-88 seeds during, 2015-16 at the Department of Seed Science and Technology, U.A.S., Bengaluru (Karnataka), India. The seeds used for storage possessing the initial seed quality parameters *viz.*, seed moisture (7.40%), seed germination (92.00%), electrical conductivity (0.2841dSm⁻¹) and total dehydrogenase enzyme activity (0.568 OD value). The results of the experiment revealed that, the higher seed germination (94.50 to 82.00%), electrical conductivity (0.296 to 0.441 dSm⁻¹) and total dehydrogenase enzyme activity (1.006 to 0.883 OD value) was maintained in seeds treated with diflubenzuron @ 2 ppm (8 mg WP per kg seeds) and stored in high density polythene bag (700 guage) at initial and end of sixteen months of storage period, respectively.

Key words : Alfalfa, cloth bag, storage period, electrical conductivity.

Introduction

Alfalfa (*Medicago sativa* L.) is popularly known as lucerne and rightly called as "Queen of Forage". It is cultivated over an area of 22 million hectares in the world with an average green fodder yield of 50 - 125 t ha⁻¹ year⁻¹ with 8 - 12 cutting frequencies and seed yield ranging from 300 - 800 kg ha⁻¹(Asaadi *et al.*, 2014).

Seed production in forage crops is very scanty and there is huge demand for seeds of forage crops. In order to make avail the quality seed produced in the previous year, an effective seed storage techniques are need to be developed. Nowadays seeds storage in different packaging materials after treating with suitable chemicals to prolong their viability for the future use is gaining importance. Seed is the nucleus of life and is subjected to continuous ageing once, it has reached physiological maturity. This phenomenon results in an irreversible change in seed quality ultimately affecting viability. The quantitative deterioration during storage is mainly attributed to period of storage (Delouche *et al.*, 1973).

One of the major constraints encountered in seed production is the lack of technology to carry over the seeds until the next planting season. Of the several factors, which affect the seed quality in storage is the environment favourable for insect multiplication and other seed deterioration changes. Therefore to find out an effective method, which can control the seed deterioration changes and insect activity besides providing an environment for maintaining the viability and vigour of seeds, the studies were under taken in alfalfa to know the seed longevity after treating seeds with different chemicals and seed packaging materials under ambient condition for Bengaluru region (Zone 5: Eastern Dry Zone).

Materials and Methods

The present study is an attempt to more information on the nature of seed viability during storage under ambient conditions. The seeds were packed in cloth bag (C_1), super fine grain bag (C_2) and High Density polythene bag (700 guage) (C_3) and treated with neem oil 100 ml per kg seeds (T_2), diflubenzuron @ 2 ppm (8 mg WP per kg seeds) (T_3), thiram @ 6 g per kg seeds (T_4) and control (T_1) stored under ambient conditions for sixteen months at the Department of Seed Science and Technology, College of Agriculture, U.A.S., GKVK,

^{*}Author for correspondence : E-mail: arvindkrathod09@gmail.com

Bengaluru (Karnataka), India. At each bimonthly intervals, samples were tested for seed quality parameters *viz.*, moisture content (%), seed germination (%), electrical conductivity and total dehydrogenase enzyme activity.

The moisture content of seed was determined by the oven dry method as per the ISTA procedure (Anonymous, 2014). After drawing samples from different treatments, they were subjected to moisture determination immediately. At each stage of harvest, 10 pods were harvested and threshed to estimate the fresh and dry weight of seeds. The seeds were dried at 103°C for 17 hours and then samples were taken out from the drying chamber, cooled to room temperature and their dry weight was recorded. From the primary data, moisture percentage in the seed was computed as detailed below

Seed moisture (%) =
$$\frac{\text{dry weight of the seed}}{\text{Fresh weight of the seed}} \times 100$$

Four replications of twenty five seeds were weighed up to two decimal places. The seeds were washed thoroughly with distilled water. The surface sterilized seeds were soaked in 25 ml of distilled water and incubated for 24 hours. Then, the steeped water from the soaked seeds was collected and electrical conductivity of the leachate was measured in the digital conductivity meter model. After subtracting the EC of the distilled water from the value obtained from the seed leachate, the actual EC due (the leachate) to electrolyte was measured and expressed in dSm⁻¹ at $25 \pm 1^{\circ}$ C (Presley, 1958).

Results and Discussion

The loss of seed viability and vigour during storage is a quite natural phenomenon occurring during deterioration process. The loss of seed during storage condition may range from 0 to 100 per cent under unhygienic condition. The mechanisms causing deterioration is yet to be elucidated. Changes which occur during storage are associated with deterioration such as delayed germination, reduced seedling growth rates, decreased tolerance to adverse conditions and loss of germinability (Abdul – Baki and Anderson, 1973) are some of the physiological manifestations of seed deterioration.

Seed viability in storage is determined not only by the period of storage, but also the type of container used, initial seed moisture content, seed germination, storage environment and seed treatments. Therefore, an understanding of how best the seeds can be stored under ambient temperature, relative humidity at relatively low cost, with minimum deterioration in quality for periods extending over one or more seasons will be of immense use to seed industry and farming community.

Influence of seed treating chemicals and packaging materials and their interaction on seed quality parameters during storage

The packaging materials and the seed treating chemical influenced seed quality significantly with the advancement of storage period. Among the seed quality attributes the linear/steady increase in seed moisture content of alfalfa seeds were noticed in super grain bag and high density polythene bag (700 guage) as compared to the cloth bag (table 1). Whereas, the significantly higher fluctuation in seed moisture content noticed after fourth and sixteen months in cloth bag (7.76 to 10.77%). Whereas, the lower moisture content was recorded in super grain bag (7.55 and 8.02 %, respectively) followed by high density polythene bag (7.54 and 8.01%), respectively). This might be due to moisture fluctuation was more in cloth bag because of its moisture pervious nature compared to super grain and high density polythene bag. It may be due to the seeds stored in high density polyethylene (700 gauge) bag and treated with diflubenzuron @ 2 ppm (8 mg WP per kg seeds) perhaps helps to reduce the absorption of atmospheric moisture and chemicals during storage. The results are in agreement with the findings of Shanmugavel et al. (1995) in soybean and Reshma et al. (2009) in hedge lucerne.

The seed moisture fluctuation was minimal among the seed treatment chemicals. However, the highest moisture (8.08 and 9.36%) was recorded in control followed by neem oil 100 ml kg⁻¹ seeds (7.91 and 9.16%), diflubenzuron @ 2 ppm (8 mg WP kg⁻¹ seeds) (7.76 and 8.69%). Whereas, the lowest seed moisture content (7.74 and 8.52%) was recorded in thiram (a) 5 gm kg⁻¹ seeds, at the end of sixth and sixteenth month of storage, respectively. This may be due to when the seeds treated with chemicals, it cover the pores in the seed coat and prevents the entry of both moisture as well as insect infestation and provide protection from physical damage which can occur during handling and storage (West et al., 1985). These results are also in accordance with the findings of Maurya et al. (2002) and Shanmugavel et al. (1995) in soybean.

Among the interaction effect, the lowest seed moisture content (7.58 to 7.84%) was recorded in seeds treated with diflubenzuron @ 2 ppm (8 mg WP per kg seeds) and stored in high density polythene bag (C_3T_3), followed by in C_3T_4 : High Density polythene bag 700 guage + Thiram @ 6 g per kg seeds (7.61 to 7.86%,

Table 1: Effect of seed packaging materials and seed treating chemicals on seed moisture content of alfalfa (Medicago sativa L)
Cv. RL-88.	

Treatments	Seed moisture content (%)												
1. outmonty	Initial	2	4	6	8	10	12	14	15	16			
Packaging materials (C)													
C ₁ : Cloth bag	7.42	7.55	7.76	8.30	8.80	8.81	9.46	9.89	10.26	10.77			
C ₂ : Super fine grain bag	7.40	7.48	7.55	7.67	7.75	7.76	7.88	7.92	7.99	8.02			
$\overline{C_3}$: High Density polythene bag (700 guage)	7.41	7.47	7.54	7.65	7.73	7.73	7.85	7.90	8.00	8.01			
S.Em±	0.02	0.08	0.05	0.04	0.04	0.04	0.05	0.05	0.05	0.05			
CD (P=0.01)	NS	NS	0.21	0.14	0.15	0.14	0.18	0.20	0.19	0.19			
Chemical (T)	•												
T ₁ : Control	7.41	7.54	7.70	8.08	8.36	8.38	8.74	8.91	9.12	9.36			
T ₂ : Neem oil 100 ml/kg seeds	7.42	7.50	7.64	7.91	8.12	8.12	8.50	8.69	8.92	9.16			
$\overline{T_3}$: Diflubenzuron @ 2 ppm (8 mg WP per kg seeds)	7.40	7.47	7.55	7.76	7.96	7.96	8.16	8.38	8.54	8.69			
T_4 : Thiram @ 5 gm per kg seeds	7.40	7.48	7.57	7.74	7.94	7.94	8.19	8.29	8.42	8.53			
S.Em±	0.03	0.10	0.06	0.04	0.05	0.04	0.06	0.06	0.06	0.06			
CD (P=0.01)	NS	NS	NS	0.16	0.17	0.17	0.21	0.23	0.22	0.22			
Interactions (CXT)	•							•					
C ₁ T ₁	7.43	7.57	7.93	8.73	9.23	9.28	10.13	10.53	10.96	11.63			
C ₁ T ₂	7.43	7.56	7.85	8.44	8.94	8.94	9.74	10.19	10.64	11.32			
C ₁ T ₃	7.40	7.53	7.63	8.08	8.58	8.58	8.98	9.58	9.93	10.32			
C ₁ T ₄	7.40	7.53	7.64	7.96	8.46	8.46	9.01	9.26	9.52	9.81			
$\overline{C_2T_1}$	7.40	7.51	7.59	7.71	7.89	7.91	8.03	8.11	8.18	8.21			
$\overline{C_2T_2}$	7.40	7.49	7.56	7.69	7.75	7.75	7.93	7.95	8.03	8.05			
$\overline{C_2T_3}$	7.40	7.47	7.53	7.63	7.68	7.68	7.76	7.80	7.88	7.93			
$\overline{C_2T_4}$	7.40	7.47	7.54	7.64	7.69	7.69	7.79	7.82	7.89	7.91			
C ₃ T ₁	7.40	7.54	7.60	7.80	7.95	7.95	8.06	8.10	8.21	8.25			
C_3T_2	7.43	7.45	7.53	7.61	7.68	7.68	7.85	7.94	8.09	8.10			
$\overline{C_3T_3}$	7.39	7.42	7.50	7.58	7.63	7.63	7.73	7.77	7.83	7.84			
$\overline{C_3T_4}$	7.40	7.45	7.52	7.61	7.66	7.66	7.76	7.79	7.85	7.86			
S.Em±	0.05	0.17	0.11	0.07	0.08	0.08	0.10	0.11	0.10	0.10			
CD (P=0.01)	NS	NS	NS	0.27	0.30	0.29	0.37	0.41	0.38	0.38			
CV	0.32	1.10	0.71	0.45	0.48	0.46	0.57	0.62	0.57	0.55			

Note: Initial reading mentioned are one month after sample treatment; NS : non-significant.

respectively). Whereas, the highest seed moisture (8.73 to 11.63 %, respectively) recorded in C_1T_1 : cloth bag + no treatment as control the end of initial and sixteenth months after storage period. Perhaps helps to reduce the absorption of atmospheric moisture and chemicals during storage. The results are in agreement with the findings of Shanmugavel *et al.* (1995) in soybean.

Seed germination decline meager rate throughout the storage period (table 2). The highest germination (%) was noticed in high density polyethylene bag (700 gauge) (93.56 and 80.75%), was on par with super grain bag (93.19 and 79.69%) and progressively rapid decline noticed in cloth bag (89.25 and 68.69%) after fourth and sixteen month after storage period, respectively. The

decline in germination percentage may be attributed to ageing effect leading to depletion of food reserves and decline in synthetic activity of embryo apart from death of seed because of fungal invasion, fluctuating temperature, relative humidity and storage packaging materials in which seeds were stored. Increased accumulation of total peroxide and leakage of electrolytes was due to ageing of seeds. Germination was higher in polyethylene bag which may be attributed to low rate of respiration compared to cloth bag. These findings are in accordance with the results obtained by Hunje *et al.* (1990) in cowpea, Channakeshava *et al.* (2001) in maize, and Gupta and Aneja (2004) in soybean.

The seed treatment with diflubenzuron @ 2 ppm (8

Table 2: Effect of seed packaging materials and seed treating chemicals on seed germination of alfalfa (Medicago sativa L.) Cv	1.
RL-88.	

Treatments	Seed germination (%)												
incatinents	Initial	2	4	6	8	10	12	14	15	16			
Packaging materials (C)							•	•					
C ₁ : Cloth bag	92.81	91.88	89.25	87.00	85.19	81.63	78.31	75.50	73.44	68.69			
C ₂ : Super fine grain bag	93.94	93.44	93.19	91.69	90.38	88.63	84.63	84.00	81.25	79.69			
C_3 : High Density polythene bag (700 guage)	94.38	94.06	93.56	92.38	91.50	89.69	85.50	84.38	82.63	80.75			
S.Em±	0.98	0.85	0.61	0.62	0.90	1.45	1.19	0.72	0.79	1.06			
CD (P=0.01)	NS	NS	2.36	2.39	3.48	5.57	4.57	2.77	3.05	4.08			
Chemical (T)													
T ₁ : Control	93.25	92.58	90.42	88.67	87.08	84.67	79.08	78.58	76.00	72.92			
T ₂ : Neem oil 100 ml/kg seeds	93.75	93.17	92.33	90.00	88.67	86.25	82.00	80.33	78.67	76.00			
$\overline{T_3}$: Diflubenzuron @ 2 ppm (8 mg WP per kg seeds)	94.08	93.58	92.83	91.67	90.42	88.25	85.42	83.58	81.33	78.75			
T_4 : Thiram @ 5 gm per kg seeds	93.75	93.17	92.42	91.08	89.92	87.42	84.75	82.67	80.42	77.83			
S.Em±	1.13	0.98	0.71	0.72	1.04	1.67	1.37	0.83	0.92	1.22			
CD (P=0.01)	NS	NS	NS	NS	NS	NS	5.28	3.20	3.52	4.71			
Interactions (CXT)													
C_1T_1	92.50	91.50	87.25	84.00	81.50	77.75	73.75	70.50	67.75	61.00			
C_1T_2	92.25	91.25	89.25	85.50	83.25	80.25	77.00	74.00	72.00	68.25			
C ₁ T ₃	93.25	92.25	90.50	89.50	88.25	85.00	81.25	79.25	77.50	73.00			
	93.25	92.50	90.00	89.00	87.75	83.50	81.25	78.25	76.50	72.50			
C_2T_1	93.25	92.75	91.75	90.50	89.00	87.50	81.50	82.75	80.75	79.25			
C_2T_2	94.50	94.00	93.75	91.75	90.50	88.25	84.00	83.50	80.25	78.75			
C_2T_3	94.50	94.25	94.00	92.75	91.50	89.75	87.00	85.25	82.50	81.25			
C_2T_4	93.50	92.75	93.25	91.75	90.50	89.00	86.00	84.50	81.50	79.50			
C ₃ T ₁	94.00	93.50	92.25	91.50	90.75	88.75	82.00	82.50	79.50	78.50			
C ₃ T ₂	94.50	94.25	94.00	92.75	92.25	90.25	85.00	83.50	83.75	81.00			
C ₃ T ₃	94.50	94.25	94.00	92.75	91.50	90.00	88.00	86.25	84.00	82.00			
C ₃ T ₄	94.50	94.25	94.00	92.50	91.50	89.75	87.00	85.25	83.25	81.50			
S.Em±	1.96	1.70	1.23	1.24	1.81	2.90	2.38	1.44	1.59	2.12			
CD (P=0.01)	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS			
CV	1.04	0.91	0.67	0.69	1.02	1.67	1.43	0.89	1.00	1.39			

Note: Initial reading mentioned are one month after sample treatment.

mg WP kg⁻¹ seeds) found significant difference with respective seed germination after fourth month of storage period. The highest seed germination (92.83 to 78.75%) was recorded in diflubenzuron @ 2 ppm (8 mg WP kg⁻¹ seeds) and which was on par with thiram @ 5 gm kg⁻¹ seeds (92.42 to 77.83%). Whereas, the lowest seed germination (90.42 to 72.92%) was recorded in control at the end of fourth and sixteenth months of storage period, respectively.

The interaction of packaging materials and seed treatment chemicals found non-significant with respective seed germination (%) at the end of sixteen month of storage.

The electrical conductivity in cloth bag recorded highest (0.345 dSm⁻¹ and 0.632 dSm⁻¹) followed by super fine grain bag and lowest electrical conductivity was noticed in high density polythene bag 700 guage (0.295 dSm⁻¹ and 0.473 dSm⁻¹) recorded at the end of two and sixteenth months of storage period, respectively. Increase in electrical conductivity may be attributed to permeability of the seed membrane as seed ages, many substances such as sugars, free amino acids, organic acids etc. will leach out in the presence of water, disruption of membrane integrity and increase in free fatty acid level and free radical production by lipid peroxidation leads to leakage of electrolytes due to ageing of seed. Similar findings obtained by Verma and Gupta (1975) in soybean,

 Table 3 : Effect of seed packaging materials and seed treating chemicals on electrical conductivity of alfalfa (*Medicago sativa* L.) cv. RL-88.

Treatments			F	lectric	al cond	uctivity	y (dS ⁻¹ n	n)		
Months	Mar- 15	May- 15	Jul- 15	Sep- 15	Nov- 15	Jan- 16	Mar- 16	May- 16	Jun- 16	Jul- 16
	Initial	2	4	6	8	10	12	14	15	16
Packaging materials (C)										-
C ₁ : Cloth bag	0.319	0.345	0.357	0.378	0.392	0.483	0.516	0.535	0.552	0.632
C ₂ : Super fine grain bag	0.296	0.296	0.308	0.335	0.356	0.363	0.442	0.464	0.476	0.474
C_3 : High Density polythene bag (700 guage)	0.295	0.295	0.292	0.322	0.349	0.359	0.418	0.460	0.467	0.473
S.Em±	0.005	0.006	0.005	0.007	0.004	0.004	0.005	0.005	0.006	0.006
CD (P=0.01)	0.020	0.022	0.019	0.029	0.017	0.016	0.021	0.019	0.022	0.024
Chemical (T)				-				-		
T ₁ : Control	0.311	0.315	0.348	0.363	0.389	0.433	0.507	0.531	0.526	0.580
T ₂ : Neem oil 100 ml/kg seeds	0.310	0.313	0.317	0.348	0.392	0.388	0.481	0.496	0.492	0.521
T_3 : Diflubenzuron @ 2 ppm (8 mg WP per kg seeds)	0.296	0.310	0.314	0.320	0.340	0.386	0.405	0.453	0.481	0.499
T_4 : Thiram @ 5 gm per kg seeds	0.296	0.311	0.296	0.349	0.342	0.399	0.440	0.465	0.496	0.505
S.Em±	0.006	0.007	0.006	0.009	0.005	0.005	0.006	0.006	0.006	0.007
CD (P=0.01)	NS	NS	0.022	0.033	0.019	0.019	0.024	0.022	0.025	0.028
Interactions (CXT)										
C ₁ T ₁	0.340	0.349	0.359	0.386	0.453	0.520	0.558	0.624	0.634	0.788
C ₁ T ₂	0.340	0.348	0.360	0.403	0.475	0.488	0.548	0.531	0.543	0.652
C ₁ T ₃	0.297	0.341	0.354	0.362	0.319	0.445	0.479	0.489	0.502	0.541
C ₁ T ₄	0.297	0.341	0.354	0.361	0.319	0.476	0.478	0.497	0.528	0.548
C_2T_1	0.297	0.298	0.342	0.355	0.362	0.415	0.477	0.479	0.452	0.452
C ₂ T ₂	0.295	0.294	0.295	0.343	0.355	0.319	0.448	0.478	0.481	0.466
C ₂ T ₃	0.294	0.294	0.295	0.299	0.349	0.355	0.416	0.447	0.490	0.486
C_2T_4	0.296	0.297	0.298	0.343	0.356	0.363	0.424	0.449	0.479	0.492
C ₃ T ₁	0.295	0.297	0.342	0.349	0.350	0.363	0.487	0.489	0.490	0.501
$\overline{C_3T_2}$	0.294	0.295	0.295	0.298	0.344	0.357	0.446	0.478	0.450	0.447
$\overline{C_3T_3}$	0.296	0.294	0.294	0.298	0.350	0.356	0.320	0.423	0.449	0.441
$\overline{C_3T_4}$	0.294	0.294	0.235	0.342	0.351	0.357	0.416	0.447	0.479	0.475
S.Em±	0.010	0.011	0.010	0.015	0.009	0.009	0.011	0.010	0.011	0.012
CD (P=0.01)	0.040	0.044	0.038	0.057	0.033	0.033	0.042	0.038	0.043	0.048
CV (%)	6.86	7.30	6.16	8.60	4.74	4.27	4.78	4.10	4.51	4.72

Note: Initial reading mentioned are one month after sample treatment; NS : non-significant.

Kathiravan *et al.* (2008) in lablab and Basavegowda *et al.* (2013) in chickpea.

Electrical conductivity was recorded highest (0.348 and 0.580 dSm⁻¹) in control, followed by thiram @ 5 gm kg⁻¹ seeds (0.296 and 0.505 dSm⁻¹). Whereas, the lowest electrical conductivity (0.314 and 0.499 dSm⁻¹) was recorded in diflubenzuron @ 2 ppm (8 mg WP kg⁻¹ seeds) at the end of fourth and sixteenth month of storage period, respectively. This variation could be due to membrane degradation, damaged by insects, many substances and leakage of electrolytes over a period of time. The results

find support from the similar findings obtained by Maurya *et al.* (2002) in soybean.

The electrical conductivity was highest (0.340 to 0.788 dSm⁻¹) in control (seed treatment as and stored in cloth bag) which was on par with C_1T_2 : cloth bag + neem oil 100 ml kg⁻¹ seeds (0.340 to 0.652 dSm⁻¹). Whereas, the lowest electrical conductivity (0.296 to 0.441 dSm⁻¹) was found in C_3T_3 . High Density polythene bag 700 guage + Diflubenzuron @ 2 ppm (8 mg WP per kg seeds) at the end of initial and sixteenth months after storage period.

Table 4 : Effect of seed packaging materials and se	ed treating chemicals on tota	al dehydrogenase enzyme activity of alfalfa
(Medicago sativa L.) cv. RL-88.		

Treatments	Total dehydrogenase enzyme activity (OD Value @ A480 nm)										
Months	Mar- 15	May- 15	Jul- 15	Sep- 15	Nov- 15	Jan- 16	Mar- 16	May- 16	Jun- 16	Jul- 16	
	Initial	2	4	6	8	10	12	14	15	16	
Packaging materials (C)											
C ₁ : Cloth bag	0.961	0.921	0.926	0.868	0.832	0.729	0.721	0.698	0.685	0.662	
C ₂ : Super fine grain bag	1.033	0.988	0.993	0.935	0.898	0.876	0.813	0.785	0.746	0.730	
C_3 : High Density polythene bag (700 guage)	0.991	0.945	0.950	0.893	0.856	0.834	0.875	0.837	0.838	0.792	
S.Em±	0.013	0.017	0.011	0.009	0.009	0.009	0.010	0.010	0.010	0.011	
CD (P=0.01)	0.050	0.065	0.044	0.036	0.036	0.036	0.038	0.037	0.039	0.041	
Chemical (T)											
T ₁ : Control	0.973	0.923	0.928	0.866	0.833	0.778	0.779	0.745	0.731	0.712	
T ₂ : Neem oil 100 ml/kg seeds	0.980	0.930	0.939	0.880	0.829	0.793	0.784	0.757	0.732	0.714	
T_3 : Diflubenzuron @ 2 ppm (8 mg WP per kg seeds)	1.021	0.987	0.988	0.940	0.907	0.851	0.829	0.807	0.799	0.780	
T_4 : Thiram @ 5 gm per kg seeds	1.005	0.966	0.970	0.909	0.878	0.829	0.819	0.785	0.762	0.706	
S.Em±	0.015	0.020	0.013	0.011	0.011	0.011	0.011	0.011	0.012	0.012	
CD (P=0.01)	0.046	0.063	0.051	0.041	0.041	0.042	0.043	0.043	0.045	0.047	
Interactions (CXT)											
C ₁ T ₁	0.960	0.909	0.914	0.853	0.820	0.710	0.711	0.692	0.679	0.656	
C ₁ T ₂	0.970	0.921	0.930	0.871	0.820	0.730	0.726	0.701	0.689	0.666	
C ₁ T ₃	0.956	0.936	0.937	0.888	0.856	0.746	0.735	0.707	0.695	0.671	
C ₁ T ₄	0.957	0.919	0.922	0.862	0.831	0.728	0.711	0.691	0.678	0.655	
$\overline{C_2T_1}$	0.972	0.921	0.926	0.865	0.832	0.803	0.790	0.746	0.715	0.702	
C ₂ T ₂	0.974	0.925	0.934	0.875	0.824	0.815	0.802	0.790	0.725	0.712	
$\overline{C_2T_3}$	1.101	1.061	1.062	1.014	0.981	0.952	0.832	0.813	0.801	0.785	
$\overline{C_2T_4}$	1.084	1.043	1.048	0.987	0.956	0.934	0.829	0.791	0.742	0.722	
$\overline{C_3T_1}$	0.986	0.937	0.942	0.881	0.848	0.819	0.837	0.797	0.800	0.780	
$\overline{C_3T_2}$	0.996	0.944	0.954	0.894	0.844	0.835	0.825	0.780	0.784	0.763	
$\overline{C_3T_3}$	1.006	0.965	0.967	0.918	0.885	0.856	0.922	0.900	0.903	0.883	
C ₃ T ₄	0.975	0.935	0.939	0.878	0.847	0.825	0.917	0.872	0.866	0.743	
S.Em±	0.026	0.034	0.023	0.019	0.019	0.019	0.020	0.019	0.020	0.021	
CD (P=0.01)	0.101	0.131	0.088	0.072	0.072	0.072	0.075	0.075	0.078	0.081	
CV (%)	5.27	7.15	4.78	4.15	4.32	4.61	4.88	5.03	5.36	5.79	

Note: Initial reading mentioned are one month after sample treatment.

The total dehydrogenase enzyme activity decreased with the advancement of storage period in all packaging materials. Highest dehydrogenase enzyme activity (0.935 OD value and 0.730 OD value) was recorded in super grain bag (C_2) and lowest shoot length was noticed in cloth bag (C_1) (0.868 OD value and 0.662 OD value) recorded at the end of sixth and sixteenth month of storage period, respectively. This shows that viability of seeds decreased with increase in storage period.

The total dehydrogenase enzyme activity decreased with the advancement of storage period in all treatments.

The highest total dehydrogenase enzyme activity (1.021, 0.940 and 0.780 OD value) was recorded in diflubenzuron @ 2 ppm (8 mg WP kg⁻¹ seeds), followed by neem oil 100 ml kg⁻¹ seeds (0.980, 0.880 and 0.714 OD value) and lowest total dehydrogenase enzyme activity (0.973, 0.866 and 0.712 OD value) was recorded in control at initial, sixth and sixteenth month of storage period, respectively. These results are in accordance with the findings of Reshma *et al.* (2009) in hedge alfalfa.

The total dehydrogenase enzyme activity was found highest (1.006 and 0.801 OD value) in seeds treated

with Diflubenzuron @ 2 ppm (8 mg WP per kg seeds) and stored in high density polyethylene bag which was on par with C_2T_3 . Super fine grain bag + Diflubenzuron @ 2 ppm (8 mg WP per kg seeds) (1.101 to0.785 OD value). Whereas, the lowest total dehydrogenase enzyme activity was found in C_1T_1 cloth bag + no treatment (0.960 to 0.656 OD value) at the end of initial and sixteenth months after storage period.

Acknowledgement

The authors would like to thank Indian Grass land and Fodder Research Institute (IGFRI), Southern Regional Research Station, Dharwad for providing certified seeds of alfalfa variety RL-88 for laboratory experiments.

References

- Abdul-Baki, A. A. and J. D. Anderson (1973). Vigour determination of soybean seeds by multiple criteria. *Crop Sci.*, 13(2): 630-633.
- Anonymous (2014). International rules for seed testing by International Seed Testing Association. Seed Sci. & Tech., 27:215.
- Asaadi, M., D. Bahareh and P. Bahman (2014). Determination the best time to harvest and seed rates in order to increase the yield production of alfalfa (*Medicago sativa* L.) seed. *Intl. J. Bio-sci.*, 5(5): 100-106.
- Basavegowda, Gururaj Sunkad and Hosamani Arunkumar (2013). Effect of commercial cold storage conditions and packaging materials on seed quality of chickpea (*Cicer Arietinum* L.). *Global J. Sci. Frontier Res.*, **13(2)**: 22-28.
- Channakeshava, B. C., K. P. Ramaprasanna and B. K. Ramachandrappa (2001). Influence of seed storage containers on seed quality and storability in African tall

fodder maize (*Zea mays* L.). *Mysore Journal of Agricultural Sciences*. **35**: 236-240.

- Delouche, J. C., T. T. Rushing and C. C. Baskin (1973). Accelerated ageing technique for predicting the relative storability of crop seed lots. *Seed Sci and Technol.*, **1** : 427-452.
- Gupta, A. and R. Aneja (2004). Seed deterioration in soybean varieties during storage physiological attributes. *Seed Res.*, **32(1)**: 26-32.
- Hunje, R. V., G. N. Kulkarni, S. D. Shashidhara and B. S. Vyakaranahal (1990). Effect of insecticide and fungicide treatment on cowpea seed quality. *Seed Res.*, 18 : 90-92.
- Kathiravan, M., A. Vijayakumar and C. Vanitha (2008). Effect of dry dressing treatments and containers on seed quality parameters in lablab (*Lablab purpureous* L.) under natural ageing conditions. *Indian J. Agric. Res.*, 42(1): 62-66.
- Maurya, C. L., C. P. Poonam Singh and V. P. Kanaujia (2002). Effect of packaging material and storage period on seedling growth and vigour of soybean. *Seed Tech. News*, **32(1)**: 209.
- Presley, H. T. (1958). Relation to protoplast permeability of cotton seed viability and pre-disposition to seedling disease. *Pl. Dis. Rep.*, **42** : 52.
- Reshma, C., P. Srimathi and K. Parameshwari (2009). Influence of pre-storage treatments on storability of hedge lucerne (*Desmanthus virgatus*) cv. TNDV 1 Seeds. *Madras Agric. J.*, **96(1 & 6)** : 76-79.
- Shanmugavel, S., A. Varier, M. Dadlani, Varier Anuradha and Dadlani Malavika (1995). Physiological attributes associated with seed ageing in soybean (*Glycine max* (L.) Merrill) cultivars. *Seed Res.*, 23: 61-66.
- Verma, R. S. and P. C. Gupta (1975). Storage behaviour of soyabean varieties vastly differing in seed size. *Seed Res.*, 3:39-44.