



## Effect of seed treatments and storing period on physiological and biochemical parameters of tomato during storage

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### ABSTRACT

Achievement of higher production and productivity in any crop, good quality seed is one of the essential requirements in production and supply. Seeds undergo deterioration resulting in a decline in germination and vigour during storage. It is essential to keep the quality of seed during storage. With this view, the present investigation had been undertaken to determine the trends in seed deterioration of tomato was observed by storing with six treatments in seed testing laboratory after harvesting from field in 2020-2021 at Seed Testing Laboratory, Department of Seed Science and Technology, BCKV, Mohanpur, Nadia, West Bengal, India. Seeds were harvested from control plot for each replication. Then the properly dried seeds of tomato were primed with marigold petal extract @ 0.75% ( $T_1$ ); Neem leaf powder @ 75g kg<sup>-1</sup> seed ( $T_2$ ); Thiram @ 2g kg<sup>-1</sup> + Imidachloprid @ 7.5ml kg<sup>-1</sup> ( $T_3$ ); Thiram @ 2g kg<sup>-1</sup> + Polymer @ 7ml ( $T_4$ ) and Bromocresol solution @ 0.75% ( $T_5$ ). Control (without treated) dry seeds were considered as  $T_0$ . Treated seeds were dried properly and maintain at 6% seed moisture content. Then the seeds were kept in 700 gauge polythene packet. Different physiological and biochemical parameters were recorded just after harvesting as well as at every two months interval up to ten months of storage. Among the different seed treatments,  $T_3$  (Thiram + Imidachloprid) was the best as germination percentage, vigour index, soluble protein content of seeds, total carbohydrate content recorded highest and lowest value of electrical conductivity was recorded in  $T_3$  (Thiram + Imidachloprid) when average over durations. The interaction between treatments and durations, a change in germination behaviour was found to be noticed in seed treatments from second months after storage onwards, which showed that from second months after storage onwards  $T_3$  (Thiram + Imidachloprid) recorded maximum potential of germination and vigour at every two months interval upto ten months of storage. But at ten months after storage germination percentage dropped below Indian Minimum Seed Certification Standard for each seed treatment including control, though it dropped below 70% for control at eight months after storage. Protein and carbohydrate content remained always highest or second highest; electrical conductivity was lowest throughout the different storage periods when the seeds were treated with  $T_3$  (Thiram + Imidachloprid).

**Keywords:** Carbohydrate, protein, seed deterioration, storage, tomato, treatments, vigour

During storage, seed deterioration is a gradual and unpreventable process resulted in considerable losses. Seeds tend to loose viability and vigour during storage and information regarding storability of seed lots from harvesting to sowing in next season and also for carry over purposes (Ray and Bordolui, 2022). In storage, many physico-chemical factors like initial seed quality, moisture content of the seed, temperature, physical and chemical composition of seed, relative humidity, gaseous exchange, packaging materials and storage structure regulate the viability and vigour of seeds. As seed is hygroscopic in nature, seed quality is mainly deteriorated by variation in moisture content, relative humidity and temperature (Ray and Bordolui, 2020). Beside this, fungi and insect attacks are the main reasons for deteriorating the seed quality and reduction in germination ability. During storage, several methods have been approved such as seed treatment with suitable bio agent, chemicals, plant products and storing in safe packaging material to check the qualitative and quantitative wabtes due to several abiotic and biotic factors (Kumari *et al.*, 2017).

It has been ascertained by Gupta (2003) and Jakhar (2003), that pre-storage treatments protect the seed from microbial infestation and also enhances the storage potential of seeds. Seed quality can be maintained by using fungicide, insecticide, botanicals and seed colouring agent and the procedure of best quality tomato seed can be stored by treating them with botanicals or chemicals in ambient conditions. Bhattacharya and Basu (1990) reported that pea seed retained higher vigour and viability in ordinary storage upto nine months as compared to control through dry dressing with CaOCl<sub>2</sub> at 3 g kg<sup>-1</sup> seed. To arrest seed quality loss in storage, seed treatments with thiram and carboxin found successful. These were not only effective against a large number of pathogens (Subramanya *et al.*, 1988), but also were thought to maintain the quality of seed during storage (Shekaramurthy *et al.*, 1994; Bordolui *et al.*, 2021). Koteswar-Rao *et al.* (1962) reported that chilli seeds treated with thiram had increase germination and without treated seeds showed lowest germination at ten months after storage. Arati (2000) noted that chickpea

maintained higher germination and vigour compared to control after ten months of storage as seeds were treated with neem leaf powder. The effect of seed colouring agent on the seed quality of soybean and tomato was observed by Tonapi *et al.* (2006). Das *et al.* (2016) recommended that Bromocresol purple and Congo red can safely be used for seed colouring in paddy seeds. Hence, the present study was taken up to evaluate the response of the genotype, *i.e.*, BCT-25 towards different seed treating chemicals as well as botanicals during storage.

## MATERIALS AND METHODS

The experiment was accomplished in seed testing laboratory, Department of Seed Science and Technology, Bidhan Chandra Krishi Viswavidyalaya, West Bengal during 2020-2021 following Complete Randomized Design with three replications. The seed material for the present investigation was comprised of one tomato genotype *viz.*, BCT-25. Seeds were harvested from control plot for each replication. Then the properly dried seeds of tomato were treated with marigold petal extract @ 0.75% (T<sub>1</sub>); Neem leaf powder @ 75g kg<sup>-1</sup> seed (T<sub>2</sub>); Thiram @ 2g kg<sup>-1</sup> + Imidachloprid @ 7.5ml kg<sup>-1</sup> (T<sub>3</sub>); Thiram @ 2g kg<sup>-1</sup> + Polymer @ 7ml (T<sub>4</sub>) and artificial dye *i.e.* Bromocresol solution @ 0.75% (T<sub>5</sub>). Control (without treated) seeds were considered as T<sub>0</sub>. After treatment seeds were dried properly and maintain at 6% seed moisture content. After that seeds were kept in 700 gauge polythene packet. Different physiological parameters such as shoot length (cm), root length (cm), seedling length (cm), germination (%), vigour index, fresh weight (g) of ten seedlings and dry weight (g) of ten seedlings as well as biochemical parameters such as electrical conductivity (ds m<sup>-1</sup> g<sup>-1</sup>), soluble protein content (mg g<sup>-1</sup>) (Lowry *et al.*, 1951) and total carbohydrate content (mg g<sup>-1</sup>) (by Anthrone's method) of variously treated seeds including control were recorded just after harvesting as well as at every two months interval up to ten months of storage. Germination test was done by using germination papers through between papers (BP) method (ISTA, 1985). Vigour Index was also calculated by Germination (%) × Seedling length (cm) (Abdul Baki and Anderson, 1973).

## RESULTS AND DISCUSSION

### Root length (cm)

In different durations of storage root length was varied significantly, when average was taken over the treatments; a reduction in root length was noted as the period of storage progressed, while longest root length (9.99 cm) was noted at just after harvesting (D<sub>0</sub>) and shortest (9.23 cm) at ten months after storage (D<sub>5</sub>) (Table 1). Significant influence upon root length was imposed

by seed treatments, when mean was calculated over storage durations; maximum length was noted for T<sub>2</sub> (10.02 cm), followed by T<sub>3</sub> and T<sub>1</sub>, and it was minimum for T<sub>0</sub> (9.28 cm). When the interaction effect of storage duration and seed treatment was taken into consideration, D<sub>0</sub>T<sub>2</sub> showed highest value (10.31 cm) for the parameter, though D<sub>0</sub>T<sub>2</sub>, D<sub>1</sub>T<sub>2</sub> and D<sub>2</sub>T<sub>2</sub> were statistically *at par* with each other. Over the duration, almost similar trend of declining root length could be observed for all the treatments including control.

### Shoot length (cm)

Shoot length significant variation in storage durations, when average was made over the seed treatments; D<sub>0</sub> recorded maximum length of shoot length (4.39 cm) and D<sub>5</sub> recorded minimum (3.68 cm) for the same, displaying decreasing trend in shoot length over the period of storage (Table 2). When average was computed over the durations, seed treatments performed significantly for the character; T<sub>2</sub> (4.24 cm) showed maximum shoot length, followed by T<sub>1</sub>, T<sub>3</sub>, though T<sub>2</sub>, T<sub>1</sub> and T<sub>1</sub>, T<sub>3</sub> performed similarly and it was minimum for T<sub>0</sub> (3.80cm). Umesha *et al.* (2017) in cluster bean showed increased shoot length after seed treatment with neem leaf powder. Among the interaction, D<sub>0</sub>T<sub>1</sub> measured highest shoot length of 4.61 cm, followed by D<sub>0</sub>T<sub>2</sub>, D<sub>1</sub>T<sub>2</sub>, though D<sub>0</sub>T<sub>1</sub>, D<sub>0</sub>T<sub>2</sub>, D<sub>1</sub>T<sub>2</sub> showed non-significant difference among themselves for the trait. Over the time of storage, seedling shoot length was reduced for treated seeds as well as control.

### Seedling length (cm)

Over the treatments, significant reduction was observed in seedling length due to progression of storage period; longest seedling was noted in D<sub>0</sub> (14.38 cm) and shortest in D<sub>5</sub> (12.91 cm). Average performance of seed treatments showed significant variation for the trait; maximum length of seedling was recorded for T<sub>2</sub> (14.27 cm), followed by T<sub>3</sub> and T<sub>1</sub>, while minimum was noted for T<sub>0</sub> (13.09 cm) (Table 3). Oyekale *et al.* (2012) observed that seed treatment with neem leaf powder in sesame produced higher seedling length than other treatment including control during storage. D<sub>0</sub>T<sub>2</sub> (14.89 cm) produced longest seedling, followed by D<sub>0</sub>T<sub>1</sub>, D<sub>1</sub>T<sub>2</sub>, though these were statistically non-significant among themselves, the overall interaction effect showed significant influence for the parameter. Similar to the previous parameters mentioned, declining nature of seedling length was noticed over the storage duration.

### Germination percentage

Over seed treatments, in different storage durations germination percentage noted to be significantly varied, where highest germination (93.64%) was observed just

**Table 1: Variation in root length (cm) of seedling after seed treatment over the period of storage**

	T <sub>0</sub>	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>	T <sub>4</sub>	T <sub>5</sub>	Mean D
D <sub>0</sub>	9.60	10.21	10.31	10.11	9.97	9.75	9.99
D <sub>1</sub>	9.54	9.97	10.26	10.09	9.95	9.61	9.91
D <sub>2</sub>	9.33	9.94	10.26	10.04	9.88	9.52	9.83
D <sub>3</sub>	9.25	9.58	9.92	9.83	9.31	9.27	9.53
D <sub>4</sub>	9.23	9.45	9.74	9.66	9.17	9.25	9.42
D <sub>5</sub>	8.74	9.40	9.66	9.59	9.16	8.79	9.23
Mean T	9.28	9.76	10.02	9.89	9.57	9.37	
	<b>D</b>	<b>T</b>	<b>D X T</b>				
SEm(±)	<b>0.011</b>	<b>0.011</b>	<b>0.027</b>				
LSD (0.05)	<b>0.031</b>	<b>0.031</b>	<b>0.075</b>				

Notes: D = Duration, D<sub>0</sub> = Zero month after storage (just after harvesting), D<sub>1</sub> = Two months after storage, D<sub>2</sub> = Four months after storage, D<sub>3</sub> = Six months after storage, D<sub>4</sub> = Eight months after storage, D<sub>5</sub> = Ten months after storage, T = Treatment, T<sub>0</sub> = Control, T<sub>1</sub> = Marigold petal extract, T<sub>2</sub> = Neem leaf powder, T<sub>3</sub> = Thiram + Imidachloprid, T<sub>4</sub> = Thiram + Polymer, T<sub>5</sub> = Bromocresol solution.

**Table 2: Variation in shoot length (cm) of seedling after seed treatment over the period of storage**

	T <sub>0</sub>	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>	T <sub>4</sub>	T <sub>5</sub>	Mean D
D <sub>0</sub>	4.22	4.61	4.59	4.38	4.29	4.24	4.39
D <sub>1</sub>	4.13	4.41	4.55	4.30	4.29	4.21	4.31
D <sub>2</sub>	3.87	4.35	4.41	4.29	3.99	3.99	4.15
D <sub>3</sub>	3.70	4.02	4.41	4.27	3.78	3.82	4.00
D <sub>4</sub>	3.45	3.98	3.77	4.08	3.78	3.73	3.80
D <sub>5</sub>	3.45	3.87	3.72	3.74	3.73	3.59	3.68
Mean T	3.80	4.21	4.24	4.18	3.98	3.93	
	<b>D</b>	<b>T</b>	<b>D X T</b>				
SEm(±)	<b>0.010</b>	<b>0.010</b>	<b>0.025</b>				
LSD (0.05)	<b>0.028</b>	<b>0.028</b>	<b>0.069</b>				

Notes: D = Duration, D<sub>0</sub> = Zero month after storage (just after harvesting), D<sub>1</sub> = Two months after storage, D<sub>2</sub> = Four months after storage, D<sub>3</sub> = Six months after storage, D<sub>4</sub> = Eight months after storage, D<sub>5</sub> = Ten months after storage, T = Treatment, T<sub>0</sub> = Control, T<sub>1</sub> = Marigold petal extract, T<sub>2</sub> = Neem leaf powder, T<sub>3</sub> = Thiram + Imidachloprid, T<sub>4</sub> = Thiram + Polymer, T<sub>5</sub> = Bromocresol solution.

**Table 3: Variation in seedling length (cm) after seed treatment over the period of storage**

	T <sub>0</sub>	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>	T <sub>4</sub>	T <sub>5</sub>	Mean D
D <sub>0</sub>	13.83	14.81	14.89	14.49	14.26	13.98	14.38
D <sub>1</sub>	13.67	14.38	14.81	14.39	14.24	13.82	14.22
D <sub>2</sub>	13.20	14.29	14.67	14.33	13.88	13.51	13.98
D <sub>3</sub>	12.95	13.60	14.33	14.10	13.09	13.09	13.53
D <sub>4</sub>	12.68	13.42	13.51	13.74	12.95	12.98	13.21
D <sub>5</sub>	12.19	13.27	13.38	13.34	12.90	12.39	12.91
Mean T	13.09	13.96	14.27	14.06	13.55	13.30	
	<b>D</b>	<b>T</b>	<b>D X T</b>				
SEm(±)	<b>0.015</b>	<b>0.015</b>	<b>0.037</b>				
LSD (0.05)	<b>0.042</b>	<b>0.042</b>	<b>0.104</b>				

Notes: D = Duration, D<sub>0</sub> = Zero month after storage (just after harvesting), D<sub>1</sub> = Two months after storage, D<sub>2</sub> = Four months after storage, D<sub>3</sub> = Six months after storage, D<sub>4</sub> = Eight months after storage, D<sub>5</sub> = Ten months after storage, T = Treatment, T<sub>0</sub> = Control, T<sub>1</sub> = Marigold petal extract, T<sub>2</sub> = Neem leaf powder, T<sub>3</sub> = Thiram + Imidachloprid, T<sub>4</sub> = Thiram + Polymer, T<sub>5</sub> = Bromocresol solution.

**Table 4: Variation in germination percentage (value within the bracket) after seed treatment over the period of storage**

	T <sub>0</sub>	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>	T <sub>4</sub>	T <sub>5</sub>	Mean D
D <sub>0</sub>	74.48 (92.87)	76.90 (94.88)	76.29 (94.41)	75.48 (93.74)	74.67 (93.04)	74.51 (92.89)	75.39 (93.64)
D <sub>1</sub>	67.26 (85.09)	69.44 (87.70)	69.45 (87.71)	71.88 (90.35)	71.01 (89.44)	68.90 (87.08)	69.66 (87.89)
D <sub>2</sub>	63.27 (79.80)	66.39 (83.99)	66.85 (84.58)	68.22 (86.26)	67.33 (85.18)	65.56 (82.92)	66.27 (83.79)
D <sub>3</sub>	59.98 (75.01)	63.41 (80.01)	64.02 (80.84)	65.18 (82.42)	64.66 (81.72)	62.34 (78.48)	63.27 (79.75)
D <sub>4</sub>	56.11 (68.94)	59.87 (74.83)	60.41 (75.65)	61.42 (77.14)	60.83 (76.29)	59.01 (73.52)	59.61 (74.40)
D <sub>5</sub>	53.61 (64.83)	54.78 (66.77)	54.83 (66.85)	56.34 (69.31)	55.62 (68.15)	54.40 (66.15)	54.93 (67.01)
Mean T	62.45 (77.76)	65.13 (81.36)	65.31 (81.68)	66.42 (83.20)	65.69 (82.30)	64.12 (80.17)	
	<b>D</b>	<b>T</b>	<b>D X T</b>				
SEm(±)	<b>0.047</b>	<b>0.047</b>	<b>0.116</b>				
LSD (0.05)	<b>0.134</b>	<b>0.134</b>	<b>0.328</b>				

Notes: D = Duration, D<sub>0</sub> = Zero month after storage (just after harvesting), D<sub>1</sub> = Two months after storage, D<sub>2</sub> = Four months after storage, D<sub>3</sub> = Six months after storage, D<sub>4</sub> = Eight months after storage, D<sub>5</sub> = Ten months after storage, T = Treatment, T<sub>0</sub> = Control, T<sub>1</sub> = Marigold petal extract, T<sub>2</sub> = Neem leaf powder, T<sub>3</sub> = Thiram + Imidachloprid, T<sub>4</sub> = Thiram + Polymer, T<sub>5</sub> = Bromocresol solution.

**Table 5: Variation in vigour index after seed treatment over the period of storage**

	T <sub>0</sub>	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>	T <sub>4</sub>	T <sub>5</sub>	Mean D
D <sub>0</sub>	1,284.04	1,405.54	1,406.08	1,357.93	1,326.75	1,298.91	1,346.54
D <sub>1</sub>	1,163.42	1,261.08	1,299.33	1,299.88	1,273.33	1,203.69	1,250.12
D <sub>2</sub>	1,053.40	1,200.17	1,241.12	1,236.15	1,181.97	1,120.48	1,172.22
D <sub>3</sub>	971.13	1,087.82	1,158.75	1,162.35	1,069.67	1,027.30	1,079.51
D <sub>4</sub>	874.12	1,004.51	1,021.82	1,059.69	987.66	954.29	983.68
D <sub>5</sub>	790.06	885.86	894.45	924.36	878.91	819.34	865.50
Mean T	1,022.69	1,140.83	1,170.26	1,173.40	1,119.71	1,070.67	
	<b>D</b>	<b>T</b>	<b>D X T</b>				
SEm(±)	<b>0.632</b>	<b>0.632</b>	<b>1.549</b>				
LSD (0.05)	<b>1.787</b>	<b>1.787</b>	<b>4.376</b>				

Notes: D = Duration, D<sub>0</sub> = Zero month after storage (just after harvesting), D<sub>1</sub> = Two months after storage, D<sub>2</sub> = Four months after storage, D<sub>3</sub> = Six months after storage, D<sub>4</sub> = Eight months after storage, D<sub>5</sub> = Ten months after storage, T = Treatment, T<sub>0</sub> = Control, T<sub>1</sub> = Marigold petal extract, T<sub>2</sub> = Neem leaf powder, T<sub>3</sub> = Thiram + Imidachloprid, T<sub>4</sub> = Thiram + Polymer, T<sub>5</sub> = Bromocresol solution.

after harvesting and it was decreased with the advancement in storage period, at D<sub>5</sub> germination percentage was observed to fall below 70%, i.e., minimum germination percentage for tomato seeds as prescribed by Indian Minimum Seed Certification Standards (IMSCS), 2013. Germination decreases due to ageing period, as noticed by Mandal and Basu (1986) in wheat, Dharmalingam (1995) in maize, Umesha *et al.* (2017)

in cluster bean due to natural aging. During storage, seed quality loosed due to the injury in cell membrane and chemical changes occurred inside the seed such as nucleic acid and protein accumulation (Roberts, 1972). Such degenerative changes resulted in complete disorientation of cell membranes and organelles; ultimately led to loss of viability and the death of the seed. Among seed treatments, maximum potential of germination

**Table 6: Variation in fresh weight (g) of seedlings after seed treatment over the period of storage**

	T <sub>0</sub>	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>	T <sub>4</sub>	T <sub>5</sub>	Mean D
D <sub>0</sub>	0.159	0.176	0.178	0.175	0.169	0.160	0.170
D <sub>1</sub>	0.145	0.170	0.177	0.172	0.166	0.160	0.165
D <sub>2</sub>	0.144	0.167	0.174	0.168	0.163	0.154	0.162
D <sub>3</sub>	0.143	0.160	0.171	0.167	0.155	0.150	0.157
D <sub>4</sub>	0.142	0.158	0.164	0.165	0.151	0.147	0.155
D <sub>5</sub>	0.140	0.157	0.161	0.157	0.150	0.145	0.152
Mean T	0.146	0.165	0.171	0.167	0.159	0.153	
	<b>D</b>	<b>T</b>	<b>D X T</b>				
SEm(±)	<b>0.004</b>	<b>0.004</b>	<b>0.009</b>				
LSD (0.05)	<b>0.011</b>	<b>0.011</b>	NS				

Notes: D = Duration, D<sub>0</sub> = Zero month after storage (just after harvesting), D<sub>1</sub> = Two months after storage, D<sub>2</sub> = Four months after storage, D<sub>3</sub> = Six months after storage, D<sub>4</sub> = Eight months after storage, D<sub>5</sub> = Ten months after storage, T = Treatment, T<sub>0</sub> = Control, T<sub>1</sub> = Marigold petal extract, T<sub>2</sub> = Neem leaf powder, T<sub>3</sub> = Thiram + Imidachloprid, T<sub>4</sub> = Thiram + Polymer, T<sub>5</sub> = Bromocresol solution.

**Table 7: Variation in dry weight (g) of seedlings after seed treatment over the period of storage**

	T <sub>0</sub>	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>	T <sub>4</sub>	T <sub>5</sub>	Mean D
D <sub>0</sub>	0.017	0.020	0.021	0.020	0.019	0.017	0.019
D <sub>1</sub>	0.017	0.019	0.019	0.019	0.018	0.017	0.018
D <sub>2</sub>	0.017	0.019	0.019	0.019	0.018	0.017	0.018
D <sub>3</sub>	0.016	0.017	0.019	0.018	0.017	0.016	0.017
D <sub>4</sub>	0.016	0.017	0.018	0.018	0.017	0.016	0.017
D <sub>5</sub>	0.015	0.017	0.018	0.018	0.017	0.017	0.017
Mean T	0.016	0.018	0.019	0.019	0.018	0.017	
	<b>D</b>	<b>T</b>	<b>D X T</b>				
SEm(±)	<b>0.0004</b>	<b>0.0004</b>	<b>0.001</b>				
LSD (0.05)	<b>0.001</b>	<b>0.001</b>	NS				

Notes: D = Duration, D<sub>0</sub> = Zero month after storage (just after harvesting), D<sub>1</sub> = Two months after storage, D<sub>2</sub> = Four months after storage, D<sub>3</sub> = Six months after storage, D<sub>4</sub> = Eight months after storage, D<sub>5</sub> = Ten months after storage, T = Treatment, T<sub>0</sub> = Control, T<sub>1</sub> = Marigold petal extract, T<sub>2</sub> = Neem leaf powder, T<sub>3</sub> = Thiram + Imidachloprid, T<sub>4</sub> = Thiram + Polymer, T<sub>5</sub> = Bromocresol solution.

**Table 8: Variation in electrical conductivity (dS.m<sup>-1</sup>) of seed leachates after seed treatment over the period of storage**

	T <sub>0</sub>	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>	T <sub>4</sub>	T <sub>5</sub>	Mean D
D <sub>0</sub>	0.113	0.111	0.112	0.112	0.111	0.111	0.112
D <sub>1</sub>	0.193	0.183	0.173	0.143	0.151	0.187	0.172
D <sub>2</sub>	0.350	0.324	0.317	0.288	0.293	0.337	0.318
D <sub>3</sub>	0.526	0.495	0.486	0.457	0.462	0.512	0.490
D <sub>4</sub>	0.812	0.783	0.773	0.742	0.754	0.799	0.777
D <sub>5</sub>	1.114	1.070	1.053	1.024	1.039	1.095	1.066
Mean T	0.518	0.494	0.486	0.461	0.469	0.507	
	<b>D</b>	<b>T</b>	<b>D X T</b>				
SEm(±)	<b>0.0003</b>	<b>0.0003</b>	<b>0.001</b>				
LSD (0.05)	<b>0.001</b>	<b>0.001</b>	<b>0.003</b>				

Notes: D = Duration, D<sub>0</sub> = Zero month after storage (just after harvesting), D<sub>1</sub> = Two months after storage, D<sub>2</sub> = Four months after storage, D<sub>3</sub> = Six months after storage, D<sub>4</sub> = Eight months after storage, D<sub>5</sub> = Ten months after storage, T = Treatment, T<sub>0</sub> = Control, T<sub>1</sub> = Marigold petal extract, T<sub>2</sub> = Neem leaf powder, T<sub>3</sub> = Thiram + Imidachloprid, T<sub>4</sub> = Thiram + Polymer, T<sub>5</sub> = Bromocresol solution.

**Table 9: Variation in soluble protein content (mg.g<sup>-1</sup>) of seed after seed treatment over the period of storage**

	T <sub>0</sub>	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>	T <sub>4</sub>	T <sub>5</sub>	Mean D
D <sub>0</sub>	2.829	2.834	2.833	2.832	2.833	2.831	2.832
D <sub>1</sub>	2.137	2.373	2.473	2.762	2.632	2.242	2.437
D <sub>2</sub>	1.788	2.116	2.390	2.612	2.546	1.992	2.241
D <sub>3</sub>	1.586	1.982	2.145	2.437	2.340	1.787	2.046
D <sub>4</sub>	1.451	1.732	1.963	2.215	2.195	1.505	1.843
D <sub>5</sub>	1.425	1.592	1.722	2.048	1.823	1.496	1.684
Mean T	1.869	2.105	2.254	2.484	2.395	1.975	
	<b>D</b>	<b>T</b>	<b>D X T</b>				
SEm(±)	<b>0.002</b>	<b>0.002</b>	<b>0.006</b>				
LSD (0.05)	<b>0.007</b>	<b>0.007</b>	<b>0.017</b>				

Notes: D = Duration, D<sub>0</sub> = Zero month after storage (just after harvesting), D<sub>1</sub> = Two months after storage, D<sub>2</sub> = Four months after storage, D<sub>3</sub> = Six months after storage, D<sub>4</sub> = Eight months after storage, D<sub>5</sub> = Ten months after storage, T = Treatment, T<sub>0</sub> = Control, T<sub>1</sub> = Marigold petal extract, T<sub>2</sub> = Neem leaf powder, T<sub>3</sub> = Thiram + Imidachloprid, T<sub>4</sub> = Thiram + Polymer, T<sub>5</sub> = Bromocresol solution.

**Table 10: Variation in total carbohydrate content (mg.g<sup>-1</sup>) of seed after seed treatment over the period of storage**

	T <sub>0</sub>	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>	T <sub>4</sub>	T <sub>5</sub>	Mean D
D <sub>0</sub>	2.872	2.883	2.880	2.883	2.876	2.872	2.878
D <sub>1</sub>	2.923	3.080	3.133	3.233	3.217	2.980	3.094
D <sub>2</sub>	3.077	3.223	3.307	3.423	3.390	3.180	3.267
D <sub>3</sub>	3.343	3.430	3.477	3.643	3.513	3.390	3.466
D <sub>4</sub>	3.443	3.650	3.707	3.777	3.747	3.580	3.651
D <sub>5</sub>	3.797	3.873	3.917	3.967	3.953	3.830	3.889
Mean T	3.243	3.357	3.403	3.488	3.449	3.305	
	<b>D</b>	<b>T</b>	<b>D X T</b>				
SEm(±)	<b>0.003</b>	<b>0.003</b>	<b>0.008</b>				
LSD (0.05)	<b>0.010</b>	<b>0.010</b>	<b>0.024</b>				

Notes: D = Duration, D<sub>0</sub> = Zero month after storage (just after harvesting), D<sub>1</sub> = Two months after storage, D<sub>2</sub> = Four months after storage, D<sub>3</sub> = Six months after storage, D<sub>4</sub> = Eight months after storage, D<sub>5</sub> = Ten months after storage, T = Treatment, T<sub>0</sub> = Control, T<sub>1</sub> = Marigold petal extract, T<sub>2</sub> = Neem leaf powder, T<sub>3</sub> = Thiram + Imidachloprid, T<sub>4</sub> = Thiram + Polymer, T<sub>5</sub> = Bromocresol solution.

(83.20%) was recorded for T<sub>3</sub>, followed by T<sub>4</sub>, T<sub>2</sub>, T<sub>1</sub> and minimum (77.76%) was observed in T<sub>0</sub> with significant variation, when average was made over storage durations (Table 4). Asokan *et al.* (1980) in rice seeds reported that seeds treated with fungicide and insecticide offered good protection while untreated seeds were invaded by storage fungi and recorded low germination. Seed treatment with chemicals has been reported to be very fruitful in maintaining seed quality as it controls the activities of fungi and storage pests (Gupta *et al.*, 1989). Dhyani *et al.* (1991) revealed that the chilli seed treated with thiram, captafol, topsin, aureofungin and vitavax increase germination and seedling length. When storage durations and seed treatments were interacted with each other, the highest germination (94.88%) was noticed in at D<sub>0</sub>T<sub>1</sub> and lowest

(64.83%) was observed for D<sub>5</sub>T<sub>0</sub>, for each seed treatment including control a similar trend of reducing germination percentage over the durations was noted. Among the interaction effects, a change in germination behaviour was found to be noticed in seed treatments from D<sub>1</sub> onwards, which showed that from D<sub>1</sub> onwards T<sub>3</sub> recorded maximum potential of germination at every two months interval upto ten months of storage and at D<sub>5</sub> germination percentage fell below IMSCS for each seed treatment including control, though it dropped below 70% for T<sub>0</sub> at D<sub>4</sub>. Raikar *et al.* (2011) reported that Intrigated Nutrient Management plot seeds treated with combination of thiram and malathion stored in polythene bag maintained germination and vigour upto twenty months of storage period under ambient conditions.

### Vigour index

Significant influence was observed in vigour index at different storing period, when average was recorded over seed treatments; maximum vigour index (1346.54) was observed at  $D_0$  and it was noted to be declined over the time of storage (Table 5). Vigour index was gradually decreased due to reduce rate of germination and production of poor seedling over the period of storability. Among the treatments over durations significant performance was noticed;  $T_3$  recorded highest vigour index of 1173.40, followed by  $T_2$ ,  $T_1$ , though  $T_3$  and  $T_2$  remain statistically at par with each other for recorded vigour index and it was lowest for  $T_0$  (1022.69). Kaddi *et al.* (2013) also confirmed that Thiram in combination with Imidachloprid was found to maintain significantly higher first count, germination (%), vigour index I and vigour index II as compared to control after six months of storage in cotton seeds. Significant variation was observed among the interaction effects for the trait, where highest magnitude of vigour index was noted in  $D_0T_1$  (1405.54), though  $D_0T_1$  and  $D_0T_2$  were statistically non-significant and lowest was calculated in  $D_5T_0$  (790.06). The change noted in performance of seed treatments after influenced by storage durations for the character was almost similar to germination percentage onwards and as the time of storage progressed, vigour index decreased. Thus seed treatments have a key role in securing the seeds during storage (Chen and Burris, 1993).

### Fresh weight of ten seedlings (g)

In different storage durations fresh weight of seedlings varied significantly, when mean was calculated over the seed treatments; highest fresh weight (0.170 g) was recorded just after harvesting, though  $D_0$ ,  $D_1$ ,  $D_2$  exhibited non-significant difference for the trait and over the period of storage, minute reduction in fresh weight was noted (Table 6). Significant performance was observed among the seed treatments;  $T_2$  was noted to produce maximum fresh weight (0.171 g), followed by  $T_3$ ,  $T_1$ , though non-significant difference was noted amongst them, when average was made over storage durations. Seed treatments after interacted with period of storage showed non-significant variation for the trait, although slight decline in seedling fresh weight was noticed with the progression of storage time. Longden (1976) noted highest seedling fresh weight in sugar beet after treated with thiram fungicide.

### Dry weight of ten seedlings (g)

Though in storage durations for the dry weight showed significant variation, but non-significant difference was observed among themselves, average over seed treatments; maximum dry weight of seedlings was

recorded at  $D_0$ , i.e., 0.019 g, though  $D_0$ ,  $D_1$ ,  $D_2$  gave statistically similar results. Over storage durations, seed treatments significantly influenced the dry weight of seedlings; both  $T_2$  and  $T_3$  produced highest seedling dry weight of 0.019 g, but non-significant difference was observed among various seed treatments (Table 7). Non-significant variation was observed among the interaction effect with slight reduction in seedling dry weight for every seed treatment over the period of storage. Sultana *et al.* (2015) noted minute rate of increase in seedling dry weight in okra while treated with provax and neem leaf powder during storage.

### Electrical conductivity of seed leachates ( $\text{dS m}^{-1} \text{g}^{-1}$ )

Electrical conductivity of seeds varied significantly in different durations of storage, when average was taken over the treatments; an increase in electrical conductivity was noted as the period of storage progressed, while lowest conductivity ( $0.112 \text{ dS m}^{-1} \text{g}^{-1}$ ) was noted at just after harvesting ( $D_0$ ) and highest ( $1.066 \text{ dS m}^{-1} \text{g}^{-1}$ ) at ten months after storage ( $D_5$ ) (Table 8). Significant influence upon electrical conductivity was imposed by seed treatments, when mean was calculated over storage durations; maximum conductivity was noted for  $T_0$  ( $0.518 \text{ dS m}^{-1} \text{g}^{-1}$ ), followed by  $T_5$  and  $T_1$ , and it was minimum for  $T_3$  ( $0.461 \text{ dS m}^{-1} \text{g}^{-1}$ ). The lower electrical conductivity was recorded in treated seeds over untreated seeds which may be due to fungicide and insecticide that protects the seeds from storage pathogens and insects, thus reduces the seed infection, cracks and aberrations of the seed coat and also the leaching of electrolytes. This result is in agreement with Maheshbabu and Hunje (2008) in soybean and Gowda *et al.* (2018) in chickpea. When the interaction effect of storage duration and seed treatment was taken into consideration,  $D_5T_0$  showed highest value ( $1.114 \text{ dS m}^{-1} \text{g}^{-1}$ ) for the parameter, whereas lowest ( $0.111 \text{ dS m}^{-1} \text{g}^{-1}$ ) was determined for  $D_0T_1$ ,  $D_0T_4$  and  $D_0T_5$ , though  $D_0T_1$ ,  $D_0T_4$ ,  $D_0T_5$ ,  $D_0T_2$ ,  $D_0T_3$  and  $D_0T_0$  were statistically at par with each other. Over the duration, almost similar trend of increasing electrical conductivity of seeds could be observed for all the treatments including control. Electrical conductivity of seed leachate was gradually increase with the increasing of storage period in all the seed treatments including control, which indicates increased membrane permeability and decrease in integrity of seed coat and thus cellular membrane deteriorated. During storage, loss of membrane integrity would be the prime reason for increased electrical conductivity and also supported by structural change and changes in membrane composition (Delouche and Baskin, 1973). Electrical conductivity is negatively correlated with seed quality in respect of seed germination and vigour index. Many workers have

reported negative correlation between electrical conductivity and seed quality (Kathiravan *et al.*, 2008 in lablab; Tejashwi *et al.*, 2014 in marigold).

#### Soluble protein content of seed (mg g<sup>-1</sup>)

In different durations of storage soluble protein content of seed varied significantly, when mean was made over the seed treatments; maximum soluble protein content (2.832 mg g<sup>-1</sup>) was noted at D<sub>0</sub> and a slight reduction in it was noted with the advancement in storage (Table 9). Maldonado *et al.* (2015) noted decrease in soluble protein content in sugar apple seeds as the age of seed progressed. The protein content reduced due to various factors like, increased rate of protein loss in embryonic axes and cotyledons. During seed deterioration, the damage was occurring to the protein synthesizing system and the synthesis or activation of large quantities of proteolytic enzymes (Bewley and Black, 1994). The huge loss of cell protein was observed in rice seed during storage (Prabhakar and Mukherjee, 1980); increases in protease activity were recorded in the cotyledons and embryonic axis in *Shorea robusta* seeds (Chaitanya *et al.*, 2000). Among the seed treatments, T<sub>3</sub> (2.484 mg g<sup>-1</sup>) exhibited highest value for soluble protein content of seed followed by T<sub>4</sub>, T<sub>2</sub>, while lowest was noted for T<sub>0</sub> (1.869 mg g<sup>-1</sup>) and significant variation was recorded by seed treatments, average over storage durations. Thiram is a sulphur fungicide and study indicated that sulfate resupply followed by S-deprivation rises nitrate assimilation for protein synthesis (Zhang *et al.*, 2015); sulphur application improved sesame seed yield, oil, and protein contents (Raza *et al.* 2018a, 2018b). On the other hand, imidachloprid is an insecticide, belongs to neonicotinoids family; Preetha and Stanley (2012) reported that imidachloprid and other neonicotinoid insecticides under their study revealed improve in the soluble protein content of okra and cotton. Seed treatments after influenced by durations of storage showed significant variation for the trait, where D<sub>0</sub>T<sub>1</sub> with 2.834 mg g<sup>-1</sup> soluble protein content exhibited highest value, though there was no significant difference among the seed treatments at just after harvesting and from D<sub>1</sub> onwards significant difference was observed among the interaction effects for this parameter, while D<sub>5</sub>T<sub>0</sub> (1.425 mg g<sup>-1</sup>) showed lowest content of it. Moreover, a similar trend of reduction in soluble seed protein content was observed for every seed treatment as the period of storage progressed.

#### Total carbohydrate content of seed (mg g<sup>-1</sup>)

Significant variation was noted for total carbohydrate content of seed among storage durations, when average was made over seed treatments. Over the period of storage, increase in carbohydrate content was noted with

lowest value for D<sub>0</sub> (2.878 mg g<sup>-1</sup>) and highest for D<sub>5</sub> (3.889 mg g<sup>-1</sup>) (Table 10). Maldonado *et al.* (2015) reported 72.3% rise in the sucrose content and 61% rise in the fructose content in sugar apple seeds over the period of storability under ambient condition. Soluble carbohydrates play an important role in desiccation tolerance during storage of seed; increase in sucrose levels, particularly in the contents of sugars of the oligosaccharide raffinose family, have been correlated with desiccation tolerance and longevity in orthodox seeds (Horbowicz and Obendorf, 1994; Obendorf, 1997). Significant variation was noted by seed treatments, when mean was determined over the storage durations; T<sub>3</sub> (3.488 mg g<sup>-1</sup>) exhibited highest carbohydrate content, followed by T<sub>4</sub>, T<sub>2</sub> and lowest was recorded for T<sub>0</sub> (3.243 mg g<sup>-1</sup>). Thiram is a sulphur containing fungicide and it has been confirmed by Haneklaus *et al.* (2007), Carciochi *et al.* (2020) and that sulphur application supplies regarding better nutrients assimilation and in the crop plants carbohydrate synthesis due to synergistic effects with other nutrients such as nitrogen, phosphorus and potassium. Sajjan *et al.* (2009) revealed that seeds treated with imidachloprid 600 FS @ 10 ml kg<sup>-1</sup> seeds in sunflower could be stored in more than 700 gauges polythene packet maintain seed quality upto eight months of storage without significant loss. Significant variation was observed among the interaction effect, where maximum carbohydrate content (3.967 mg g<sup>-1</sup>) was measured in D<sub>5</sub>T<sub>3</sub>, followed by D<sub>5</sub>T<sub>4</sub>, though D<sub>5</sub>T<sub>3</sub> and D<sub>5</sub>T<sub>4</sub> were statistically at par with each other, whereas minimum carbohydrate content (2.872 mg g<sup>-1</sup>) was measured in D<sub>0</sub>T<sub>0</sub> as well as D<sub>0</sub>T<sub>5</sub>, though statistically similar values were recorded by D<sub>0</sub>T<sub>0</sub>, D<sub>0</sub>T<sub>5</sub>, D<sub>0</sub>T<sub>4</sub>, D<sub>0</sub>T<sub>2</sub>, D<sub>0</sub>T<sub>1</sub> and D<sub>0</sub>T<sub>3</sub>. Similar to storage durations, here also increase in total carbohydrate content was noticed for seed treatments as the period of storability forwarded.

#### CONCLUSION

T<sub>3</sub> (Thiram + Imidachloprid) was proved to be the best as its performance of germination and vigour was highest; protein and carbohydrate content remained always highest or second highest; electrical conductivity was lowest throughout the different storage periods.

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