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"Seed storage behavior of *Embelia tsjeriam-cottam* (An important medicinal plant of India)"

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Abstract
Embelia tsjeriam-cottam Burm.f. has great medicinal properties due to that they are used in several medicines to cure different types of infections and diseases. No information is available on seed storage behavior of E. tsjeriam-cottam, which is an essential requirement for storing seeds in seed banks for germplasm conservation. Fan dried seeds shown seed germination 88.37% after three month, while up to 59% after 12-month duration with 5 °C temperature. Fan dried seeds shown significant effect of stored at 30 °C, reduced germination 44% after 9 month and 34% after 12 month. Where as one-day sun dried seeds and three-day sun dried seeds stored at 5 °C temperature lost seed germination after 12 month up to 56.5% and 51.25% respectively. Seeds stored at 30 °C decrease seed quality very fast. One day and Three day sun dried seeds losses maximum seed quality and seed germination during storage, after 12 month at 30 °C temperature where; seed germination percent were count minimum, 30% and 23.75% respectively.

Keywords: Seed, genetic resource, medicinal plants, seed storage behavior, conservation, orthodox, recalcitrant

Introduction
Embelia tsjeriam-cottam Burm.f. has immense medicinal potential. Embelin (2,5-dihydroxy-3-undecyl-2,5-cyclohexadiene-1,4-benzoquinone) is a phenolic compound found in the fruits of *Embelia tsjeriam-cottam* and it is responsible for the medicinal properties of the plant Thus the fruits are harvested at a large scale before maturity leading to depletion of population. *Embelia* dried berries are also used to treat worms infection especially tape worms. They are acrid, astringent and carminative and hence recommended for constipation, colic, dyspepsia, flatulence and piles. A paste of the seed is applied locally against ringworm and other coetaneous infections (Sivarajan & Indira 1994) [13]. Berries are rich in quinines *viz*. embelin, rapanone, homoembelin, himorapanone and vilangin (Khare 2004) [8]. System of medicine, the plant is used as an anti-fever remedy, a laxative, an anthelmintic as well as in migraine treatments. The roots of the plant are used to treat lung diseases in cattle and for the treatment of diabetic carbuncles and headaches (Gaur 1999) [6].
Seeds are fundamental to agriculture and natural ecosystem. Fundamental knowledge about mechanisms pertaining to seed development, germinability, dormancy and storability is required for improving seed performance. Information on seed longevity, desiccation, and freezing sensitivity is a prerequisite for conserving plant species that generally produce non-orthodox seeds. Seed longevity and storage behavior of many wild and semi-domesticated species from tropical regions has been documented (Hong and Ellis 1996) [7]. Based on seed storage behavior, three categories of seeds viz., orthodox, recalcitrant, and intermediate or orthodox with limited desiccation ability are recognized and widely used (Hong and Ellis 1996; Schmidt 2000) [7, 12]. Seed survival in desiccation tolerant seeds can be quantified by seed viability equations, as seed viability depends upon chemical composition of seed, moisture content and storage temperature (Roberts 1973, Ellis and Roberts 1980, Ellis *et al*. 1988, Pritchard and Dickie 2004, Hong *et al*. 2005) [11, 3, 4, 10, 5]. Orthodox seeds retain viability for several decades in gene bank condition, as 90-95% of their original water is lost during seed maturation.
No information is available on seed storage behavior of *E. tsjeriam-cottam*, which is essential for storing the seeds in seed bank for germplasm conservation. The seeds with varying (targeted) moisture content were stored in three different storage temperatures and seed germinability and seed viability was tested at three months interval up to 12 months. Keeping in view the above studies was carried out to investigate the one year seed storage behavior of medicinal plant species *Embelia tsjeriam-cottam*.

Material and method

The present work deals with storage behavior of important species *Embelia tsjeriam-cottam*. The species of *Embelia tsjeriam-cottam* collected from Lakhamandal-Goraghati valley, situated at Dehradun District of Uttarakhand, India. *Embelia tsjeriam-cottam* has one seeded fruit. The ripened pulpy fruits collected in the month between Novembers to December. Seeds were extracted from fruits by using fermentation process and dried in shadow places for pre drying.

Prior to storage, seed samples were dried in deferent ways. Seed moisture content of dried seed sample determined by air oven method. Three types of storage temperature (5±1, 20±1, and 30±1 ºC) were sat for seed storage. Seed samples were placed in moisture proof container with airtight lid. Seed storage effects by seed germination test observed in three-month interval up to 12 months. Based on drying three types of seed sample was derived. 24-hour fan dried seeds reduced their moisture content up 17.59%, while one day sun dried and three day sun dried seeds reduced their moisture content up to 16.07% and 15.22% respectively.

Table 1: Seed moisture content of *E. tsjeriam-cottam*

Fresh seed	24 hour fan air dried	One day sun dried	Three day sun dried
36.92	17.59	16.90	15.22

Results

Seed storage of *E. tsjeriam-cottam*

Three types dried seed sample i.e. 24 hour fan dried, one day sun dried and three day sun dried seeds of baybidang stored at three temperatures (5, 20, and 30 °C) for one-year duratution determined the seed storability, type of seeds and influences of factors which affects seed longevity during storage. Analysis of variance has given in Table.1 Mean squares of the seeds storage of baybidang show effects of all aspects significant. However, collective effects of seed drying, storage period and storage temperature (A*B*C) show slight not significant. Stored seeds moisture content and temperature shown significant effects during seed storage. Storage period and temperature significantly affected seeds quality during storage.

Table 2: Analysis of variance for germination of *E. tsjeriam-cottam* during storage

Source	df	Mean Square	Sig.
Seed drying types (A)	2	673.443**	.000
Storage period (b)	3	6877.798**	.000
Storage temperature (C)	2	3200.130**	.000
(A*B)	6	65.461**	.003
(A*C)	4	152.760**	.000
(B*C)	6	300.468**	.000
(A*B*C)	12	31.126^{ns}	.078
Std. Error	108	18.358	
Significant level at 0.5** ns-not significant			

Effect of various factors on seed germination

Temperature, storage period, seed moisture content and methods of seed drying affected the seed quality and longevity and the results described in table 2.

Mean germination was reduced under storage conditions. Germination significantly affected individually by the temperature and moisture content of seed at one-year storage conditions. Initial germination before storage of baybidang seeds was 92%, which was reduced up to 42% after one-year duration. Temperature play significant role and reduced germination up to 64.05, 57.7, and 47.8 percent. Drying of seed samples influenced seed storage and reduced germination in one-year storage up to 60, 55 and 53%, which affected significantly germination percentage after 12 months. Decline in germination was observed much faster in seeds after 6 months of storage. Storage period effect on mean germination after six month shown 55% and reduced up to 42% after one year. Germination on 5 ˚C temperature recorded 64% while on 20 and 30 °C germination reduced rapidly up to 57.75 and 47.85% respectively. Storage period and temperature effects seed quality more than to seed moisture content during storage. fan dried seeds reduced their germination up to 60.76, one day sun dried seeds reduced germination up to 55.31, and three day sun dried seeds reduced moisture content up to 53.31 percent.

Table 3: Effect of various factors on Germination

factor		Germination Percentage	Std. Error
Seed Drying	Fan dry	60.760a	0.618
	One day sundry	55.313b	0.618
	Three day sundry	53.583c	0.618
Storage Period	Three month	75.653a	0.714
	Six month	55.750b	0.714
	Nine month	51.889c	0.714
	Twelve month	42.917d	0.714
Storage temperature	5 °C	64.052a	0.618
	20 °C	57.750b	0.618
	30 °C	47.854c	0.618
Means within the same column and treatment followed by the same letter are not significantly different according to Duncan ($p \leq 0.05$)			

Effect of seed drying and storage periods on seed germination

Types of seed drying and storage period significantly reduce seed quality in case of *Embelia tsjeriam-cottam* seeds. Seed storage period and seed drying interaction significantly start to reduced seed germination and seed quality after 3 month. Effect of seed drying and storage period has given in table no.16.

Fan dried seeds shown seed germination percentage 82.04, 61.08, 53.08, and 46.83% after 3 month, 6 month, 9 month, and 12 month respectively. One-day sun dried seeds reduced germination 73.83, 54.41, 50.25, and 42.75% after 3 month, 6 month, 9 month, and 12 month respectively. Three-day sun dried seeds affected adversely seed Germination up to 71.08, 51.75, 50.33 and 39.16% after 3 month, 6 month, 9 month, and 12 month respectively. Significant reduction was observed in seed Germination percentage due to significant effect between seed drying and storage period. One year storage period and seed moisture content of seeds play significant role which is affect seed quality and seed germination.

Table 4: Effect of seed drying and storage period on seed germination

Seed drying	Storage period	Germination Percentage	Std. Error
fan dry	3month	82.042a	1.237
	6month	61.083b	1.237
	9month	53.083c	1.237
	12mont	46.833d	1.237
One day sundry	3month	73.833a	1.237
	6month	54.417b	1.237
	9month	50.250c	1.237
	12mont	42.750d	1.237
Three day sundry	3month	71.083a	1.237
	6month	51.750b	1.237
	9month	50.33b	1.237
	12mont	39.167c	1.237
Means within the same column and treatment followed by the same letter are not significantly different according to Duncan ($p \leq 0.05$)			

Effect of seed drying and storage temperature on seed germination

Seed drying and storage temperature significantly affect seed quality during storage. Seed storage temperature and seed drying interaction reduced seed germination and seed quality continue. Effect of seed drying and temperature of storage has given in table no.17. Fan dried seeds shown seed germination 71.53, 59.5, 51.25% on storage temperature 5, 20, and 30 °C respectively. One-day sun dried seeds reduced germination 61.31, 55.93, and 48.68% on storage temperature 5, 20, and 30 °C respectively. Three-day sun dried seeds affected by storage temperature and reduced seed Germination up to 59.31, 54.81, and 43.62% on storage temperature 5, 20, and 30 °C respectively. Significant reduction was observed in seed Germination percentage due to significant effect between seed drying and storage period.

Table 5: Effect of seed drying and storage temperature on seed germination

Seed draying	Storage temperature	Germination Percentage	S.E.?
Fan dry	5 °C	71.531a	1.071
	20 °C	59.500b	1.071
	30 °C	51.250c	1.071
One day _sundry	5 °C	61.313a	1.071
	20 °C	55.938b	1.071
	30 °C	48.688c	1.071
Three day sundry	5 °C	59.313a	1.071
	20 °C	54.813b	1.071
	30 °C	43.625c	1.071
Means within the same column and treatment followed by the same letter are not significantly different according to Duncan ($p \leq 0.05$)			

Effect of seed storage period and storage temperature on seed germination

Effect of storage period and storage temperature has been given in Table no.18. Comparative effect of both factor on seed germination and seed quality found significant. Storage period and temperature collectively effect seed quality during storage and reduced germination up to 29% after 12 month at 30 °C temperature. Seeds stored with low temperature at 5 °C reduced germination up to 55.83%. After six month duration seed germination decrease up to 63.33%, 57.83% and 46.08% at storage temperature 5, 20, and 30 °C respectively.

Three month storage duration and storage temperature effect seed quality and germination up to 78.45%, 76.58%, 73.91% at storage temperature 5, 20, and 30 °C respectively. 12 month Storage period and temperature significantly affect seed germination and seed quality during storage.

Table 6: Effect of seed storage period and storage temperature on seed germination

Storage period	Storage temperature	Germination Percentage	Std. Error
3 month	5	78.458a	1.237
	20	76.583b	1.237
	30	73.917c	1.237
6 month	5	63.333a	1.237
	20	57.833b	1.237
	30	46.083c	1.237
9 month	5	60.583a	1.237
	20	53.167b	1.237
	30	41.917c	1.237
12 month	5	55.833a	1.237
	20	43.417b	1.237
	30	29.500c	1.237
Means within the same column and treatment followed by the same letter are not significantly different according to Duncan ($p \leq 0.05$)			

Effect of seed drying, duration and storage temperature on seed germination

Effect of seed drying, storage period and storage temperature on seed germination of bearing has given in below table, no. 19. Seed drying effects with storage period and storage temperature comparatively reduced seed germination continue after 3 month to 12 month. Fan dried seeds shown seed germination after three month was 88.37, 81.50 and 76.25% at storage temperature 5, 20, and 30 °C respectively. After six month fan dried seeds shown 74.25% germination at 5 °C and 50% at 30 °C temperature. Quality of Seeds reduced very fast on high temperature rather than low temperature. Seed germination of fan-dried seeds stored at 5 °C, has found 63.75% germination after nine month, and germination decrease up to 59% after 12 month storage period. Fan dried seeds stored at 30 °C, reduced germination 44% after 9 month and 34% after 12 month shown significant effect of storage. Where as one-day sun dried seeds and three-day sun dried seeds stored at 5 °C temperature lost seed germination after 3 month up to 78.5 and 74.5% respectively. One day sun dried Seeds effected significantly during storage and reduced their germination continue after 6, 9 and 12 month up to 71.25%, 61% and 56.5% respectively at 5 °C, while at 20 °C seed germination were recorded 56.2, 51.5 and 41.2%. Germination per cent of three day sun dried seeds gradually decreased during storage period at 5, 20 and 30 °C temperature. Seed germination was recorded after six month 58.5, 57.0, 40.75% at temperature of 5, 20 and 30 °C respectively. Whereas after 9 months 55%, 50.75% and 39% seed germination was record. Three day sun dried seeds maximum lost their seed quality and seed germination during storage after 12 month at 5, 20 and 30 °C temperature where;

seed germination percent were count minimum, 51.25, 41.40, and 23.75%.

Table 7: Effect of seed drying, storage period and storage temperature on seed germination

Seed drying	Storage Period	Storage Temperature	Germination Percentage	Std. Error
Fan dry	3month	5	88.375a	2.142
		20	81.500b	2.142
		30	76.250c	2.142
	6 month	5	74.250a	2.142
		20	59.000c	2.142
		30	50.000d	2.142
	9 month	5	63.750a	2.142
		20	51.500b	2.142
		30	44.000c	2.142
	12 month	5	59.750a	2.142
		20	46.000b	2.142
		30	34.750c	2.142
One day sundry	3 month	5	78.500a	2.142
		20	75.250b	2.142
		30	74.750b	2.142
	6 month	5	71.250a	2.142
		20	56.500a	2.142
		30	47.500b	2.142
	9 month	5	61.000a	2.142
		20	51.250b	2.142
		30	42.500c	2.142
	12 month	5	56.500a	2.142
		20	41.750b	2.142
		30	30.000c	2.142
Three day sundry	3 month	5	74.500a	2.142
		20	73.000a	2.142
		30	70.750b	2.142
	6 month	5	58.500a	2.142
		20	57.000a	2.142
		30	40.750c	2.142
	9 month	5	55.000a	2.142
		20	50.750a	2.142
		30	39.250b	2.142
	12 month	5	51.250a	2.142
		20	41.500b	2.142
		30	23.750c	2.142

Means within the same column and treatment followed by the same letter are not significantly different according to Duncan ($p \leq 0.05$)

Discussion

During Seed storage periods reduction of seed quality and seed germination is high in *E. tsjeriam-cottam.* 75.65% seed germination observed after three month, whereas it remain 42.91% after 12-month. Agarwal (1980) [1] observed that maximum life span achievable by the seeds of a given species may substantially differ from that of another under similar conditions of storage. Seed longevity and viability of different plants depends on their physical and physiological characteristics (Priestley *et al*., 1985; Steadman *et al*., 1996) [9, 14]. Relative humidity (RH), temperature, and moisture content of seed effect seed storage.

Interaction Effects of temperature, seed drying and storage period on seed germination

Temperature affects seed quality and longevity of seeds during storage. It is the major cause which plays an important role to affect seed storability. A higher temperature influence the metabolism of cell, may cause a rise in the rates of several metabolic processes causing faster deterioration. Temperature with moisture content of seed breakdown metabolism very fast and causing age factors and sometimes. Seed drying method, temperature and storage period affects seed quality. Three day sun dried seeds of *Embelia tsjeriam-cottam* stored on 30 °C, lost maximum seed quality and seed germination during storage after 12 month. Whereas fan dried seeds sample stored at 5 °C give batter response. Embelia seeds are shows desiccation sensitive nature it can be says intermediate seeds reduced safe moisture content up to 14%.same type of work proposed by Bonner and Vozzo (1987) suggested storage longevity of the genus Quercus have a wide range of storability. Acorns of the red oak subgenus can be stored up to one year although viability loss can be high.

Conclusion

Three types dried seed sample i.e. 24 hour fan dried, one day sun dried and three day sun dried seeds of baybidang stored at three temperatures (5, 20, and 30 °C) for one-year duration. Fan dried seeds shown seed germination 88.37% after three month, while up to 59% after 12-month duration with 5 °C temperature. Fan dried seeds shown significant effect of stored at 30 °C, reduced germination 44% after 9 month and 34% after 12 month. Where as one-day sun dried seeds and three-day sun dried seeds stored at 5 °C temperature lost seed germination after 12 month up to 56.5% and 51.25% respectively. Seeds stored at 30 °C decrease seed quality very fast. One day and Three day sun dried seeds maximum lost their seed quality and seed germination during storage after 12 month at 30 °C temperature where; seed germination percent were count minimum, 30% and 23.75% respectively.

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