June 2009 *Volume 3 Issue 1*

Trade Science Inc.

Research & Reviews in

BioSciences

Regular Paper

RRBS, 3(1), 2009 [11-15]

Germination behaviour, viability and longevity of safflower (*Carthamus tinctorius* L.) seeds

Aloka Kumari

University Department of Botany, T.M.Bhagalpur University, Bhagalpur-812007

E-mail : aloka_kumari04@yahoo.com

Received: 27th April, 2009 ; Accepted: 2nd May, 2009

ABSTRACT

Germination behaviour, viability and longevity of the seeds were studied in an important oil-yielding crop safflower (*Carthamus tinctorius* L., Asteraceae) stored in screw capped plastic containers under ambient conditions. Immediately after harvest (May) germination percentage was lower (80%-97%) than that stored between June to October. Thereafter, the germination percentage sharply declined and the viability was completely lost after 14 months of storage. Viability tests with tetrazolium gave reasonable agreement with the result from germination tests. During winter months with relatively low room temperature, significant increase in germination percentage was observed when the seed were incubated at 28 $\pm$ 2°C. Loss of viability with ageing was always associated with reduced radicle growth and irregularities and abnormalities in hypocotyl development. 100 seed weight and seed moisture content also decreased during storage. It was concluded that we can able to stored safflower more than a year under ambient conditions.

KEYWORDS

Safflower (*Carthamus tinctorius* L.);
Germination behaviour;
Viability;
Longevity;
Seed storage.

INTRODUCTION

Seeds vary greatly in their viability and longevity depending on the plant species and varieties[36] as well as the conditions of harvest and storage[18,8]. Deterioration of the seeds with ageing results in the loss of viability and vigour during storage which is usually due to the alteration in moisture content, changes in biochemical composition and result in increased leaching of electrolytes and other low molecular weight substances during imbibitions[10,21]. Seed moisture and storage temperature are the two most important factors influencing loss of viability during storage. The status of water and its binding in relation to seed moisture determine its storage behaviour[29]. Seeds of almost all important agricultural crops need to be stored from the day of harvest till the time of next sowing. Further, it is a general practice of Indian farmers to carry over about 20% of the seeds for subsequent sowing as a safeguard against natural hazards[4]. Although longevity of seeds can considerably be increased in storing them in certain well defined conditions, in general practice the surplus seeds or even the seeds kept aside for sowing in succeeding year is stored in 'gowdowns' or in some plastic or bamboo containers devoid of ideal storage conditions.

Safflower (*Carthamus tinctorius* L.) is one of the important commercial oil seed crops. It is generally considered a rabi season crop. Safflower oil is primarily used for edible purposes due to high percentage (70%) linoleic acid[7]. Safflower is more drought and salt tolerant than other oilseed crops[9,14,37]. The crop is principally cultivated in India, California and Arizona,

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but has been grown successfully in every state west of the 100th meridian[3,12,22,23,34].

Seed production in safflower is directly related with success of pollination because the plant shows self pollination in absence of pollinators[24]. The plant is harvested in April-May and sown in September-October[37] and thus the seeds require storage for one season (one and half if they are to be carried over for the next year as a safeguard). As no information was available on the storability of safflower seeds, the present work was carried out to determine the germination behaviour, viability and longevity of safflower seeds stored in plastic pots under ambient conditions.

MATERIALS AND METHODS

Mature seeds of safflower were obtained from Directorate of Oil Seed Research Centre, Rajendranagar, Hyderabad (Variety = A-2). The seeds were stored in plastic containers with screw cap in the laboratory under ambient conditions. Seeds were germinated on moistened filter papers in open petridishes at room temperature and germinated recorded every 24 hours until there was no further germination. The seeds were watered by adding 3ml of distilled water every day. A seed was considered germinated when the radicle had broken the pericarp. During winter months (October to March) germination tests were also conducted at 28 ± 2^0C in an incubator together with the tests at room temperature. Radicle and hypocotyls lengths were measured after 72 hours of sowing (after 96 hours during winter months) by taking ten seedlings randomly from each petridish and the average length was calculated. Germination tests were conducted every month until all the seeds lost their viability completely. Moisture contents were measured by drying the seeds at 100-105^{0}C in an oven and expressed as the percentage on fresh weight basis.

Along with germination tests viability was also tested using 1% 2, 3, 5- triphenyl tetrazolium chloride (TTC). A seed was considered viable if the whole of the embryo was stained or only a part of the cotyledon remain unstained after incubating the embryo in TTC for 24 hours in dark. Whenever the radicle portion was not stained the seed was counted as nonviable. All the experiments were conducted in triplicates. A seedling vigour index was calculated by using the following formula:

Vigour index = Germination percent × (Radicle length + Hypocotyl length in cm)

Meteorological data for the period of experimentation were obtained from the Meteorological Division, Sabour Agricultural College, Bhagalpur.

RESULTS

Seed germination and viability

Results of germination tests conducted at monthly intervals from the time of harvest till the seeds completely lost viability are shown in **figure** 1. Immediately after harvest in the months of April the germination percentage was incomplete (39%). From June to October (3 month of storage) almost all the seeds (>93%) germinated at room temperature. Germination percentage decreased from October (7 months of storage) and sharply dropped to 13% in March of the following year (12 month in storage). Negligible germination was recorded in May (14 months in storage). Starting from 2nd month in storage (May) till the seventh month (October) germination started within 24 hours. However, from the month of November there was no sign of germination before 36 hours after sowing even when the seeds were incubated at 28 ± 2^0C. This was seen not only in older seeds from the previous year's harvested seeds germination could be recorded only after 48 hours of sowing during the months of November to May. Maximum germination in these cases was recorded after 96 hours of sowing.

Viability tests with TTC

In general, viability of seeds as demonstrated by TTC staining was comparable to the result of germination tests (TABLE 1). Newly harvested seeds tested in January and May showed almost 98% viability with TTC, although the actual germination percentages at room temperature (about 23.4^{0}C) were only 39% to 78% respectively. Nevertheless, at 28 ± 2^0C germination approach 83 and 78% in October - November. From February (10 months of storage) the fall in actual germination percentage was rapid and sharp whereas viability as shown by TTC test declined gradually, nearly always giving an over estimate of germination (TABLE 1).

Up to nine months of storage almost all the parts of the embryo were stained with TTC. But as the seeds

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TABLE 1: Comparison of the viability of safflower seeds as shown by the tetrazolium and seed germination tests after different periods of storage. The seeds were harvested on the last week of May ± SD

Time of testing	Pollen viability			Vigour index
	Tetrazolium test	Germination test		
		Room temperature	28±2°C	
April	94.55±2.14	39.43±5.84	-	303.61
May	97.99±1.83	78.29±2.91	-	704.61
June	95.48±1.66	91.78±1.62	-	1099.44
July	95.21±1.38	97.95±2.35	-	1283.14
August	97.74±1.66	97.99±2.76	-	1215.07
September	95.40±1.99	96.99±2.43	-	1057.19
October	92.5±2.6	94.87±0.81	80.87±3.27	948.7
November	90.67±1.73	89.69±0.57	76.69±2.41	896.9
December	88.84±4.29	85.75±3.24	78.77±1.58	565.95
January	85.3±7.53	80.42±5.65	59.68±2.89	458.39
February	69.66±5.13	46.17±5.64	32.57±8.14	286.25
March	36.17±4.27	13.07±0.88	27.96±3.45	44.43
April	15.59±1.97	08.95±7.69	-	15.21
May	6.39±1.78	03.87±1.62	-	5.03

TABLE 2: Meteriological data of Bhagalpur from April 2007 to May 2008, the storage period

Date	Temperature (°C)		Relative humidity		Rainfall
	Maximum	Minimum	7a.m.	2p.m.	
April-07	35.0	20.8	80	43	69.3
May	37.8	23.4	76	48	54.5
June	33.6	24.6	87	61	199.0
July	32.7	24.8	93	75	333.0
August	32.2	26.0	89	72	146.4
September	32.0	26.6	91	78	92.3
October	29.9	23.9	94	74	126.8
November	29.1	16.0	89	54	0.0
December	26.6	12.7	94	59	0.0
January-08	21.9	08.9	86	59	12.2
February	23.3	11.3	91	58	5.8
March	30.9	15.2	82	42	24.9
April	34.6	19.4	82	42	0.0

TABLE 3: 100 seed weight and moisture contents of safflower seeds

	Freshly harvested seeds (in gm)	Seeds after sun drying	14 months old seeds
100 seed weight	485± 14.69	381± 11.43	315± 10.99
Moisture content (% Fresh weight)	10.17± 2.12	10.82± 1.26	8.86± 0.59

began to loose their viability many of the embryos showed only partial staining. It was the radicle which first stopped to take stain followed by the cotyledons. The extreme tip of embryonal stem was seen to be stained in a few seeds up to April next year (14 months in storage), although these seeds were not able to germinate.

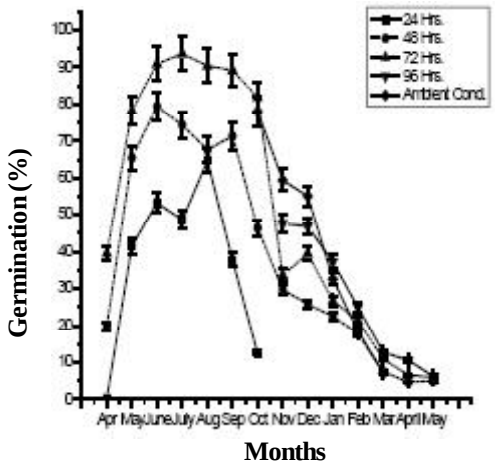

Figure 1: Percentage germination of safflower seeds every month in storage after harvest in May 2006. Bars indicate ± SD

Effect of ambient temperature

The metrological data of Bhagalpur (Bihar, India) with respect to the storage period for mean temperature, relative humidity and rainfall is given in TABLE 2. At room temperature germination percentage was considerably lower during the winter months (November to May), when the average room temperature was also lower. Effect of ambient temperature was evident in fresh harvested seeds in April and May (1st and 2nd month of storage) where a rise in temperature to 28±2°C highly improved the seed germination percentage. Even in seeds with longer storage periods (12 to 14 months) increase the temperature considerably increased the germination percentage, nearly approaching the value given by TTC tests.

Seed weight and moisture content

Seeds weight and seed moisture contents just after the harvest and after 14 months of storage are given in TABLE 3. The seeds contained about 12% moisture when put into storage but this was reduced to 8.8% after 14 months storage. Loss in weight by the seeds during storage was also evident.

Seeding growth and vigour index

A very close similarity was observed among the patterns of seedling growth, germination and vigour indices (figure 2, TABLE 1). Radicles and hypocotyls both were well developed till the month of February (11 months in storage) and thereafter their length started

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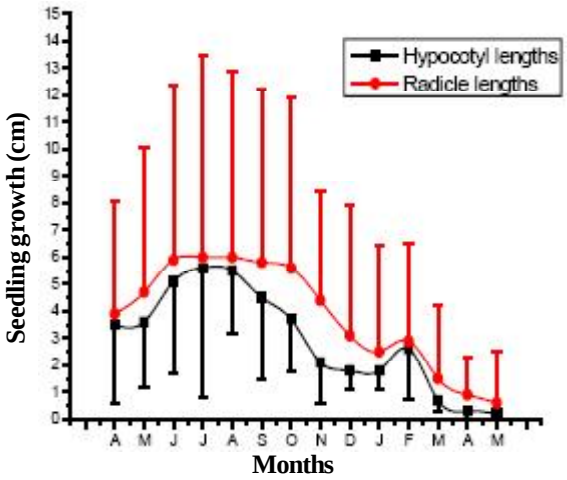

Figure 2: Seedling growth in safflower in different months of storage. Bars indicate ± SD

to decrease. From April onwards (13 month in storage) the radicle growth virtually stopped, but in many of the seeds the hypocotyls grew well, although with some irregularities and relatively large deviations even in the same experimental lot.

DISCUSSION

Ageing is a natural, irreversible phenomenon which affects viability and vigour in stored seeds and poses a serious problem in modern agricultural practice. Seeds possess highest vigour at the time of their physiological maturity[25]. Thereafter, the seeds gradually age and decline in viability and vigour. Various internal as well as external factors effect the viability of seeds during storage[1,18,26]. Of several problems encountered in storage of seeds, maintenance of viability and vigour are most important[32]. Planning for seed storage requires information on relative storability of seeds of particular species under ambient conditions in different agro-climatic zones.

The loss of viability of safflower seeds can be detected from the germination percentage, seedling vigour and staining pattern of the embryo to tetrazolium chloride. Storage of safflower seeds for one year in plastic containers decreased germination percentage to virtually negligible level. This was reflected by tetrazolium tests, except in cases where the ambient temperature effect was more prominent. Tetrazolium test has been successfully used for topographic determination of seed viability[5,30,31] as well as to evaluate viability even when the seed is in a state dormancy[15]. In this case the discrepancy, whenever observed between actual seed germination percentage and the viability percentage reflected by tetrazolium test can, to a large extent, be explained on the basis of ambient temperature effect on germination. Even after 12 months of ageing incubation of seeds in 28°C in the month of November to March sufficiently improved the germination percentage. However, on 12th month of storage and completely lost after 14th month of storage. Thereafter, the low germination of the seeds was clearly due to the irreversible loss of viability caused by natural ageing. In these observations during the first and second months of storage the seed showed lower percentage of germination at room temperature which increased and approached 97% from the 4th month of storage. This behaviour can only partially be attributed to the low room temperature during the month of October-January as increase in temperature highly improved germination percentage germination in these months. Meena et al.[25] observed initial increase in percentage germination up to six months of storage under ambient conditions in cotton indicating the existence of hard seeds. Narayanswamy and Swamy[28] found that five month old seeds had higher germination and vigour index in maize. An increase in germination percentage after a few months of dry storage is also recorded for *Caseulia axillaris*, a member of the family Asteraceae[27]. This suggests that safflower seeds may possess a short dormancy period or may require after ripening.

Amongst different factors influencing seed longevity storage environment, relative humidity and temperature contribute to a greater extent[17,19]. We have presented the meteriological data of Bhagalpur for reference. The oxidative deterioration of polyunsaturated lipid in cellular membrane leading free radicle chain reaction is considered to be primary region of ageing[20]. Poor membrane structure and leaky cells are usually associated with deteriorating and low vigour seed lots[5,11]. Sharma and Singh[33] observed that loss in viability in linseed was related with the seed weight and variety with bold seeds (higher 100 seed weight) lost the viability earlier. No varietal differences in seed weight in storage. Moisture content of linseeds stored in cloth bags varied in different months depending on relative humidity[33]. However, in this case no such fluctuation in moisture content is expected as the seeds were stored in screw capped plastic container. Many seeds retain

their viability better in sealed container than in open storage because sealed container provide a simple and convenient method of controlling seed moisture content[6].

Seedling growth is an important criterion to assess the degree of seed deterioration. The seedling vigour index of safflower decline considerably after 8th months of storage. This may be due to decreased mobilization of reserve substances during germination of the stored seeds[31,35]. It is interesting to note that loss of viability of safflower seeds was always associated within the inhibition of radicle growth. This was also reflected by TTC staining as in partially stained embryos. It was radicle which first stopped to take stain followed by cotyledons. In many seeds the hypocotyls grew readily even after 12 to 15 months of storage, when the radicle and cotyledons both remained unstained by TTC. The vigour tests are commonly evaluated according to their ability to predict some aspects of potential seed performance, particularly seedling growth rate, seedling emergence in field, plant uniformity, crop yield and storability. This observation indicates that safflower seeds have potential storability of a year or even less. The period is sufficient for the same year's sowing but it is clear that they can not be carried over for subsequent years under ambient conditions. Field performance often depends on initial seed vigour[31]. However, separate experiments are needed to strictly evaluate the case in the field.

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