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Studies on storage life, and effect of temperature and pre-sowing seed treatments on germination behaviour and maturity indices in King-chilli (*Capsicum* spp)

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ABSTRACT

King-chilli (*Capsicum* spp) is a high value spice crop of the North Eastern India. Although crop is having a great demand in national and international market due to its richness in capsaicin and oleoresin contents, however, it is grown in some pockets of the region on small scale due to specific climatic requirement, poor seed germination and traditional cultivation practices. Keeping these in view, the present investigation was undertaken to study the storage life, effect of temperature and pre-sowing seed treatment on germination behaviour and maturity indices of the crop for seed purpose. The results indicated that seeds of King-chilli remain viable and can be stored up to one year. The maximum seed germination (69.67%) was recorded at first month of the storage and then gradually decreased with increase in storage period with least (0.33%) at the 18 months after storage. The optimum temperature for seed germination was 25-30°C. The synchronous, higher germination (65.67%) and vigorous seedling (1477.58) can be raised by pre-sowing seed treatment with KNO_3 (5%) for 24 hr. The seeds of dried pod had shown highest germination (68.77%) and seedling vigour (611.37) when the pods are harvested at fully developed yellow stage. However, seed from red ripe fresh pods with 7 days resting period was found ideal for higher germination and vigour over the seeds from the dried pods. Hence, commercial growers can store the seeds of King-chilli for one year and raise vigour seedlings with maximum germination by pre-sowing seed treatment with KNO_3 (5%).

Key words: King-chilli, Maturity indices, Seed treatment, Temperature

King-chilli (*Capsicum spp.*) is one of the most important high value spice crop of Solanaceae family, grown widely in the north eastern states of India especially, Nagaland, Manipur, Mizoram and eastern part of Assam. It is known by different names as *Bhut Jolokia* in Assamese, *Naga chilli*/*King-chilli* in Nagaland and *Umrok* in Manipur. The crop is grown traditionally by local people for different uses. The crop was reported as hottest chilli in the world in 2007. In recent past, the crop is gaining importance due to demand in international market for the different value added products such as capsaicin and oleoresins. Wider genetic diversity has been observed in the crop with respect to plant, flower and pod shape and size, colour and quality traits like, aroma and capsaicin (2-4%) content. King-chilli having differences for germination behaviour over the other *Capsicum* spp. and it seems to be originated as an interspecific hybrid between *C. chinense* and *C. frutescens* (Bosland and Baral 2007).

The storage life of King-chilli seeds is very poor and farmers store the seeds in dried pod (fruits). Seed germination in King-chilli is not only slow but also has erratic behaviour which is not common in other capsicum species. Seed coats and surrounding structures may influence the ability of a seed to germinate through interference with water uptake, gas exchange, diffusion of endogenous inhibitors, or by mechanical restriction of embryo growth (Mayer and Shain 1974). In seeds that do not have hard seed coats or require scarification for water uptake, other seed parts such as the endosperm may mechanically restrict embryo expansion, thus preventing radicle emergence (Pavlista and Haber 1970). Seed invigoration techniques have been employed to achieve improvement in the rate of germination, uniformity, root growth and seed vigour (Thornton and Powell 1992) in a number of crops. Primed seeds with PEG-8000, NaCl, and KNO_3 produced more vigorous seedlings than untreated seeds of tomato (Farooq *et al*. 2005). Besides, seed priming also improve the stress tolerance (Yadav *et al*. 2011, Hassen *et al*. 2017) in *Capsicum* sp. Gibberellins induce the production of enzymes that degrade the stored carbohydrates (starch) and proteins. These enzymes are secreted from the cells into the aleurone endosperm where starch is converted to sugars until enzymes digest the cell wall in the endosperm.

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Moreover, the potassium regulates the activities of 40 or more enzymes. It is responsible for strengthening the cell walls, which results in a plant resistance to disease. It facilitates the synthesis and translocation of sugars, starches and oils. Seed germination is also influenced by external factors like temperature, light, moisture and O_2 concentration (Watkins and Cantliffe 1983).

In the region, crop is sown during February-March by picking the seeds from the dried pods or fully mature fresh pod as well in the nursery and direct seeding is also prastised in *jhum* field. The main harvesting time of the crop is during August and September. Although, the crop is having great demand locally as well as in international market, but it is grown in limited scale due to some limitations such as specific climatic requirements, poor and erratic seed germination, traditional cultivation practices etc. Keeping these in view, the present investigation was undertaken to 1) study the storage life of seeds; 2) study the effect of temperature on seed germination; 3) study the effect of pre-sowing seed treatment on the germination behaviour of King-chilli seeds; and 4) study the maturity indices in King-chilli for seed purpose.

MATERIALS AND METHODS

This experiment was conducted during 2012-2014 at Division of Horticulture, ICAR Research Complex for NEH Region Umiam, Meghalaya, India. To study the storage life, King-chilli seeds were collected from the red ripe pods grown under the polyhouse in first week of April, 2012. The pods were dried and stored in different moisture proof containers up to 18 months of the storage at room temperature. For germination study, seeds at 1, 6, 12 and 18 months of storage were taken out; surface sterilized with Captan and then was placed on wet germinating paper in petriplate at room temperature. Five seeds were sown in each petriplate with 10 petriplate in each replication. The moisture was maintained through day interval irrigation using distilled water. The observation was taken as germination percentage.

To study the effect of temperature on seed germination, the month old dried seeds were sown in pots (5 seeds/pot) and transferred in growth chamber at 20^0C, 25^0C, 30^0C and 35^0C and observations were recorded as days taken to seedling emergence and germination percentage.

To study the effect of pre-sowing seed treatments on germination behaviour of King-chilli, one year old seeds were used. Total twelve treatments, i.e. T_1: GA_3 (50 ppm), T_2: GA_3 (100 ppm), T_3: GA_3 (200 ppm), T_4: KNO_3 (2%), T_5: KNO_3 (3%), T_6: KNO_3 (5%), T_7: Disodium hydrogen phosphate (HNa_2O_4P) 10^{-1}, T_8: Disodium hydrogen phosphate 10^{-3}, T_9: Potassium dihydrogen phosphate (H_2KO_4P) 10^{-1}, T_{10}: Potassium dihydrogen phosphate 10^{-3}, T_{11}: Zn ($ZnSO_4$ 1%), and T_{12}: control. The shade dried treated seeds were sown in pots having sterilized growing medium of sand, soil and FYM each in 1:1:1(v/v) proportion. Five numbers of seeds were sown in each pot and total 20 pots were used for each treatment. Seedling vigour tests were conducted on six randomly selected seedlings of each treatment at 30 days after sowing. The vigour test evaluation was carried out between February-March 2013.

The seedling vigour index-I (SVI-I) was determined by using the formula given by Abdul Bakshi and Anderson (1973) as below:

$$\text{SVI} - \text{I} = \text{Average shoot length (cm)} + \text{Average root length (cm)} \times \text{Germination (\%)}$$

Seedling vigour index-II (SVI-II) was calculated using following formula:

$$\text{SVI} - \text{II} = \text{Germination (\%)} \times \text{Average seedling dry weight (mg)}$$

Speed of germination index (SGI) was calculated as described in the Association of Official Seed Analyst (AOSA 1983) as follows:

$$\text{SGI} = \frac{\text{Number of germinated seed} + ... + \text{Number of germinated seeds}}{\text{Days of first count} + ... + \text{Days of final count}}$$

To study the maturity indices in King-chilli for the seed production, the seeds of week old (7 days) harvested dried pods of three maturity stages, i.e. red ripe, yellowing and turning stage were taken for the analysis during June-July, 2014. The seeds were treated with Captan and five seeds were sown in pots. Total 100 seeds were used for each treatment with four replications having five pots each. The observation were taken as days taken to first emergence, shoot length (cm), root length (cm), number of leaves, dry matter (%), SVI-I and SVI-II were taken at 30 days after seed sowing. The mean value was calculated from the six randomly selected seedlings in each replication.

The observed data were analyzed using Statistical Tool for Agricultural Research (STAR) software and treatment means were separated by the Duncan Multiple Range Test (DMRT) at $P<0.05$ level of significance.

RESULTS AND DISCUSSION

King-chilli crop is grown mostly under open condition and also under the low cost polyhouse. In the present investigation crops were grown under low cost polyhouse, it produced on an average of 128.33±10.41 pods/plant with average yield of 669±65.60 g/plant. The fully developed pod size ranged from 5.5 to 6.0 cm with average weight of 3.63±0.42 g. Similarly, the number of seeds per pod ranged from 37.0 to 45.0 and test weight (100 seeds) was 0.47±0.01 g.

Storage life and germination of King-chilli seeds

The results on storage life of the King-chilli seeds have shown the significant decrease in the germination percentage with increase in the storage life from the 3 months onwards up to 18 months (Fig 1). Maximum germination (69.67%) percentage was recorded at one month of the storage followed by 3 months of storage (67.63%) while it was least (0.33%) at the 18 months after storage. The results on germination of seeds at different stage of storage life have revealed that the seeds of King-chilli remain viable

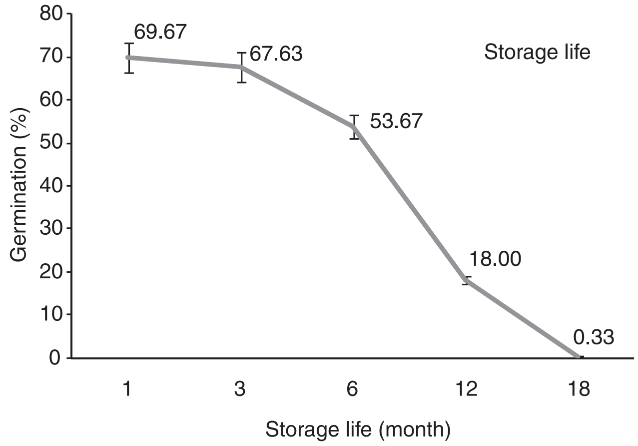

Fig 1 Effect of seed storage life on germination (%) in King-chilli.

for one year. The decrease in the germination percentage of the seeds with the proceedings of seed storage life, significantly after three month may be due to drying of the semi-permeable layer in the seed coat comprising of suberin (Beresniewicz *et al*. 1995) and the mechanical resistance possessed by endosperm on emergence of radicle (Watkins and Cantliffe 1983).

Effect of temperature on seed germination

The significant effect of temperature was observed for both days taken to emergence and germination percentage (Fig 2). The temperature has shown the positive correlation (r= 0.816) with the germination percentage while it was negative (r = -0.922) with the days taken to emergence. The maximum germination and earliest seedling emergence was recorded at 30°C. However, minimum germination and late emergence was recorded at 20°C (Fig 2). For nursery production, the optimum temperature for King-chilli was 25-30°C. Significant effect of temperature on seedling emergence was also observed by Samarah *et al*. (2016) in pepper. Temperature and humidity are most important factors responsible for germination of the seeds. The earliest germination at 30°C may be due to increase in rate of water absorption by the seeds and reduction in the strength of endosperm. Watkins and Cantliffe (1983) also observed that in capsicum seeds, the puncture force decreased faster

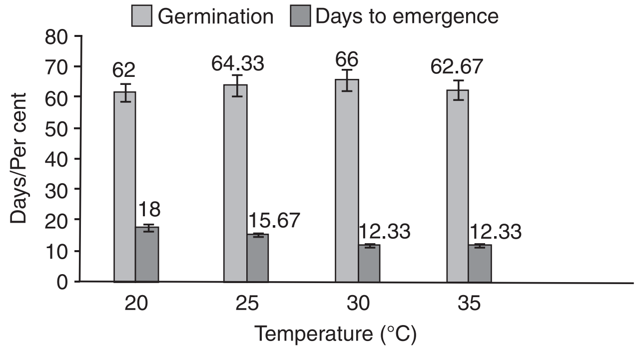

Fig 2 Effect of temperature on germination and days to emergence.

when seeds were imbibed at 25°C than at 15°C. Similarly, Bosland *et al*. (2000) indicated that day/night temperature fluctuations of 30º/15º C (86º/59º F) at 16 hr of light and 8 hr of darkness is biologically the best temperature and lighting range for chilli germination.

Effects of pre-sowing seed treatments

The results of pre-sowing seed treatments (Table 1) have shown the significant effect on germination and vigour of the seedling. The presoaked seeds have shown early and faster emergence over the control. This may be due to greater activities of the enzymes of glyoxylate cycle involved in lipid to sugar conversion during germination (Pandita *et al*. 2007). The faster and improved emergence of pepper seed by presoaking (hydropriming) was also observed by Samarah *et al*. (2016). Among the treatments, the earliest emergence (9.33 days) was observed with treatments GA_3 200 ppm followed by GA_3 100 ppm (Table 1). Significant effect of GA on seedling emergence was also reported by Tzortzakis (2009) in endive (*Cichorium intybus*). Gibberellins stimulate germination by inducing hydrolytic enzymes that weaken the barrier tissues such as the endosperm or seed coat, inducing mobilization of seed storage reserves and stimulating expansion of the embryo (Bewley and Black 1994). GA may also stimulate germination via the transition from embryonic to vegetative development, in part mediated by the chromatin remodeling factor PICKLE (PKL) (Henderson *et al*. 2004).

The pre-sowing seed treatments have also shown significant effect on root and shoot growth and number of leaves in the seedlings. Among the treatments, KNO_3 (5%) recorded highest values (Table 1). However, there was no significant effect of GA_3 on root length over control. This may be due to negative correlation by GA_3 (Nautiyal *et al*. 1985) and positive correlation with KNO_3 (Kattimani *et al*. 1999). Similar findings were also reported by Maiti *et al*. (2013) in tomato and chilli from seed priming with KNO_3. Many nitrogen-containing compounds, including NO gas, nitrite (NO_2^-), nitrate (NO_3^-), nitrogen dioxide, ammonium, azide and cyanide, promote breaking dormancy and improving seed germination in many species, possibly as a means of sensing soil N availability (Bethke *et al*. 2007). According to Bewley and Black (1994), KNO_3 raises the ambient oxygen levels by making less oxygen available for citric acid cycle.

Furthermore, seeds treated with Zn (1%) have also shown significant effect on shoot and root length contributing to the vigour of the seedlings over the control (Table 1). Similar findings were also reported by Stratu and Costica (2015) in *Cucumis melo*. The pre-sowing treatment with zinc increases the concentration of Zn in endosperm and contributes to the increase in shoot length through auxin metabolism.

Likewise, highest dry matter content (10.16%) was observed from KNO_3 (3%). Similar findings were also reported by Chaudhary *et al*. (2008) in chilli. Increase in dry matter content may be due to effect of K and N

Table 1 Effect of seed treatments on germination behavior of King-chilli seeds

Treatment	Days to seedling emergence	Shot length (cm)	Root length (cm)	No. of leaves (cm)	Dry matter (%)	Germination (%)	SVI-I	SVI-II	SGI
GA_3 (50 ppm)	10.67^{bcd}	8.60^{g}	3.90^{e}	2.90^{ef}	9.45^{bcd}	33.00^{g}	412.50^{gh}	372.90^{i}	0.49^{e}
GA_3 (100 ppm)	9.67^{cd}	10.80^{de}	4.35^{e}	3.55^{d}	9.76^{ab}	32.50^{g}	492.38^{fg}	507.00^{g}	0.44^{f}
GA_3 (300 ppm)	9.33^{d}	10.25^{ef}	4.10^{e}	3.45^{d}	9.58^{bc}	29.10^{h}	417.59^{gh}	445.23^{h}	0.50^{e}
KNO_3 (2%)	15.00^{a}	13.47^{b}	8.10^{a}	4.05^{c}	10.16^{a}	32.50^{g}	701.03^{e}	591.50^{f}	0.68^{c}
KNO_3 (3%)	14.67^{a}	13.65^{b}	8.40^{a}	4.10^{c}	9.59^{bc}	44.50^{d}	981.23^{b}	765.40^{d}	0.54^{d}
KNO_3 (5%)	11.33^{bcd}	15.75^{a}	6.75^{b}	5.60^{a}	8.96^{d}	65.67^{a}	1477.58^{a}	2331.29^{a}	1.12^{a}
HNa_2O_4P (10^{-1})	12.33^{b}	12.40^{bc}	5.40^{cd}	2.65^{f}	9.61^{bc}	48.50^{c}	863.30^{c}	1000.02^{b}	0.74^{b}
HNa_2O_4P (10^{-3})	10.67^{bcd}	8.90^{fg}	3.75^{e}	3.40^{d}	9.38^{bcd}	29.17^{h}	369.00^{h}	460.04^{gh}	0.43^{f}
H_2KO_4P (10^{-1})	11.33^{bcd}	10.90^{cde}	4.55^{de}	4.60^{b}	9.68^{ab}	36.50^{f}	563.93^{f}	770.15^{ef}	0.56^{d}
H_2KO_4P (10^{-3})	11.67^{bc}	11.25^{cde}	4.25^{de}	4.55^{b}	9.32^{bcd}	52.50^{b}	813.75^{cd}	834.75^{c}	0.76^{b}
Zn (1%)	11.67^{bc}	12.1^{bcd}	6.25^{bc}	3.05^{e}	9.13^{cd}	40.23^{e}	738.22^{de}	700.00^{e}	0.54^{d}
Control	15.33^{a}	7.75^{g}	3.66^{e}	2.77^{ef}	8.15^{e}	16.50^{i}	188.27^{i}	277.20^{j}	0.24^{g}
Mean	11.67	11.32	5.29	3.72	9.4	38.31	668.23	754.64	0.58
CV (%)	6.50	3.42	4.43	1.95	2.33	3.66	4.16	1.57	1.58
LSD (P<0.05)	2.25	1.56	0.94	0.29	0.88	1.55	110.77	40	0.04

* Means with the same letter in column are not significantly different at P<0.05 level.

in the translocation of reserved food and assimilation of carbohydrates in the seedlings. Fournier *et al*. (2005) also observed that, when the concentration of K^+ in the nutrient solution increases there is also increase in the fresh and dry matter in sunflower.

The treatments were also found significant for total germination percentage of the seeds (Table 1). Among the treatments, highest germination (65.67%) was recorded from KNO_3 (5%) and it was least in control (16.50%). Chaudhary *et al*. (2008) also reported highest germination percent by priming of fresh and aged seeds of chilli with KNO_3. The significant effect on germination percentage may be due to a better metabolic response of the seed to the KNO_3 and H_2KO_4P.The significant effect of KNO_3 may be possibly through oxidized forms of nitrogen causing a shift in respiratory metabolism to the pentose phosphate pathway (Robert and Smith 1977).

Among the treatments, seedling vigour index-I, II and speed of germination index were highest in the treatment T_6 (KNO_3, 5%) while it was least in control, i.e. untreated seeds (Table 1). The similar findings were also reported by Chaudhary *et al*. (2008) in *Capsicum annuum* and Torres *et al*. (2013) in Habanero chilli (*Capsicum chinense* Jacq.). The increase in the seedling vigour index-I (SVI-I) by the application of KNO_3 is mainly due to increase in germination percentage as well as a length of the seedling which is having positive correlation, similarly SVI-II is having positive correlation with germination and dry weight of the seedling. Seed treatment with KNO_3 may have influenced the increase in seedling growth and development by faster multiplication of cells due to better hydration of colloids and cell division. The increase in the speed of germination may be due to faster initiation of activities within the seeds due to the imbibed (K+) ions. Sowmya *et al*. (2012) also observed faster emergence and vigour seedlings in cucumber by seed priming with KNO_3 which increases biochemical activities (amylase, catalase and peroxidase) in the seeds.

Maturity indices in King-chilli

Under maturity indices in King-chilli for seeds, the period of seedling emergence from fresh pod was 11-17 days in red stage, 13-19 days in yellow stage and 15-22 days from turning stage. There was difference of two days in the first emergence of seeds at each stage. Likewise, the seeds from dried pods were emerged during 14-22 days of red stage, 15-22 days of yellow stage and 16-22 days periods of turning stage. Moreover, from mean data of seeds of both fresh and dried pods the early emergence was recorded from the red (11.33 and 14.33 days) stage followed by yellow stage (12.67 and 14.67 days), respectively. The early emergence of seeds from red stage (dried and fresh) may possibly be due to the more vigour of the seeds of red stage over the others. However, the early emergence of the seed of fresh pods over the dry may be due to improvement in physiological potential of the seeds by post-harvest resting and lower concentration of inhibitors ABA. Pereira *et al*. (2014) also reported in the improvement of physiological potential of the seeds by post-harvest resting of the fruits for 10 days in pepper.

From fresh pods, the highest germination percentage was obtained from the red stage followed by yellow stage and turning stage (Table 2). This may be due to decline in the seed water content while the dry matter continue to be accumulated and moisture content remained high even

Table 2 Maturity indices in King-chilli for seed purpose

Stages	Days to seedling emergence	Shot length (cm)	Root length (cm)	No of leaves	Dry matter (%)	Germination (%)	SVI-I	SVI-II
Seeds of fresh pods								
Red	11.33^{b}	7.36^{a}	4.79^{a}	3.00^{a}	6.46^{a}	88.08^{a}	1070.17^{a}	3200.0^{a}
Yellow	12.67^{b}	6.35^{a}	4.63^{b}	2.33^{ab}	5.76^{a}	85.67^{b}	940.66^{b}	813.83^{b}
Turning	15.33^{a}	4.12^{b}	3.89^{c}	2.00^{b}	4.18^{b}	82.31^{c}	659.30^{c}	576.18^{c}
Mean	13.11	6.61	4.43	2.44	5.47	85.35	890.04	1530.0
CV (%)	5.08	1.79	0.76	13.64	7.31	0.90	5.90	2.59
LSD (P<0.05)	1.51	0.27	0.08	0.75	0.88	1.76	13.70	120.0
Seeds of dried pods								
Red	14.33^{b}	6.56^{a}	4.29^{a}	3.67^{a}	6.92^{a}	57.56^{b}	624.53^{a}	949.6^{a}
Yellow	14.67^{c}	6.25^{b}	2.64^{b}	2.33^{b}	5.66^{b}	68.77^{a}	611.37^{a}	489.9^{b}
Turning	16.0 ^{a}	5.57^{c}	1.69^{c}	2.0^{c}	5.11^{c}	59.64^{b}	432.99^{b}	340.0^{c}
Mean	15	6.12	5.28	2.67	5.9	61.44	556.29	593.20
CV (%)	2.72	0.99	2.87	10.65	0.95	1.83	4.76	6.80
LSD (P<0.05)	0.93	0.14	0.34	1.3	0.13	2.57	21.45	70.0

*Means with the same letter in column are not significantly different at P<0.05 level. Percentage were also recorded from the seeds of both dried and fresh pod of the red stage pods.

after maximum dry matter has been accumulated, as related in several species that produce seeds inside a fleshy fruit (Bewley and Black 1994). In these species the accumulation of osmotic solutes, primarily sugars, creates a low water potential inside the fruit that maintains high seed water content, but below the threshold required for germination (Welbaum and Bradford 1988).

However, among the seeds of dry pod, the highest germination was observed from the seeds of yellow stage followed by turning stage and least in red stage (Table 2). This may possibly be due to more accumulation of ABA in red ripe pods over the yellow and turning stage. Moreover, the establishment of dormancy during seed maturation is regulated by interactions between GA and ABA, whereby ABA induces dormancy and GA promotes germination (Holdsworth *et al*. 2008).

The mean germination of seeds from fresh pods was higher over the seeds of dried pods (Table 2). The decline in germination percentage of the seeds of dry pod may be due to loss of seed viability or mechanical resistance imposed by the endosperm on the radicle emergence. Watkins *et al*. (1983), reported mechanical resistance imposed by the endosperm on the radicle emergence of seeds rather than seed coat in pepper.

Further, the highest shoot length, root length, number of leaves and dry matter followed by yellow stage (Table 2). This may be due to more development of the seeds in red stage pods. Vidigal *et al*. (2011) also reported higher seedling vigour in sweet pepper harvested at red stage (75 days after anthesis).

However, the highest seedling vigour index both SVI-I and SVI-II was recorded from the seeds of red stage followed by yellow stage and turning stage (Table 2). This may be due to better shoot length, root length and dry weight of the seedlings. Similar findings were also reported by Vidigal *et al*. (2011) and Pereira *et al*. (2014) in sweet pepper. Thus, King-chilli pods for seed purpose should be harvested at fully developed red stage for the better germination and vigour of the seedlings. Santos *et al*. (2015) in Habanero pepper which is similar to King-chilli has also showed higher values for seed germination and vigour in the seeds obtained from the fruits harvested at 70 days after anthesis and subjected to rest for 7 days.

From above findings, it can be concluded that the seeds of King-chilli remain viable and can be stored up to one year. The optimum temperature for the seed germination is 25-30^{0}C. The synchronous, higher germination and vigorous seedling can be raised by pre-sowing seed treatments with KNO_3 (5%) for 24 hours. To store the seeds (dried pod) for next season farmers should harvest the King-chilli pods at fully developed yellow stage for the better germination and vigour of the seedlings. However, seed from fully red ripe fresh pods with 7 days resting period is ideal for current planting with higher germination and vigour.

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