



International Journal of Research in Agronomy

E-ISSN: 2618-0618
P-ISSN: 2618-060X
© Agronomy
NAAS Rating (2026): 5.20
www.agronomyjournals.com
2026; 9(5): 340-344
Received: 09-02-2026
Accepted: 13-03-2026

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Germination responses of *Argemone mexicana* to temperature, light conditions and seed burial depth

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DOI: <https://www.doi.org/10.33545/2618060X.2026.v9.i5e.5524>

Abstract

The study was conducted to assess the impact of abiotic factors, specifically temperature, light/darkness, and burial depth, on germination of fresh *Argemone mexicana* seeds during 2024 at Department of Seed Science and Technology, CCS HAU Hisar. The seeds were exposed to alternating temperatures of 15/20, 20/25 and 25/30 °C, along with two light regimes: 16 hour of light/8 hour of darkness and complete darkness. Results indicated that the highest germination rate (22.67%) occurred at 20/25 °C, while the lowest (9.33%) was at 25/30 °C. Strong seed dormancy was observed, with the highest proportion of hard seeds (88.67%) at 25/30 °C. At 15/20 °C, light conditions did not affect the proportion of hard seeds, with 82.66% recorded under both regimes. However, at higher temperatures, more hard seeds were found under complete darkness. The maximum seedling length (3.56 cm), dry weight (0.81 mg), Vigour Index-I (81.54), and Vigour Index-II (18.84) were observed at 20/25 °C, while the lowest values were at 25/30 °C. Germination and seedling growth were better under the light/darkness cycle compared to complete darkness. Radicle emergence began on 10th at 20/25 °C, on 14th at 15/20 °C and on 17th at 25/30 °C. No field emergence occurred due to strong morpho-physiological dormancy. This information is useful for weed scientist to understand the biology and effective weed management of *Argemone mexicana* which is objectionable weed of mustard crop.

Keywords: *Argemone mexicana*, burial depth, dormancy, germination, objectionable weed

Introduction

A wide range of annual and perennial weeds compete with crops, resulting in decreased yields. However, objectionable weeds pose a particularly serious problem in seed production. These weeds closely resemble the seed crop in appearance, making them difficult and costly to remove once they become mixed with the crop. Different objectionable weeds have been identified in various field crops such as *Phalaris minor* and *Convolvulus arvensis* in wheat, wild oat (*Avena fatua*) in oat, wild rice (*Oryza sativa*) in rice, *Argemone mexicana* in mustard, wild sunflower (*Helianthus spp.*) in sunflower, *Cichorium intybus* in berseem and *Cuscuta spp.* in Lucerne. According to the Indian Minimum Seed Certification Standards (Anonymous, 2013) ^[2] there is a maximum allowable limit for objectionable weed seeds per plants, with *Argemone mexicana* seeds not exceeding 2 per kilogram in foundation seeds and 5 per kilogram in certified seeds. *A. mexicana*, a member of the Papaveraceae family is commonly known as Mexican prickly poppy or Satyanashi. It grows naturally in barren fields and wastelands and is not cultivated or harvested like typical field crops. The plant differs significantly from mustard in all aspects. Due to its thorny nature including spiny leaves and capsules, handling the plant is difficult. The initial leaves are simple, alternate and arranged in a rosette, with green surfaces and white ribs. They are sessile, spatulate in shape, tapering at the base, about 6-8 cm long and 1 cm wide with four prominent teeth ending in short spines. The stem is covered with short hairs and a few long yellowish spines. Mature leaves are simple, alternate, sessile, blue-green, thick and leathery. The leaf blade is pinnately lobed, obovate, slightly clasping at the base, measuring 6-20 cm in length and 3-8 cm in width. Lobes are oblong and convoluted, with spiny teeth and pinnate venation. Whitish spines are present particularly on the underside while both leaf surfaces are glabrous. The flowers are large measuring 4-7 cm in diameter, with six bright yellow and rounded petals. They are borne singly either terminally or in the leaf axils, forming a solitary sessile inflorescence. The fruits are prickly, oblong to ovoid capsules measuring about 5 cm in length

and 2 cm in diameter, covered with thick yellowish spines. At maturity, the capsule splits open from the apex and is divided into five chambers containing numerous seeds. Capsules begin to develop from March onward with seeds enclosed inside. When dry, the capsules open at the top but the seeds are not released until the still-attached capsules bend downward. The seeds are produced in large numbers and are small, black and approximately 1.7–2 mm × 1.6 mm in size. The seeds resemble the seeds of *Brassica juncea* and thus can be adulterated with the black mustard and rendering it poisonous. Seeds are numerous, spherical, covered in a fine network of veins, brownish black and about 1 mm in diameter. Both *A. mexicana* and mustard seeds look almost alike and unless critically examined the difference is difficult to detect.

Different weeds have different ecology and need different environment for growth and development. As weeds are part of dynamic agricultural ecosystem, the knowledge of weed ecology is essential for sustainable management of weeds. The germination of weed seeds depend on various factors such as temperature, light, moisture and pH of growing medium. Water availability and temperature are key factors influencing seed germination. The optimum temperature for most seeds is between 15 and 30°C. By generating information on weed ecology we can know about requirement of weeds for their growth and development. This understanding can aid in developing effective cultural management practices for problematic weeds by either suppressing their germination or promoting it at a stage when seedlings can be easily controlled (Opena *et al.*, 2014).

These seeds can grow into plants that compete with crops for resources like sunlight, water and nutrients, ultimately affecting crop yields. Their presence can significantly impact seed quality and quantity, highlighting the importance of effectively managing and controlling these specific weeds to ensure optimal seed production. Light plays a crucial role in regulating dormancy termination and subsequent germination in several weed species. In natural settings, the light environment, primarily sensed by photoreceptors of the phytochrome family, provides vital cues for initiating germination under suitable environmental conditions. The spectral quality and intensity of light allow weed seeds to detect their burial depth, the presence of a light-intercepting canopy and disturbances caused by soil cultivation. The initiation of seed germination is strongly influenced by light and alternating temperature conditions, which are recognized as key environmental factors (Baskin and Baskin, 1988; Vandellook *et al.*, 2008) [3, 16]. Photochromes within an imbibed seed enable the assessment of competition intensity within its environment (Shinomura, 1997) [15]. Temperature, through its impact on seed physiology, plays a crucial role in breaking dormancy and has been noted to affect the rate and percentage of germination among different species (Presotto *et al.*, 2014) [13]. Deeper burial of seeds within the soil profile frequently results in reduced success rates for emergence and establishment, primarily due to the increased energy needed to reach the soil surface. The emergence depth may be influenced by the size of the seed, with larger seeds potentially having greater energy reserves, enabling them to emerge from deeper depths compared to smaller seeds.

Seed dormancy is an inherent characteristic that hinders germination, even under favourable environmental conditions (Batlla and Benech-Arnold, 2007; Fennor and Thompson, 2005) [4, 5]. Keeping the above aspects in mind, the present study was planned to study germination responses of *Argemone mexicana* to temperature, light conditions and seed burial depth.

Materials and Methods

Freshly harvested seeds of *A. mexicana* were collected from research farms and study was conducted at the laboratories of Department of Seed Science and Technology, Chaudhary Charan Singh Haryana Agricultural University, Hisar (India) during 2023-24. To study the effect of temperature and light on germination of *A. mexicana*, the seeds were exposed to the alternate temperatures of 15/20, 20/25 and 25/30 °C with two light regimes *i.e.*, 16 hours light/ 8 hours darkness and exposure to complete darkness for 24 hours.

Following observations were recorded

Germination (%)

The germination test was conducted through top of paper method (TP) in which three replications were placed at alternating temperature of 15/20, 20/25, and 25/30 °C in the germinator. Throughout the experiment, a 16/8 h light–dark cycle was maintained, with the lower temperature applied for 16 hours and the higher temperature for 8 hours. Illumination was provided by a 20-W cool white fluorescent bulb. Germination tests under light conditions were carried out in transparent Petri dishes. Seeds were monitored daily after the emergence of both the radicle and plumule until maximum germination was reached. The seeds were treated with GA₃ @ 250 ppm to break the dormancy. The germination was calculated using following formula suggested by Weise and Binning (1987):

$$\text{Germination (\%)} = \left[\frac{\text{Number of normal seedlings}}{\text{Total number of seedlings per replicate}} \times 100 \right]$$

Radicle emergence (%)

The radicle emergence test was conducted by using top of paper method. Weed seeds in three replicates were randomly placed in Petri dishes lined with adequately moistened Whatman No. 1 filter paper. The dishes were then kept in a controlled germinator set to alternating temperature regimes of 15/20 °C, 20/25 °C and 25/30 °C. The seeds that exhibited at least 2 mm radicle emergence were calculated and recorded at 24-hour interval, starting from the initiation of radicle emergence till maximum radicle emergence.

$$\text{Radicle emergence (\%)} = \left[\frac{\text{No. of seeds with 2 mm radicle length} \times 100}{\text{Total no. of seeds placed}} \right]$$

Seedling length (cm)

The seedling length including both root and shoot was measured in centimetres using a scale for 10 randomly selected normal seedlings from three replicates of the standard germination test. The mean length of these 10 seedlings was then calculated for final analysis. When germination was below 10%, the average seedling length was determined accordingly, following ISTA (2019) [6].

Seedling dry weight (mg)

During the germination test, 10 randomly selected normal seedlings were dried in a hot air oven at 80 ± 1 °C for 24 hours. The mean dry weight of these 10 seedlings was then recorded and expressed in milligrams (mg). In cases where germination was below 10%, the average weight of the germinated seedlings was calculated accordingly, following ISTA (2019) [6].

Hard seeds (%)

Seeds that remained hard and impermeable to water at the end of

the test period were classified as hard seeds. The percentage of hard seeds was calculated by dividing the number of hard seeds by the total number of seeds per replicate and expressing it as a percentage. Their presence was further confirmed using the tetrazolium (viability) test.

Vigour indices

The Vigour Index-I and Vigour Index-II were calculated by the methods suggested by Abdul-Baki and Anderson (1973) ^[1]:

Vigour Index-I = Standard germination (%) × Average seedling length (cm)

Vigour Index-II = Standard germination (%) × Average seedling dry weight (mg)

To assess the effect of burial depth, twenty-five seeds in three replications were sown in plastic pots (2 feet in length and half feet deep) at various depths *i.e.*, 4, 6 and 8 cm in soil which were put under open environment and following field parameters were recorded:

Speed of emergence

The number of seedlings emerged were counted on each day up to seedling establishment. The speed of emergence was calculated as described by Maguire (1962).

$$\text{Speed of emergence} = \left[\frac{\text{No. of seedlings emerged}}{\text{Day of first count}} + \dots + \frac{\text{No. of seedlings emerged}}{\text{Day of final count}} \right]$$

Field emergence (%)

Field emergence was assessed by counting the total number of seedlings after completion of emergence or when there was no further increase in total count of seedlings.

Statistical Analysis

Statistical analysis of data collected during the study was done by using the factorial complete randomized (CRD) design as described by Panse and Sukhatme (1985) ^[12]. All values described as the mean of the replicates with the evaluation of least significant difference (LSD) at 5% level of significance using the online statistical tool (OPSTAT) developed by Sheoran (2010) ^[14].

Results and Discussions

The results revealed that maximum germination (22.67%) was observed at alternate temperature of 20/25 °C followed by 15/20 °C (12.00%) and minimum (9.33%) was observed at 25/30 °C. More germination (17.33%) was observed under 16 hours light followed by 8 hours darkness as compared with exposure to complete darkness for 24 hours (12.00%). Maximum germination (28.00%) was recorded at 20/25 °C 16 hours light followed by 8 hours darkness while minimum (8.00%) was recorded at 25/30 °C under complete darkness. It is found that optimum temperature for *A. mexicana* lies in the range of 20-30 °C in light. Since *A. mexicana* is winter annual herb, it requires low temperature for its germination and growth. The results are supported by Karlsson *et al.* (2003) ^[7]. Minimum germination was found in *A. mexicana* among all the four weed species taken because they have strong morphological dormancy

according to Nikolaeva (1977) ^[10] and Baskin and Baskin (1998) ^[3]. Maximum hard seeds (88.67%) were found at alternate temperature of 25/30 °C, followed by 15/20 °C (82.66%). At alternate temperature of 15/20 °C, no effect of light/ darkness was observed and same percentage of hard seed (82.66%) was found in both the situations. But at higher alternate temperatures more hard seeds were recorded under complete darkness. Overall, seeds exposed to complete darkness for 24 hours recorded more hard seeds over the seeds exposed to 16-hour light/8-hour darkness indicating role of light in combating the dormancy in *A. mexicana* seeds.

Maximum seedling length (3.56 cm) was observed at an alternating temperature of 20/25 °C whereas minimum seedling length (2.45 cm) was observed at 25/30 °C. Between the light regimes, longer seedling length (3.06 cm) was observed under exposure to 16-hour light/8-hour darkness as compared to complete darkness (2.77 cm). The maximum seedling length (3.77 cm) was recorded at 20/25 °C under 16-hour light/8-hour darkness while minimum (2.25 cm) was recorded at 25/30 °C under complete darkness.

The maximum seedling dry weight (0.81 mg) was recorded at 20/25 °C, followed by 15/20 °C (0.66 mg) and minimum seedling dry weight (0.44 mg) was recorded at 25/25 °C. More seedling dry weight (0.79 mg) was observed under 16-hour light/8-hour darkness than complete darkness (0.56 mg). Maximum Vigour Index-I (81.54) was observed at 20/25 °C followed by 15/20 °C (32.75). The temperature that exhibited the minimum Vigour Index-I (23.02) was 25/30 °C. More Vigour Index-I (56.62) was recorded under 16-hour light/8-hour darkness than under complete darkness (34.93). Maximum Vigour Index-I (105.30) was recorded at 20/25 °C under 16-hour light/ 8 hours darkness. Similarly, maximum Vigour Index-II (18.84) was observed at 20/25 °C followed by 15/20 °C (7.91). Minimum Vigour Index-II (5.35) was recorded at 25/30 °C. More Vigour Index-II (14.32) was recorded under 16-hour light/8-hour darkness than under complete darkness for 24 hours (7.08). Maximum Vigour Index-II (25.54) was recorded at 20/25 °C under 16-hour light/8-hour darkness.

Radicle emergence was started on 14th day at 15/20 °C and on 17th day at 25/30 °C while it was started after 10th day at 20/25 °C. Maximum radicle emergence (24.17%) was observed at 20/25 °C followed by 15/20 °C (9.24%) and minimum recorded at 25/30 °C (4.13%). More radicle emergence (14.55%) was recorded 16-hour light/8-hour darkness than under complete darkness (10.49%). Maximum radicle emergence was obtained on 18th day 15/20 °C and 20/25 °C but radicle emergence was continued up to 24th day at higher alternate temperature *i.e.*, 25/30 °C. Delayed radicle protrusion was found in *A. mexicana* which may be due to presence of growth inhibitors. The presence of alkaloids such as sanguinarine, prior to germination may lead to decreased germination and delayed radicle protrusion in *A. mexicana*. Similar result was favoured by Kucera *et al.* (2005) ^[8] in eudicot seeds. No field emergence was observed for *A. mexicana* at 4, 6 and 8 cm depth. The seed remain dormant and did not emerge from the deeper depth in the required period of time. The freshly harvested seeds of *A. mexicana* showed strong dormancy and tend to remain persistent for several years. The results are in conformity with Manalil and Chauhan, 2021 ^[9]. The seeds showed physiological or morpho-physiological dormancy. This information may be used by weed scientist that may be helpful in management of this weed.

Table 1: Effect of temperature and light/darkness on germination, hard seeds and seedling length of *Argemone mexicana* seeds

Alternate Temperatures (T)	Germination (%)			Hard seeds (%)			Seedling length (cm)		
	L ₁	L ₂	Mean	L ₁	L ₂	Mean	L ₁	L ₂	Mean
15/20 °C	13.33	10.66	12.00	82.67	82.67	82.67	2.75	2.71	2.73
20/25 °C	28.00	17.33	22.67	61.33	76.00	68.67	3.77	3.36	3.56
25/30 °C	10.66	8.00	9.33	88.00	89.33	88.67	2.66	2.25	2.45
Mean	17.33	12.00		77.33	82.67		3.06	2.77	
LSD (P=0.05)	T=1.67, L=1.36, T × L=2.36			T=4.32, L=3.53, T × L=6.11			T=0.54, L=NS, T × L= NS		

Where L₁ =16 hours followed by 8 hours darkness and L₂= exposure to complete darkness for 24 hours

Table 2: Effect of temperature and light/darkness on seedling dry weight and vigour indices of *Argemone mexicana* seeds

Alternate Temperatures (T)	Seedling dry weight (mg)			Vigour Index-I			Vigour Index-II		
	L ₁	L ₂	Mean	L ₁	L ₂	Mean	L ₁	L ₂	Mean
15/20 °C	0.79	0.52	0.66	36.50	29.00	32.75	10.27	5.55	7.91
20/25 °C	0.91	0.70	0.81	105.30	57.79	81.54	25.54	12.13	18.84
25/30 °C	0.66	0.44	0.55	28.05	18.00	23.02	7.15	3.55	5.35
Mean	0.79	0.56		56.62	34.93		14.32	7.08	
LSD (P=0.05)	T=0.07, L=0.06, T × L=NS			T=3.95, L=3.22, T × L=5.58			T=0.76, L=0.62, T × L=1.07		

Where L₁ =16 hours light followed by 8 hours darkness and L₂= exposure to complete darkness for 24 hours

Table 3: Effect of temperature, light/darkness and duration on radicle emergence (%) of *Argemone mexicana* seeds

Temperature (T)	15/20 °C		20/25 °C		25/30 °C		Mean
Duration (D) ↓ Light (L) →	L ₁	L ₂	L ₁	L ₂	L ₁	L ₂	
10 th day	0.00	0.00	1.33	0.00	0.00	0.00	0.22
11 th day	0.00	0.00	10.67	1.33	0.00	0.00	2.00
12 th day	0.00	0.00	16.00	10.67	0.00	0.00	4.44
13 th day	0.00	0.00	20.00	21.33	0.00	0.00	6.89
14 th day	6.67	0.00	22.67	22.67	0.00	0.00	8.67
15 th day	10.67	5.33	24.00	22.67	0.00	0.00	10.44
16 th day	13.33	9.33	29.33	24.00	0.00	0.00	12.67
17 th day	16.00	10.67	36.00	24.00	2.00	0.00	14.78
18 th day	16.00	13.33	38.67	24.00	4.00	4.00	16.67
19 th day	16.00	13.33	38.67	24.00	4.67	4.33	16.83
20 th day	16.00	13.33	38.67	24.00	6.00	7.67	17.61
21 st day	16.00	13.33	38.67	24.00	9.00	8.67	18.28
22 nd day	16.00	13.33	38.67	24.00	10.67	10.33	18.83
23 rd day	16.00	13.33	38.67	24.00	12.33	11.67	19.33
24 th day	16.00	13.33	38.67	24.00	16.67	12.00	20.11
Mean	10.58	7.91	28.71	19.64	4.36	3.91	
LSD (P=0.05)	T=0.79, L=0.65, D= 1.77 T × L=1.12, T × D=3.07, L × D=2.51, T × L × D=4.34						
Mean	T(15/20 °C) =9.24, T (20/25 °C) =24.17, T(25/30 °C) =4.13, L ₁ = 14.55, L ₂ =10.49						

Where L₁ =16 hours light followed by 8 hours darkness and L₂= exposure to complete darkness for 24 hours

Conclusion

The optimum temperature for germination of *Argemone mexicana* seeds is alternating temperature of 20/25 °C under 16 hours light/8 hours darkness. Strong morpho-physiological dormancy was also recorded in seeds. No radicle emergence was recorded before 10th day. The maximum radicle emergence was recorded at alternate temperature of 20/25 °C under 16 hours light followed by 8 hours darkness with maximum peak on 18th day. No field emergence was recorded in seeds at any depth, due to strong physiological or morpho-physiological dormancy.

Acknowledgment

The authors would like to express their gratitude to Department of Seed Science and Technology, CCS Haryana Agricultural University, Hisar for providing the necessary facilities to conduct the research.

Finding

This research paper did not receive any specific funding or financial support. The research work was conducted as part of Department of Seed Science and Technology, CCS HAU Hisar's

ongoing research activities and with available resources.

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