

## Postprint: Seed Germination Responses of the Invasive Plant *Xanthium italicum* to Environmental Factors

**Authors:** He Ying, Ma Miao

**Date:** 2018-03-08T00:00:00+00:00

### Abstract

Under controlled indoor conditions, the response of *Xanthium italicum* seed germination to environmental factors (external temperature, light, soil moisture, soil salinity, and soil pH) was investigated. The results indicated that *Xanthium italicum* possesses a relatively broad tolerance amplitude to all environmental factors; seeds germinated across temperatures of 10-35°C, with an optimal temperature of 25°C. The seed germination stage exhibited no strict light requirement, with germination rates exceeding 93% under both light and dark conditions. *Xanthium italicum* demonstrated extremely strong tolerance to soil moisture; in soil environments with relative humidity ranging from 30% to 100%, over 70% of seeds could germinate normally. Its seed germination rate decreased with increasing soil salinity concentration, yet one-third of seeds could still germinate in a soil NaCl environment of 280 mmol/L. Its seeds showed strong tolerance to soil pH; in soil environments with pH values of 4-10, seed germination rates were all above 90%. *Xanthium italicum* exhibited extremely strong tolerance to temperature, light, soil moisture, salinity, and soil pH during the seed germination stage, indicating that cool plateau mountainous areas, hot basin deserts, open lands, closed forests and fields, arid deserts, humid river valley wetlands, normal soils, and saline soils are all suitable habitats for *Xanthium italicum* seed germination. Given the broad spectrum of its suitable habitats and the severity of its ecological risk, comprehensive strict monitoring of *Xanthium italicum* is recommended across all habitat types, with immediate eradication upon detection.

## Full Text

### Preamble

ACTA ECOLOGICA SINICA

Vol. 38, No. 4, Feb. 2018

DOI: 10.5846/stxb201701120098

### Responses of Seed Germination of the Invasive Plant *Xanthium italicum* to Environmental Factors

HE Ying, MA Miao

College of Life Sciences, Shihezi University, Shihezi 832003, China

### Abstract

We investigated the responses of *Xanthium italicum* seed germination to environmental factors—including temperature, light, soil moisture, soil salinity, and soil pH—under controlled laboratory conditions. Our results demonstrated that *X. italicum* exhibits broad tolerance to environmental fluctuations. Seeds germinated across a wide temperature range of 10–35°C, with 25°C being optimal. Germination percentages exceeded 93% under both light and dark conditions, indicating no strict light requirement during germination. Seeds showed strong tolerance to soil moisture, with many germinating at relative humidities of 30%–100%. Although germination percentage decreased with increasing soil salinity, one-third of seeds still germinated even at high salinity levels of 280 mmol/L NaCl. Most seeds germinated across a wide pH range, with germination percentages above 90% at soil pH 4–10. These findings reveal that *X. italicum* possesses strong adaptability to temperature, light, soil moisture, salinity, and pH during germination. Seeds can vigorously germinate in drastically different habitats, including frigid high-altitude mountains, hot basin deserts, open terrain, forest canopy, arid deserts, humid river valleys, saline-neutral soils, and alkaline soils. Given the species' serious ecological hazard and the universality of its suitable habitats, we recommend strict monitoring across all habitat types and immediate eradication upon detection.

**Keywords:** invasive plant; *Xanthium italicum*; seed; germination; tolerance

## 1. Experimental Materials

Experimental materials consisted of fully mature *Xanthium italicum* fruits collected in October from Kuitun City, Xinjiang (84°53 E, 44°27 N). The soil at the collection site had a pH of 7.84 and electrical conductivity of 202 dS/m. Fruits were quickly transported to the laboratory after collection and air-dried. Uniform-sized fruits were selected for experiments. In this study, “fruits” refer to the involucre-enclosed infructescences of *X. italicum*, each containing two achenes (one large, one small) with one seed per achene. Fruits were soaked in water at room temperature. Because *X. italicum* exhibits asynchronous germination

between upper and lower seeds—with upper seeds having longer dormancy and delayed germination compared to lower seeds—we eliminated this effect by clipping the beak tip of the involucre. When lower seeds germinated, cotyledons emerged through the small hole left by beak removal, pushing the involucre and ungerminated upper seed above the soil surface. Therefore, germination statistics counted only one seed per fruit.

## 2. Experimental Methods

Germination experiments were conducted in colorless transparent plastic germination boxes (20 cm × 12 cm × 5 cm) filled with nutrient soil. Boxes were placed horizontally with a burial depth of 1 cm (except for light quality treatments). Evaporation losses were compensated by adding distilled water via weighing to maintain soil humidity at 70% (except for soil moisture treatments). Temperature was controlled at 20°C during the day and 15°C at night (except for temperature treatment groups). All boxes were placed in constant-temperature illumination incubators with light intensity of 450 mol · m<sup>2</sup> · s<sup>-1</sup>.

### 2.1 Temperature Effects on *Xanthium italicum* Seed Germination

Constant-temperature illumination incubators were set to six temperature gradients: 10°C, 15°C, 20°C, 25°C, 30°C, and 35°C for seed germination experiments.

### 2.2 Light Quality Effects on *Xanthium italicum* Seed Germination

Three different light quality treatments were applied. In germination boxes placed horizontally, fruits were positioned with their upper surface level with the soil surface but not covered by soil. The control group used unwrapped boxes to simulate fruits on open ground. Green transparent plastic film was used to wrap boxes for simulating green-light-rich environments under forest canopy or field conditions. Black plastic bags wrapped boxes to simulate complete darkness in buried soil environments.

### 2.3 Soil Moisture Effects on *Xanthium italicum* Seed Germination

Nutrient soil was prepared at seven moisture gradients with relative humidities of 30%, 40%, 50%, 60%, 70%, 80%, and 90% for seed germination experiments.

### 2.4 Soil Salinity Effects on *Xanthium italicum* Seed Germination

Six NaCl concentration gradients were established: 0 (CK), 80, 160, 200, 240, and 280 mmol/L. Soil relative humidity was maintained at 70%, and fruits were placed for germination experiments.

### 2.5 Soil pH Effects on *Xanthium italicum* Seed Germination

Solutions of pH 2, 4, 6, 7, 8, 10, and 12 were prepared and thoroughly mixed with equal amounts of nutrient soil. Soil relative humidity was maintained at

70%, and fruits were placed for germination experiments.

## 2.6 Measurement Methods

Germination was defined as cotyledons emerging above the soil surface. Moldy seeds were considered non-viable. Germination rate and germination index were calculated as follows:

Germination rate = (Number of germinated seeds / Total seeds)  $\times$  100%

Germination index =  $\Sigma(G / D)$ , where G is the number of germinated seeds on day t and D is the germination day.

Germination duration was observed and recorded. The number of germinated seeds was counted daily.

## 3. Data Processing

SPSS 19.0 software was used for statistical analysis of experimental data. Repeated measures ANOVA was performed on cumulative germination rate data. One-way ANOVA was conducted on germination index and cumulative germination rate data on day 15 under different treatments, followed by Duncan's multiple range test to detect significant differences between treatments. Origin 9.0 was used for data visualization.

## 4. Results

### 4.1 Effects of Temperature on *Xanthium italicum* Seed Germination

Both germination index and cumulative germination rate showed highly significant differences among temperature conditions and germination stages ( $p < 0.001$ ), with a highly significant interaction between germination time and temperature. Seeds germinated across a broad temperature range (10-35°C). Although initial germination was slower at 10°C (22.2% germination index), both germination index and rate peaked at 25°C (19.07 and 100%, respectively). When temperature reached 30°C, germination rate remained high at 96%, but both indices began to decline significantly ( $p < 0.05$ ). At 35°C, germination rate could still maintain 71%. The earliest initial germination occurred at 25°C, while other temperatures showed varying degrees of delayed germination. Higher temperatures favored earlier germination, and the delay effect became more pronounced with greater deviation from optimal temperature. At low temperatures (10-20°C), seed germination was inhibited, with delays of 3-8 days.

Table 1. Variance analysis of cumulative germination index of *X. italicum* under different temperatures in 1-15 days

Table 2. Variance analysis of cumulative germination percentage of *X. italicum* under different temperatures in 1-15 days

[Figure 1: see original paper] Figure 1. Effect of temperature on germination index of *X. italicum*

[Figure 2: see original paper] Figure 2. Effect of temperature on cumulative germination percentage of *X. italicum*

#### 4.2 Effects of Light Quality on *Xanthium italicum* Seed Germination

Light quality had highly significant effects on seed germination ( $p < 0.001$ ), with highly significant differences in germination index and cumulative germination rate among stages and a highly significant interaction between germination time and light quality. Seeds showed high germination potential across all three light treatments. However, germination speed was significantly faster in darkness than in other treatments ( $p < 0.05$ ). Germination rates under darkness, green light, and full-spectrum light were 96%, 93%, and 69%, respectively, indicating high germination rates in both dark and open-ground conditions, with fastest germination in darkness. Seeds under forest canopy showed lower germination rates.

Table 3. Variance analysis of cumulative germination index of *X. italicum* under different light conditions in 1-15 days

Table 4. Variance analysis of cumulative germination percentage of *X. italicum* under different light conditions in 1-15 days

[Figure 3: see original paper] Figure 3. Effect of light on germination index of *X. italicum*

[Figure 4: see original paper] Figure 4. Effect of light on cumulative germination percentage of *X. italicum*

#### 4.3 Effects of Soil Moisture on *Xanthium italicum* Seed Germination

Soil moisture had highly significant effects on seed germination ( $p < 0.001$ ). Significant differences existed in germination index and cumulative germination rate among different stages, with a highly significant interaction between germination time and soil moisture. Seeds germinated across all moisture conditions. Even at 30% relative humidity, germination index reached relatively high levels (10.63-11.26). At 50% relative humidity, germination index peaked (14.72). Germination rate remained high at 71% under waterlogged conditions (100% relative humidity). Seeds showed strong tolerance to both drought and waterlogging extremes.

Table 5. Variance analysis of cumulative germination index of *X. italicum* under different soil moisture treatments in 1-15 days

Table 6. Variance analysis of cumulative germination percentage of *X. italicum* under different soil moisture treatments in 1-15 days

[Figure 5: see original paper] Figure 5. Effect of soil moisture on germination index of *X. italicum*

[Figure 6: see original paper] Figure 6. Effect of soil moisture on cumulative germination percentage of *X. italicum*

#### 4.4 Effects of Soil Salinity on *Xanthium italicum* Seed Germination

Soil salinity concentration had highly significant effects on seed germination ( $p < 0.001$ ). Significant differences existed in germination index and cumulative germination rate among different stages, with significant interaction between time and salinity. Seeds exhibited broad salinity tolerance during germination. At 80 mmol/L NaCl, germination index and rate decreased but final germination showed no significant difference from the control ( $p > 0.05$ ). At 80-200 mmol/L, although both indices decreased significantly compared to the control, final germination rate remained above 71%. At high salinity of 280 mmol/L, germination was significantly inhibited with slower speed, yet one-third of seeds could still germinate.

Table 7. Variance analysis of cumulative germination index of *X. italicum* under different soil salinity treatments in 1-15 days

Table 8. Variance analysis of cumulative germination percentage of *X. italicum* under different soil salinity treatments in 1-15 days

[Figure 7: see original paper] Figure 7. Effect of NaCl concentrations on germination index of *X. italicum*

[Figure 8: see original paper] Figure 8. Effect of NaCl concentrations on cumulative germination percentage of *X. italicum*

#### 4.5 Effects of Soil pH on *Xanthium italicum* Seed Germination

Soil pH had highly significant effects on seed germination ( $p < 0.001$ ). Significant differences existed in germination index and cumulative germination rate among different stages, with highly significant interaction between germination time and pH. Seeds showed strong pH tolerance. Germination speed and rate increased with pH from 2 to 7, reaching maximum rates above 90% at pH 4-10. Beyond pH 10, both indices began to decline, yet germination rate remained above 71% even at pH 12. The optimal pH range was 4-10.

Table 9. Variance analysis of cumulative germination index of *X. italicum* under different soil pH values in 1-15 days

Table 10. Variance analysis of cumulative germination percentage of *X. italicum* under different soil pH values in 1-15 days

[Figure 9: see original paper] Figure 9. Effect of soil pH value on germination index of *X. italicum*

[Figure 10: see original paper] Figure 10. Effect of soil pH value on cumulative germination percentage of *X. italicum*

## 5. Discussion

Biological invasion has become a serious threat to China's ecological and biological security, causing annual economic losses exceeding 100 billion RMB. Weak public awareness of ecological protection and inadequate government prevention measures are primary causes. Effective prevention and control of alien species

have become hot topics among governments, academia, and the public worldwide. *Xanthium italicum* is one of the fastest-spreading invasive alien species in China, with Bole and Changji as its diffusion centers in Xinjiang.

Population establishment is crucial for successful invasion, and seed germination provides the foundation for rapid population growth. Environmental factors directly affect seed germination capacity and determine invasion speed and population size. This study shows that while temperature, light, soil salinity, and pH affect *X. italicum* seed germination, the species exhibits remarkable ecological adaptability.

The wide temperature amplitude (10–35°C) with optimal germination at 25°C indicates seeds can germinate from early spring to midsummer, meeting temperature requirements in both hot basin deserts and cool high-altitude regions. High germination rates (>93%) in both darkness and full-spectrum light demonstrate photoneutral seeds that germinate equally well on open ground or buried in soil, differing from other Asteraceae invaders that require light. This contributes to rapid population expansion. However, green light significantly inhibited germination (69% vs. 96% in darkness), possibly an adaptive strategy to avoid competition under dense canopies.

The broad soil moisture tolerance (30–100% relative humidity) indicates seeds can germinate in both arid deserts and humid river valleys, another key factor for rapid expansion. Regarding salinity, germination rate decreased with increasing salt concentration, yet one-third of seeds germinated at 280 mmol/L NaCl, showing stronger salt tolerance than native *Xanthium sibiricum*. Spring snowmelt and rainfall in northern Xinjiang leach surface salts, potentially facilitating germination in saline soils.

Soil pH tolerance was also broad, with >90% germination at pH 4–10 and >71% even at pH 12, consistent with studies on native *Xanthium* species. Xinjiang's extensive saline-alkali soils, with low plant richness and weak disturbance resistance, may become potential distribution areas for *X. italicum*.

Given the species' broad habitat suitability and severe ecological risk, we recommend strict monitoring across all habitat types and immediate eradication upon detection. Since green light significantly inhibits germination, planting early-germinating species to create canopy cover could suppress *X. italicum* seed germination in invaded areas.

## References

- [1] Xinjiang Agricultural Sciences, 2012, 49(1): 86-100.
- [2] Biodiversity, 2001, 9(4): 430-438.
- [3] Xinjiang Agricultural Sciences, 2012, 49(5): 879-886.
- [4] Andreani S, Barboni T, Desjobert JM, Paolini J, Costa J, Muselli A. Essential oil composition and chemical variability of *Xanthium italicum* Moretti from Corsica. Flavour & Fragrance Journal, 2012, 27(3): 227-236.

- [5] Shao H, Huang XL, Wei XY, Zhang C. Phytotoxic effects and a phytotoxin from the invasive plant *Xanthium italicum* Moretti. *Molecules*, 2012, 17(4): 4037-4046.
- [6] Tsankova ET, Trendafilova AB, Kujungiev AI, Galabov AS, Robeva PR. Xanthanolides of *Xanthium italicum* and their biological activity. *Zeitschrift Für Naturforschung C: A Journal of Biosciences*, 1994, 49(1/2): 154-155.
- [7] Kovács A, Vasas A, Forgó P, Réthy B, Zupkó I, Hohmann J. Xanthanolides with antitumour activity from *Xanthium italicum* Moretti. *Zeitschrift Für Naturforschung C: A Journal of Biosciences*, 2007, 64(5/6): 343-349.
- [8] *Allelopathy Journal*, 2004, 13(2): 189-199.
- [9] *Cereal Research Communications*, 2009, 37(S1): 77-80.
- [10] *Plant Ecology*, 2010, 206(2): 309-319.
- [11] *Plant Biosystems*, 2010, 144(1): 47-52.
- [12] Beijing Flora. Beijing Press, 1993: 1504-1505.
- [13] *Chinese Journal of Plant Protection*, 2007, (2): 57-59.
- [14] *Chinese Journal of Plant Protection*, 2015: 36.
- [15] *Journal of Ecology and Rural Environment*, 2014: 233-233.
- [16] *Chinese Journal of Plant Protection*, 2015, 32(2): 36-41.
- [17] *Acta Ecologica Sinica*, 2010, 30(13): 3433-3440.
- [18] *Proceedings: Biological Sciences*, 2002, 269(1487): 151-155.
- [19] *Acta Ecologica Sinica*, 2013, 33(6): 1711-1716.
- [20] *Acta Ecologica Sinica*, 2009, 29(4): 1947-1953.
- [21] *Journal of Arid Land Resources and Environment*, 2012, 26(12): 54-58.
- [22] *Journal of Desert Research*, 2005, 25(9): 2389-2398.
- [23] *Xinjiang Plant Quarantine Pests*. China Quality Inspection Press, 2012: 112-113.
- [24] *Journal of Chongqing University of Technology*, 2012, 32(12): 3825-3833.

*Note: Figure translations are in progress. See original paper for figures.*

*Source: ChinaXiv – Machine translation. Verify with original.*