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# Seed Germination of Three Dominant Artemisia Species in Loess Hilly Region: Alternating Temperature, Osmotic Potential and Light Effects

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## Abstract

In this study, we evaluated the effects of alternating temperature, osmotic potential and light in the achene germination of three dominant Artemisia species (*A.scoparia*, *A.gmelinii* Web. and *A.giraldii* Pamp.) on arable old land after being abandoned in Loess hilly region of Northern Shaanxi, in order to provide information about germination requirements which could be use for conservation studies. Results showed that *A.scoparia* had wider alternating temperature extremes than two other species, and *A.giraldii* Pamp. and *A.gmelinii* Web. had lower optimum alternating temperature (25 °C / 20 °C) than *A.scoparia*. (30 °C / 25 °C). Osmotic potential affected germination of all species significantly, but *A.scoparia*. had better germination results than others in every treatment. In light treatments, although *A.giraldii* Pamp. and *A.gmelinii* Web. had lower germination proportion in white light, they had bigger germination proportion in darkness than *A.scoparia*.

**Keywords:** arable old land after being abandoned; Artemisia; germination; seed storage; mucilaginous layer

## 1. Introduction

Artemisia is the annual or perennial shrub, undershrub or grass of Compositae (Jane O., 2007), and it is the most important component of vegetation covering on the Loess Plateau, specially on the arable old land after being abandoned in Loess hilly region. As dominant species there, Artemisia has a very important aspect on the local vegetation construction and succession (Cheng J.M et al, 2001). So, study on Artemisia spreading on arable old land after being abandoned in Loess hilly region plays a very important role of developing vegetation recovery and management strategies for the Loess Plateau. During the past research of ecological restoration in this area (WANG K.Q., WANG B.R., 2002), most researchers focused on Leguminosae (ZHAO X.Y et al, 2005), Gramineae and water/fertility conditions (WEN et al, 2005), and some of them focused on typical steppe (LIU Z.G., LI Z.Q., 2004) and moving dunes of Ordos (GUO K., 2000; XIAO C.W., et al, 2001), however, reports about Artemisia inhabiting on the arable old land after being abandoned in Loess hilly region are limited, and few of them did research on the growth characters and ecological mechanisms of Artemisia directly, but just explained them as some companion species in some period of vegetation succession in local region (JIAO J.Y., et al, 2005; HOU F.J., et al, 2002). All three Artemisia species studied in this report have dominant ecological status at local area. *A.scoparia* is a dominant species at the primary period of vegetation succession on local arable old land after being abandoned, and affects the water content and soil fertility a lot at habitats (DU F., et al, 2006). *A.gmelinii* Web. spreads widely locating North Shannxi as the abundance distribution center, and trend to form dominant species at the metaphase of succession. *A.giraldii* Pamp. spreads widely at middle-high altitude area and grows with other species well than others. So, all three Artemisia species have their own characters and this study of them could help us to explain ecological meaning of them.

Besides, seed germination is a very important stage of plant life history, high germination rate always means high seedling rate and resource occupancy. In this way, seed germination studies are key tools in conservation programs because they can be used for management programs and species reintroduction. Specially for *Artemisia* species, since they used to produce a mass of seed, high germination rate decides if they could occupy large part of natural resources and being a dominant species. And for seed germination, temperature, water potential and light are three most important resources. So, the aim of our study was to determine the effect of alternating temperature, osmotic potential and light upon the seed germination of three *Artemisia* species (*Artemisia gmelinii* Web., *Artemisia scoparia*. and *Artemisia giraldii* Pamp.). To fulfill this purpose, we formulated the following questions: (1) Will alternating temperature extremes affect achene germination of those species? (2) Will osmotic potential inhibit achene germination of those species? (3) Will those *Artemisia* species show either a positive photoblastic or an indifferent behavior to light? We expect the results of the experiments could illustrate the germination characters and mechanisms of those three *Artemisia* species affected by those three environmental resources, and offer a scientific way for us to ecological restoration at local region.

## 2. Methods and Materials

### 2.1 Study Site

The study area is located at Gaoqiao village, Ansai county, arable old land after being abandoned in Loess hilly region in Northern Shannxi. This area belongs to mediate temperate semiarid continental monsoon climate, and the annual mean temperature is 7.7~10.6, the dry degree is 1.2 (DU, 2004). The annual mean hours of sunshine is 2397.3h, the annual total quantity of radiation is 117.74kcalcm<sup>-2</sup>. The mean annual precipitation is about 513mm, and the highest rainfall occurs from June to August (60%~80% of the annual total, RAN L.G., et al, 2006). The soil type is loess, which originate from loess parent materials (ZHANG J.T., 2005). Based on the system of national vegetation regionalization, this area is classified in the forest steppe. We collected seeds of three *Artemisia* species in this area and the table 1 is the details about the locations of collection (Measured at August, 2006).

Table 1 Land abandoned time, geographic locations, weight of thousand seeds, seed water content and seed size of three *Artemisia* species. Seed size is indicated(n=20)

|                                   | <i>A. gmelinii</i> Web.   | <i>A. scoparia</i>        | <i>A. giraldii</i> Pamp.  |
|-----------------------------------|---------------------------|---------------------------|---------------------------|
| Land abandoned time (year)        | 30                        | 3                         | 24                        |
| Altitude (m)                      | 1277±4.6                  | 1144±4.4                  | 1306±5.8                  |
| Location                          | E109° 11.880'N36° 39.662' | E109° 11.539'N36° 39.370' | E109° 11.915'N36° 39.538' |
| Slope                             | 31.6°                     | 9°                        | 52.9°                     |
| Weight of thousand seeds (g±S.E.) | 0.132±0.0009              | 0.032±0.0009              | 0.079±0.0016              |
| Seed water content (%±S.E.)       | 6.089±0.0980              | 6.258±0.1641              | 4.897±0.0447              |
| Seed size (mg±S.E.)               | 0.082±0.02                | 0.073±0.1                 | 0.079±0.05                |
| Imbibition rate (%±S.E.)          | 1665.192±14.3236          | 922.548±41.1536           | 1212.147±9.9188           |

## 2.2 Materials and Methods

### 2.2.1 Seed collection

Achenes of each *Artemisia* species were collected from 40 mature fruits from 20 individuals from Loess hilly region in Northern Shaanxi in November 2006(see Table 1). In the laboratory,

achenes were extracted from the fruits and put into paper bags and stored in darkness under natural temperature ( $\leq 5$  °C) and dry condition until March 2007. All achenes were used for experiments in which the effect of light, temperature and osmotic potential solutions were evaluated.

### **2.2.2 Alternating temperature treatments**

To determine the effect of alternating temperature upon the germination of achenes of three *Artemisia* species, experiment with a complete random design with five replicates per treatment were made. For each replicate of each treatment, 100 achenes were sown inside Petri dish (120 mm in diameter) on the surface of No.1 Whatman filter paper wet by distilled water. Dishes were incubated in growth chambers at five alternating temperatures ranging from 10 °C / 5 °C to 30 °C / 25 °C, every 5 °C (i.e. 10 °C / 5 °C, 15 °C / 10 °C, 20 °C / 15 °C, 25 °C / 20 °C and 30 °C / 25 °C) with a thermoperiod of 14/10 h and a 12 h photoperiod.

### **2.2.3 Osmotic potential solutions treatments**

To determine the effect of osmotic potential solutions at five concentrations (0, -0.2, -0.4, -0.6, -1.2 MPa) in white light at 25 °C upon the germination of achenes of three *Artemisia* species, experiments were made with a complete random design with five replicates. Each replicate consisted of 100 achenes sown inside Petri dishes (120 mm in diameter) on the surface of No.1 Whatman filter paper. The treatments were as follows: (1) white light with distilled water only (0 MPa), (2) white light with osmotic potential solution at -0.2 MPa, (3) white light with osmotic potential solution at -0.4 MPa, (4) white light with osmotic potential solution at -0.6 MPa, (5) white light with osmotic potential solution at -1.2 MPa. The equation of osmotic potential has reported by Burlyn and Kaufmann (Burlyn E.M., Merrill R.K., 1973), and the equation of germination rate has reported by Wang M. (WANG M., 2003).

### **2.2.4 Light treatments**

With the aim to assess the light effect on the germination of achenes of three *Artemisia* species, experiments were made with a complete random design with five replicates per treatment. Each replicate consisted of 100 achenes sown inside Petri dishes (120 mm in diameter) on the surface of No.1 Whatman filter paper wet by distilled water. Light treatment were incubated in growth chambers at 25 °C / 20 °C alternating temperature with a thermoperiod of 14/10 h and a 12 h photoperiod. Darkness treatment were wrapped with two aluminum foil sheets and incubated in growth chambers at the same alternating temperature. All experiments were followed for 20 days and germination was considered once the radicle emerged from the testa. The response variables were the proportion of germinated achenes at the end of each experiment.

### **2.2.5 Statistical analysis**

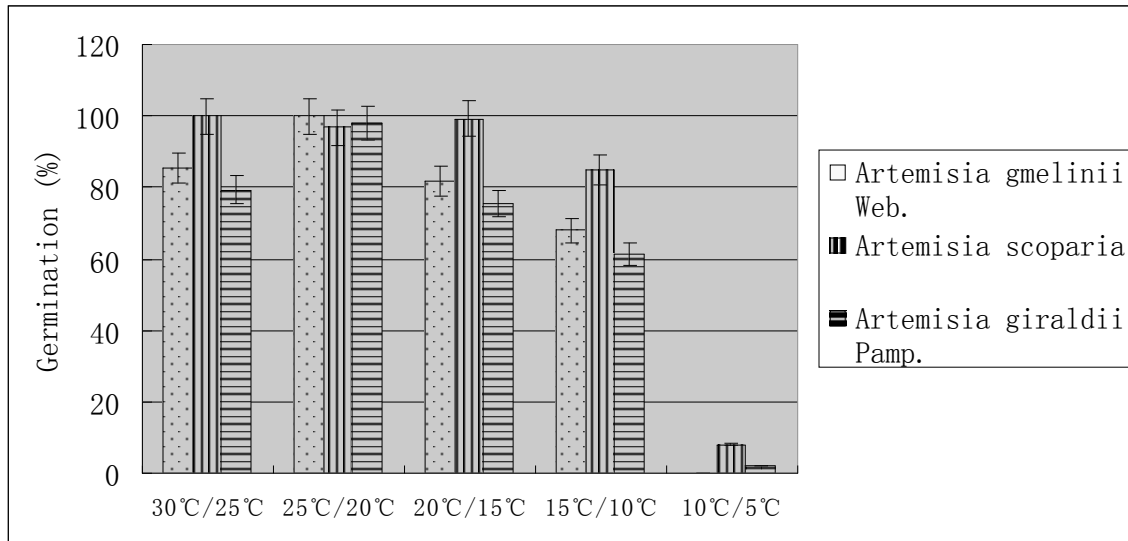
For all experiments of alternating temperature, osmotic potential solutions and light, data were tested for statistical significance using an ANOVA (Zar, J.H., 1996) with the statistical program SPSS 15.0.

## **3. Results**

### **3.1 Alternating temperature treatments**

The optimal temperature for achene germination differed with species ( $P < 0.0001$ ). Maximum germination proportion of *A. gmelinii* Web. and *A. giraldii* Pamp. was found at 25 °C / 20 °C,

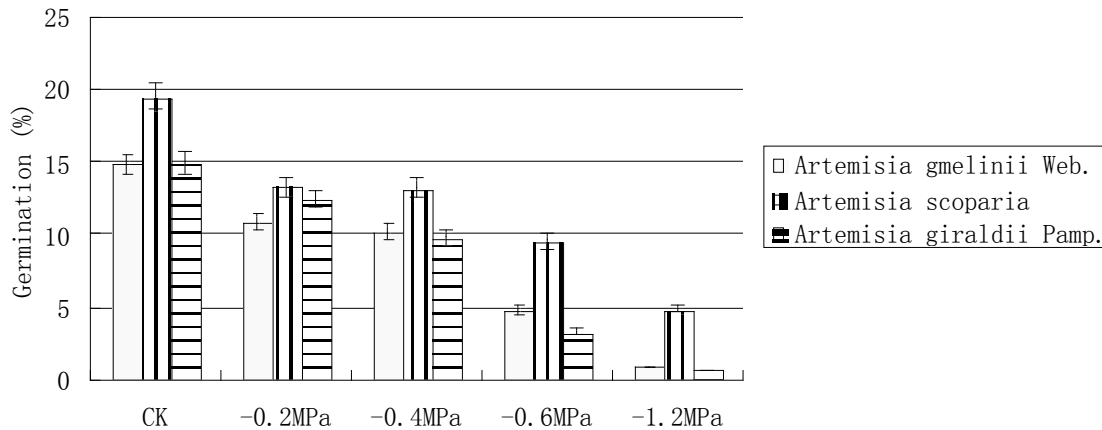
but for *A. scoparia*, 30 °C / 25 °C is the best germinating temperature (Fig. 1).



**Fig. 1** Proportion of three *Artemisia* species achenes germinated under different alternating temperature treatments from 30 °C / 25 °C to 10 °C / 5 °C with a thermoperiod of 14/10 h and a 12 h photoperiod.

### 3.2 Osmotic potential solutions treatments

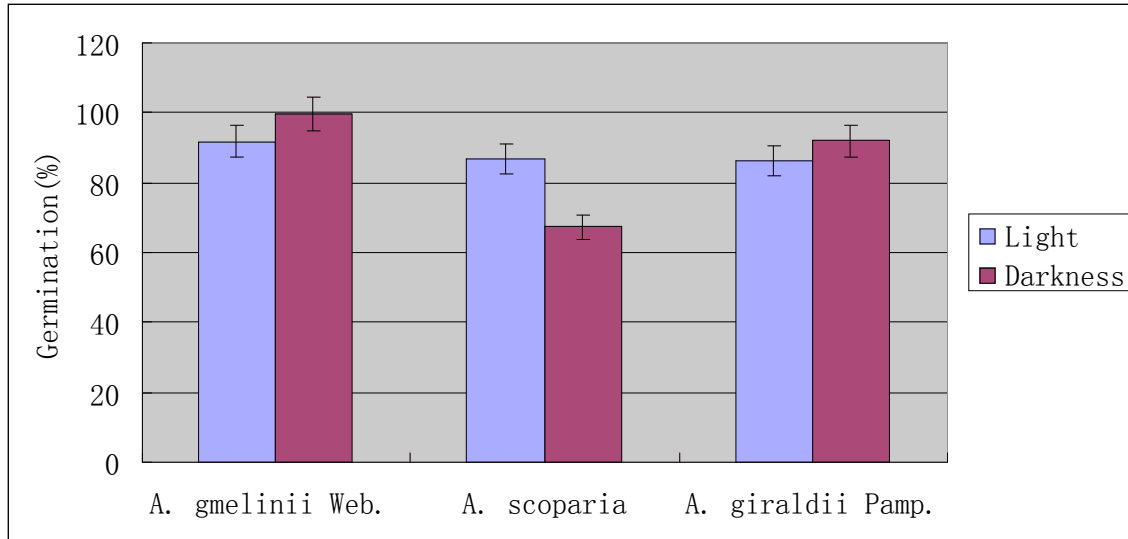
The optimal osmotic potential for achene germination differed with species ( $P < 0.0001$ ). And among three *Artemisia* species, *A. scoparia* had the highest germination proportion. And *A. gmelinii* Web. had higher germination proportion than *A. giraldii* Pamp. when osmotic potential was lower than -0.4 MPa (Fig. 2).



**Fig. 2** Germination proportion of three *Artemisia* species within a osmotic potential range from 0 MPa to -1.2 MPa

### 3.3 Light treatments

Results of achene germination of *A. gmelinii* Web. showed no differences between illumination and nonillumination (Fig. 3) treatments at 25 °C / 20 °C alternating temperature ( $P > 0.1$ ), but for *A. scoparia*, the differences was significantly ( $P < 0.0001$ ), and for *A. giraldii* Pamp., significant differences was found ( $P < 0.05$ ).



**Fig. 3** Germination proportion of three *Artemisia* species between white light and darkness at 25 °C / 20 °C alternating temperature.

#### 4. Discussion

Diurnal temperature is large in Loess hilly region in Northern Shaanxi (Ma X.H., Jiao J.Y., 2005), so, research of alternating temperature treatments of dominant plant species has more practical meaning than constant temperature treatments. Results obtained with respect to alternating temperature treatments indicate although the achenes of *Artemisia gmelinii* Web. and *Artemisia giraldii* Pamp. had lower optimum germinating temperature (25 °C / 20 °C) than *Artemisia scoparia*. (30 °C / 25 °C), achenes of *Artemisia. Scoparia*. had higher germination proportion than two other *Artemisia* species, specially at low temperature treatments ( $\leq 15$  °C / 10 °C). The results demonstrate that probably temperature couldn't limit the distribution range of *Artemisia scoparia*., and it could has a wider response to temperature and germinate earlier than other two species when the environment temperature is still low. This response of temperature could help *Artemisia scoparia*. to occupy environmental resources earlier, and this may be crucial for population formation.

Water is an important factor which affects seed germination obviously. Too much or less surrounding water content could inhibit germination of seed, specially when the surrounding water content is low, germination rate and morphodifferentiation of seed would change a lot according to the increase of drought stress. For example, Inhibition rate and germination rate of seed of *Caragana.Korshinskii*. and *Hedysarum.scoparium*. decreased when drought stress increased (ZENG Y.J., et al, 2002), Ren studied on several desert vegetation and got the same results (REN J., et al, 2002). Further study showed that low water content could cause the decrease of amylase activity and soluble sugar content, and increase of membrane permeability, hydroxyproline and soluble protein content (KONG X.S., et al, 1998). In this study, the addition of different concentration osmotic potential solutions at three *Artemisia* species reduced germination proportion significantly. *A.scoparia* had better drought adaption and higher germination proportion at each treatment than two other *Artemisia* species, which means it had a wider range of osmotic potential and could germinate better than two others when the water content of environemt is low. Besides, in higher osmotic potential solution treatments, *A.giraldii*

*Pamp.* had higher germination proportion than *A.gmelinii Web.*, but in lower osmotic potential solution treatments, germination rate of *A.giraldii Pamp.* decreased obviously and lower than *A.gmelinii Web.*, which means germination of *A.giraldii Pamp.* needs more water.

Results obtained with respect to light treatments demonstrate that the achenes of *Artemisia Scoparia.* had a positive photoblastic response, similar to some arid and semiarid dominant species (De la Barrera, E., Nobel, P.S., 2003). But *Artemisia gmelinii Web.* and *Artemisia giraldii Pamp.* had better germination proportion in darkness treatments than in light treatments, suggesting that they can germinate if they come slightly buried in the soil where humidity may be available for a longer period of time than in the soil surface. On this aspect, *Artemisia gmelinii Web.* and *Artemisia giraldii Pamp.* had better germinating strategy than *Artemisia Scoparia.* But all those *Artemisia* species had small achenes and they usually spread on the soil surface, so positive photoblastic response of achenes can control the timing of germination in their natural habitat (P. Ortega-Baes, M. Rojas-Aréchiga., 2007), which may be crucial for population formation in the further growing period.

Some research reported that smaller and more spheric shape could form a more steady soil seed storage and larger seed bank (LIU Z.M., et al, 2003; Bowers, J.E., 2000.). Also a short-term persistent bank has been demonstrated in arid and semiarid area (De Viana, M.L., 1999). Small seed not only means more production with little resources, but also reduces the venture of been eaten (Thompson K., 1990; Thompson K., 1987). On the other hand, small seed could occupy a larger abundance distribution range and space than bigger one, and easier to survive at different stage of succession (GUO Q., et al, 2000). All three *Artemisia* species in this study have small achenes (Table 1), and results showed that achenes of *A.scoparia* were more like sphericity than two others which means it could form a bigger soil seed storage. Achenes of three *Artemisia* species are mucilaginous, which has powerful water-imbibition ability. Huang has reported the seed coat and pericarp of *Artemisia* are concrescence while the out most layers of the pericarp are covered by a layer which becomes mucilaginous when wetted and increases in weight 589 times by absorbing water (HUANG Z.Y., et al, 2001). The pellicle is unlikely to be a source of nutrition for the embryo, but is a means of sustaining the hydrated state of the embryo under the inhospitable conditions for germination in arid or semiarid habitats (HUANG Z.Y., Daphne J.O., 2002). In this study, we found *A.gmelinii Web.* has the thickest mucilaginous layer and the highest imbibition rate (1665.192%, See table 1) among all three *Artemisia* species, which means it has a high water-imbibition ability in arid or semiarid habitats to keep seed wet and a great area/cubage rate to short the transport time of water from seed capsule to embryo (HUANG Z.Y., Y. Gutterman, 1999). Mucilaginous also could make achene become heavier and more adhesive, which increase the settle rate of seed in dry and windy habitats (HUANG Z.Y., Y. Gutterman, 1998). Results of seed water content and imbibition rate showed that *A.scoparia* is more likely becoming the dominant community at the primary period of local vegetation succession but *A.gmelinii Web.* and *A.giraldii Pamp.* had more steady status in the succession. And this laboratory germination results can be applied to propagation projects that would support conservation programs within the study site.

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