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Short communication

Environmental control of seed dormancy and germination in the short-lived *Olimarabidopsis pumila* (Brassicaceae)

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ABSTRACT

The purpose of our study was to better understand seed germination ecology of the spring annual short-lived *Olimarabidopsis pumila*, which grows in the Gurbantonggut Desert, China. Seeds underwent after-ripening at 4, 20 ± 2 (room temperature) and 30 °C. After dormancy was broken, germination capacity was a function of temperature and presence of light. For the temperature range studied (4–30 °C), germination capacity was significantly higher between 15 and 25 °C than at temperatures above or below them. Alternating temperatures of 20/10, 25/10 and 25/15 °C were favorable for germination. Although GA_3 did promote dark germination of seeds, GA_3 did not replace light for germination. Seeds germinated to >75% if light exposure time was over 8 h and temperature and moisture conditions were favorable. Seeds were able to germinate at relatively low water potentials (83% at −0.41 MPa), but decreasing water potentials produced detrimental effects on germination percentage and rate. Thus, dormancy characteristics and germination behavior of *O. pumila* seeds ensure that germination occurs in the desert only when soil moisture conditions are favorable for seedling establishment and survival.

1. Introduction

The Gurbantunggut Desert, the second largest desert in China, is located in the Junggar Basin in northwest Xinjiang province (44°15′–46°50′ N, 84°50′– 91°20′ E), where climate is typically temperate continental. There are more than 200 short-lived plant species in the desert. The ephemeral flora is a particular group in the desert flora of China (Mao and Zhang, 1994). Ecophysiological studies on these species are meager, as compared to taxonomic, floristic, and phenological studies (Wu, 1979; Mao and Zhang, 1994; Ma et al., 2006).

One of the members of the ephemeral flora is *Olimarabidopsis pumila* (Stephan) Al-Shehbaz et al. (1999), whose morphological characteristics and life history are similar to those of *Arabidopsis thaliana*. Previously, this species was named *Arabidopsis pumila* and was considered to be a close relative *Arabidopsis thaliana* (Wang and Li, 2001). *Oliarabidopsis pumila* has a life cycle of approximately 60 days, flowering in April and fruit maturing during mid- and late-May (Ma et al., 2006). However, the seed germination stage of its life cycle in relation to natural environmental factors is still not well understood. Thus, the purpose of our study is to determine the dormancy breaking and germination requirements of seeds of this short-lived, annual species.

2. Materials and methods

2.1. Seed collection and measure

Ripe seeds were collected from >100 *O. pumila* plants growing in a dry sandy field on the foot of the Jiangjun Mountain on the south side of the Gurbantunggut Desert in the Junggar Basin (44°11.77′ N, 86°5.16′ E) on 26 May 2006.

After cleaning, we measured the initial water content and a 1000-seed weight. The moisture content of 500 seeds was determined by the high-constant-temperature oven method, 103 °C for 17 h (ISTA, 1999), and calculated on a fresh weight basis. We weighed four replicates of 200 seeds each by using an analytical balance (accuracy of 0.1 mg) (FA10004A, Shanghai Jielun Electronia Apparatus Manufactory, Shanghai, China) and calculated 1000-seed weight on the basis of mean 200-seed weight. Seeds were stored open in our laboratory at room temperature until used.

2.2. General germination test

All germination tests of fresh or stored seeds were performed on moist filter paper (Whatman No. 1) in 9-cm-diameter glass Petri

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dishes in temperature- and light-controlled incubators (HPG-280B Illuminating Incubators, Har'erbin Electronia Apparatus Manufactory, Har'erbin, China). The light source was cool white fluorescent tubes (14 h light: 10 h dark photoperiod, ca. 40 μmol m^{-2} s^{-1} photosynthetically active radiation (PAR)) measured with LI-1400 Data Logger (LI-COR Inc., Nebraska, USA). Four replicates of 50 seeds each were tested for germination on top of filter paper (previously moistened with 5 ml distilled water). Seeds were considered to be germinated when the radicle emerged. Germination was monitored at 1- or 2-day intervals until no seeds germinated for more than 5 days, unless specified otherwise. Final germination percentages were determined after 14 days.

2.3. Effect of dry storage on breaking dormancy

To assess effects of storage temperature on dormancy break, freshly mature seeds (with 5.6% moisture content) were hermetically stored (dry storage) in laminated aluminum foil packets and placed in the dark at 30 °C for 8 weeks. After 8 weeks of dry storage, seeds were tested for germination in 14-h light/10-h dark photoperiod under constant temperature regimes (4, 10, 15, 20, 25 and 30 °C) and alternating temperatures (30/15, 25/15, 25/10 and 20/10 °C). Similarly, some seeds (with 5.6% moisture content) were stored hermetically in a plastic box wrapped with two-layer black cloth at 30 °C for 2, 4, 8, 12 and 16 weeks (continuous darkness), respectively, then transferred to 4 °C and stored for 16, 14, 12, 8 and 4 weeks. Subsequently, germination tests were conducted at 20 °C in light and darkness. Seeds stored open at room temperature (RT) (20 ± 2 °C) were used as controls.

2.4. Minimum light exposure

After dry storage at 30 °C for 8 weeks, seeds which were on wet filter paper were exposed to light for 0, 5, 10, 20, 30 and 45 min and 1, 2, 4, 6, 8, 10, 12 and 14 h. We did not allow them to imbibe in darkness before we exposed them to light. After each light exposure, seeds were wrapped with aluminum foil and incubated in darkness at 20 °C for 14 days. Seeds were sown on moist filter paper (previously moistened with 5 ml distilled water, which was periodically added) in 9-cm Petri dishes. Incubation took place in incubators described above at 20 °C; four replications of 50 seeds each were tested for germination.

2.5. Sensitivity of seeds to GA_3

Response to GA_3 of *O. pumila* seeds dry-stored for 8 weeks at 30 °C was tested. A range of GA_3 concentrations (0.05, 0.1, 0.3, 0.5, 1.0 and 2.0 mmol/l GA_3) was prepared following the method of Derkx and Karssen (1994). Seeds were imbibed for 24 h in GA_3 solutions and transferred to other dishes with the filter paper moistened by the same GA_3 solution (i.e., 0.05, 0.1, 0.3, 0.5, 1.0 and 2.0 mmol/l GA_3, respectively) at 20 °C in darkness for 14 days. GA_3 solution was added to the Petri dishes if necessary during incubation.

2.6. Germination responses to water stress

Solutions of Polyethylene Glycol 8000 (PEG) were prepared to simulate water potentials of 0, −0.41, −0.98, −1.27, −2.04, −4.89 and −5.10 MPa at 20 °C (Michel, 1983). Ten milliliters of PEG solution was poured into each Petri dish, and a piece of gauze was placed on the surface of the liquid. Each Petri dish was wrapped with transparent plastic foil to maintain constant humidity. Controls were sown on a piece of gauze in Petri dishes with 10 ml Mini-Q water (i.e., distilled water was purified by the purifier (SIMSV0000) manufactured by Shanghai Minipore Industrial Co. Ltd., China). Petri dishes were kept under the same temperature and light conditions as described above. The time necessary (in days) to reach 50% of the final germination percentage (T_{50}) was calculated by the linear interpolation from the two germination values closest to median germination. Incubation was terminated after 21 days.

2.7. Data analyses

Germination percentages were arcsine-transformed using the SPSS 12.0 software package before analysis. One-way factorial ANOVA and Duncan multiple comparison tests were used to statistically analyze the data.

3. Results

The initial water content and a 1000-seed weight of *O. pumila* seeds were 5.6 ± 0.3% and 77.9 ± 1.8 mg, respectively. Obviously, *O. pumila* seeds were at low water content and very light at shedding.

At constant temperatures of 4, 10, 15, 20, 25 and 30 °C, freshly matured *O. pumila* seeds germinated to <10% in both light and darkness. After seeds were hermetically stored at 30 °C for 8 weeks, they germinated to >80% in light when incubated at 20, 25, 25/10 and 25/15 °C; however, final germination did not exceed 23% at 30 °C. Also, seeds could germinate to 71% and 81% at 20/10 and 20/15 °C, respectively, with the exception of 29% at 30/15 °C (Table 1). *O. pumila* seeds stored for 8 weeks at RT, the control, germinated to 84 ± 3% at 20 °C in light.

As for *O. pumuila* seeds stored for any period of time at single constant (4 °C or 30 °C) or combined temperature (4 °C plus 30 °C), their dark-germination percentages were all ≤29%, and storage effects of both single 30 °C and combination of 30 °C and 4 °C on light germination at 20 °C were not significantly different ($P > 0.05$) (Tables 1 and 2). Interestingly, light exposure strongly affected the germination of after-ripened seeds. After the seeds were exposed to light for 0, 5, 10, 20, 30 and 45 min and 1, 2, 4, 6, 8, 10, 12 and 14 h, they germinated to 11 ± 3%, 17 ± 2%, 35 ± 2%, 43 ± 5%, 41 ± 4%, 56 ± 3%, 67 ± 4%, 72 ± 2%, 73 ± 6%, 73 ± 1%, 76 ± 2%, 84 ± 3%, 83 ± 4% and 86 ± 4% in the dark, respectively.

GA_3 did not replace light for the germination of *O. pumila* seeds which had after-ripened at 20 °C in the dark although GA_3 enhanced dark germination. Non-dormant *O. pumila* seeds treated with 0.05, 0.1, 0.3, 0.5, 1.0 and 2.0 m mol/l GA_3, germinated to 19 ± 3%, 33 ± 4%, 56 ± 6%, 61 ± 4%, 57 ± 4% and 59 ± 5%, respectively, in the dark; controls germinated to 87 ± 2% and 21 ± 3% in light and dark, respectively. Although final germination

Table 1
Germination of fresh and stored *Olimarabidopsis pumila* seeds at different temperatures.

Temperature (°C)	Germination of stored seeds for 8 weeks at 30°C (%) (SD)		Germination of fresh seeds (%) (SD)	
	Light	Dark	Light	Dark
4	19.0 (2)^{d}	6.0 (3)	3 (0.7)	0
10	33.0 (2)^{c}	5.0 (1)	7 (2.1)	3 (0.5)
15	67.0 (2)^{b}	7.0 (2)	5 (0.7)	4 (0.6)
20	91.0 (3)^{a}	13.0 (2)	9 (0.7)	7 (1.5)
25	84.0 (2)^{a}	11.0 (4)	4 (2.1)	2 (1)
30	23.0 (1)^{cd}	7.0 (2)	6 (2.1)	2 (1)
20/10	71.0 (2)^{b}	6.0 (1)		
25/10	81.0 (3)^{ab}	11.0 (1)		
25/15	87.0 (2)^{a}	14.0 (2)		
30/15	29.0 (4)^{c}	9.0 (1)		

Means with the same superscript letters in the same column are not significantly different at $p < 0.05$ according to Duncan's multiple comparisons test.

Table 2
Germination percentages of *Olimarabidopsis pumila* seeds incubated at 20 °C in light and in dark for 14 days, through 0–16 weeks of different combined storage at 30 °C and 4 °C.

Dry storage duration at 30°C (weeks)	Dry storage duration at 4°C (weeks)	Light		Dark	
		Germination (%)	SD	Germination (%)	SD
16	0	83[a]	3	17[c]	4
12	4	84[a]	2	21[b]	3
8	8	87[a]	2	29[a]	5
4	12	71[b]	2	21[b]	5
2	14	74[b]	3	19[c]	2
0	16	73[b]	3	24[b]	2

Means with the same superscript letters in a column are not significantly different at $p < 0.05$ according to Duncan's multiple comparisons test.

percentages of seeds treated by GA_3 were higher than that of the control in the dark, they were significantly lower than that of the control in light.

Final germination percentages decreased with decreasing water potential. Percentage germination declined severely to 43% at −1.27 MPa. Moreover, light germination was completely inhibited at −5.10 MPa (Fig. 1). Also, the germination rate slowed down with decreasing water potential. With the exception of failure to germinate at −5.10 MPa, T_{50} values were 3.25 ± 0.5, 6.5 ± 1.7, 7.75 ± 1.0, 10.25 ± 1.3, 11.75+2.1, 12.5 ± 2.7 at −0.41, −0.98, −1.27, −2.04 and −4.89 MPa PEG, respectively.

4. Discussion

The freshly matured seeds of *O. pumila* were dormant, and dormancy was broken during dry storage (Tables 1 and 2). After dormancy was broken, *O. pumila* seeds germinated to higher percentages in light than in darkness (Tables 1 and 2), and the favorable temperatures were between 15 and 25 °C, followed by the alternating temperature 25/10 and 25/15 °C; the optimal was 20 °C. With an increase or decrease in temperature, germination was markedly inhibited. As shown by the present results, dry storage at 4 °C, 20 °C (room temperature) and 30 °C could release *O. pumila* seeds from dormancy although the effect of 4 °C on the loss rate of dormancy was slower than at room temperature or 30 °C. This fact suggested that *O. pimila* seeds must be exposed to dormancy-breaking temperatures before they will become nondormant, and dormancy release was temperature-dependent.

Like *O. pumila*, Sharif-Zadeh and Murdoch (2001) found that loss of dormancy (after-ripening) of *Cenchrus ciliaris* L. seeds was affected by storage temperature. Moreover, our results are consistent with knowledge gained from germination studies in dry Central Asian steppes (Wesche et al., 2006), North American mid-latitude steppes and other regions (Baskin and Baskin, 1988, 1998). For example, optimum temperatures for after-ripening of seeds of the winter annual *Arabidopsis thaliana* were 25/15, 30/15 and 35/20 °, although some after-ripening occurred at 5 and 15/6 °C (Baskin and Baskin, 1986), and seeds require light for germination (Baskin and Baskin, 1983). In these cases, dry storage is known to allow after-ripening and to break physiological dormancy. In our study, *O. pumila* grows in the arid Gurbantunggut Desert, where the average temperature of summer is above 20 °C, which favors after-ripening.

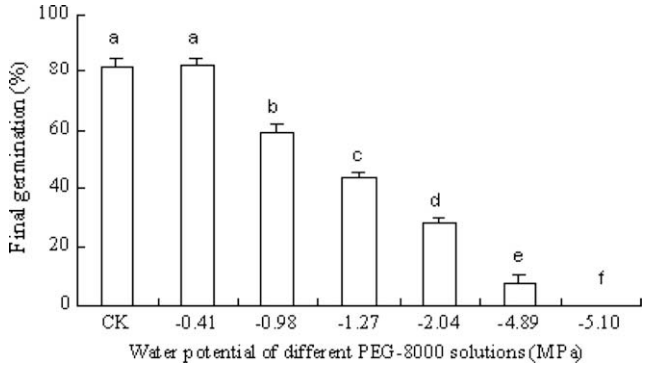

Fig. 1. Germination (%) (mean ± SD) of *Olimarabidopsis pumila* seeds at water potentials of 0 (CK), −0.41, −0.98, −1.27, −2.04, −4.89 and −5.10 MPa at 20 °C. Values with the same superscript letters are not significantly different at $p < 0.05$ according to Duncan's multiple comparison test.

When seeds of *O. pumila* were stored either hermetically or open, they underwent after-ripening, but even after they became nondormant they required light for germination. Furthermore, even if seeds were exposed to light, they could only germinate well at certain temperatures. The results indicated that *O. pumila* seeds were sensitive to light although effects of light quality and light intensity were not clear. In some previous studies, seeds of many species are photosensitive, such as *Pistia stratiotes* (Fu, 1957), *Rumex crispus* (Baskin and Baskin, 1985), *Potentilla recta* and *Barbarea vulgaris* (Baskin and Baskin, 1988). The light requirement for germination does not restrict germination to any particular season. In fact, nondormant seeds will germinate at any time during the growing season, if light and soil moisture are not limiting.

Also, GA_3 did not promote dark germination of nondormant *O. pumila* seeds to the level equal to that of the control in light, indicating that GA_3 could not replace light for germination. As reported by other studies, GA_3 can significantly promote seeds of wild type (Derkx and Karssen, 1994) and ga1-3 mutant of *Arabidopsis thaliana* (Sun and Kamiya, 1994; Tyler et al., 2004), *Lactuca sativa* cv. Grand Rapids (Fountain and Bewley, 1976) and *Sisymbrium officinale* (Derkx and Karssen, 1993) to germinate in darkness. Moreover, other bioactive gibberellins (GAs) (e.g., GA_4, GA_7 or GA_{4+7}) significantly increase dark germination, such as *Cucumis sativus* (Staub et al., 1989) and *Cucumis melo* (Edelstein and Kigel, 1990). Thus, sensitivity to GAs depends on the species.

Water stress has a significant effect on the germination of *O. pumila* seeds, and germination percentage decreased with a decrease in water potential. The decreasing speed and percentage of germination with water stress are commonly found in desert plants (Gutterman, 1993; Huang et al., 2001; Villagra and Cavagnaro, 2006). Most studies report optimum germination at osmotic potentials above −1 MPa (Song et al., 2005; Tobe et al., 2005). The limited resistance to water stress plays a very important role in the survival of the species growing in arid habitats.

Olimarabidopsis pumila grows in the Gurbantunggut Desert, where natural environments are variable in time and space. Seeds of annual short-lived *O. pumila* begin to germinate in late March, and mature and disperse in late May (Ma et al., 2006). The 36.9% of total precipitation (202.5 mm, data from local meteorological service) occurs between June and September, when average temperature is above 22 °C. As shown by the results in our study, seeds of *O. pumila* are dormant at shedding must after-ripen before they can germinate in favorable environments. In autumn, *O. pumila* seeds undergo partial or complete after-ripening and may germinate. However, we have not observed seedling emergence in autumn but in spring in the field for 3 years. We infer that there are two major reasons for lack of germination in autumn: (1) only a small quantity (52 mm) of rain falls during this period, and (2) the top soil layer becomes very dry due to strong evaporation in autumn in the Gurbantunggut desert. Water moisture in top soil is about 0.027(v/v) (<2%) in autumn (Zhang et al., 2007). Therefore, *O. pumila* seeds fail to germinate until exposed to favorable soil moisture conditions the next late March and early April. Even though some nondormant *O. pumila* seeds can germinate in

autumn, the resultant seedlings have not sufficient time to attain enough size to survive the rigorous conditions in winter. So we concluded that dormancy breaking and germination requirements of *O. pumila* seeds are adaptive because they ensure that germination occurs in the desert only when soil moisture conditions are favorable for seedling establishment and survival.

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References

Al-Shehbaz, I.A., O'Kane Jr., S.L., Price, R.A., 1999. Genetic placement of species excluded from *Arabidopsis*. Novon 9, 296–307.

Baskin, J.M., Baskin, C.C., 1983. Seasonal changes in the germination responses of buried seeds of *Arabidopsis thaliana* and ecological interpretation. Botanical Gazette 144, 540–543.

Baskin, J.M., Baskin, C.C., 1985. Does seed dormancy play a role in the germination ecology of *Rumex crispus*? Weed Science 33, 340–343.

Baskin, J.M., Baskin, C.C., 1986. Temperature requirements for afterripening in seeds of nine winter annuals. Weed Research 26, 375–380.

Baskin, C.C., Baskin, J.M., 1988. Germination ecophysiology of herbaceous plant species in a temperature region. American Journal of Botany 75, 286–305.

Baskin, C.C., Baskin, J.M., 1998. Seeds: Ecology, Biogeography, and Evolution of Dormancy and Germination. Academic Press, San Diego, CA.

Derkx, M.P.M., Karssen, C.M., 1993. Changing sensitive to light and nitrate but not to gibberellins regulates seasonal dormancy patterns in *Sisymbrium officinale* seeds. Plant, Cell and Environment 16, 469–479.

Derkx, M.P.M., Karssen, C.M., 1994. Are seasonal dormancy patterns in *Arabidopsis thaliana* regulated by changes in seed sensitivity to light, nitrate and gibberellin. Annals of Botany 73, 129–136.

Edelstein, M., Kigel, J., 1990. Seed germination of melon (*Cucumis melo*) at sub- and supra-optimal temperatures. Scientia Horticulturae 45, 55–63.

Fountain, D.W., Bewley, J.D., 1976. Lettuce seed germination. Plant Physiology 58, 530–536.

Fu, J.R., 1957. Sensitivity to light of *Pistia stratiotes* (Araceae) seeds. Chinese Science Bulletin 19, 590.

Gutterman, Y., 1993. Seed Germination in Desert Plants. Springer, Berlin.

Huang, Z.Y., Gutterman, Y., Hu, Z.H., Zhang, X.S., 2001. Seed germination in *Artemisia sphaerocephala* II. The influence of environmental factors. Acta Phytoecologica Sinica 25, 240–246 (in Chinese with English abstract).

Ma, S.J., Liu, X.F., Tan, D.Y., 2006. The phenology of ephemeral plants of eight Brassicaceae and its adaptation to the desert environment. Journal of Xinjiang Agricultural University 29, 1–4 (in Chinese with English abstract).

Mao, Z., Zhang, D.M., 1994. The conspectus of ephemeral flora in northern Xinjiang. Arid Zone Research 11, 1–26 (in Chinese with English abstract).

Michel, B.E., 1983. Evaluation of the water potentials of solutions of polyethylene glycol 8000 both in the absence and presence of other solutes. Plant Physiology 72, 66–70.

Sharif-Zadeh, F., Murdoch, A.J., 2001. The effects of temperature and moisture on after-ripening of *Cenchrus ciliaris* seeds. Journal of Arid Environments 49, 823–831.

Song, J., Feng, G., Tian, C.Y., Zhang, F.S., 2005. Strategies for adaptation of *Suaeda physophora, Haloxylon ammodendron* and *Haloxylon persicum* to a saline environment during seed-germination stage. Annals of Botany 96, 399–405.

Staub, J.E., Wehner, T.C., Tolla, G.E., 1989. The effects of chemical seed treatments on horticultural characteristics in cucumber (*Cucumis sativus* L.). Scientia Horticulturae 38, 1–10.

Sun, T.-P., Kamiya, Y., 1994. The arabidopsis *GA1* locus encodes the cyclase *ent*-kaurene synthetase A of gibberellin biosynthesis. Plant Cell 6, 1509–1518.

Tobe, K., Zhang, L., Omasa, K., 2005. Seed germination and seedling emergence of three annuals growing on desert sand dunes in China. Annals of Botany 95, 649–659.

Tyler, L., Thomas, G.T., Hu, J.H., Dill, A., Alonso, J.M., Ecker, J.K., Sun, T.-P., 2004. DELLA Proteins and gibberellin-regulated seed germination and floral development in *Arabidopsis*. Plant Physiology 135, 1008–1019.

Villagra, P.E., Cavagnaro, J.B., 2006. Water stress effects on the seedling growth of *Prosopis argentina* and *Prosopis alpataco*. Journal of Arid Environments 64, 390–400.

Wang, S.M., Li, X.Y., 2001. Distribution of the genus of *Arabidopsis* in China. Journal of Shihezi University (Natural Science) 5, 102–108 (in Chinese with English abstract).

Wu, C.Y., 1979. The regionalization of Chinese flora. Acta Botanica Yunnanica 1, 1–22 (in Chinese with abstract).

Zhang, J., Zhang, Y.M., Zhou, Z.B., Wu, N., Wang, H.L., Cheng, R.Y., 2007. Study on daily change of soil moisture content under the effect of microbiotic crusts in the Guerbantonggut Desert, xinjiang, China. Arid Zone Research 24, 661–668.