

RELATIONSHIP BETWEEN GERMINATION AND SEED SIZE IN ALPINE SHRUBS IN TIBETAN PLATEAU

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Abstract

Seed germination is related to seed size in many herbs species, but few studies investigated the relationship between germination and seed size in shrubs. We examined the correlation of seed size and germination percentage, germination rate and germination persistence time for 29 alpine shrubs in the eastern Qinghai-Tibetan Plateau, China. Results showed that seed size was significant negatively correlated with coefficients of variation ($r = -0.709$, $p < 0.001$), with germination percentage ($r = -0.682$, $p < 0.001$), and with germination rate ($r = -0.512$, $p = 0.004$). Coefficients of variation was significant positively correlated with germination percentage ($r = 0.525$, $p = 0.003$) and germination rate ($r = 0.522$, $p = 0.004$). It revealed that the smaller-seeded species present a bigger variance in seed size than the larger-seeded species. Smaller-seeded species were apt to fast and concentrative germination strategy, but, larger-seeded species were apt to slow and stochastic germination strategy. It appears that smaller-seeded species advance their germination fitness by adaptive variance of seed size.

Introduction

Seed size is a central component of plant life histories (Harper, 1977; Baskin & Baskin, 1998; Fenner & Thompson, 2005; Moles *et al.*, 2005) and its importance to plant fitness and growth strategy are widely appreciated (Foster, 1986; Westoby *et al.*, 1992; Leishman & Westoby, 1994; Moles & Westoby, 2006; Moles *et al.*, 2007). Seed size has long been regarded as an important aspect in life-history strategy of plants. Seed size variations usually occur within species under heterogeneous habitats and different evolutionary pressure (Westoby *et al.*, 1992; Moles & Westoby, 2006). Seed size has important evolutionary and ecological meaning within and among species (Moles *et al.*, 2005). Differences in seed size among species are related to differences in seed production (Smith & Fretwell, 1974) and seedling establishment and growth (Leishman & Westoby, 1994; Jakobsson & Eriksson, 2000), with seed size underlying a trade-off between these traits (Muller-Landau, 2003). Seed size can be considered as a key factor that influences the dynamics of interspecific interactions and the mechanisms of coexistence in plant community (Rees & Westoby, 1997), because it significantly impacts seed dispersal, seed germination (Moles & Westoby, 2006) and seedling recruitment (Moles & Westoby, 2004), which, indeed, is strongly correlated with plant abundance in many communities (Muller-Landau, 2003; Murray *et al.*, 2005). So, it is important to study the relationships of seed size and seed germination with particular focus on interspecies within communities.

Although many studies have been conducted on the relationship between seed size and germination in species of forest and grassland, very few empirical studies have been conducted to test this relationship for shrub species. In this study, we selected 29 shrubs which are important component species in alpine grassland community of the Qinghai-Tibetan Plateau of China to study seed germination characteristics. The specific objectives of this investigation on 29 shrubs from the alpine area were to

test (1) the relationship between seed size and coefficients of variation; and (2) the relationships between seed size and germination characteristics.

Materials and Methods

Study site: The region of this study is located on the eastern Qinghai-Tibetan plateau (101°–103° E, 34°–35° 70'N). The altitude ranges from 2,800 to 4,200 m, and the climate is cold Humid-Alpine with mean annual rainfall of 450–780 mm. Mean annual temperature is 1.2°C with –10.7°C in January and 11.7°C in July, and there are on average 270 frost days a year (Wu & Du, 2007). The vegetation types mainly contain alpine meadow, alpine shrubs and alpine forest (Wu, 1995).

Seed collection and germination: In this study, 29 shrub species which were dominant and main component shrubs were collected from the alpine area in the eastern Qinghai-Tibetan Plateau (Table 1). Ripe seeds were collected from as many sources as possible to get an adequate representation (more than 20 individual samplings in three to five different populations) of the whole community of typical alpine meadow on the eastern Qinghai-Tibetan Plateau (Gannan, Gansu Province), from August to October 2005. Sarcocarp was wiped off artificially for all seeds. Enveloped seeds were spread on tables and allowed to air-dry to a constant mass at room temperature (approximately 15°C) before being weighed. Seed mass was defined as the weight of the embryo and endosperm, plus the seed coat. Structures having the function of contributing to dispersal were not included as part of the seed mass. Seeds were pooled per species and we selected stochastically three subsamples of 100 seeds from three population samples ($N = 900$ seeds). The average weight of the nine subsamples was used as seed mass variable. All seeds from different subsamples and populations per species were mixed for germination experiment. They were moist-cold stratified (about $\pm 4^\circ\text{C}$)

to break dormancy for a month before germination experiment. The germination experiment was started on early of March (starting season of germination), in 2006. Seeds were placed in covered Petri dishes (9 cm diameter) on a double layers of moistened filter paper, then placed in temperature chambers at a diurnal fluctuation (of 25°C day, 12 h; 5°C night, 12 h) with 12-h photophase and 12-h darkness periodically and a relative humidity of about 70%. The temperature and light condition of the germination trial resembled natural conditions of April and May and it approximated to the daily maximum and minimum temperature in 5 cm-deep soil. All of the

species had three replicates of 50 seeds. Every day, the percentage of seeds germinated was recorded, newly emerged seedlings were removed from the Petri dishes, and seeds were regularly watered with distilled water. A seed was considered as germination when the radicle was visible. The experiment of seed germination lasted 60 days. After germination finished, we test seed viability with the TTC test methods (Scharpf 1970), 1 g of 2, 3, 5-triphenyl tetrazolium chloride (TTC) was dissolved in 100 ml of distilled water to make a 1% solution of the tetrazolium salt.

Table 1. Mean seed size (\pm S.D. mg/per seed) and their family for studied 29 shrub species in this experiment. Nomenclature and assignment to families follows Wu (1995).

Species	Seed size (\pm S.D. mg/seed)	Family
<i>Aster poliothamnus</i>	0.26 \pm 0.009	Asteraceae
<i>Berberis aggregata</i>	3.773 \pm 0.165	Berberidaceae
<i>Berberis diaphana</i>	8.66 \pm 0.146	Berberidaceae
<i>Berberis dictyoneura</i>	8.41 \pm 0.008	Berberidaceae
<i>Berberis verna</i>	3.485 \pm 0.105	Berberidaceae
<i>Caragana brevifolia</i>	2.929 \pm 0.817	Leguminosae
<i>Caragana densa</i>	8.659 \pm 0.109	Leguminosae
<i>Caragana jubata</i>	9.177 \pm 0.134	Leguminosae
<i>Caragana jubata</i> var. <i>recurva</i>	5.915 \pm 0.064	Leguminosae
<i>Caryopteris tangutica</i>	0.659 \pm 0.025	Verbenaceae
<i>Hippophae neurocarpa</i>	10.54 \pm 0.255	Elaeagnaceae
<i>Hippophae rhamnoides</i>	9.47 \pm 0.225	Elaeagnaceae
<i>Hippophae thibetana</i>	13.474 \pm 0.43	Elaeagnaceae
<i>Lеспедеза davurica</i>	4.189 \pm 0.165	Leguminosae
<i>Lonicera rupicola</i> var. <i>syringantha</i>	0.735 \pm 0.034	Caprifoliaceae
<i>Osteomeles schwerinae</i> var. <i>microphylla</i>	8.621 \pm 0.134	Rosaceae
<i>Potentilla fruticosa</i>	0.657 \pm 0.048	Rosaceae
<i>Potentilla glabra</i> var. <i>mandshurica</i>	0.028 \pm 0.001	Rosaceae
<i>Potentilla parvifolia</i>	0.325 \pm 0.018	Rosaceae
<i>Rhododendron thymifolium</i>	0.043 \pm 0.002	Ericaceae
<i>Rhododendron anthopogonoides</i>	8.603 \pm 0.165	Ericaceae
<i>Rhododendron rubropilosum</i>	0.146 \pm 0.008	Ericaceae
<i>Salix oritrepha</i>	0.143 \pm 0.004	Salicaceae
<i>Sibiraea laevigata</i>	0.109 \pm 0.007	Rosaceae
<i>Spiraea mollifolia</i>	0.103 \pm 0.005	Rosaceae
<i>Spiraea mongolica</i>	0.139 \pm 0.008	Rosaceae
<i>Spiraea rosthornii</i>	0.032 \pm 0.008	Rosaceae
<i>Syringa oblata</i>	8.759 \pm 0.107	Oleaceae
<i>Syringa reticulata</i>	10.266 \pm 0.106	Oleaceae

Statistical analysis: Three germination parameters viz. Coefficients of variation (C.V.), Germination percentage (%) and Germination rate were used. These parameters were also calculated by the following formulas:

- (1) Coefficients of variation (C.V.) = S.D. of seed mass \times 100 / mean seed mass
- (2) Germination percentage (GP) = number of germinating seeds / number of viable seeds
- (3) Germination rate (GR) = $(A_1 + A_2 + A_3 + \dots + A_n) / (A_1 T_1 + A_2 T_2 + A_3 T_3 + \dots + A_n T_n) \times 100$

An is the numbers of germinating seeds in the n day, T_n is the corresponding days of germination (Brown & Mayer, 1988; Wu & Du, 2007).

Both ecological correlates of seed size and germination were taken into account in the present study. Results of germination characteristic were presented as

mean \pm SD. Data of seed size and germination percentage and germination rate were logtransformed before statistical analysis to ensure homogeneity of variance (Leishman *et al.*, 1995). Spearman's correlation coefficient (r) was used to explain their correlations. All analyses were performed with the SPSS 12.0 procedure.

Results

The coefficients of variation was significant negatively correlated to seed size ($r = -0.709$, $p < 0.001$; Fig. 1). There were significant negatively correlations between seed size and germination percentage ($r = -0.682$,

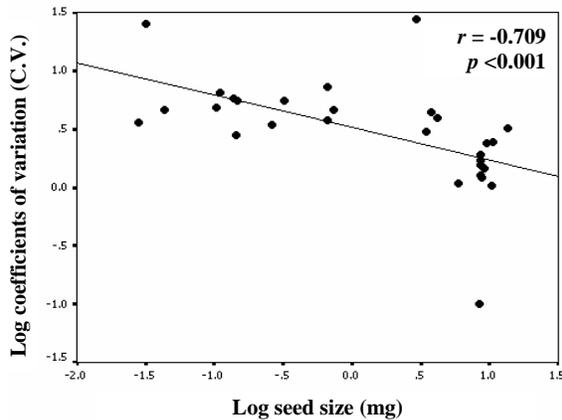


Fig. 1. The relationship between log seed size and log coefficients of variation (C.V.) for 29 shrub species in this study.

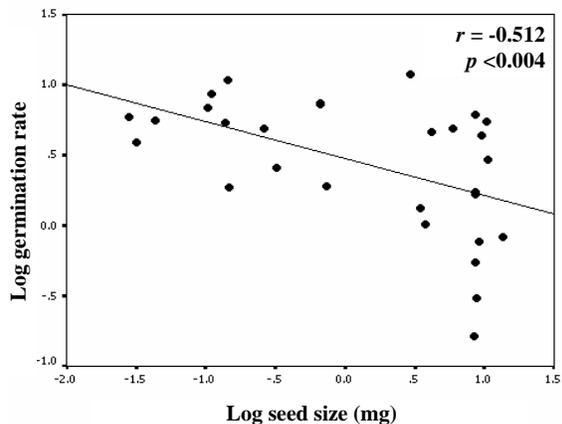


Fig. 3. The relationship between log seed size and log germination rate for 29 shrub species in this study.

Discussion

Seed mass is the main factor determining germination characteristics (Parciak, 2002). We found that there is a significant negative correlation between C.V. and mean seed mass. It revealed that the smaller-seeded species present a bigger variance in seed size than the larger-seeded species for these studied species. In other words, it support that the larger-seeded species tend to be more invariable in seed size evolution than the smaller-seeded species. Meanwhile, there were significant positive correlations between C.V. and germination percentage, germination rate. It suggests that smaller-seeded species advance their germination fitness by adaptive variance of seed size. Our results revealed that smaller-seeded species which had bigger C.V. were apt to fast and concentrative germination strategy because of higher germination rate and shorter germination time. In opposite, larger-seeded species which had less C.V. were apt to slow and stochastic germination strategy because of lower

$p < 0.001$; Fig. 2), and between seed size and germination rate ($r = -0.512$, $p = 0.004$; Fig. 3). Result showed that coefficients of variation was significant positively correlated to germination percentage ($r = 0.525$, $p = 0.003$) and germination rate ($r = 0.522$, $p = 0.004$).

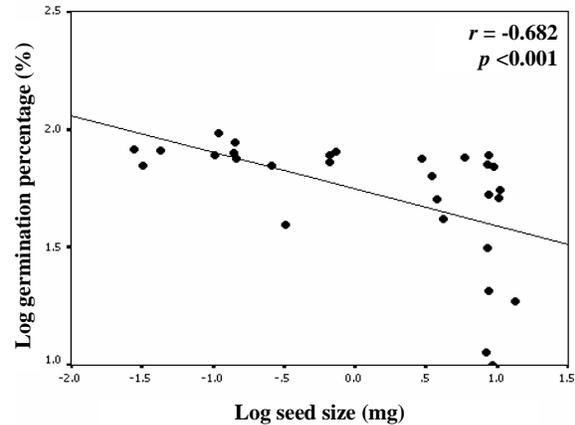


Fig. 2. The relationship between log seed size and log germination percentage (%) for 29 shrub species in this study.

germination rate and longer germination persistence time. This may be determined by co-evolution effects for seed size and plant life-history. Because smaller-seeded species usually produced smaller seedlings and they had lesser competition ability than larger seedlings from larger-seeded species (Westoby *et al.*, 1992; Moles & Westoby, 2006). So, Smaller-seeded species have to adopt fast and concentrative germination strategy when they meet potential circumstances.

Traditionally, larger and heavy seeds have better germination percentage (Wu & Du, 2007), emergence, seedling survival and growth (Armstrong & Westoby, 1993; Bonfil, 1998) than smaller seeds because of their advantage in seed reserves, which is consistent with the idea that large seed size confers a competitive advantage, as assumed by the competition-colonization model (Rees *et al.*, 2001). However, this study showed that larger-seeded species present lower germination percentage than smaller-seeded species for these shrubs, which was opposite to other studies in this area (Wu & Du, 2007). We consider that there are two main reasons. First, the smaller-seeded species can germinate in a broader range of microsites than larger-seeded species (Pearson *et al.*, 2003). The smaller-seeded species will face selection for low discrimination in habitat required to stimulate germination because they face the greatest risk of mortality, either through high susceptibility to pathogens or through small seed reserves, and because greater seed production decreases the relative cost of low discrimination (Smith & Fretwell, 1974); Second, the larger-seeded species of these shrubs presented a larger dormancy proportions because of thick seed capsule. Meanwhile, lower germination rate and longer germination persistence time of larger-seeded species in this study also indicated a probability of lower germination percentage during a determinate time. So, it is suggested that lower germination percentage of larger-seeded species also may be caused by experimental time

scale. Additionally, it is well known that environmental factors were also crucial for seed germination (Baskin & Baskin, 1998; Fenner & Thompson, 2005). It also can be considered as a co-evolutionary strategy to the extreme environmental conditions in alpine area. Alpine plants are exposed to high levels of stress and perturbation because of the extreme environmental conditions (Körner, 2003). The smallest-seeded species will have a greater density of propagules and germination and a less seedling survival in potential recruitment sites. However, the larger-seeded species have a greater seedling establishment, but a less density of propagules and germination (Moles & Westoby, 2004), even if under favourable conditions. This relationship would allow less discriminating germination behaviour to evolve because the increased risk of post-germination mortality would be balanced by an increased likelihood that at least one seed would be dispersed to each potentially favourable establishment site (Pearson *et al.*, 2003). The shift of environmental factors may alter this relationship between seed size and germination percentage, because seed germination strategy was a co-evolution results with their habitats (Westoby *et al.*, 1992). We suggest that interspecific correlations of seed size and germination characteristics should be taken into account in comparative studies integrated with potential variational habitats.

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