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## Relationship of *Sophora davidii* seed size to germination, dormancy, and mortality under water stress



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#### ABSTRACT

Sophora davidii is an important leguminous shrub that is widely used for revegetation in areas experiencing karst rocky desertification in southwest China. Seed size is a prominent life history trait affecting seed fate. Laboratory experiments were conducted to evaluate the effect of osmotic potential and seed size on germination, dormancy, and mortality. The osmotic potentials required for 50% inhibition of maximum germination of large, medium, and small size seeds were -0.836, -0.786, and -0.812 MPa, respectively. The highest seed dormancy (70%) was observed in small seeds at -1.0 MPa and the lowest seed mortality (6%) in large seeds at 0 MPa. Increasing osmotic potential caused a decrease in the germination, large seed dormancy, and small seed mortality. Under mild water stress, medium seed germination, large seed dormancy, and small seed size significantly affected seed germination, dormancy, and mortality, as well as the interactions between them. Our findings suggest that seed size plays an ecologically important role in the fate of *S. davidii* seeds by improving medium or small seed germination and reducing medium or small seed mortality in severe habitats of the karst rocky desert-ification region.

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#### 1. Introduction

The seed is the reproductive body of spermatophytes, and its resource contribution to seedling development has a direct effect on offspring fitness and the renewal of vegetation communities (Ramírez-Valiente et al., 2009; Susko and Cavers, 2008). Generally, larger size seeds have the advantages of higher seed germination and seedling establishment, while smaller size seeds have the advantages of escaping animal and bird predation and forming soil seed banks. and exhibit greater potentiality in vegetation renewal (Donaldson et al., 2014; Garner and Witkowski, 1998; Hanley et al., 2003; Hwang et al., 2014; Sobral et al., 2013). Under various kinds of stress, different size seeds present different fates in their life history and have different contributions to vegetation renewal. Seed fate is highly related to seed germination, dormancy, and mortality (Chauhan, 2013; Liebman et al., 2014), and correlations have been found among seed size, seed fate, and environmental conditions (Arellano and Peco, 2012; Yang et al., 2012; Zhu et al., 2004). In many species, the plant may produce few large seeds but many more small seeds because small seeds will be less vulnerable to predators and thus may have a higher chance of

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survival (Halpern, 2005; Susko and Cavers, 2008). However, large seeds usually show higher germination rates and lower dormancy rates compared with medium and small seeds (Benard and Toft, 2008; Volis and Bohrer, 2013). Information about the correlation between seed size and seed fate in various environmental conditions provides a biological basis for the conservation and reintroduction of many species.

Sophora davidii is an important leguminous scrub that is widely distributed in southwest karst areas in China (Fan et al., 2013; Gong et al., 2013; Xu et al., 2008; Zhang et al., 2006). Karst environments are fragile and very sensitive to environmental and climate changes, and the soil is often under a considerable degree of water stress (Qi et al., 2013; Yang et al., 2013). S. davidii is used as fodder, fuel wood, and in herbal medicines (Ohyama et al., 1996; Qi et al., 2013; Tanaka et al., 2000; Tobe et al., 2005; Vandelook and Van Assche, 2010). As a multipurpose species with a variety of economic and environmental uses, it has been used for fodder improvement and ecological reconstruction because of its high nutritional value and high stress tolerance. However, both the seed size of wild S. davidii populations in grams and the number of seeds produced have rapidly shrunk because of habitat destruction and overexploitation. Previous studies have focused on breaking seed dormancy and seedling physiological traits (Benard and Toft, 2008; Gad and Kelan, 2012; Li et al., 2009; Wu et al., 2008; Wu et al., 2012; Wu et al., 2009), and reported that a high seed dormancy rate, fragile seedlings and low seedling establishment are the major reasons for population degradation. Despite this information, the consideration of

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#### Table 1

Results of two-way ANOVA showing the effects of seed size and osmotic potential treatments on seeds fate in Sophora davidii.

	df	Seeds germination		Seeds dormancy		Seeds mortality	
		F-value	P-value	F-value	P-value	F-value	P-value
Treatments	5	68.134	< 0.001	39.359	< 0.001	14.940	< 0.001
Seed weight	2	12.571	< 0.001	27.271	< 0.001	10.363	0.0015
Treatments × seed weight	10	7.372	< 0.001	27.794	< 0.001	52.620	< 0.001
Error	54						

seed size and seed fate in various environmental conditions may be useful for efficient germination and adequate regeneration.

Consequently, in the present study, we examined the patterns of seed fate in relation to seed size under water stress conditions. Our major objectives were to assess correlations between seed size and seed fate features, such as seed germination, dormancy and mortality, and whether these correlations depended on water stress.

#### 2. Materials and methods

A 80

60

40

#### 2.1. Seed collection

In October 2012, freshly matured seeds of S. davidii were collected from natural populations in the karst mountain region (26°21'N, 105°49'E; 1026 m a.s.l.) near Guanling Town, southwest China. The seeds were then dried in a greenhouse for 2 days, bulked, cleaned, and stored in a dry closed cotton bag at 5  $^\circ C$  and 10% relative humidity until used in experiments (Chauhan, 2013; Hwang et al., 2014).

 $G(\%) = -0.412x^3$ 

#### Germination (%) 20 0.836 MI 0 0.0 -0.2 -0.4 -1.0 -0.6 -0.8 Osmotic petential (MPa) С 80 G(%) = $-0.7685x^{3} + 8.8373x^{2} - 36.537x + 86.667$ = 0.959 $R^2$ 60 Germination (%) 40 20 0.812 MP 0 0.0 -0.2 -0.4 -0.6 -0.8 -1.0

 $+4.7282x^{2} - 20.074x + 59.833$ 

 $R^2 = 0.999$ 

Osmotic poential (MPa)

#### 2.2. Seed size

Individual viable seeds were weighed to the nearest 0.0001 g on a microbalance. Based on this, all seeds were further graded manually into large, medium, and small sizes. The classification standard was as follows: seeds more than 1 standard deviation over the mean weight were defined as large size seeds; those within 1 standard deviation of the mean were considered medium size, and those less than 1 standard deviation below the mean were considered small (Benard and Toft, 2008). The seeds needed for the germination test were sampled in a stratified random design to ensure that a full range of seed sizes was selected from the population. The variation in our experiment was guite high (coefficient of variation = 26.594%) with a seven-fold variance in weight (ranging from 0.0031 g to 0.0214 g, mean  $\pm$  standard deviation =  $0.0119 \pm 0.0032$  g) between large and small seeds.

#### 2.3. Germination. dormancy. and mortality under water stress

The effects of water stress on germination were assessed by incubating 50 seeds of S. davidii in each of five 9-cm-diameter Petri dishes with 5 ml solutions having osmotic potentials of 0, -0.2, -0.4, -0.6, -0.8, and -1.0 MPa (250 seeds in total). The solutions were prepared by dissolving polyethylene glycol 6000 in distilled water. The dishes were placed in an incubator under alternating day/night temperatures of 30/20 °C with a 12 h photoperiod. Seed germination was recorded 60 days after the start of the experiment as no seeds germinated in a preliminary experiment in the growth chamber after this period. A seed was considered germinated when the radicle visibly protruded from the seed coat (Chauhan, 2013).



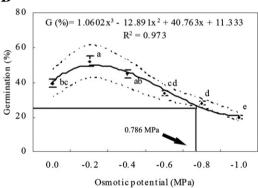


Fig. 1. Effect of seed size and osmotic potential (MPa) on seed germination in Sophora davidii. Bold letters "A," "B," and "C" represent small, medium, and large size seeds, respectively. Solid lines represent the cubic curve model fitted to the germination data for S. davidii and dashed lines represent 95% confidence interval. Vertical bars represent ± standard error of the mean if greater than the symbol size. Different lowercase letters indicate significant difference at different osmotic potentials. The germination of small and large seeds decreased persistently as the osmotic potentials increased. For medium seeds, the germination rate decreased following a sigmoid curve with an inflection point at -0.2 MPa. The osmotic potential for 50% inhibition of the maximum germination of *S. davidii* was -0.836 MPa for small seeds, -0.786 MPa for medium seeds, and -0.812 MPa for large seeds.

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