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ORIGINAL SCIENTIFIC PAPER

Effect of seed priming on germination of common bean (*Phaseolus vulgaris* L.) under drought stress

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Abstract
The aim of this study was to determine the effect of hydropriming (dH_2O; PT1) and osmopriming (-0.8 MPa PEG; PT2) on germination of two common bean cultivars ('Biser' and 'Trešnjevac') under induced drought stress. The germination capacity was tested on the germination paper soaked with dH_2O (T1) and -0.6 MPa PEG (T2) for nine days. The results showed that there was no significant difference between two common bean cultivars in terms of germination success, but there were significant differences between pretreatments (PT1 and PT2) and treatments (T1 and T2) in terms of germination success for these cultivars. Seeds of both cultivars germinate significantly faster on dH_2O (T1) when pretreatment is PEG -0.8.MPa (PT2), but dH_2O (PT1) is a better seed priming technique to use under induced drought stress (-0.6 MPa PEG; T2).

Key words: drought tolerance, hydropriming, PEG, water stress

Introduction
Common bean (*Phaseolus vulgaris* L.) is the most important grain legume for direct human consumption worldwide and has an important role in human nutrition (Murube et al., 2021). Drought stress is one of the abiotic stressors caused by climate change that limits the germination, growth and productivity of crops worldwide (Nadeem et al., 2019) and has a significant impact on food security, especially in developing countries (Hussaini et al., 2021). Drought affects various aspects of legume growth and development, and it is well known that rapid germination and establishment of seedlings provides competitive advantages over weeds and improves yields (Nadeem et al., 2019; Lei et al., 2021). Therefore, researches are focused on improving drought tolerance and production of common bean through selection for various physiological and genetic traits (Hussaini et al., 2021; Lei et al., 2021). Priming is one of the pre-sowing treatments that results in a physiological state that enables the seed to germinate more efficiently (Dhal et al., 2019). Polyethylene glycol (PEG) is a popular osmopriming agent that can mitigate the negative effects of abiotic stress and hydropriming is a simple and economical technique useful in areas with adverse environmental conditions including heat and drought stress (Waqas et al., 2019).

The aim of this study was to determine the effect of hydropriming and osmopriming on germination of two common bean cultivars ('Biser' and 'Trešnjevac') under induced drought stress.

Material and methods
This research was conducted at the University of Zagreb Faculty of Agriculture, Department of Seed Science and Technology, in 2021. Seeds of two common bean cultivars 'Biser' and

'Trešnjevac', bought on the market, were used. One hundred seeds of each cultivar were soaked in distilled water (dH_2O; PT1) and 100 seeds in -0.8 MPa polyetilene glycol (PEG 8000; PT2) for eight hours. Then the seeds were taken out, washed under running water, then under distilled water and dried between paper towels. One hundred seeds without seed priming treatment represented the control. The laboratory test for germination of common bean seeds was conducted as per the ISTA (1993) Rules. The seeds were germinated on germination paper (Munktell 21/N, 580x580mm, 80g/qm) in 10-cm-diameter Petri dishes (Steriplan®, DURAN®, DWK Life Sciences GmbH, Germany). Fifty seeds from each pretreatment [dH_2O (PT1), -0.8 MPa PEG (PT2)] and control were placed on filter paper soaked with dH_2O (T1) and -0.6 MPa PEG (T2) (Table 1) in five replicates (five Petri dishes) with 10 seeds each. Petri dishes were placed in a germination chamber at a constant temperature (22 °C ± 1 °C) with a photoperiod of 16 h of light and 8 h of darkness. The number of germinated seeds (seeds with root size ≥ 2 mm) was determined every 24 h for nine days.

Table 1. Seed priming treatments (pretreatments) and treatments used in reserch

Pretreatment		Treatment	
PT1	100 seeds dH_2O	T1	50 seeds on dH_2O
		T2	50 seeds on PEG -0.6 MPa
PT2	100 seeds PEG -0.8 MPa	T1	50 seeds on dH_2O
		T2	50 seeds on PEG -0.6 MPa

PT1 – hydropriming (dH_2O); PT2 – osmopriming (-0.8 MPa PEG); T1 –germination – (dH_2O); T2 – drought stress (-0.6 MPa PEG);

Statistical analysis

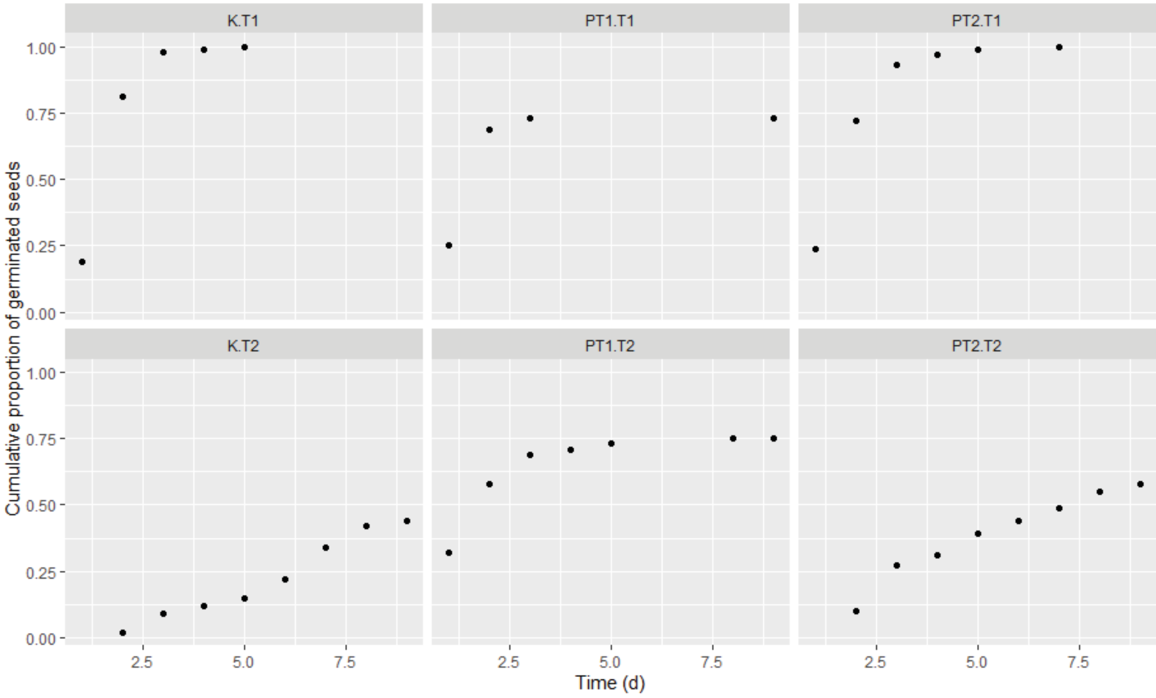
The differences among cultivars, seed priming treatments (pretreatments) and treatments in terms of seed germination were tested by survival analysis using Kaplan-Meier method with log-rank test (Kaplan and Meier, 1958) as implemented in R package ′survival′ (Therneau and Grambsch, 2000; Therneau, 2015). Mean germination time was estimated as restricted mean survival time with upper limit equals 9. The impact of cultivars, seed priming treatments (pretreatments) and treatments on germination success was tested by Cox proportional-hazards model (Cox, 1972) using ′coxph′ function from R package ′survival′ (Therneau and Grambsch, 2000; Therneau, 2015).

Results and discussion

The results of Kaplan-Meier analysis with log-rank test showed that there was no significant difference between two common bean cultivars ('Biser' and 'Trešnjevac') in germination success, but there were significant differences ($P < 0.05$) between seed priming treatments in germination success for these cultivars.

Seed from control treatment (K; without seed priming treatment) had the longest mean germination time (4.83 days), while seed in PT2 (-0.8 MPa PEG) had a mean germination time of 4.30 days, whereas seed in PT1 (dH_2O) had the shortest mean germination time (3.72 days). According to the Cox regression model for germination success, there were significant differences ($P < 0.01$) between seed priming treatment PT1 (dH_2O), which resulted in a relatively higher germination success of 40% compared to the control (K), and there were no significant differences between PT2 (-0.8 MPa PEG) and the control (K). In the T2 (-0.6 MPa PEG) treatment, germination time was 71% significantly ($P < 0.001$) longer than in T1 (dH_2O), unless the seed was previously primed with dH_2O (PT1 + T2). Germination is generally slower with T1 (dH_2O) compared to T2 (-0.6 MPa PEG), except in combination with PT1 (dH_2O) when there is almost no difference between the treatments (Graph 1).

This study provides useful information on seed germination of common bean cultivars ('Biser' and 'Trešnjevac') under hydropriming (dH_2O) and osmopriming (-0.8 MPa PEG). Although osmopriming has more advantages than hydropriming, especially better response to stress (Marthandan et al., 2020), hydropriming improved germination of common bean cultivars under drought stress in our study. In agreement with studies on other crop species, seed hydropriming improved germination parameters of chickpea (Kuar et al., 2002) and Malaysian Indica rice (Kalhori et al., 2018) under drought stress and of sunflower under salt and drought stress conditions (Moghanibashi et al., 2012). Hydropriming is a simple, cheap and environmentally friendly technique and therefore can be used to improve seed performance of common beans under different conditions and in field trials.



Graph 1. Cumulative proportion of germinated seeds by category (pretreatment-treatment combinations) and days (over two cultivars).
K – control; PT1 – hydropriming (dH_2O); PT2 – osmopriming (-0.8 MPa PEG); T1 – treatment 1 – (dH_2O); T2 – treatment 2 (-0.6 MPa PEG).

Conclusion

It can be concluded that there was no significant difference between two common bean cultivars ('Biser' and 'Trešnjevac') in terms of germination success and that seeds of both cultivars germinate significantly faster on dH_2O (T1) when seed priming treatment is PEG - 0.8.MPa (PT2), but dH_2O (PT1) is a better seed priming treatment under induced drought stress (T2; -0.6 MPa PEG).

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