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Isabgol (*Plantago ovata* Forsk) seed germination and emergence as affected by environmental factors and planting depth

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Abstract

Isabgol (*Plantago ovata* Forsk) seed germination and emergence in response to drought (Polyethylene glycol 8000) and salinity stress (NaCl), temperature, pH and planting depth were studied in laboratory and greenhouse experiments. Base, optimum and ceiling germination temperature were estimated as 3.35, 21.24 and 35.04 ^{o}C, respectively. Isabgol seed is rather tolerant to low water potential and high salt stress. Salinity stress up to 200 mM had no effect on Isabgol seed germination, but the germination decreased by increasing the salt concentration. The drought and salinity required for 50% inhibition of maximum germination were 328 mM and -1.24 MPa, respectively. High percentage of seed germination (>93%) was observed at pH=4-6 and declined to 52-58% at alkaline (pH 7-9) pH. Maximum seedling emergence occurred when the seeds were planted on the soil surface and decreased with increasing the depth of planting; no seed emerged from depth of 3 cm.

Keywords: Germination; pH; Salinity stress; Soil depth; Temperature; Water stress.

Introduction

Isabgol (*Plantago ovata* Forsk) is an annual herb that grows to a height of 30-40 cm. Leaves are born alternately opposite, linear or linear lanceolate on the stem. A large number of flowering shoots arise from the base of the plant. Flowers are numerous, small, and white. The seeds are enclosed in capsules that open at maturity. Isabgol is native to the Mediterranean region, and is found in the surrounding areas of India, Pakistan and Iran (Sharma, 2004).

Seeds and husks of isabgol have various formulation characteristics like binding, disintegrating and sustaining properties and are used widely in pharmacology as laxative, cure inflammation of the mucous membrane of gastro-intestinal and genitor-urinary tracts, chronic constipation, dysentery, duodenal ulcers and piles (Bannayan et al., 2008; Nekonam and Razmjoo, 2007; Khinchi et al., 2011).

Seed germination and seedling emergence are critical stages in the life cycle of crops. These processes determine uniformity, crop stand density, degree of weed infestation, and the efficient use of the nutrients and water resources available to the crop and ultimately affect the yield and quality of the crop (Bench Arnold, 2006; Gan et al., 1996). Seed germination is affected by a wide range of environmental factors, such as temperature, salt, water, oxygen concentration, and pH (Romo and Haferkamp, 1987; Balbaki et al., 1999; Karan et al., 1985; Swarn et al., 1999; Lu et al., 2006; Saeidi, 2008; Esmaeili et al., 2009).

Temperature is one of the major environmental factors determining the success of seed germination and early seedling growth. It affects both the capacity and rate of seed germination (Sincik et al., 2004). Other major factors that affect seed germination are water potential and salinity. Researches showed that water and salt stress can delay and reduce or prevent germination (Guan et al., 2009; Joshi et al., 2005; Kebreab and Murdoch, 2000). In addition to these factors, germination is also affected by pH and planting depth (Soltani et al., 2006; Guan et al., 2009; Norsworthy and Oliveira, 2006).

The purpose of this research was to study the effects of temperature, osmotic stress and salt stress, pH, and planting depth on isabgol seed germination and seedling emergence.

Materials and Methods

Temperature experiment

Seed germination was determined in the incubators at constant temperatures of 5, 10, 20, 25, 30 and 35 ^{o}C on 3 replicates of 50 seeds in moist paper towels in darkness. Seeds were observed twice daily and considered germinated when the radicle was approximately 2 mm long or more.

Estimates of time taken for cumulative germination to reach 50% of its maximum at each replicate (D_{50}) were interpolated from the germination progress curve versus time. Germination rate (R_{50}, h^{-1}) was then calculated as (Soltani et al., 2002):

$$R_{50}=1/D_{50} \tag{1}$$

Salt and osmotic stress experiments

Seeds were germinated in distilled water (0) and in 50, 100, 150, 200, 300 and 400 mM sodium chloride solutions. Aqueous solutions with osmotic potential of 0, -0.2, -0.4, -0.6, -0.8, -1.0, -1.2 and -1.4 MPa were prepared by using polyethylene glycol 8000 in distilled water, as described by Michel (1983). The seeds then were incubated at 20 ^oC in darkness.

pH experiment

The effect of pH on seed germination of isabgol was studied in the laboratory using buffer solutions of pH 4, 5, 6, 7, 8 and 9 prepared according to the method described by Shaw et al. (1991). The seeds were incubated at 20 ^oC in darkness.

Planting depth experiment

Thirty seeds of isabgol were placed on soil surface in 15 cm-diameter plastic pots and then covered with soil to achieve planting depths of 0, 1, 2, 3 and 4 cm. Plants were watered throughout the study as needed to maintain optimal moisture for seed germination. Plants were considered to have emerged when cotyledons could be visibly discerned and monitored daily for 15 d after planting.

Statistical analyses

All experiments were conducted with the use of a completely randomized design with three replicates.

The Segmented model (Equation 2) was used to quantify the response of germination rate to temperature and to estimate cardinal temperatures (Soltani et al., 2006).

$$f(T)=(T - T_b) / (T_o - T_b) \quad \text{if} \quad T_b < T \leq T_o \tag{2}$$
$$f(T)=(T_c - T) / (T_c - T_o) \quad \text{if} \quad T_o < T < T_c$$
$$f(T)=0 \quad \text{if} \quad T \leq T_b \ \text{or} \ T \geq T_c$$

Where T is the temperature, T_b, T_o and T_c are the base, optimum and ceiling temperatures, respectively.

Planting depth data were fitted to a linear regression model, whereas salt and osmotic stress data showed a sigmoidal trend and a three-parameter logistic model was fitted to data (Equation 3):

$$G(\%)=G_{max} / (1 + (x / x_{50})\text{^}Grate) \tag{3}$$

Where G is the total germination (%) at concentration x, $Gmax$ is the maximum germination (%), x_{50} is the NaCl or osmotic potential required for 50% inhibition of the maximum germination and $Grate$ indicates the slope of the curve in x_{50}. Statistical Analysis System (SAS) was used for analyzing data (SAS Institute, 1989).

Results and Discussion

Maximum germination was observed between 10 and 25 ^{o}C and was about 84-92% (Figure 1a). A marked decrease in germination occurred when temperature was outside this range. At 5 and 30 ^{o}C, the germination percentage was 78 and 46%, respectively; both lower than all other temperatures and seeds did not germinate at a constant temperature of 35 ^{o}C. These results suggest that Isabgol seeds can germinate over a range of temperatures (especially at low temperatures) and can reach more than 50% germination (ranging from 78-92%).

Figure 1b shows the relationships between germination rate and temperature. The influence of the temperature on germination rate was described by a Segmented model. Crops have three cardinal temperatures which are the base, ceiling, and optimal temperatures. Germination rate increases from the base to optimum temperature and then decreases to a ceiling temperature (Ghaderi et al., 2008). Base, optimum and ceiling temperatures were estimated as 3.35, 21.24 and 35.04 ^{o}C, respectively. The cardinal temperatures for germination have been determined for different plants (Mwale et al., 1994; Ramin, 1997; Phartyal et al., 2003; Jami Al-Ahmadi and Kafi, 2007).

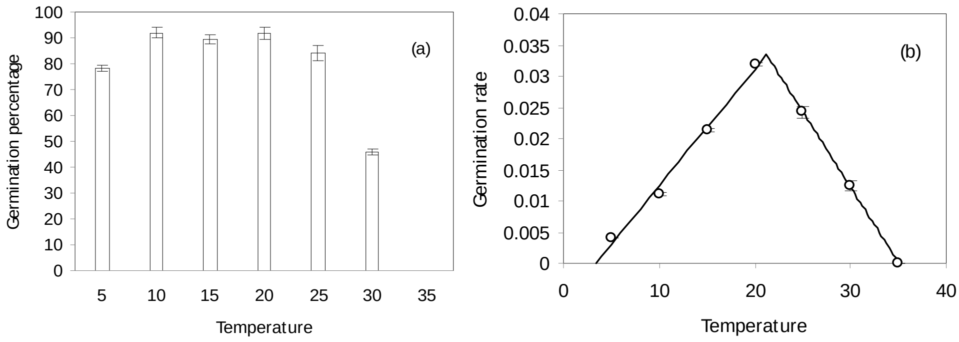

Figure 1. Effect of constant temperature on (a) germination percentage and (b) germination rate of Isabgol. Bars represent standard error of the mean.

Salt tolerance during germination and early seedling stages is critical for the establishment of plants. Seeds germination isabgol was significantly affected by salinity and increase in salinity causes a decrease in germination (Figure 2). Seed germination did not decrease significantly from 0-200 mM, but beyond this point declined with increasing salinity following a sigmoidal trend (Figure 2). The concentration required for 50% inhibition was 328 mM NaCl. These results suggest that even at high soil salinity, isabgol seed may germinate. Salinity might negatively affect some important physiological processes in plants. Additionally, sodium ions can alter soil structure and fertility by replacing calcium and magnesium in anion exchange process, and this leads to nutrient and water stress (Rao et al., 2008).

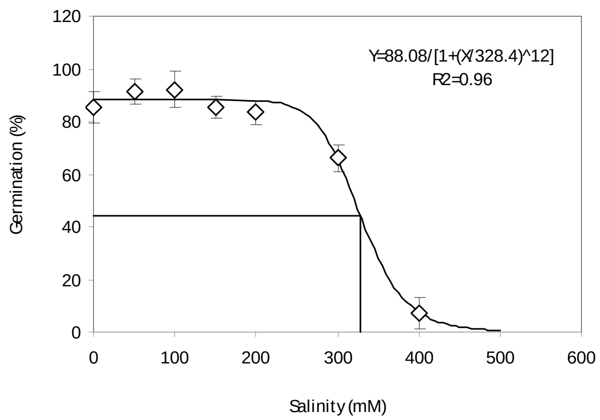

Figure 2. Effect of NaCl concentration on germination of Isabgol. Bars represent standard error of the mean.

A similar trend to that of salinity was observed for the water stress. Germination percentage decreased from 93-53% as osmotic potential decreased from 0 to -1.2 MPa, and was completely inhibited at osmotic potential of -1.4 MPa (Figure 3). The osmotic potential required for 50% inhibition of maximum germination was -1.24 MPa. Germination decreased sharply as osmotic potential decreased beyond -1.0 MPa. These findings indicate that isabgol is rather resistance to drought stress.

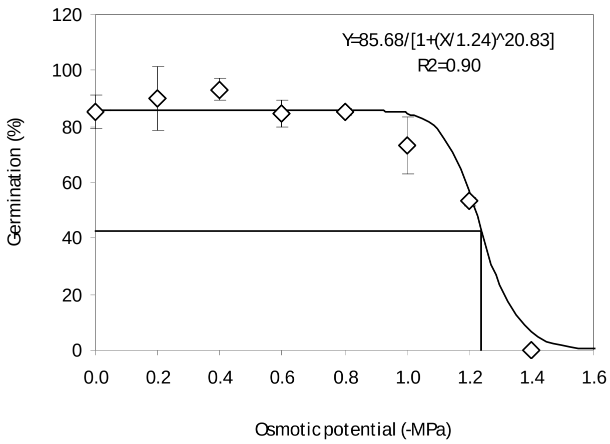

Figure 3. Effect of osmotic potential on germination of Isabgol. Bars represent standard error of the mean.

The germination occurred in a range of pH between 4 and 9 in which the maximum germination was observed between 4 and 6 (87-93%) (Figure 4). Germination started to decline as pH increased from 7 to 9 (52-61%). Based on these results, Isabgol tends to germinate better in acidic soils compared with alkaline soil environments.

Seedling emergence of Isabgol was influenced by planting depth of seed (Figure 5). Emergence of Isabgol decreased rapidly with increased planting depth. Isabgol seeds on the soil surface had the highest emergence (80%), and no seedlings emerged from seeds placed at a depth of 3 cm. Emergence declined at a rate of 27% per cm of planting depth. Decreasing in seedling emergence at deeper soil depths has been reported (Gan et al., 2003; Shanmuganathan and Benjamin, 1992; Soltani et al., 2006; Yousefi-Daz, 2005) and usually is related to light and seed size. Benvenuti (1995) reported that very little light is transmitted below a 4 mm depth in all soil types. Therefore, this research indicates that light is not a requirement for Isabgol seed germination because 54% of seeds germinated at a depth of 1 cm and also, in the light/dark experiment, there are not differences between germination in the dark and light (data not shown). Lower seedling emergence of seeds at deep depths may be linked to seed size and reservation (Mennan and Ngouajio, 2006). Because of the small size of Isabgol seeds (about 1.8-2 mg seed mass), the decrease emergence of Isabgol seeds could be related to the inadequate energy reserve to support germinated seedling growth, which make it difficult to emerge even from a planting depth of 3 cm.

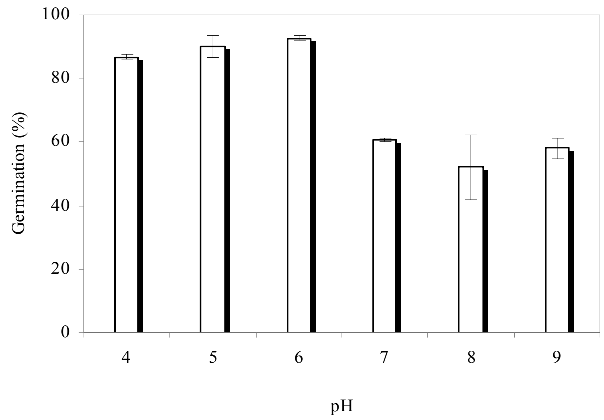

Figure 4. Effect of buffered pH solutions on germination of Isabgol. Bars represent standard error of the mean.

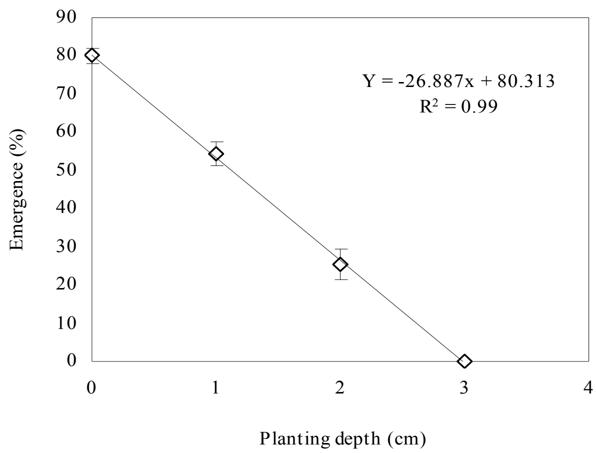

Figure 5. Effect of planting depth of seed in soil on emergence of Isabgol. Bars represent standard error of the mean.

Clear understanding of the germination response of seeds to environmental factors and agronomic aspects are useful in screening crop tolerance to stress, identifying geographical areas where a crop can germinate and establish successfully and developing management models for the prediction of timing of crop development processes. These studies revealed that Base, optimum and ceiling germination temperature was estimated as 3.35, 21.24 and 35.04 ^{o}C, respectively and Isabgol seed is rather tolerant to low osmotic potential and high salt stress, and is able to germinate over a broad range of pH level. But, planting depth affects emergence of isabgol seed and emergence decreased linearly with increased planting depth, and no seedling emerged from depth of 3 cm.

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