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SEED GERMINATION AND EARLY SEEDLING GROWTH OF PEA (*Pisum sativum* L.) IN RESPONSE TO SEED PRIMING

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Abstract

The early growth ability of plants strongly influences their growth and yields. Seed priming is widely used for better germination and emergence of plants both under stressful and non-stressful conditions. The purpose of this study was to determine the effects of seed priming on some growth parameters of Pisum sativum seedlings and to identify the most suitable priming techniques. For this reason, a laboratory experiment was carried out in randomized blocks with three repetitions, in which pea seeds were primed for 6 hours with distilled water and with two concentrations and combinations of salicylic acid and calcium carbonate (CaCO3). The treatment non-priming was used as a control. The results showed that the priming treatments had significant effects on all studied parameters (p<0.05). Seed priming with both concentrations of calcium carbonate (1 g/l and 2 g/l) and hydro-priming (with water) showed the best priming effects on root and shoot length, root and shoot fresh weight, and seedling vigour index. Therefore, these priming techniques, which are cheap and ecological, can be used by farmers for the successful establishment of pea crops.

Key words: *calcium carbonate, germination seed, pea, salicylic acid, seedlings.*

INTRODUCTION

Pea (*Pisum sativum* L.) is one of the most important food legumes worldwide due to its multiple uses: forage dry matter, green grains, green fodder, flour, straw and green manure. It is a rich source of protein because 100 grams of green pea (grains) contain 6.55 g of protein, 0.84 g of lipids and 0.92 g of mineral salts (Roman et al., 2011).

In Romania, pea seeds are generally sown in autumn or early spring, thus being exposed to cold stress. This low-temperature stress during germination can lead to suppression of germination and low seedling vigour. Also, other factors can lead to a decrease in pea production, such as fungal infections, the small number of nodules, the slow process of grain filling, and to overcome these problems, Gour et al. (2019) recommend the seed priming method.

The decisive factors that ensure successful crop establishment are uniform germination and seedlings vigour, because these factors contribute to the uniform growth and maturity of the plants and finally to a high yield (Finch-Savage & Bassel, 2016; Drăghici et al., 2021; 2022). Therefore, the main purpose of the farmer is to stimulate some essential stages in ensuring high yields by improving germination and seed vigour (Karim et al., 2020).

Priming is a simple method used for rapid germination and emergence that is applied before sowing and consists of partially hydrating the seeds with different priming agents, followed by drying the seeds. According to Elkoca (2014), the priming method permits partial seed hydration so that pre-germination metabolic activities proceed but primary root protrusion is prevented. This is a simple, low-risk, low-cost technique that can be useful for farmers (Dessalew et al., 2022).

Notable results for various plant species regarding improving the germination and seedling emergence by different priming agents (i.e. polyethylene glycol, abscisic acid, glycinbetaine, gibberellic acid, salycilic acid, KNO_3, etc.) or plant extracts, were reported previously (Hussain et al., 2015; Bonea, 2016; Kumari et al., 2017; Bonea, 2018; 2020; Jia et al., 2020).

However, the success of seed priming depends on several factors such as priming agents, duration of priming and seed condition. Therefore, optimizing the priming technique for a specific crop is an area worth exploring to improve crop productivity under various environmental conditions (Karim et al., 2020).

Some previous studies have reported that hydro-priming is an agronomically efficient procedure with the potential to increase tolerance to many environmental stress conditions (Plazek et al., 2018; Rhaman et al., 2021).

Salicylic acid has an important role in regulating many plant processes increasing tolerance to abiotic stress (Noreen et al., 2009; Ma et al., 2017).

Many studies found that seed priming with salicylic acid improves the germination and viability of seeds in wheat (Soare et al., 2015), rice (Shatpathy et al., 2018), faba bean (Anaya et al., 2018), okra (Rhaman et al., 2021).

Calcium is an essential nutrient for plant growth, having various structural roles in cell walls and membranes and in key enzyme activities (White & Broadley, 2003). Calcium also has a central role in many defense mechanisms that are induced by stress (Cousson, 2009).

Several studies have reported that $CaCl_2$ (as a calcium source) used as a seed priming agent in different plant species stimulated germination and vegetative growth under drought or salinity conditions (Aroubandi, 2016; Kaczmarek et al., 2017; Gao &Yan, 2020), but little information exists concerning the effect of $CaCO_3$ on seed germination and seedling growth.

In the context of the previously mentioned, this study was carried out to evaluate the effects of some priming treatments on growth parameters of pea seedlings.

MATERIALS AND METHODS

Plant material

The pea seeds (cv. Kelvedon) were purchased from Mefim Agro SRL (Craiova, RO).

An experiment was carried out at the Faculty of Agronomy, Breeding of plants laboratory in 2022. The seeds were sterilized with 5% sodium hypochlorite solution for five minutes and rinsed with distilled water, then dried at 22±2°C for 24 hours.

Seed Priming Protocol

For priming, pea seeds were subjected to hydro-priming (distilled water) and priming with 1 g/l and 2 g/l of salicylic acid (SA); 1 g/l and 2 g/l of calcium carbonate ($CaCO_3$) and combinations of 1 g/l and 2 g/l $CaCO_3$ + 1 g/l and 2 g/l SA for 6 hours, then dried to the original moisture content.

Non-priming treatment was used as a control.

Germination test

Three replicates, each with 30 seeds for each treatment variant, were germinated on two layers of filter paper in plastic pots. 5 ml of distilled water was added to each pot for seven days. The plastic pots were kept at room temperature (22±2°C).

Data Collection

Seed germination was recorded every 24 h for 7 days, and seeds were considered germinated when their radicle was at least 2 mm long.

The following germination and seedling vigour parameters were measured using the following equations:

Germination percentage (%) = (Nr. germinated seeds/Total nr. of seeds) × 100

The seedling vigour index (SVI) was calculated according to Abdul-Baki & Anderson (1973) as:

Seedling vigour index (SVI) = Seedling length × G (%),

Where G, is the germination percentage

Seven days after seed priming, ten normal seedlings were randomly selected from each plastic casserole to record seedling growth parameters, i.e. root length and shoot length, and their fresh weight.

The length was measured with a ruler and the fresh weight was determined using a precision balance.

Statistical analysis

Statistical analysis was performed using the analysis of variance (ANOVA), and treatment means were compared using Duncan's multiple comparison tests at a 5% level of probability.

RESULTS AND DISCUSSIONS

According to the results, all studied parameters (germination percentage, root length, shoot length, root fresh weight, shoot fresh weight, and seedling vigour index were significantly affected by different priming treatments (Table 1).

Significantly higher germination percentages were recorded priming pea seeds with calcium carbonate in both concentrations of 2 g/l and 1 g/l (98% and 95%), in the treatment non-priming (95%) and in the treatment with hydro-priming (87%), and a significantly lower germination percentage was recorded in the treatment with 2 g/l salicylic acid (12%), compared to other treatments (Figure 1).

Table 1. ANOVA table for effects of priming treatments on studied parameters

Parameter	MS	F-value
Germination percentage	2928.420	36.99*
Root length	5.779	8.47*
Shoot length	10.053	16.38*
Root fresh weight	0.007	15.87*
Shoot fresh weight	0.015	12.31*
Seedling vigour index	354910.100	17.19*

* - Significant at 5% level of significance; MS - Mean sum of squares.

Anaya et al. (2018) pointed out that a lower concentration of salicylic acid can improve seed germination by increasing gibberellic acid biosynthesis. At the same time, a high concentration of salicylic acid can stop seed germination as a result of enhancing ABA synthesis (Wu et al., 1998) or can increase oxidative stress that leads to the degradation of leaf pigments (Rhaman et al., 2021).

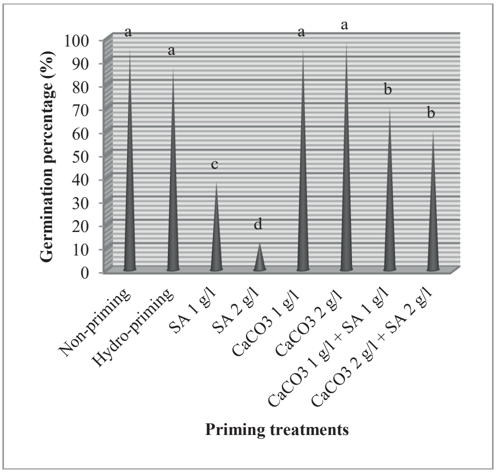

Figure1. Germination percentage of pea seeds in different priming treatments. Different letters show significant differences at the 5% probability level

Significantly, maximum root length (4.76 cm) was recorded in seed primed with 1 g/l calcium carbonate as compared to other priming treatments, followed by hydro-priming (3.97 cm) and priming by 2 g/l calcium carbonate (3.81 cm). The lowest root lengths (0.83 and 1.27 cm) were observed when priming seeds with both concentrations of salicylic acid (1 and 2 g/l) (Figure 2).

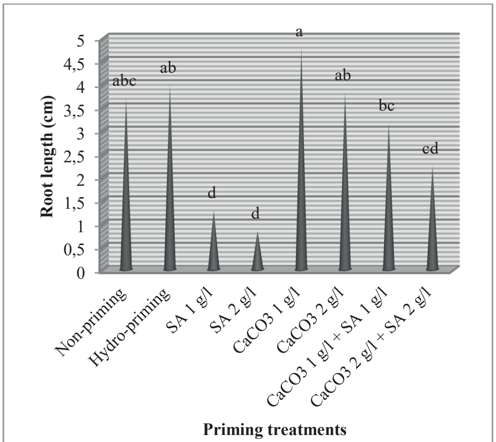

Figure 2. Root length of pea seed in different priming treatments. Different letters show significant differences at the 5% probability level

Significantly highest shoot lengths were recorded in the seed primed with 2 g/l calcium carbonate (4.74 cm), hydro-priming (4.24 cm) and priming with 1 g/l calcium carbonate (4.23 cm). Shoot lengths significantly lower, compared to other priming treatments, were recorded in the seed priming with 2 g/l salicylic acid (0.33 cm), the priming with 1 g/l salicylic acid (0.59 cm) and the priming with the combination of 2 g/l salicylic acid + 2 g/l calcium carbonate (0.63 cm) (Figure 3).

For the fresh weight of the root, significantly high values (0.173 g) were recorded in the treatment with 1 g/l calcium carbonate (0.173 g) followed by the treatment with 2 g/l calcium carbonate (0.129 g), and the lower value was recorded in the treatment with 2 g/l salicylic acid (0.031 g) compared to other treatments (Figure 4).

Significantly higher shoot fresh weights were recorded in seed primed with both concentrations of 1 and 2 g/l calcium carbonate (0.182 g and 0.165 g, respectively) and in hydro-priming treatment (0.158 g), whereas lower values were found in the non-priming treatment and in other priming treatments (Figure 5).

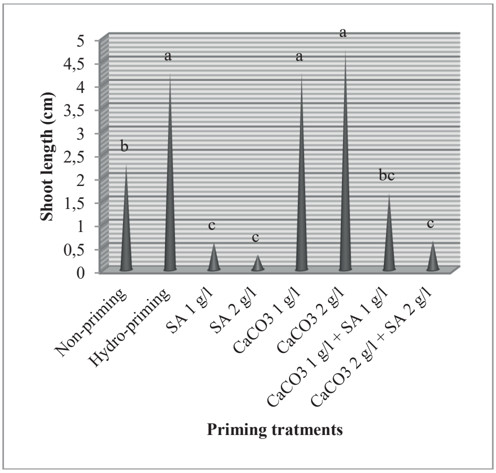

Figure 3. Shoot length of pea seed in different priming treatments. Different letters show significant differences at the 5% probability level

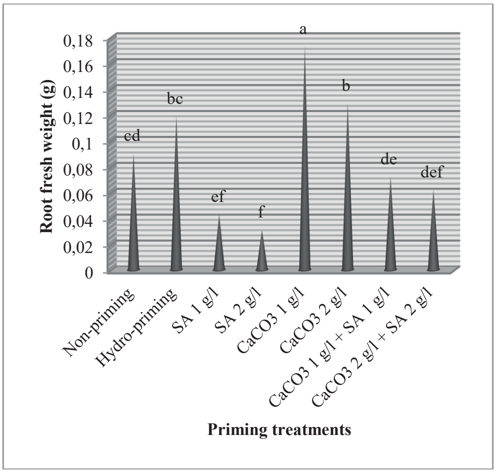

Figure 4. Root fresh weight of pea seed in different priming treatments. Different letters show significant differences at the 5% probability level

Significantly, maximum seedling vigour indices were recorded in seed priming with calcium carbonate in both concentrations of 1 and 2 g/l (855.3 and 843.5, respectively) followed by hydro-priming treatment (711.0) as compared to other treatments. The lowest seedling vigour index was observed in priming with 2 g/l salicylic acid (13.8) (Figure 6).

Hao et al. (2020) reported that high seed vigour would determine the potential for rapid and uniform seed emergence and can increase yield by up to 20%.

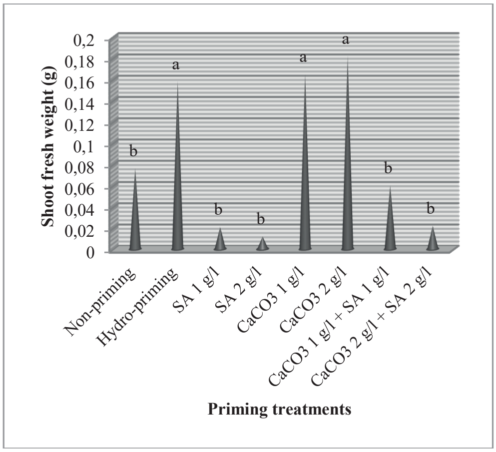

Figure 5. Shoot fresh weight of pea seed in different priming treatments. Different letters show significant differences at the 5% probability level

According to Yanglem et al. (2016), this could be due to the removal of harmful substances such as inhibitory hormones and higher seed leachates at the time of seed invigoration.

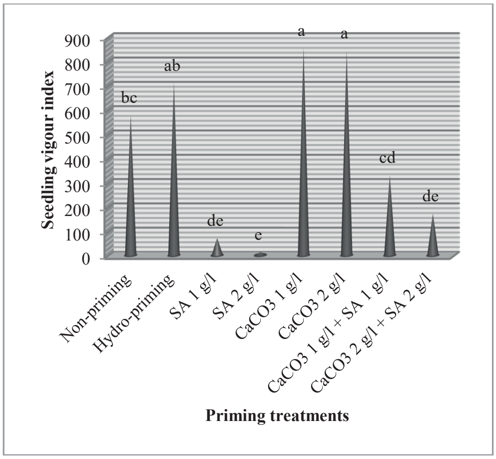

Figure 6. Effect of different seed priming treatments on seedling vigour index. Different letters show significant differences at the 5% probability level

A positive effect of calcium carbonate was also observed by Sadak and Talaat (2021) who reported that seed priming with 20 mg/l and 40 mg/l calcium carbonate significantly promoted wheat growth under normal and stressed conditions. Belur et al. (2010) reported that higher seed quality parameters in soybean could be obtained by pre-sowing seed treatment of 1% calcium carbonate, compared to hydro-priming treatment.

Other studies have shown that priming wheat seeds with combinations of Ca^{2+} and salicylic acid increased the production of phenolics compounds, thus leading to better seed vigour (Yücel & Heybet, 2016).

CONCLUSIONS

The results of the present experiment showed that different priming treatments had significant effects on the germination percentage and seedling growth of pea.

Priming with both concentrations of calcium carbonate (1 g/l and 2 g/l) and hydro-priming showed the most significant effects on root and shoot length, root and shoot fresh weight, and seedling vigour index.

These priming techniques, which are cheap and ecological, can be used by farmers to improve the growth of pea seedlings when the plants are not growing properly as they expected.

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