Effect of gibberellic acid on the seed germination of *Lavandula angustifolia* Mill.

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ABSTRACT

Lavandula angustifolia Mill. is a perennial plant, which is part of the *Lamiaceae* family, it is known as a medicinal and aromatic plant, however in recent years it can be found as landscaping, cut flower, or even in the art of culinary. This majestic plant propagation by seeds is a slow, hard difficult stage, for this reason, new methods are needed. Biostimulants could improve seed germination percentage and time, and also could improve productivity and growth of the plants. This study aimed at the effect of GA₃ on two commercial cultivars of *Lavandula angustifolia* Mill. Lavender seeds were subjected to three concentrations of gibberellic acid (100, 200, 300 ppm), under controlled conditions in the growing chamber. Under our experimental conditions GA₃ improved the germination percentage and time of lavender seeds. It can be concluded that biostimulators, as gibberellic acid could have an effect on dormancy breaking of *Lavandula* seeds.

Keywords: biostimulants, dormancy, GA₃, germination, lavender.

INTRODUCTION

Today there is a growing interest in the *Lavandula angustifolia* Mill. species industry, but it is also recognized in academia, health sciences, cosmetics as well as art. The aromatic Mediterranean plant has been known and used since ancient times (Hanamanthagouda *et al.*, 2010; Pohrib and Nistor, 2012). Lavender is a beautiful addition to almost any garden (Berringer Bader, 2012). The *Lavandula* genus includes more than 45 species and about 400 of cultivars (Abdelkader *et al.*, 2011; Koulivand *et al.*, 2013). It is native to the Mediterranean Basin, from southern Europe to North and East Africa and countries from the Middle East to southwest Asia and southeast India (Lambardi *et al.*, 2013; Lis-Balchin, 2017).

More and more farmers are choosing to grow this wonderful plant because it is a plant that does not require a lot of care and attention, but at the same time, it is also considered a profitable crop.

Lavender can be propagated by seeds, vegetative with cuttings of shoots, or layering. Sowing is done in greenhouses (in boxes), solarium, in March, or outside in the nursery, in May. The optimum temperature for seed germination is 18°C, the plants sprout in 14–20 days from sowing (Toma and Petra, 2020).

However, the germination of *Lavandula* seeds is a problem in growing these plants. The germination rate depends on the quality of the seeds, their dormancy percentage, and the degree of dormancy of each seed (Chavagnat, 1977). Seed dormancy is a bottleneck for some plant species, necessary to complete germination (Bewley, 1997). Lavender propagated by generative propagation (seeds) is used more for plant breeding. Although this propagation is cheaper, it is not used much in large crops because the germination capacity is low and not uniform.

Germination biostimulators can influence germination percentage and release dormancy, the dormant state of lavender seeds (Yang *et al.*, 2020). According to previous research seed dormancy and the growth of these seed-derived plants can be influenced by the use of hormones/growth promoters (Giannoulis *et al.*, 2020; Jelačić *et al.*, 2008; Liopa-Tsakalidi *et al.*, 2011). The fruits of the lavender comprise four nodules, located at the base of the persistent calyx of brown, grayish to black color. Lavender seeds are hard, smooth, shiny and covered with a weakly permeable membrane; they, therefore, germinate slowly and over a long period. They are very small, elongated, about 1 mm thick and 2 mm long (Butnaraş, 2016; Vârban and Vârban, 2017). According to Toma and Jitareanu (2007), GA₃ is one of the most used gibberellins. Gibberellin treatments can completely replace vernalization; moreover, vernalization can induce plant sensitivity to the action of gibberellins (Chouard, 1960; Levy and Dean, 1998). Brewster and Butler (1989) studied the effect of gibberellic acid (GA₃) on *Allium cepa* species.

Performing treatments before vernalization, the results aimed to increase the number of flowers in some genotypes studied. Also, Naamni *et al.*, (1980) have also been performed numerous studies on the administration of gibberellic acid (spraying, immersion, injection). Doses and methods of application greatly influence the response of plants to these treatments.

Gibberellic acid (GA₃) plays a key role in releasing dormancy and promoting seed germination (Kucerna *et al.*, 2005). GA₃ is an essential biostimulator, for several plant development processes, this includes germination, plant growth, and effective induction of flowering; it significantly reduces the stratification requirement of slow germinating seed species (Kitchen and Meyer, 1991). It is also a natural regulator, which is used in agriculture and horticulture, having a considerable impact on plant growth (Cornea-Cipcigan *et al.*, 2020; Nasri *et al.*, 2014). Positive effects of GA₃ on seed germination have been demonstrated in multiple ornamental plants (Lee *et al.*, 2016; Urbanova and Leubner-Metzger, 2018; Nasri *et al.*, 2014), but also in lavender seeds (Liopa-Tsakalidi *et al.*, 2011; Singh and Srivastava, 1990; Chavagnat, 1977).

The dormancy of lavender seeds is a complicated problem at the moment, as the seeds germinate hardly and slowly. Regarding this, the experiment aimed to find new and efficient methods to increase the germination rate of *Lavandula angustifolia* Mill. seeds.

MATERIALS AND METHODS

The study was conducted in 2018 at the University of Agricultural Sciences and Veterinary Medicine Cluj-Napoca the Institute for Advanced Horticultural Research of Transylvania, Romania.

We have selected two commercial lavender cultivars highly cultivated in Romania: *Lavandula angustifolia* 'Codreanca' approved in 1992, considered to be one of the cultivars that are very well suited to the climatic conditions in our country, being frost resistant and having a volatile oil concentration of 1.40-1.48% (Butnaraş, 2016). On a floral stem, there are 5-6 small flowers arranged in terminal spiky inflorescences, their color is a dark blue to purple, and they give off a pleasant fragrance.

Lavandula angustifolia 'Sevtopolis' is native to Bulgaria. Of all the cultivars approved in Bulgaria, 'Sevtopolis' has the highest values for basic chemical components such as *Lavandulyl acetate*, *Lavandulol*. 'Sevtopolis' is 40-60 cm tall. The flowering period is in May-July, which requires a suitable sunny and semi-shady environment. In the above-mentioned cultivars, it has been observed that 6-7, even 8 flowers per 7 cm of floral stem and their color is light blue-violet to silver-blue.

In this experiment, the influence of the biostimulator GA₃ (gibberellic acid) on the generative propagation of lavender was investigated. The experiment was bifactorial design with the following factors (Table 1).

Combining the two experimental factors resulted in eight experimental variants. Twenty seeds of each experimental variant were placed in each Petri dish and GA₃ (gibberellic acid) was added in different concentrations, 0 ppm (Control), 100 ppm, 200 ppm, 300 ppm. For the concentrations, 1 mg, 2 mg, and 3 mg of gibberellic acid respectively were weighed and 10 ml of distilled water was added to each. Each day seeds germinated were counted and noted.

Calculation of ppm (parts per million) concentrations:

$$(X \text{ mg} / Y \text{ ml}) \times 1000 = Z \text{ ppm.}$$

$$(1 \text{ mg} / 10 \text{ ml}) \times 1000 = 100 \text{ ppm.}$$

$$(2 \text{ mg} / 10 \text{ ml}) \times 1000 = 200 \text{ ppm.}$$

$$(3 \text{ mg} / 10 \text{ ml}) \times 1000 = 300 \text{ ppm.}$$

Germination percentages of lavender seeds were calculated according to the Timson germination rate test (Timson, 1965), modified by Khan and Ungar (1984):

$\sum G/t$; where G is the percentage of seeds that germinated after 1-day intervals and t is the total germination time.

GP = Seed germination/Total seeds $\times$ 100 (Liopa-Tsakalidi *et al.*, 2011; Vicente *et al.*, 2004).

Table 1. Lavender seed germination

Factor A –Cultivar with 2 graduations	Factor B – GA ₃ with 4 graduations
a ₁ – <i>Lavandula angustifolia</i> 'Codreanca'	b ₁ – 0 ppm (Control)
	b ₂ – 100 ppm
	b ₃ – 200 ppm
	b ₄ – 300 ppm
a ₂ – <i>Lavandula angustifolia</i> 'Sevtopolis'	b ₁ – 0 ppm (Control)
	b ₂ – 100 ppm
	b ₃ – 200 ppm
	b ₄ – 300 ppm

Lavender seeds were germinated in a growth chamber (EKOCHL 700), where the humidity was between 60–70 % and the temperature was between 18–20°C.

The results of the experiment were statistically analyzed using Paired Two Sample t-test for Means (<0.05), with Microsoft Excel software and post hoc comparisons were made using a Tukey HSD test.

RESULTS AND DISCUSSIONS

Seeds of the two lavender cultivars (*Lavandula angustifolia* 'Codreanca' and *Lavandula angustifolia* 'Sevtopolis') have been placed to germination with GA₃ biostimulator on 20.03.2018. Within seven days, on 26.03.2018, the first seeds started germinating in Petri dishes to which gibberellic acid was added. At each concentration the number of germinated seeds was different, but in the Petri dishes for the control seeds germination only started after 11 days, on 30.03.2018 and in smaller numbers.

Observations on the germination of lavender seeds lasted 35 days and in the end differences in germination between treatments could be observed. It could also be observed visually that the lavender seeds treated with gibberellic acid were better developed and grew at a faster rate.

Gibberellic acid plays an important role in seed germination (Ritchie and Gilroy, 1998) through a multiple biostimulator mechanism (Fincher, 1989). However, the stimulatory effects of gibberellic acid on seed germination are not similar in all crop species (Bell *et al.*, 1995). There are many reports on how gibberellic acid could influence seed germination. According to the results presented by Liopa-Tsakalidi *et al.*, (2011) seed germination rate increased in lavender when GA₃ solutions were added. A study by Chavagnat (1977), on dormancy and seed germination, showed that GA₃ favorably influenced seed germination when applied at the dose of 200 ppm. Other studies carried out by Janowska and Stanecka (2011), confirm the effectiveness on leaf longevity when preserving *Zantedeschia* treated with GA₃ solution with a high concentration of 400 mg/l.

Gibberellic acid had an increased number of the germinated seeds in both lavender cultivars, compared to control (Figure 1 and 2). However, seed germination was treatment dependent, seeds under the effect of GA₃ had an increased number of germinations, and also the under the treatment it was recorded quicker germination.

First seeds germinated were observed at the GA₃ treatments, which were recorded after 7 days, on contrary, the lavender seed in the control treatment started germinating after 11 days.

Figure 1. Number of seeds germinated at 'Codreanca' cultivar

Figure 2. Number of seeds germinated at 'Sevtopolis' cultivar

Analysing Figure 3 and 4 it can be observed that the highest germination percentage in both cultivars it was recorded at the 300ppm treatment (Figure 5d) of gibberellic acid. In the cultivar 'Codreanca' the germination percentage reaches 95% and, in the cultivar, 'Sevtopolis' the germination percentage was 90%.

The seeds under 200 ppm GA₃ treatment (Figure 5c) reported an increased germination percentage followed by the GA₃ 100 ppm treatment (Figure 5b). Thus, at GA₃ 200 ppm treatment the 'Codreanca' cultivar reached a germination percentage of 85% and the 'Sevtopolis' cultivar 75%, and at 100 ppm gibberellic acid treatment the Romanian cultivar 'Codreanca' had a germination percentage of 80% and the Bulgarian cultivar 'Sevtopolis' a germination percentage of 65%. In contrast the lavender seeds in the control treatment (Figure 5a) had a much lower germination percentage, in 'Codreanca' only 30% and in 'Sevtopolis' 35%.

Similar results were obtained by Roşca *et al.*, (2016) in *Allium* 'Purple Rain', who reported that with increasing GA₃ concentrations up to 500 ppm the best germination increases were observed.

Figure 3. Germination percentage at 'Codreanca' cultivar

Figure 4. Germination percentage at 'Sevtopolis' cultivar

Cornea-Cipcigan *et al.*, (2020) has experimented to evaluate the influence of gibberellic acid (50, 100, or 150 mg/L), on the germination earliness of seeds, along with the growth and ornamental quality of selected *Cyclamen* species. They were found a significant difference in germination percentage (GP), mean germination time (MGT) and seedling vigor index (SVI), within different GA₃ supplementation and greenhouse sunlight intensity (short and long days).

Figure 5. Seed germination of *Lavandula angustifolia* 'Codreanca' and 'Sevtopolis';
a–Control; b–100 ppm; c–200 ppm; d–300 ppm

CONCLUSIONS

From the present study can be concluded that lavender seeds under the effect of GA₃ at high concentrations above 100 ppm resulted in higher germination percentages than untreated variants in both *Lavandula angustifolia* cultivars. Thus, was observed that GA₃ treatment reported a germination percentage of 80-95% at 'Codreanca' and 65-90% at 'Sevtopolis' (depending on the treatment), compared to 35% and 30% in the untreated variants (control). Due to the high germination percentage, we can state that the dormancy, the resting state of the seeds was positively influenced by GA₃. However, the excess of GA₃ can also negatively influence plant growth, as plants develop and grow at a higher rate, which can lead to quicker mortality.

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