RESEARCH PAPER



## Determination of the Efficiency of Moringa Leaf Extract Treatments on Seedling Vigor in Marigold

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### Abstract

The effect of different planting depths on emergence characteristics was investigated in this study carried out using seeds of Marigold (*Tagetes patula*) collected from nature. In addition, the most effective dose that can be used in priming treatments was tried to be determined by examining the effects of moringa leaf extract treatments applied at different doses in terms of emergence rate, emergence time, survival seedling rate, true leaf emergence time, emergence speed index, coefficient of velocity of emergence and vigor index values. After the T test, it was determined that different planting depth treatments created statistically significant differences especially in the survival seedling rate. It has been observed that the fact that the planting is exposed causes an increase in the survival seedlings rate. In addition, following hydropriming and moringa treatments, the highest seedling emergence rate at both planting depths was achieved with hydropriming and moringa leaf extract (4 g L<sup>-1</sup>) treatment. Moreover, moringa leaf extract (8 g L<sup>-1</sup>) treatments exhibited the earliest emergence compared to other treatments. All treatment groups were statistically different compared to the control group in terms of emergence speed index, coefficient of velocity of emergence and vigor index values in both planting depths, and HP and M3 treatments were the treatment groups that gave the best results. As a result of the study, it has been determined that surface sowing can be recommended in marigold cultivation and moringa treatments improve the emergence characteristics.

#### 1. Introduction

There are approximately 50 species in the *Tagetes* genus. These species are especially widespread in the United States, Argentina and Chile. In addition to being terrestrial, subaquatic and perennial species within the genus, they also have a lot of diversity in terms of flower and leaf characteristics. It is known that the leaves of the species in the Tagetes genus, like other members of the family, have secretory cavities containing fragrant and essential oils (Hinojosa Espinosa and Schiavinato, 2022).

In ornamental plant cultivation, marigold is grown in many different aspects all over the world. It has features such as being suitable for pot, field and garden cultivation, having a long flowering period (summer and autumn), being used as a cut flower and containing a wide variety of species within the family (Priyanka et al., 2013; Singh et al., 2016). Tagetes is a species that has recently become widely used in sustainable agriculture and organic farming practices, with its many useful components such as thiophenes, alkaloids, polyacetylenes, fatty acids, flavonoids and terpenes. It is used as a low-cost product because it has antimicrobial and nematicidal effects and uses plant material of natural (biological) origin (Santos et al., 2015). In addition to these advantageous aspects, it is grown in many different species such as melon (Mavi and Atak, 2016), bell pepper (Mavi, 2016), tomato (Mavi and Uzunoğlu, 2020a), gourd (Mavi, 2020) and pepper (Mavi and Uzunoğlu, 2020b). With its allelopathic effect, it is used as an organic priming material in pre-sowing seed treatments.

Pre-sowing seed treatments aim to increase germination and emergence rates, provide uniform emergence, as well as improve seedling quality and increase the plant's resistance to stress factors. While marigold cultivation is generally carried out at 25°C, it is known that temperatures above 35°C prevent germination (Drennan and Van Staden, 1989). While the optimum temperature value is 25°C, low temperatures may have a negative effect on the emergence rate in early plantings. Like other species in the family, marigold is known as a species that requires light during emergence. The aim of the study is to break the dormancy of the species regarding its need for light during emergence by applying different planting depths. In addition, it was aimed to determine the effective dose for use in organic priming treatments with moringa leaf extracts used in different doses.

#### 2. Material and Methods

The study was conducted in Hatay Mustafa Kemal University Seed Physiology Laboratory and unheated glass greenhouses between January 2023 and March 2023. Marigold (*Tagetes patula*) seeds, which are used as ornamental plants in the campus area, were used in the study. Plants whose seeds are taken have orange flower color and a layered structure. The thousand-seed weight obtained from the plant was determined as 1.76 g. It was observed that the seed colors were black and the seed width was 0.27 mm and the seed length was 18.36 mm.

Five different treatments doses obtained from hydropriming, which is a pre-sowing seed treatments, extract obtained from Tagetes erecta petals, which is an agent used in organic priming treatments, and Moringa leaf extracts, were used on marigold seeds.

Hydropriming treatments: The seeds were planted after keeping them at 25°C for 24 hours between filter papers moistened with 10 ml distilled water in 9 cm petri dishes (Gündüz et al., 2019).

*Tagetes erecta* treatments: The extracted Tagetes erecta petals were brewed in pure water at the rate of 4 g per 1 Liter of distilled water for 3-4 minutes and this brewing tea was used in seed treatments. After the prepared extract cooled, the seeds were moistened with 10 ml of extract between filter papers in 9 cm petri dishes and kept at 25°C for 24 hours (Mavi, 2016).

Moringa treatments: Moringa leaf extracts prepared as brewing tea as in tagates at 5 different treatments doses  $(1-2-4-8-16 \text{ g L}^{-1})$  are added to the seeds in each petri dish, 10 ml, and inoculated 24 hours between filter papers in 9 cm petri dishes. The treatments were carried out by keeping it at 25°C. Treatments and abbreviations for marigold seeds before planting are given in Table 1.

After priming treatments, all treatments and control group seeds were planted in 3×25 repetitions×seeds in peat: perlite (3:1) growing medium, each repetition in separate vials (195×103×63 mm). To examine the effect of planting depth on emergence for each treatment and control group, the vials were covered with a peat:perlite (3:1) mixture at two different planting depths, 0.2 cm and 1.0 cm.

During the seedling emergence test, seedling emergence and actual leaf emergence were counted for 30 days and the vials were kept in the greenhouse environment. During this period, the minimum temperature was measured as 7°C and the minimum temperature average was 10°C, the maximum temperature was 37°C and the maximum temperature average was 24°C. The total average temperature value was determined as 17°C. The rate of surviving seedlings was obtained by counting the seedlings that remained alive for 30 days and increasing the percentage. At the end of the census, mean emergence rate (%) and mean emergence calculated based time were on 3×25 repetitions × plants. Mean emergence time and true leaf appearance were calculated according to the values obtained from daily counts made during the

Table 1. Treatments and abbreviations for marigold seeds before planting.

Treatments	Abbreviations
Control	К
Hydropriming	HP
Tagetes erecta priming	TAG
Moringa leaf extract (1 g L <sup>-1</sup> )	M1
Moringa leaf extract (2 g L <sup>-1</sup> )	M2
Moringa leaf extract (4 g L <sup>-1</sup> )	M3
Moringa leaf extract (8 g L <sup>-1</sup> )	M4
Moringa leaf extract (16 g L <sup>-1</sup> )	M5

seedling emergence trial (Orchard, 1977). At the emergence stage, emergence speed index and coefficient of velocity of emergence were also determined (Kader, 2005). Vigor index value was calculated as the emergence speed index × coefficient of velocity of emergence. In the experiment, percentage values were subjected to angle transformation before statistical analysis, and real values were used in figures and tables. Statistical analysis of all data was analysed using the Duncan multiple comparison test in the SPSS 17.0 package program in order to reveal the differences between the treatments. Differences were determined at the p<0.05 significance level. In addition, in order to observe the difference created by different planting depths on emergence characteristics, T test was performed on the averages of all treatments in the same package program and data with a significance value less than 0.05 were taken into account.

#### 3. Results and Discussion

After the emergence test performed at both planting depths, the highest emergence rate was obtained from HP treatments at both planting depths. Among Moringa and TAG treatments, it was determined that M3 treatments gave the highest results for both planting depths. While the emergence rate of control group seeds was determined to be 11% at 1 cm planting depth, this rate was 21% at 0.2 cm planting depth, and it is seen that the emergence rate increased by 10% by changing the planting depth without any treatments. At 0.2 cm planting depth, emergence rates were determined as 39% after HP treatments and 31% after M3 treatments. When the results are compared, the treatments increased the emergence rates varying between 2-18% at 0.2 cm planting depth. At 1 cm planting depth, the emergence rate increased by 6-22% (Figure 1).



Figure 1. Effects of treatments and different planting depths on mean emergence rate (%) and surviving seedling rate (%) in marigold (K: Control, HP: Hydropriming, TAG: *Tagetes erecta* priming, M1: Moringa leaf extract (1 g L<sup>-1</sup>), M2: Moringa leaf extract (2 g L<sup>-1</sup>), M3: Moringa leaf extract (4 g L<sup>-1</sup>), M4: Moringa leaf extract (8 g L<sup>-1</sup>), M5: Moringa leaf extract (16 g L<sup>-1</sup>).

Conway et al. (2003) stated that while the initial viability of the genotype coded E-1236 in African marigold was 89%, the viability increased to 96% after osmotic priming and solid matrix priming treatments. Ilbi et al. (2020) in their study on T. erecta and T. patula species, after the standard germination test of 20°C dark / 30°C light (16 hours / 8 hours), the average germination rate varied between 78-90%, while the highest emergence rate (90%) was obtained by keeping T. patula seeds at 5°C for 2 weeks. In their study on 3 different species, Kumar and Sharma (2012) examined the effects of different pre-planting practices and different planting environments and stated that the germination rate values for Tagetes species varied between 14.4-42.3%. It was observed that the highest germination rate value was obtained from the treatments of keeping it in the dark at 20°C for 2 days. While some of the literature results had higher outputs, they were similar to the results of Kumar and Sharma (2012). In this study, a low stress temperature of approximately 17°C was used. It is aimed to guarantee emergence with seed treatments, especially in early plantings in the spring period. In addition, it is seen that the effectiveness rate of the treatments is better understood when working with seed groups with low vitality values. As a result of the calculations of the survival seedling rate after the emergence test, it was determined that the effectiveness of the planting depth on the living seedling rate created statistically significant differences, while the treatments were not statistically significant in the living seedling rate (Figure 1).

After the emergence test on marigold, it was observed that while the average planting depths caused differences in the mean emergence time, they did not create significant differences in the true leaf appearance. At a planting depth of 0.2 cm, the earliest emergence time value was in the M5 treatments with 4.47 days in the earliest group, while the longest emergence time was in the control group with 8.37 days. At 1 cm planting depth, the earliest emergence was obtained from the M4 treatments with 4.4 days, while the groups with the longest emergence time were the control group with 6.34 days and the M5 treatments with 6.48 days (Figure 2). Afzal et al., (2009) stated that



Figure 2. Effect of different pre-sowing seed treatments and planting depths on the mean emergence time and true leaf appearance in marigold. (K: Control, HP: Hydropriming, TAG: *Tagetes erecta* priming, M1: Moringa leaf extract (1 g L<sup>-1</sup>), M2: Moringa leaf extract (2 g L<sup>-1</sup>), M3: Moringa leaf extract (4 g L<sup>-1</sup>), M4: Moringa leaf extract (8 g L<sup>-1</sup>), M5: Moringa leaf extract (16 g L<sup>-1</sup>).

Table 2. Effect of treatments and different planting depths on emergence speed index, coefficient of velocity of emergence and vigor index in marigold.

Treatments	0.2 cm		1 cm			
Treatments	ESI	CVE	Vigor index	ESI	CVE	Vigor index
К	0.74±0.4 d	12.78±0.7 b	285.76±142.0 c	0.44±0.1 c	15.78±1.0 b	168.83±25.6 c
HP	1.97±0.1 a	19.69±1.1 a	762.07±54.9 a	1.77±0.3 a	19.37±2.1 ab	656.40±124.0 a
M1	0.91±0.1 cd	18.24±2.2 a	372.92±74.9 bc	1.30±0.1 ac	22.10±1.5 a	504.76±60.4 ac
M2	1.04±0.1 bd	18.35±1.2 a	418.13±47.1 bc	1.29±0.4 ac	21.65±1.8 a	506.29±143.9 ac
M3	1.67±0.3 ac	19.97±2.2 a	627.49±134.1 ab	1.63±0.2 ab	20.40±0.2 ab	624.37±66.4 ab
M4	1.31±0.3 ad	21.17±0.8 a	498.17±105.7 ac	1.01±0.3 ac	22.75±0.5 a	395.75±110.2 ac
M5	1.73±0.3 ab	22.58±1.3 a	648.80±97.8 ab	0.74±0.3 bc	16.11±2.3 b	275.52±123.6 bc
TAG	1.48±0.0 ad	18.21±0.7 a	556.91±12.4 ac	0.86±0.4 ac	18.81±0.4 ab	331.59±148.2 ac

(ESI: Emergence speed index, CVE: Coefficient of velocity of emergence, K: Control, HP: Hydropriming, TAG: Tagetes erecta priming, M1: Moringa leaf extract (1 g L<sup>-1</sup>), M2: Moringa leaf extract (2 g L<sup>-1</sup>), M3: Moringa leaf extract (4 g L<sup>-1</sup>), M4: Moringa leaf extract (8 g L<sup>-1</sup>), M5: Moringa leaf extract (16 g L<sup>-1</sup>).

Table 3. Determination of the effectiveness of different planting depths on emergence characteristics in marigold by T test.

Depth	Emergence rate (%)	Mean emergence time (day)	True leaf appearance (day)	Surviving seedling rate (%)	ESI	CVE	Vigor index
1	31	5.55	28.06	97	1.36	18.87	521.3
2	27	5.19	28.51	91	1.13	19.62	432.9
Sig	0.157	0.234	0.982	0.03	0.359	0.771	0.244

\*In the depth column, number 1 indicates 0.2 cm planting depth, number 2 indicates 1 cm planting depth. Data with a significance value (sig) less than 0.05 are statistically significant in terms of depth. ESI: Emergence speed index, CVE: Coefficient of velocity of emergence.

halopriming treatments increased the seedling emergence rate, reduced emergence time and increased seedling quality. In another study, it was determined that germination time varied between 1.75-2.84 days after pre-sowing seed treatments (Ilbi et al., 2020). After the seedling emergence test, the emergence speed index, coefficient of velocity of emergence and vigor index values were calculated to determine the emergence power of the seed groups. When the changes in treatments and planting depths were examined, HP and M3 treatments were the treatments groups that gave the best results in terms of all values, while the results of 0.2 cm planting depth were seen to give better results compared to 1 cm planting depth (Table 2). HP treatments gave the highest results, with the emergence speed index value being 1.97 at 0.2 cm planting depth and 1.77 at 1 cm planting depth. The highest coefficient of velocity of emergence value was measured from the M5 treatments with a value of 22.58 at 0.2 cm planting depth, while this value was obtained from the M4 treatments with 22.75 for 1 cm planting depth. When the Vigor index values are examined, HP and M3 treatments are seen as the prominent treatments groups (Table 2.) It is known that different planting depth treatments cause changes in emergence rate in the Asteraceae family. A T Test was also conducted to observe the difference of different planting depths on the emergence characteristics of marigold, a member of the Asteraceae family (Table 3).

In the T Test performed by taking the mean of 24 replicates of 8 treatments, it was observed that while the mean emergence rate of 0.2 cm surface planting was 31%, this rate decreased to 27% in 1 cm deep planting. In addition, it has been found important to have a high rate of survival as well as a high survival rate in ornamental plant cultivation. While the rate of surviving seedlings at 0.2 cm planting depth was 97%, the rate of surviving seedlings at 1 cm planting depth decreased to 91%. As a result of the T test, planting depth was found to be statistically significant (p<0.05). Among all the features, the prominent feature was the survival rate of seedlings with a significance level of 0.03.

#### 4. Conclusion

When all the results were evaluated, it was seen that there may be changes in the effectiveness levels of the treatments as the variation between repetitions increased with the increase in planting depth. Decreasing the planting depth is important to increase the effectiveness of the treatments and the emergence rate. In addition, starting to work with a seed batch with low viability proves the effectiveness of the planting depth and treatments. It is thought that the fluctuations seen in the test results may be due to the low test temperature, maturity differences between seeds collected from nature and high seed variation. When all the results are evaluated, it is seen that M3 treatments stands out in terms of moringa leaf extract and can be used as an effective dose in future studies. Although the emergence rate of control group seeds is low in early stage cultivation at low temperatures, it is seen that this rate can be increased by 22% with treatments. Thus, it is thought that early seedlings obtained at low temperatures make a significant difference for the ornamental plant breeding sector.

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