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# THE EFFECTS OF LOCATION AND THE APPLICATION OF DIFFERENT MINERAL FERTILIZERS ON SEED YIELD AND QUALITY OF POT MARIGOLD (*Calendula officinalis* L.)

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**ABSTRACT**

**The four-replicate trial with a local variety Domaći oranž (*Local orange*) was set up according to the randomised complete-block design in four locations (Gorobilje, Arilje, Pančevo and Kačarevo) and two variants of fertilizing (200 kg ha$^{-1}$ KAN with 27 % of nitrogen and 400 kg ha$^{-1}$ NPK 15:15:15) and the control without fertilizing. According to the three factorial analysis of variance for all observed traits it was determined that there were very significant differences within growing locations and fertilizing variants and their interaction (L x F). The highest seed yield (672.84 kg ha$^{-1}$) was detected in the variant with 400 kg NPK ha$^{-1}$. The significantly lower seed yield (579.84 kg ha$^{-1}$) was obtained in the variant with KAN at the rate of 200 kg ha$^{-1}$ and the control variant (344.88 kg ha$^{-1}$). The highest total seed germination of 91.84% and the 1000-seed weight (6.83 g) were obtained in the variant with 400 kg NPK ha$^{-1}$. Total seed germination (85.87) and the 1000-seed weight (5.82 g) obtained in the control were significantly higher than total seed germination (83.31%) and the 1000-seed weight (5.42 g) obtained in the variant with 200 kg ha$^{-1}$ KAN.**

**Key words: pot marigold, location, mineral fertilizers, seed, germination, 1000-seed weight.**

## INTRODUCTION

Marigold (*Calendula officinalis L.*) is a decorative and medicinal plant. It belongs to the family of Asteracea, sub-family Asteroidea (*Tubuliflorae*) (Kojić and Pekić, 1995). It is used more in folk medicine than scientific medicine (Tucakov, 1996). This species contains a sufficient amount of active substances, such as: saponosoides, essential oils, flavonides, sterols, polysaccharides, sesquiterpene lactones, etc. (Kovačević, 1995). Flower heads (*Calendula flos*) and seeds (*Calendula semen*) are most often used in the pharmaceutical industry. It has anti-viral, anti HTV and anti-oxidant effects (Muley et al., 2009)

In order to provide a high-quality raw material of high and stable yields, pot marigold is increasingly plantation grown. The fertilization with N, P and K fertilizers is of the greatest significance for the balanced nutrition in the seed crop production. The application of different fertilization treatments had a considerable effect on the different vegetative growth properties of *Calendula officinalis* L. plants compared to the unfertilized control (Hussein et al., 2011). Numerous studies (Golcz et al., 2006; Dzida and Jarosz, 2006; Biesiada and Kuś, 2010) have shown that nitrogen fertilization results in a significant increase in quantity and quality of herbal plant yields. According to Rahmani et al., 2009, nitrogen had a significant effect on all plant properties of calendula (1000-seed weight, seed yield, head diameter and the number of seeds per head) achieved after application of 90 kg N ha-1. Also, the results showed that applications of N fertilizer increased seed yield of calendula, because nitrogen, which is a primary constituent of proteins, is extremely susceptible to loss when considering that average recovery rates fall in the range of 20 to 50% for dry matter production systems in plants. Nitrogen is a major nutrient that influences plant yields and protein concentrations. When the amount of available soil N limits the yield potential, additions of N fertilizers can substantially increase plant yields (Olson and Swallow, 1984; Grant et al., 1985).

The aim of the present study was to determine to which extent different types of mineral fertilizers and growing locations affect the pot marigold seed yield and quality.

## MATERIALS AND METHODS

The local variety of pot marigold Domaći oranž was observed. The trial was set up on different types of soils in the Zlatibor region (Gorobilje and Arilje) and the southern Banat region (Pančevo and Kačarevo) during 2006 and 2007. The soil in Gorobilje, i.e. Arilje, is brown earth, i.e. alluvium, respectively. Marshy black soil prevails in Pančevo, while chernozem is present in Kačarevo. Chemical properties of the studied types of soil were presented over locations (Table 1). Mean monthly air temperatures and precipitation sums for 2006 and 2007 in locations of Pančevo and Kačarevo obtained from the state meteorological station Banatski Karlovac (latitude - 45° 03', longitude - 21° 02' and altitude - 89 m AMSL) are presented in Figures 1 and 3. On the other hand, Figures 2 and 4 present meteorological data for the locations of Arilje and Gorobilje registered by the state meteorological station Požega (latitude - 43° 50', longitude - 20° 02', and altitude - 310 m AMSL).

**Table 1**. Chemical properties of the soil over studied locations

| **Location**<br>**Soil type** | **Kačarevo**<br>**Chernozem** | **Pančevo**<br>**Marshy black** | **Gorobilje**<br>**Alluvium** | **Arilje**<br>**Brown earth** |
|---|---|---|---|---|
| pH(KCl) | 7.30 | 6.56 | 6.30 | 6.89 |
| $CaCO_3$ (mg $kg^{-1}$) | 12.30 | 1.41 | 3.81 | 4.78 |
| Humus (%) | 3.14 | 2.97 | 3.00 | 2.11 |
| $P_2O_5$ (mg $kg^{-1}$) | 20.50 | 4.10 | 9.70 | 8.47 |
| $K_2O$ (mg $kg^{-1}$) | 17.00 | 37.30 | 35.50 | 29.36 |

Two variants of fertilizing and the control (C) without fertilizing were applied in all locations during both years. The N fertilizer KAN (27% N) at the rate of 200 kg $ha^{-1}$ was applied in the first variant ($F_1$) prior to sowing, while mixed NPK (15:15:15) fertilizer at the rate of 400 kg $ha^{-1}$ was applied in the second variant ($F_2$) during autumn primary tillage.

The four-replicate trials were set up according to the randomised complete-block design. The elementary plot size amounted to 20 $m^2$. Sowing was done on optimum dates during the third decade of April, while sowing rate was 8 kg seeds $ha^{-1}$.

Harvest was done in the full seed maturity stage. The harvested seed was dried down to 10 % moisture and cleaned from admixtures. Seed germination was tested in Petri dishes on filter paper in four replicates, each consisting of 100 seeds, constantly at 20 °C. The first, i.e., second count was done after seven, i.e. 14 days, respectively, according to the ISTA Rules (Handbook 2006). The seed yield was estimated in kg per hectare, and the 1000-seed weight was determined.

The test of significance of differences among estimated average values of observed factors (year, location and variants of fertilizing) was done by the application of the model of the analysis of variance for factorial trials set up according to the randomised complete-block design. All evaluations of significance were performed on the basis of the F- and LSD-test at the probability levels of 5% and 1%.

## RESULTS AND DISCUSSION

The mean monthly air temperatures, obtained from both meteorological stations, were insignificantly higher during the 2007 growing season than during the 2006 growing season (Figures 1 and 2). The mean monthly precipitation sums in April, June and August in 2006 were significantly higher than the mean for the same months in 2007, while the mean monthly precipitation sums in March, May, September and November in 2007 were higher than the mean for the same months in 2006 (Figures 3 and 4). Climate conditions during the growing season were favourable for pot marigold cultivation in both years of investigation.

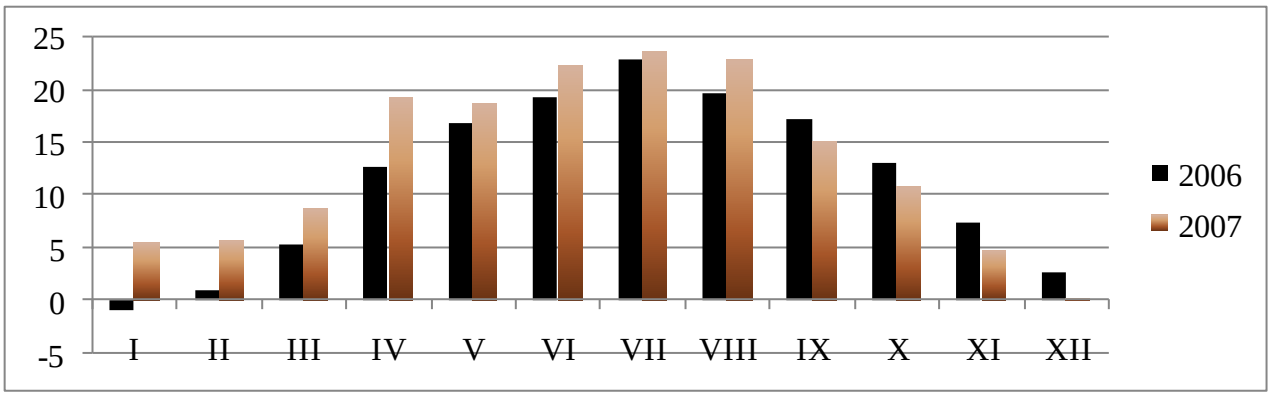


**Figure 1.** Mean monthly air temperatures (°C) recorded by the meteorological station Banatski Karlovac (Pančevo and Kačarevo)

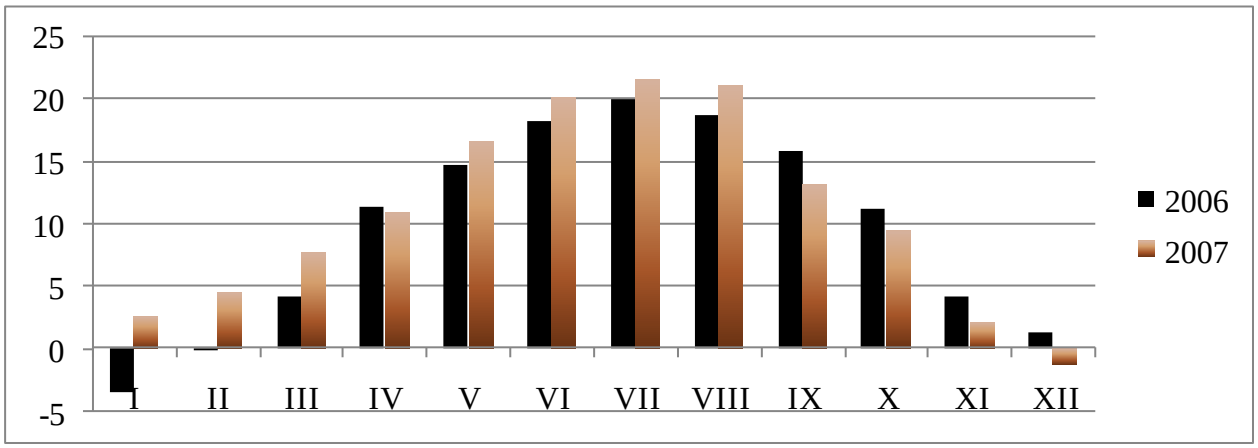


**Figure 2.** Mean monthly air temperatures (°C) recorded by the meteorological station Požega (Arilje and Gorobilje)

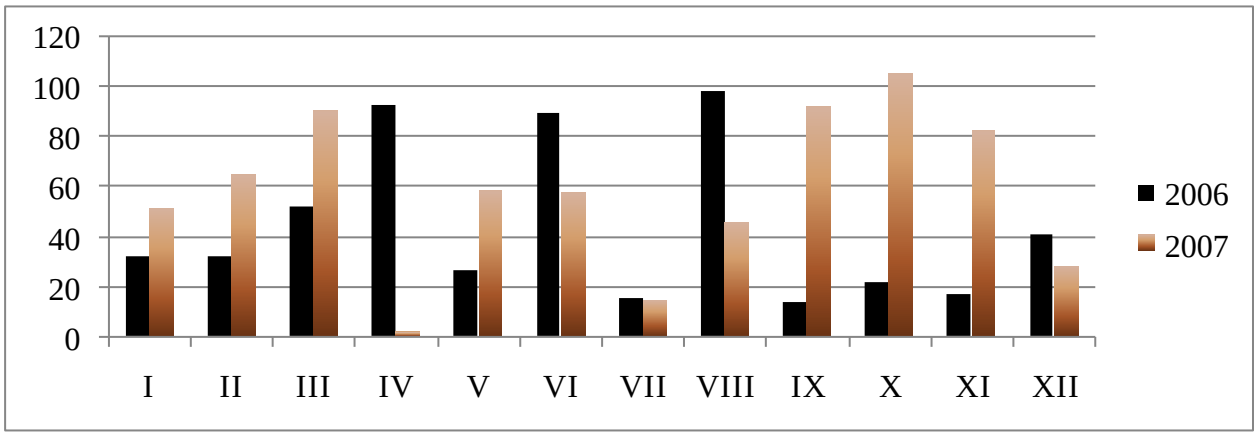


**Figure 3.** Mean monthly precipitation sums (mm) recorded by the meteorological station Banatski Karlovac (Pančevo and Kačarevo)

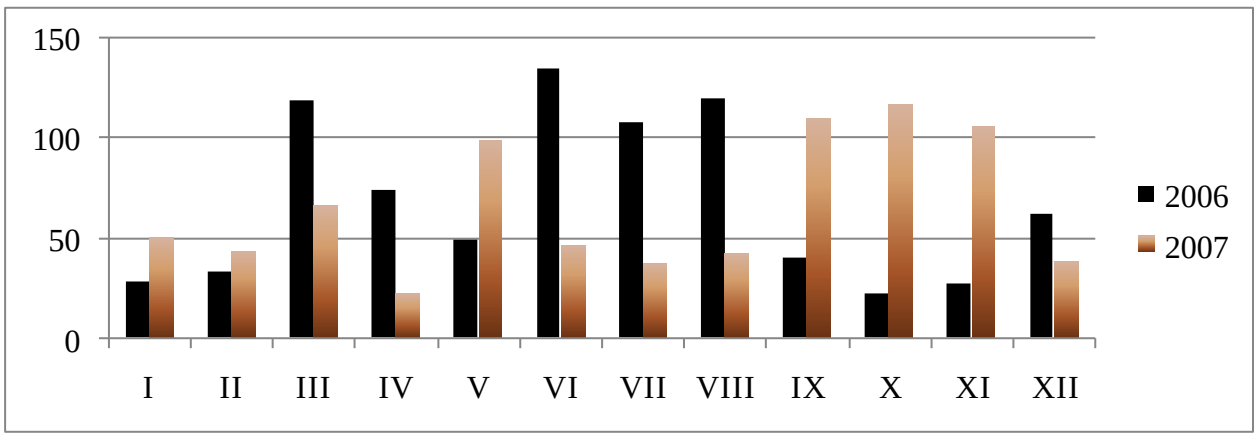


**Figure 4.** Mean monthly precipitation sums (mm) recorded by the meteorological station Požega (Pančevo and Kačarevo)

According to the three factorial analysis of variance for all observed traits, very significant differences within growing locations and variants of fertilizing and their interaction (L x F) were determined. Very significant differences in total germination between studied years were established, while there were no significant differences for other traits over years. The Y x L, Y x F and Y x L x F interactions were very significant only for the trait 1000-seed weight (Table 2).

**Table 2.** Three factorial analysis of variance for seed yield, first account, total seed germination and 1000-seed yield of pot marigold

| | | | **Seed germination** | | |
|---|---|---|---|---|---|
| | | **Seed yield** | **First count** | **Total germination** | **1000-seed weight** |
| Source of variation | d.f. | | F value | | |
| Replication | 3 | 0.0217 | 1.4547 | 2.3278 | 1.2469 |
| Year (Y) | 1 | 0.3183 | 3.1243 | 16.4685** | 0.8653 |
| Location (L) | 3 | 66.0981** | 230.2583** | 343.4380** | 205.2433** |
| Y x L | 3 | 0.1407 | 0.3349 | 0.4398 | 8.6497** |
| Fertilizing (F) | 2 | 514.3688** | 69.5336** | 107.4187** | 161.6098** |
| Y x F | 2 | 0.0987 | 2.8581 | 2.6514 | 11.2552** |
| L x F | 6 | 4.0173** | 6.9606** | 5.0710** | 19.9920** |
| Y x L x F | 6 | 0.2242 | 1.6484 | 2.3886 | 6.0701** |
| Error | 69 | 1776.882 | 8.335 | 5.708 | 0.001 |
| Total | 95 | | | | |

** P≤1%

There was no significant difference in average seed yields obtained in 2006 (535.00 kg $ha^{-1}$) and in 2007 (530.10 kg $ha^{-1}$). The seed yield significantly varied over locations and types of soil. The higher seed yield was obtained under conditions without the fertilizer application on chernozem in Kačarevo and alluvium in Arilje than on marshy black soil in Pančevo and brown earth in Gorobilje. Chernozem and alluvium are lighter, looser soils richer in humus and therefore they are more favourable for the production of pot marigold and its seed. The highest, i.e. lowest seed yield, on the average, was recorded in Kačarevo (581.91 kg $ha^{-1}$), i.e. in Gorobilje (429.21 kg $ha^{-1}$), respectively. The seed yield in Gorobilje was significantly lower than seed yields in other locations (Table 3). Fertilizing significantly affected the seed yield increase, hence the highest yield (672.84 kg $ha^{-1}$) was detected in the variant with 400 kg NPK $ha^{-1}$. The significantly lower seed yield (579.84 kg $ha^{-1}$) was obtained in the variant with KAN at the rate of 200kg $ha^{-1}$. The seed yield (344.88 kg $ha^{-1}$) recorded in the control was significantly lower than seed yields obtained in both variants with the fertilizer application. The results are in accordance with results obtained by Doddagoudar et al., (2002) in China aster. The higher seed yield obtained with fertiliser NPK 15:15:15 may be attributed to a greater number of flowers, filled seeds and a greater 1000-seed weight. This was mainly due to availability of adequate quantity of nutrients for better filling up of seeds, which resulted in the increased seed weight. These findings are in agreement with the findings of Mantur (1988) and Doddagoudar et al., (2002) in China aster, Hugar (1997) in gaillardia, Sigedar et al., (1991) and Shivakumar (2000) in marigold and Akkannavar (2001) in ageratum. In contrast to the application of nitrogen fertilizer KAN, the distribution of NPK 15:15:15 contributes to the incorporation of not only nitrogen into the soil, but also of phosphorus and potassium, which are most important for plant growth and flowering. They also play a key role in the production of higher flower and seed yield of ornamentals (Kashif, 2001).

The average seed yield of the L x F interaction varied from 290.00kg $ha^{-1}$ (control in Gorobilje) to 733.37kg $ha^{-1}$ (in the variant with 400 kg NPK $ha^{-1}$ in Kačarevo). Seed yields recorded in Kačarevo, Arilje and Pančevo in the variant with 400 kg NPK $ha^{-1}$ were significantly higher than remaining yields obtained in the L x F interaction (Table 3).

**Table 3**. Average seed yield and 1000-seed weight of pot marigold over years, locations and fertilizing variants

| | | Seed yield (kg ha$^{-1}$) | | | 1000-seed weight (g) | | |
|---|---|---|---|---|---|---|---|
| **Location** | **Fertilizing** | **2006** | **2007** | **Average** | **2006** | **2007** | **Average** |
| | **C** | 280.50 | 299.50 | **290.00** | 4.37 | 4.53 | **4.45** |
| **Gorobilje** | **F1** | 471.00 | 452.25 | **461.62** | 3.93 | 4.26 | **4.09** |
| | **F2** | 539.75 | 532.25 | **536.00** | 6.11 | 6.21 | **6.16** |
| Average | | **430.42** | **428.00** | **429.21** | **4.80** | **5.00** | **4.90** |
| | **C** | 372.00 | 365.75 | **368.87** | 6.77 | 5.00 | **5.88** |
| **Arilje** | **F1** | 599.25 | 618.25 | **608.75** | 4.56 | 4.77 | **4.66** |
| | **F2** | 724.25 | 720.00 | **722.12** | 6.89 | 6.98 | **6.93** |
| Average | | **565.17** | **568.00** | **566.58** | **6.07** | **5.58** | **5.82** |
| | **C** | 342.50 | 337.25 | **339.87** | 6.01 | 6.06 | **6.03** |
| **Pančevo** | **F1** | 623.75 | 611.75 | **617.75** | 5.64 | 5.81 | **5.72** |
| | **F2** | 703.00 | 696.75 | **699.87** | 6.76 | 6.89 | **6.82** |
| Average | | **556.42** | **548.58** | **552.50** | **6.14** | **6.25** | **6.19** |
| | **C** | 384.25 | 378.00 | **381.12** | 6.83 | 7.05 | **6.94** |
| **Kačarevo** | **F1** | 643.00 | 619.50 | **631.25** | 6.71 | 7.73 | **7.22** |
| | **F2** | 736.75 | 730.00 | **733.37** | 7.42 | 7.42 | **7.42** |
| Average | | **588.00** | **575.83** | **581.91** | **6.99** | **7.40** | **7.19** |
| | **C** | **344.65** | **345.12** | **344.88** | **5.99** | **5.66** | **5.82** |
| Average | **F1** | **584.25** | **575.44** | **579.84** | **5.21** | **5.64** | **5.42** |
| | **F2** | **675.94** | **669.75** | **672.84** | **6.79** | **6.87** | **6.86** |
| Average (Y) | | **535.00** | **530.10** | | **6.00** | **6.06** | |
| | | **L** | Lsd $_{0.05}$ = 24.28 | Lsd $_{0.01}$ = 32.23 | **L** | Lsd $_{0.05}$ = 0.18 | Lsd $_{0.01}$ = 0.24 |
| | | **F** | Lsd $_{0.05}$ = 21.02 | Lsd $_{0.01}$ = 27.92 | **F** | Lsd $_{0.05}$ = 0.16 | Lsd $_{0.01}$ = 0.21 |
| | | **Y x L** | Lsd $_{0.05}$ = 42.05 | Lsd $_{0.01}$ = 55.83 | **Y x L** | Lsd $_{0.05}$ = 0.26 | Lsd $_{0.01}$ = 0.34 |
| | | | | | **L x F** | Lsd $_{0.05}$ = 0.32 | Lsd $_{0.01}$ = 0.42 |
| | | | | | **Y x Đ** | Lsd $_{0.05}$ = 0.22 | Lsd $_{0.01}$ = 0.30 |
| | | | | | **Y x L x F** | Lsd $_{0.05}$ = 0.45 | Lsd $_{0.01}$ = 0.59 |

The average 1000-seed weight (6.06 g) recorded in 2007 was significantly higher than the average 1000-seed weight (6.00 g) obtained in 2006. Furthermore, the average 1000-seed weight varied from 4.90 g in Gorobilje to 7.19 g in Kačarevo. As in the case of other studied traits, the highest 1000-seed weight (6.83 g) was obtained in the variant with 400 kg NPK ha$^{-1}$. The 1000-seed weight (5.82 g) recorded in the variant without fertilizing was significantly higher than the 1000-seed weight (5.42 g) in the variant with 200 kg KAN ha$^{-1}$. The value of the Y x L interaction was the highest for the 1000-seed weight (7.40 g) obtained in the location of Kačarevo in 2007 and was significantly higher than remaining values of this trait of the Y x L interaction. The Y x F interaction showed that the 1000-seed weight obtained in the variant with 400 kg fertilizers ha$^{-1}$ in 2007 (6.87 g) did not significantly differ from the 1000-seed weight (6.79 g) obtained in 2006. All other values of Y x F interactions for the 1000-seed weight were significantly lower than the stated values. The value of the 1000-seed weight of the L x F interaction varied from 4.09 g in Gorobilje in the variant with 200 kg KAN ha$^{-1}$ to 7.42 g in Kačarevo in the variant with 400 kg NPK ha$^{-1}$. The Y x L x F interaction showed that the highest 1000-seed weight of 7.73 g was obtained in the second variant with 200 kg KAN ha$^{-1}$ in Kačarevo in 2007. This value did not significantly differ from the value of 7.42 g recorded in the variant with 400 kg NPK ha$^{-1}$ in Kačarevo in 2006 and 2007. The value of the Y x L x F interaction was significant only for the 1000-seed weight in comparison to values of remaining traits (Table 2).

The location, fertilizing variants and their interactions affected the first count (Table 2). The average value of this trait varied from 70.21% (Gorobilje) to 91.00% (Kačarevo). The first count ranged from 79.15 % (200 kg ha$^{-1}$) to 87.43% (400 kg NPK ha$^{-1}$). Similarly to the seed yield, the first count (93.50 %) was the highest in the variant with 400 kg NPK ha$^{-1}$ in the location of Kačarevo. The first count in the same fertilizing variant in the locations of Arilje and Pančevo did not significantly differ from the first count obtained in Kačarevo, while all other values of the L x F interaction were significantly lower. The average first count (90.25%) obtained in the variant with 200 kg KAN ha$^{-1}$ in Kačarevo did not significantly vary from the variant with 400 kg NPK ha$^{-1}$ (93.50%, Table 4).

Total seed germination (95.58%) determined in Kačarevo was significantly higher than total seed germination in other studied locations. The lowest total germination (74.42%) obtained in Gorobilje was significantly lower than total seed germination established in Arilje (90.58%) and Pančevo (87.45%). The highest total seed germination of 91.84% was obtained in the variant with 400 kg NPK ha$^{-1}$. Total seed germination obtained in control (85.87%) was significantly higher than

**Tablo 4.** Average values of the first count and total seed germination of pot marigold over years, locations and fertilizing variants

| First count (%) | | | | | Total germination (%) | | |
|---|---|---|---|---|---|---|---|
| **Location** | **Fertilizing** | **2006** | **2007** | **Average** | **2006** | **2007** | **Average** |
| | **C** | 68.50 | 69.75 | **69.12** | 71.50 | 74.00 | **72.75** |
| **Gorobilje** | **F1** | 67.50 | 68.75 | **68.12** | 70.50 | 72.00 | **71.25** |
| | **F2** | 73.00 | 73.75 | **73.37** | 78.25 | 80.25 | **79.25** |
| Average | | **69.67** | **70.75** | **70.21** | **73.42** | **75.42** | **74.42** |
| | **C** | 89.75 | 85.00 | **87.37** | 92.75 | 89.75 | **91.25** |
| **Arilje** | **F1** | 79.00 | 81.75 | **80.37** | 83.75 | 86.50 | **85.12** |
| | **F2** | 90.75 | 93.00 | **91.87** | 93.50 | 97.25 | **95.37** |
| Average | | **86.50** | **86.58** | **86.54** | **90.00** | **91.17** | **90.58** |
| | **C** | 80.00 | 81.25 | **80.62** | 83.25 | 86.75 | **85.00** |
| **Pančevo** | **F1** | 76.75 | 79.00 | **77.87** | 81.50 | 84.50 | **83.00** |
| | **F2** | 90.25 | 91.75 | **91.00** | 93.50 | 95.25 | **94.37** |
| Average | | **82.33** | **84.00** | **83.16** | **86.08** | **88.83** | **87.45** |
| | **C** | 89.00 | 89.50 | **89.25** | 94.75 | 94.25 | **94.50** |
| **Kačarevo** | **F1** | 87.50 | 93.00 | **90.25** | 90.75 | 97.00 | **93.87** |
| | **F2** | 94.50 | 92.50 | **93.50** | 98.25 | 98.50 | **98.37** |
| Average | | **90.33** | **91.67** | **91.00** | **94.58** | **96.58** | **95.58** |
| | **C** | **81.81** | **81.37** | **81.59** | **85.56** | **86.19** | **85.87** |
| Average (F) | **F1** | **77.68** | **80.62** | **79.15** | **81.62** | **85.00** | **83.31** |
| | **F2** | **87.12** | **87.75** | **87.43** | **90.87** | **92.81** | **91.84** |
| Average (Y) | | **82.21** | **83.25** | | **86.02** | **88.00** | |
| | | **L** | Lsd $_{0.05}$= 1.66 | Lsd $_{0.01}$ = 2.21 | **L** | Lsd $_{0.05}$ = 1.38 | Lsd $_{0.01}$ = 1.83 |
| | | **F** | Lsd $_{0.05}$= 1.44 | Lsd $_{0.01}$ = 1.91 | **F** | Lsd $_{0.05}$ = 1.19 | Lsd $_{0.01}$ = 1.58 |
| | | **Y x L** | Lsd $_{0.05}$= 2.88 | Lsd $_{0.01}$ = 3.82 | **Y x L** | Lsd $_{0.05}$ = 2.38 | Lsd $_{0.01}$ = 3.16 |

total seed germination (83.31%) recorded in the variant with 200 kg KAN ha$^{-1}$. In Kačarevo, total seed germination of 98.37% in the variant with 400 kg NPK ha$^{-1}$ was significantly higher that other obtained values of the L x F interaction (Tab 4). The higher seed quality (1000-seed weight and germination) observed in the variant with fertilizer NPK 15:15:15 might be due to a proper development of seed and also a higher number of filled seed weight per flower, which in turn might have supplied adequate food reserves during germination. This kind of improvement in seed quality attributes was also reported by Mantur (1988), Doddagoudar et al., (2004) and Gnyandev (2006) in China aster, Hugar (1997) in gaillardia, Shivakumar (2000) in marigold and Akkannavar (2001) in agaratum.

## CONCLUSION

The significant effect of the location, year, fertilizing and the L x F interaction was determined for all observed traits on the basis of the three factorial analysis of variance. Very significant differences over years of investigation were determined for total seed germination, while there was no significant difference over years for remaining traits. The Y x L, Y x F and Y x L x F interactions were very significant only for the trait 1000-seed weight.

The highest seed yield, first count, total count and the 1000-seed weight were recorded in Kačarevo, while the lowest values of all studied traits were obtained in Gorobilje.

Higher average values of all observed traits were obtained in the plots treated with mineral fertilizers than in control plots. The application of NPK fertilizer at the rate of 400 kg ha$^{-1}$ resulted in the highest average seed yield, 1000-seed weight, first count and total seed germination in the location of Kačarevo.

According to the Y x L x F interaction, it was determined that the highest 1000-seed weight was obtained in the first fertilizing variant in Kačarevo in 2007. This value did not significantly differ from the 1000-seed weight obtained in the second fertilizing variant in Kačarevo in 2006 and 2007.

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