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INVESTIGATION OF SUPER ABSORBENT POLYMERS AND ZINC SULFATE ON THE YIELD AND YIELD COMPONENTS OF *Calendula officinalis* L.

Fatemeh Nejatzadeh-Barandozi✉

Department of Horticulture, Faculty of Agriculture, Islamic Azad University, Khoy Branch, Khoy, Iran

ABSTRACT

Calendula officinalis L. is one of the most valuable medicinal plants of *Asteraceae* family and is widely used in the pharmaceutical, health, and food industries. To evaluate the effect of different treatments of super absorbent and zinc sulfate fertilizer on the yield and yield components of *Calendula officinalis* L., this experiment was conducted at the Islamic Azad University of Khoy. This was a factorial experiment using as the first factor super absorbent polymers at four levels to the soil (0, 4, 6, 8 g/kg) and as the second factor, 4 zinc sulfate concentrations of 0, 25, 50, and 70 mg per pot (based on a randomized complete block design with three replications). The results showed that super absorbent polymers had significant effect on all yield traits, but Zn sulfate fertilizer had only a significant effect on flower diameter, number of stems, total biomass with seeds, total biomass yield, seed yield, and harvest index. The interaction of the super absorbent polymers and zinc sulfate fertilizer increased the plant height, total biomass with seeds and flowers, seed weight, and flower and seed harvest index, whereas the diameter of flowers, number of stems, dry flower yield, flower number per m^2, and seed yield were not affected. Super absorbent polymers with zinc sulfate fertilizer had the greatest effect on most yield traits and had a positive effect on increasing the yield components of *Calendula officinalis* L., which improved the grain yield. The use of super absorbent polymers can improve the water holding capacity of the soil, which reduces the need for water and improves the yield in arid and semi-arid areas.

Key words: pot marigold, harvest index, components, biomass

INTRODUCTION

Calendula officinalis L. was cultivated as an ornamental plant until its medicinal properties were discovered. The cultivation of this plant began in Europe in the 12th century in the monasteries' gardens for their healing effect [Prance and Nesbitt 2005]. The plant is cultivated in Germany, Australia, Czech Republic, Slovakia, Switzerland, Hungary, and most recently in Egypt and Syria [Omidbeigi 2015]. It is a perennial of the *Asteraceae* family and primarily grows in the Mediterranean, the Middle East, and Central Europe [Smsam Shariat and Moatar 2014]. The purpose of the cultivation of this plant is to produce drugs from active ingredients found in its flowers, especially in petals [Koohestani et al. 2009]. The flowers of this plant, in addition to being used in the food industry (for flavoring and coloring), also contain ingredients used in paints (including nylon paints) and pharmaceuticals (in the preparation of different types of creams and lotions). Its seeds have an oil content of 15–20% and a calendic acid content

✉ fnejatzadeh@yahoo.com

of 45–60%. Antioxidant, antimicrobial, antifungal, and other medicinal properties of the plant have been reported [Noori Pour et al. 2010]. In addition, organic extract of *C. officinalis* has antiviral activity against AIDS (HIV) [Barvenik 1994].

Calendula officinalis contains bitter compounds, organic acids, and resins. It also contains a 3% concentration of a substance called callandoline, gum, glaze, 0.02% albumin, essential oil, saponin, and an unknown yellow-colored substance [Noori Pour et al. 2010, Omidbeigi 2015]. The saponin in this plant produces an active substance that decomposes due to hydrolysis, and substances such as oleanolic acid and a bitter substance called calendin can also be obtained from it [Noori Pour et al. 2010]. New studies have shown that dry petals of this plant contain stigmasterol, cistosterol and cholesterol, as well as various acids such as lauric acid, palmitic acid, stearic acid, acid penta desilic, myristic acid, faradiol, and arnidiol [Noori Pour et al. 2010]. Its pigments contain beta-carotene, lycopene, rubixanthine, violaxanthine, and phytosterol [Noori Pour et al. 2010].

The active ingredients of this plant are made and stored in its flowers, and the most important of them are water soluble flavonoids (0.04–0.1%). Other materials include (3%) carotenoids soluble in water and fat, soluble mucilage, and vitamin E [Omidbeigi 2015]. *Calendula officinalis* extract is a complex combination of volatile oils (essential oils), glycosides, triiodenes, and xanthophylls with terpenoves [Noori Pour et al. 2010]. The newly opened blooms contain about 0.0004% of the essential oil, which includes the azulenogenic sesquiterpenes [Noori Pour et al. 2010]. Recent research suggests that seeds of *C. officinalis* contain oil. If this oil is extracted under the influence of cold pressure, it will have anti-inflation effects [Omidbeigi 2015]. Recently, in Europe, much attention has been paid to this plant as an oil producer. The industrial importance of the product has expanded after discovery that its seeds have 5–20% oil, with more than 60% of this oil being composed of calendic acid [Noori Pour et al. 2010, Omidbeigi 2015].

Low water use efficiency is a major problem in agriculture, and the most important solution for increasing the efficiency of water usage requires hydrogels enriched with the plant's nutrients. Super absorbent hydrogels or hydrophilic polymer gels are hydrogels that can absorb extra amounts of water [Elgendy et al. 2001, El-Hady and Wanas 2006]. The super absorbent particles are swollen until they reach their equilibrium volume. Super absorbent properties include high water absorption capacity, high absorption rate, and gel strength. The correction of a plant root environment by polymers also results in an increased water retention in plant growth medium, improved soil texture, increased irrigation interval requirements, increased water penetration, reduced erosion and runoff, and faster germination and growth of plants [Barvenik 1994, Abedi-Koupai et al. 2008].

Yazdani and Kerman [2007] investigated the effect of four amounts of super absorbent polymer (0, 70, 150, and 225 kg/ha) and three irrigation intervals (6, 8, and 10 days) on soybean growth and yield of the L11 cultivar. The results of this research showed that application of 225 kg of super absorbent polymer per hectare showed the best effect on soybean growth and performance under all irrigation conditions (normal irrigation and drought stress conditions).

In Egypt, Elhadi et al. [2006] mixed two anionic and cationic hydrogels of acrylamide in a 2 : 3 ratios and used it at three levels (2, 3, and 4 g) for each pot. Then they cultivated cucumber, with four irrigation regimes (50, 70, 85, and 100%). Their purpose was to investigate the effect of hydrogels on water and fertilizer use efficiency. They observed that with decreasing irrigation from 100% to 85%, despite the presence of hydrogels, cucumber production increased.

On the other hand, zinc deficiency is a common nutritional problem in calcareous and saline soils, which reduces the yields. This element plays an important role in activating the antioxidant enzymes and using carbon in the biosynthesis of the active ingredients of plants and can thus affect their antioxidant properties [Misra et al. 2006, Allah Dadi and Mozazzan Qamsari 2015]. Therefore, the aim of this study was to investigate the effect of super absorbent polymer and zinc sulfate on the yield and components of *C. officinalis*. In order to identify the zinc sulfate fertilizers, it is appropriate to move towards the research and use of these fertilizers and ultimately the stability of the system. The crop was stepped up to save water consumption efficiency while consuming water.

MATERIALS AND METHODS

In order to evaluate the effect of different treatments of super absorbent and zinc sulfate fertilizer on the yield and yield components of *Calendula officinalis* L., this experiment was conducted at the Islamic Azad University of Khoy located at 46°6' E and latitude 38°2' N and elevation 1350 m. The average annual rainfall and precipitation area is 12.45°C and 310 mm, respectively. A factorial experiment was conducted using as the first factor 200 A super absorbent at four levels of soil (0, 4, 6, and 8 g/kg) and as the second factor, 4 zinc sulfate concentrations of 0, 25, 50, and 70 mg per pot (based on a randomized complete block design with three replications). Before carrying out the test, a sample of soil was selected and transferred to the soil laboratory, the results of which are given in Table 1. Also, the amount of application of hydrogel was 0, 4, 6 and 8 g per pot. In this experiment, black plastic pots with an elevation of 18 cm were used. In order to prepare the medium, 3 kg of soil were mixed with the amount of superabsorbent and chemical fertilizer of zinc sulfate. Super absorbent polymer type A 200 was used as a product of Iran Polymer and Petrochemical Research Institute. Some of the characteristics of this polymer are given in Table 2. Weed practices were also carried out during the growing season, during which no pest or disease was observed. Due to the unlimited growth of this plant, flowers and seeds were harvested. Beginning and the formation of flowers was in mid-June, and harvesting continued until mid-October, and the beginning of seed formation was in early July and continued until the end of October. The measured traits were plant height, number of branches, number of flowers per m^2, flower diameter, seed weight, dry flower yield, seed yield, total biomass with flower, total biomass with seeds, flower harvest index and seed harvest index. Drying of flowers and other parts of the plant was done in the shade at a maximum of 40°C. Seed and flower harvest index was determined by accounting the total biomass yield and seed and flower yield.

Table 1. Result of physicochemical analysis of the soil

Texture	pH	EC (ds/m)	Available P (mg/kg)	Available K (mg/kg)	Total N (%)	Organic carbon (%)	Percent saturated	Lime
Lomi	7.35	2.92	6.32	215	0.08	0.85	41.95	18.02

Table 2. Characteristics of super absorbent polymer A200

Moisture content (%)	Density (g/cm^3)	pH	Particle size (mμ)	Maximum lifespan (years)	Water absorption capacity (gr/gr)		
					distilled water	water	NaCl solution 0.9%
5–7	1.4–1.5	6–7	50–150	7	220	190	45

Statistical analysis of data

At the end, statistical analysis was performed using SPSS software and for comparing the means, Duncan's multi-domain test was used at a probability level of 5%.

RESULTS AND DISCUSSION

Yield. The results of analysis of variance dry stem yield (Tab. 3) showed significant difference in super absorbent factor, while the factor of zinc sulfate fertilizer and the interaction of two factors were not significant. Results of the comparison of the mean of treatments (Tab. 4) also showed that the highest dry stem yield was related to the 4 g super absorbent treatment and the lowest dry matter yield was related to the control treatment. Treatments of zinc sulfate fertilizer did not differ significantly and all were placed in statistical group a. Correlation analysis (Tab. 5) showed a positive and significant correlation between this trait and other traits. Dry flower yield had the highest correlation (0.88%) with seed yield. Also, the results of analysis of variance for 1000 grain weight (Tab. 5) showed a significant difference in super absorbent factor, but zinc sulfate factor did not make a significant difference in 1000-weight weight. The interaction of two factors also had a significant effect on this trait. The results of the comparison of means (Tab. 6) did not show any significant difference in consumption of super absorbent material. Also, zinc sulfate fertilizer was not significantly different. The results from variance analysis of a number of flowers per m^2 showed that super absorbent factor had significant effect on this trait, while zinc sulfate fertilizer and the interaction of two factors did not have a significant effect on this trait (Tab. 5). Results of the mean comparison also indicated that the highest number of flowers was related to 6 g super absorbent and 25 mg zinc sulfate fertilizer, respectively. The results of variance analysis for seed yield showed that there was a significant difference between two factors of super absorbent and zinc sulfate fertilizer in Table 5, but the interaction between two factors was not significant. The results of the comparison of mean in Table 4 did not show significant differences in the amount of super absorbent consumed. Also, no significant difference was observed between levels of zinc sulfate fertilizer. This trait showed a positive and high correlation with other traits, thus the highest correlation was observed with dry flower yield (0.88%) and flower number (0.88%) (Tab. 7).

Table 3. Analysis of variance of super absorbent polymer treatment and zinc sulfate fertilizer on the traits studied in (*Calendula officinalis* L.)

Source of variations	df	Means of squares					
		plant height	flower diameter	number of stems	dried flowers yield	total biomass yield with seed	total biomass yield with flower
Replication	3	1.72 ns	0.15 ns	0.70 ns	210.01 ns	450.47 ns	4812.36*
Super absorbent polymer (A)	3	145.55**	2.02**	4.42**	2407.51**	23165.55**	8061.72*
Zinc sulfate (B)	3	10.12 ns	0.77**	7.71*	2.23 ns	2598.66*	10048.12*
(A × B)	9	21.75*	0.01 ns	0.03 ns	204.24 ns	2458.30*	5156.75*
Error	14	30.00	0.41	1.71	541.97	2357.21	6008.50
CV (%)	–	12.80	13.30	11.64	21.35	8.55	17.41

ns, * and **: non-significant and significant at the 5 and 1% levels of probability, respectively

Table 4. Comparison of the mean of super absorbent polymer treatment and zinc sulfate fertilizer with Duncan's multiple domain test at 0.05%

Treatment	Level of treatment	Plant height	Flower diameter	Number of stems	Dried flowers yield	Total bio-mass yield with seed	Total bio-mass yield with flower
Super absorbent polymer (g/kg of soil)	0	35.86 b	4.01 b	10.12 b	80.18 b	489.2 c	392.35 a
	4	46.90 a	4.91 a	12.01 a	124.24 a	636.75 a	463.92 a
	6	45.65 a	4.88 a	11.86 a	120.76 a	588.30 ab	473.92 a
	8	42.45 ab	5.40 a	11.02 ab	110.42 a	553.42 b	447.33 a
Zinc sulfate (mg per pot)	0	43.35 a	4.18 c	10.02 c	109.21 a	550.41 d	423.78 b
	25	43.37 a	4.98 a	11.82 a	108.60 a	577.47 a	465.10 a
	50	42.06 a	4.60 a	10.69 a	108.51 a	556.27 a	424.18 a
	70	42.20 a	4.30 b	10.21 b	108.10 a	554.42 b	423.80 b

In each column, the same letters indicate that there is no significant difference between the meanings

Table 5. Analysis of variance of super absorbent polymer treatment and zinc sulfate fertilizer on the traits studied in (*Calendula officinalis* L.)

Source of variations	df	Means of squares				
		weight of one thousand seeds	number of flowers per m^2	seed yield	flower harvest index	seed harvest index
Replication	3	14.35*	161336.205 ns	128.31 ns	11.46*	5.73 ns
Super absorbent polymer (A)	3	20.96**	3290221.829**	13820.50**	41.55**	132.61*
Zinc sulfate (B)	3	3.02 ns	241802.371 ns	2454.20*	38.15*	18.40 ns
(A × B)	9	9.90*	216298.598 ns	2516.87 ns	45.93**	99.97*
Error	14	8.43	514576.83	4551.08	6.81	60.22
CV (%)	–	16.35	13.10	23.10	10.56	15.19

ns, * and **: non-significant and significant at the 5 and 1% levels of probability, respectively

Table 6. Comparison of super absorbent polymer treatment and zinc sulfate fertilizer using Duncan's multiple domain test at 0.05%

Treatment	Level of treatment	Weight of one thousand seeds	Number of flowers per m^2	Seed yield	Flower harvest index	Seed harvest index
Super absorbent polymer (g/kg of soil)	0	15.12 b	43.73 ab	221.12 b	20.64 b	44.81 a
	4	19.50 a	45.241 b	323.20 a	26.31 a	54.67 a
	6	18.47 ab	46.627 a	320.27 a	26.06 a	54.62 a
	8	17.95 ab	46.460 a	302.70 ab	25.03 a	49.84 a
Zinc sulfate (mg per pot)	0	17.25 a	5051.8 b	281.71 a	23.22 b	50.41 a
	25	18.12 a	5553.3 a	301.39 a	25.77 a	51.86 a
	50	17.41 a	5353.4 a	281.71 a	25.13 a	50.11 a
	70	17.28 a	5352.3 a	281.59 a	23.25 b	50.08 a

In each column, the same letters indicate that there is no significant difference between the meanings

Table 7. Simple correlation coefficients of the studied traits in (*Calendula officinalis* L.)

Traits examined	Plant height	Flower diameter	Number of stems	Dried flowers yield	Total biomass yield with seed	Total biomass yield with flower	Weight of one thousand seeds	Number of flowers per m^2	Seed yield	Flower harvest index	Seed harvest index
Plant height	1										
Flower diameter	0.88**	1									
Number of stems	0.65*	0.88**	1								
Dried flowers yield	0.77**	0.83**	0.72**	1							
Total biomass yield with seed	0.48*	0.66*	0.80**	0.72**	1						
Total biomass yield with flower	0.52*	0.76**	0.82**	0.53*	0.80**	1					
Weight of one thousand seeds	0.71**	0.63*	0.61*	0.70**	0.59*	0.42*	1				
Number of flowers per m^2	0.74**	ns 0.18	0.84**	0.86**	0.82**	0.82**	0.52*	1			
Seed yield	0.71**	0.84**	0.80**	0.88**	0.78**	0.62*	0.53*	0.88*	1		
Flower harvest index	0.47*	0.37*	0.18 ns	0.74**	0.23 ns	0.21 ns	0.52*	0.36*	0.53*	1	
Seed harvest index	0.67*	0.68*	0.46*	0.68*	0.27 ns	0.21 ns	0.29 ns	0.59*	0.80**	0.59*	1

ns, * and **: non-significant and significant at the 5 and 1% levels of probability, respectively

Harvest index. The results of analysis of variance for harvest index (flower) showed a significant difference between the two factors super absorbent and zinc sulfate fertilizer (Tab. 5). Comparisons of the mean for super absorbent (Tab. 6) showed that the amount of super absorbent does not have a significant effect on the flower harvest index, but there is a significant difference between levels of zinc sulfate factor. In general, the highest flowering index according to Table 6 was related to 4 g of super absorbent treatments and 25 mg of zinc sulfate, respectively. Also, the results of variance analysis showed that super absorbent had a significant effect on seed yield index, while zinc sulfate did not have any significant effect. Interaction of these two factors was also significant. Comparison of mean treatments in Table 6 showed that the highest seed yield index was related to 4 g of super absorbent and 25 mg zinc sulfate, which had a significant difference with control, but did not show significant difference with other treatments.

The results showed that super absorbent had a significant effect on all traits, but zinc sulfate fertilizer had only a significant effect on flower diameter, number of stems, total biomass with seeds, total biomass yield, seed yield and harvest index. The interaction of super absorbent and zinc sulfate fertilizer increased the plant height, total biomass with seeds and flowers, seed weight and flower and seed harvest index, while the diameter of flowers, number of stems, dry flower yield, flower number per m^2 and seed yield were not affected. Of course, in most of the traits, super absorbent with zinc sulfate fertilizer had the highest amount and had a positive effect on increasing the yield components of (*Calendula officinalis* L.), which improved the seed yield. Generally, it can be concluded that the use of super absorbent polymer due to improved root conditioning through gravity water absorption in a relatively short time after irrigation and also preventing soil compaction creates a very suitable plant environment and the plant absorbs water and salts in this condition. Regarding the nutritional effects of this polymer, it can be stated that these compounds, by increasing air in the soil, cause better performance of some types of chemical fertilizers and also better activity of microorganisms in soil, or because of their negative charge in hydrated state, it is possible to absorb some ions positive to the soil [Abedi-Koupai et al. 2008]. Based on our results, the application of this polymer at 4 g per kg of soil provides the best conditions for *C. officinalis*. Also, when using zinc sulfate fertilizer with this polymer, it can increase the water use efficiency. Also, the amount of hydrogel consumption is very important and should not be overused. In the case of *C. officinalis*, the economic yield is the dry matter production per unit area, thus fertilizer management should be achieved in such a way to achieve maximum economic yield [Ameri et al. 2012]. Due to the fact that zinc plays an important role in activating the antioxidant enzymes and using carbon in the biosynthesis of the active ingredients of plants, it can affect their antioxidant properties and its deficiency, which is one of the common nutritional problems in salty and calcareous soils that reduce the products [Martin et al. 2005, Misra et al. 2006]. Therefore, application of zinc sulfate alone or in combination with other fertilizers can have a positive effect on the improvement of quantitative and qualitative properties and yield of medicinal plants. Considering the necessity of producing medicinal plants in agricultural systems and the need to pay attention to the cultivation of these plants in low-input systems, it seems that simultaneous use of zinc sulfate fertilizer and irrigation is an appropriate option for increasing the production of medicinal plants. Ghasemi Ghahsareh [2005] used super absorbent for *Fikusa Benjamin*. The highest mean in all measured parameters was related to irrigation intervals of 4 days, and the mean values decreased with increasing irrigation intervals. Based on the results of this research, for reducing the cost of irrigation and considering the cost of polymer, the application of 0.8% polymer and irrigation intervals of 4 days for these plants were suggested. Allah Dadi and Moazzan Ghamsari [2015] surveyed the effect of four levels of super absorbent polymer (0, 100, 200 and 300 kg/ha) and three distances (5, 7, and 9 days' irrigation intervals) on growth and yield of corn cultivar Sinagel Cross 704. The experiments showed that increasing the height and yield of corn using high levels of super absorbent polymer, did not result in a significant difference between of irrigation intervals of 3 days without using polymer and irrigation intervals of 7 days by the application of 200 kg/ha [Allah Dadi and Moazzan Qamsari 2015]. Kuhestani et al. [2009] stated that with increasing the drought stress and decreasing available water content, the effect of superabsorbent hydrogels increased the yield increase. Therefore, the researchers concluded that the effect of super absorbent polymers on lower levels of moisture is more perceptible. Thus, although the soil moisture content of clay soils in clay pores is much higher than that of soft tissue soils, it seems that due to diminishing the amount of moisture content for the plant, the use of super adsorbent in soil with heavy texture has a much greater effect than in a light soil. Clay soils, as compared to soils with lighter texture, such as loam and sandy loam, require more compression than super absorbent due to larger compression [Yazdani et al. 2007, Koohestani et al. 2009].

Super absorbent polymers affect the water penetration rate, density, structure, compactness, texture and crust hardiness of soil, aggregate anchorage, evaporation, soil infiltration and aeration, size and the number of aggregates, water tension, available water [Abedi-Koupai 2006], soil crispiness and finally cause better water management practices in soil. Abilities such as nutrient release and soil nitrification, increase the nutrient absorption, osmotic moisture of soil and decrease the transplanting stresses, cause an improvement in plant growth reaction and increase in yield and reduction in growth and production costs of plant. By absorbing hundred times of its origin weight, super absorbent can be used as a cultural medium itself or even can be used alone as a rooting medium. Furthermore, it reduces the impact pressure in turfs, usage of pesticide (i.e. herbicides, fungicides), absorbs soluble fertilizer and releases it in time and it also improves drainage when used as a soil amendment [Joao et al. 2007]. Tayel and El-Hady [1981] estimated that whereas the gel increased the total porosity, the micro pores relative to the total or the macro ones, void ratio, water holding pores, water retention, available water, and hydraulic resistively decreased the soil bulk density, quickly drained pores, hydraulic conductivity, mean pore diameter, intrinsic permeability, transmissivity and evaporation. Additionally, polymers are effective in correction of aggregation, prohibiting the capillary water soar, decreasing cumulative evaporation and improving in growth, efficiency in vast range of plant species [Sivapalan 2006]. Dorraji et al. [2010] reported that increasing the polymer levels results in reduction of soil electrical conductivity. They noticed that after 0.6% polymer application in sandy, loam and clay soil, electrical conductivity declined by 15.3, 20 and 16.9%, respectively compared to control.

Results of this study showed that the use of super adsorbent polymer can improve the water holding capacity in the soil, which reduces the need for water, which is difficult in arid and semi-arid areas with limited water resources. Also, application of super absorbent polymer with improved ventilation, porosity increase, maintenance of nutrients, increase of permeability and porosity and modulation of soil temperature created more favorable conditions for plant growth, which resulted in an increased flower and seed yield. Therefore, since the soil moisture content is usually relatively low in the planting areas, thus moisture stresses often occur and, considering the heavy texture of the soil in arid and semi-arid regions, it can therefore be applied with the application of super absorbent polymer. In addition to improving the growth characteristics and increasing yield, by expediting the onset of the plant's life, reduces the length of the planting period until harvest, and also improves the yield.

CONCLUSIONS

In general, the results of this experiment showed that the effects of zinc sulfate fertilizer on *C. officinalis* were hoped; this study has been confirmed in a few studies on these fertilizers on medicinal plants. The superiority of optimum treatments of zinc sulfate fertilizers suggests that the application of zinc sulfate fertilizers in sustainable agricultural systems, while improving the soil structure, provides optimal preparation of water and macro- and micronutrients, which leads to increased plant yields. Therefore, it can be said that super absorbent polymer with Zn fertilizer has favorable synergistic status and both provide better conditions for growth and increase of yield in medicinal plant especially in *C. officinalis.*

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