Research Article

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Application of selenium and silicon to alleviate short-term drought stress in French marigold (*Tagetes patula* L.) as a model plant species

https://doi.org/10.1515/chem-2020-0183 received June 3, 2020; accepted October 31, 2020

Abstract: Selenium (Se) and silicon (Si) are the beneficial elements that may significantly modify plants' tolerance to various types of biotic and abiotic stress. They may be particularly important due to the current climate changes. The aim of model experiments was to assess how selenium and silicon could alleviate short-term drought stress in French marigold (Tagetes patula L. "Pascal"). Tagetes plant species are economically important annual plants and are also very popular decorative flowering species in city parks due to its beautiful colored flowers and resistance on drought stress. Silicon was applied in the form of silica sol and choline-stabilized orthosilicic acid (ch-OSA). Selenium was applied in the form of sodium selenate (Na₂SeO₄). They were tested at the following concentrations (mg dm⁻³ of NS): silica sol – level I (23.25), level II (31.0); ch-OSA – level I (0.21), level II (0.63); and Se – level I (0.4), level II (0.8). The experiment showed that silicon had stimulating effect on the biometric parameters of control plants cultivated under an optimal water regime. When the plants treated with selenium were exposed to stress, the values of their biometric parameters were generally higher than in the plants treated with silicon. Both silicon and selenium significantly modified the gas exchange parameters. During the growing season, the net photosynthesis activity (P_N) , stomatal conductance (g_s) , and transpiration rate (E) tended to decrease, but they increased significantly when selenium and silicon were applied. In general, the factors significantly modified the plants' content of macro- and micronutrients as well as the proportions between them. Both selenium and silicon alleviated the short-term drought stress in French marigolds as a model plant, but when silicon was applied, the positive effect was modified by the source and its concentration.

Keywords: trace elements, French marigold, stress, drought, alleviation

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1 Introduction

The availability of water in soil is a major environmental factor that may limit the growth and the yield of crops [1]. Water stress may be caused by a real water deficit in soil (drought) or excessive salinity of the root zone. Drought causes abiotic stress, which results in lower growth and development rates, flower abortion, and lower yield during plants' maturation and reproduction [2]. Drought stress also reduces water potential, dry weight, root development, and photosynthetic parameters. Water scarcity may also result in stem stretching or increased movements of stomata [3]. Drought stress causes physiological and biochemical changes in plants, the accumulation of some compounds, e.g. sugars (oligosaccharides, sucrose, trehalose, sorbitol), sugar alcohols (mannitol), amino acids (proline), and amines (glycine, betaine, polyamides) [1]. These compounds function

osmolytes, antioxidants, and scavengers, which help to avoid and tolerate stress. Changes in these metabolites at the cellular level are related with the protective functions of the cell and the maintenance of cellular structures. Drought stress may also significantly modify plant nutrition [4] - these changes may result from adaptation to adverse environmental conditions.

Selenium is an element that may provide protection from stress factors such as heavy metals, UV radiation, low and high temperature, and drought stress. Several plant species have been examined through exposure to drought stress and the role of selenium against this stress [5]. Studies have shown that the application of selenium during drought stress increases the activity of both enzymatic and nonenzymatic antioxidants [6-8]. Although details of the selenium pathway in cells exposed to drought stress have not been fully investigated, it is known that selenium affects the cell water balance [8]. As drought stress affects the photosynthetic activity, especially PSII [9], the photosynthetic response should also be examined. Like selenium, silicon is not an essential element for plants, but it has beneficial effect at low concentrations [10,11]. It positively affects photosynthesis and nutrition when it is applied at larger amounts [12]. Like selenium, silicon provided protection from drought stress. The mitigation effect also results from proper water management in cells. According to some researchers, the fact that silicon increases the hydraulic conductivity of roots is more important than its role in the transpiration process [13]. Studies on different plant species revealed the positive effect of silicon on the photosynthetic activity [14-16]. However, the mechanism of its protective role is still unknown and requires detailed investigations. Although the beneficial role of both elements in photosynthesis has already been proved [17], there is little knowledge about their synergistic role in drought stress, especially their effect on the water cell potential. The application of elements with beneficial effect, e.g., Se or Si, may modify the chemical composition of plants. For example, drought stress may increase the content of potassium, sodium, calcium, magnesium, and iron in plants, whereas silicon may reduce the content of these elements [4]. The application of selenium may modify the uptake of K, Fe and Zn [18], and Ca [19]. Selenium is also known to reduce the uptake of metals [20,21].

The aim of this study is to compare the effect of selenium and silicon, which are abiotic stress-mitigating elements, on the reaction of French marigolds (Tagetes patula L. "Pascal") as a model exposed to a short-term drought stress. Tagetes plant species are economically important annual plants and are also very popular decorative flowering species in city parks due to its beautiful colored flowers and resistance on drought stress [22,23].

2 Material and methods

2.1 Plant material and growth conditions

Vegetation experiments were carried out between May and July in unheated greenhouse. Their purpose was to determine the effect of the application of silicon and selenium in relieving drought stress of marigold (T. patula L. 'Pascal') as a model plant. Experiment was established in a systematic design with eight replications (a replication was one single plant). The greenhouse was equipped with a modern climate control system. The following ambient conditions were maintained throughout the experiment: temperature of 20-24°C and RH of 65% to 75%.

The seeds were sown individually in rockwool pits made with fingers (17.05). The rockwool was soaked in a standard nutrient solution 48 h before experiments. After 2 weeks, plants were placed in rockwool blocks hydrated with the nutrient solution (Grodan, $100 \times 100 \times$ 65 mm), while after another 12 days, they were transferred to a special hydroponic system with recirculation of nutrient solution.

2.2 Plant nutrition

The nutrient solution for plant fertigation is composed of the following (mg dm⁻³): N-NH₄ < 10; N-NO₃, 150; P-PO₄, 50; K, 150; Ca, 150; Mg, 50; Fe, 3.00; Mn, 0.5; Zn, 0.44; Cu, 0.03; and B, 0.011. The pH was 5.50, and the EC was 1.8 mS cm⁻¹. The following fertilizers were used to prepare nutrient solutions: potassium nitrate (13% N-NO₃, 38.2% K), calcium nitrate (14.7% N-NO₃, 18.5% Ca), monopotassium phosphate (22.3% P, 28.2% K), potassium sulfate (44.8% K, 17% S), magnesium sulfate (9.9% Mg, 13% S), Librel FeDP7 (7% Fe), manganese sulfate (32.3% Mn), copper sulfate (25.6% Cu), borax (11.3% B), and sodium molybdate (39.6% Mo). Nitric acid (38%) was used to regulate the pH value. The sources of silicon were as follows: silica sol (200 g SiO₂ dm⁻³, Optysil, Intermag Olkusz) and choline-stabilized orthosilicic acid (ch-OSA; 0.6% Si; Actisil; Yara Poland). ch-OSA was

obtained from Bio Minerals N.V., Destelbergen, Belgium. The source of selenium was sodium selenate purum p.a. (Na₂O₄Se₃; Sigma-Aldrich). They were tested at the following concentrations (mg dm⁻³ of NS): silica sol – level I (23.25), level II (31.0); ch-OSA – level I (0.21), level II (0.63); and Se – level I (0.4), level II (0.8). In all combinations, nutrient solution was dosed six times daily per 5 min in each watering cycle. Drought stress was induced in selected combinations for 55 h (from July 17 to July 19) until all the plants tested had wilting symptoms of aerial parts. Then, irrigation was re-enabled for 9 days.

2.3 Measurement of gas exchange parameters

Portable photosynthesis system Ci 340 aa (CID BIOSCIE-NCE Inc., Camas, USA) was applied to measure the following gas exchange parameters: net photosynthesis activity (P_N) , stomatal conductance (g_s) , transpiration rate (E), and intercellular CO_2 concentration (C_i) . The constant conditions were maintained in the measuring chamber to obtain comparable results. So, the following parameters were sustained at the stable level: CO2 inflow concentration (400 μ mol (CO₂) mol⁻¹), photosynthetic photon flux density (PPFD) 1,000 μ mol (photon) m⁻² s⁻¹, chamber temperature 24°C, and relative humidity 50 \pm 3%. Investigations were conducted during midday light conditions between 10:00 and 14:00 h. Two gas exchange parameters investigations were performed before and after drought stress.

2.4 Biological measurements

The relative water content (%) [24] was determined for the samples collected before and after drought stress (July 2 and July 25, respectively). On the last day of the experiment (July 28), the weight of plants (individually shoots and inflorescences) was measured.

2.5 Chemical analysis

Chemical analyses were performed for the aboveground parts of plants. Samples (individually shoots and inflorescences) were dried for 48 h at 45-50°C to a stable mass and then ground. Before mineralization, the plant material was dried for 1h at 105°C. To assay the total forms of N, P, K, Ca, Mg, and Na, the plant material (1g) was digested in the concentrated (96%, analytically pure) sulfuric acid (20 cm³) with the addition of hydrogen peroxide (30%, analytically pure) [25]. For analyses of total Fe, Mn, Zn, and Cu, the plant material (2,5 g) was digested in a mixture of concentrated nitric (ultra-pure) and perchloric acids (analytically pure) at a 3:1 ratio (30 cm^3) . To assay the Si, the plant materials (0.5 g)were microwave digested in the mixture of HNO₃, HClO₄, and HF (10, 1, and 0.25 cm³, respectively). HF acid residue was neutralized by adding 3 cm3 4% H₃BO₃. After mineralization, the following determinations were performed: N-total was determined using the distillation method according to Kjeldahl in a Parnas Wagner apparatus; P was colorimetrically determined with ammonia molybdate; K, Ca, Mg, Na, Fe, Mn, Zn, and Cu were determined using flame atomic absorption spectroscopy (FAAS, on a Carl Zeiss Jena apparatus 5); and Si was determined using the atomic emission spectroscopy method with microwave nitrogen plasma (on an Agilent MP-AES 6200 apparatus). Earlier in the laboratory, the accuracy of the used methods of chemical analyses and the precision of analytical measurements of heavy metals were tested by means of the analysis of the reference material of branched flour (Pseudevernia furfuracea), certified by the IRMM (institute for reference materials and measurements) in Belgium [26].

2.6 Statistical analysis

The data were analyzed using Statistica 13.0 (StatSoft Inc., Tulsa, OK, USA). The results of chemical analyses and plant yielding measurements were processed using ANOVA and the Duncan test ($\alpha = 0.05$).

Ethical approval: The conducted research is not related to either human or animal use.

3 Results and discussion

3.1 Yield, DM (Dry Matter), and RWC (Relative Water Content)

The experiment showed that the factors under analysis significantly influenced differences in the inflorescence mass (Table 1). When silicon was applied, the mass of inflorescences increased in both the plants grown under the optimal water regime and those exposed to drought stress. The mass also increased along with the intensity of selenium nutrition. There were significant differences in the mass of green parts – it was the lowest in the control plants (I), i.e., 78 g plant⁻¹, and significantly higher in the plants treated with silicon. Selenium mitigated the effects of drought stress, whereas silicon caused the mass of shoots to increase, but the difference was not statistically significant. The total biomass of the aerial parts was the smallest in the control sample with optimal irrigation, but it was significantly greater in other combinations, even those exposed to stress. The factors under analysis significantly differentiated the plants' characteristics, both the height and width. The values of both parameters improved when silicon was applied to the plants grown under a standard water regime. The height of the plants exposed to the drought stress increased only when Se-I was used. There were similar relationships observed in the plant width. The number of inflorescences was the smallest in the control sample exposed to the drought stress. However, it did not differ significantly from the control sample grown under the standard water regime. Silicon significantly increased the number of inflorescences in the samples grown under the optimal water regime. The source of Si used for the treatment caused differences in the plants exposed to the drought stress.

Selenium also had a positive stimulating effect on the number of inflorescences. The diameter of the stems was the longest in the combinations with the largest mass of aerial parts. Silicon increased the diameter of the shoots. However, the changes were significant only for the plants

grown under optimal irrigation. The largest diameter of the shoots was observed in the Se-II combination.

The trace elements used in the experiments caused an increase in the relative water content (RWC) in the leaves of the plants grown under optimal water conditions (Table 2). Both of the tested sources (except silica sol-II) used for the silicon treatment of the plants subjected to drought stress significantly increased the RWC, compared with the control sample. The selenium treatment had no positive effect. Before the drought stress was induced, the Se-II and silicon treatments (except ch-OSA II) had significantly increased the dry matter content. When the plants were exposed to the stress, only Se-I significantly modified this parameter in relation to the control sample.

So far studies have shown that silicon affects the water management of plants and improves the efficiency of water consumption. It stimulates enzymatic and nonenzymatic antioxidative defense systems [10]. It affects the production of antioxidants and thus reduces the amount of photo-oxidative damage. It also inhibits the generation of free radicals and helps to maintain the integrity of chloroplast membranes [27]. Silicon may be involved in metabolic or physiological activities of higher plants exposed to drought stress [28]. Silicon treatment decreased the content of thiobarbituric acid reactive substances (TBARS) and the activities of acid phospholipase (AP) and lipoxygenase (LOX) in plants exposed to drought stress [29]. Silicon affects the rate of transpiration by silicating the surface of leaves [30] and reducing the lumen of stomata [31].

It was observed that the silicon treatment of sorghum plants exposed to drought stress improved their cellular

Table 1: The influence of the factors on the biometric parameters of Tagetes patula L. "Pascal"

Treatments		Fresh weight (g)			Height (cm)	Width (cm)	Number of inflorescences	Stem	
		Inflorescence	Stem + leaves	Total			(quantity plant ⁻¹)	diameter (cm)	
Standard	Control	15.0a	78.0a	93.0a	30.5a	34.7a	11.9ab	7.00a	
watering	Silica sol-II	29.0b	105.0bc	134.0bc	33.0abc	40.3cde	17.1ef	8.13b	
	ch-OSA-II	29.0b	107.0bc	136.0c	33.9c	39.4cd	18.4f	8.00b	
Water stress	Control	17.0a	94.0b	111.0b	31.2abc	35.8ab	10.4a	7.09a	
	Se-I	18.0a	115.0c	133.0bc	38.1d	43.1e	13.2bcd	7.84ab	
	Se-II	22.0b	110.0bc	132.0bc	33.6bc	42.0de	14.9cde	8.00b	
	Silica sol I	19.0a	92.0ab	111.0ab	31.5abc	34.9a	12.4abc	7.63ab	
	Silica sol II	21.0b	96.0b	117.0b	31.4abc	37.3abc	12.6abcd	7.38ab	
	ch-OSA I	20.0b	96.0b	116.0b	30.9ab	37.9abc	15.1de	7.13a	
	ch-OSA II	20.0b	103.0bc	123.0b	31.7abc	38.9bcd	13.5bcd	7.63ab	

Values within columns described with the same letter do not differ significantly at $\alpha = 0.05$.

Table 2: The influence of the factors under study on the relative water content (RWC) and dry matter content (DM) in Tagetes patula L. "Pascal" leaves

Treatments		RWC (in %)	DM (in %)		
		Before stress	After stress	Before stress	After stress	
Standard watering	Control	81.27a	73.30a	8.02a	6.95ab	
	Silica sol-II	83.53ab	83.75c	10.10b	7.24a	
	ch-OSA-II	84.01ab	88.83d	8.66a	8.47c	
Water stress	Control	81.37a	80.01b	8.02a	6.44a	
	Se-I	84.58ab	82.00b	8.55ab	8.01bc	
	Se-II	82.35a	75.26a	9.52b	6.66a	
	Silica sol I	82.91a	86.10c	9.77b	6.41a	
	Silica sol II	83.53b	80.89b	10.10b	7.32ab	
	ch-OSA I	81.84a	86.80c	12.20c	6.40a	
	ch-OSA II	84.01ab	85.17c	8.66a	7.26ab	

Values within columns described with the same letter do not differ significantly at $\alpha = 0.05$.

hydration [14]; meanwhile in studies conducted on rice (Oryza sativa L.) with varied levels of drought tolerance, it was found that under wet conditions, silicon had no effect on the growth and yielding of plants [4]. Our research showed the opposite tendency. The drought stress reduced the dry weight, root traits, and water potential. In this study, the opposite effect was observed for the DM and RWC. In the previous study, the highest relative water content (RWC) was found in the plants that received foliar silicon treatment [32]. In this study, during the growing season, the RWC also increased in the plants fed with silicon. This increase may have been caused by improved osmoregulation and reduced transpiration [33]. In the study that researched the effect of silicon on the growth and mineral nutrition of maize (Zea mays cv. DK 647 F₁) grown under varied water conditions, it was found that induced water stress reduced the total dry matter content (DM), chlorophyll content, and RWC, but it increased proline accumulation and electrolyte leakage in maize plants [34]. Our study revealed similar tendencies for the DM content at the second term, but the opposite tendencies for the RWC. Silicon treatments improved the physiological parameters, but their levels remained significantly lower than in the control sample, except for the electrolyte leakage and root-shoot ratios, which were higher [34]. In this study, there were similar tendencies observed for the RWC, but the opposite for the DM content. Water-deficient sorghum plants treated with silicon was characterized by the reduced dry matter content [14]. In consequence, the plants' cellular hydration improved despite the stress. Silicon treatment and water deficit resulted in an increased silicon concentration in potato leaves [35]. The researchers observed higher

proline concentrations under water stress and higher silicon availability in the soil, which indicates that silicon may influence osmotic adjustment.

Similar silicon, selenium exhibits antioxidative properties, and it can remove reactive oxygen species under environmental stress, especially water stress [36]. According to Emam et al. [8], plants treated with selenium produce more phenols and flavonoids. Glutathione peroxidase is an important enzyme in the plant's system of defense from oxidants. Selenium is an essential element in the structure of this enzyme. Selenium may stimulate the plant's antioxidative system and thus improve tolerance to oxidative stress caused by drought [7]. Selenium ions regulate and ensure the optimal water balance in cells. This is the main mechanism that is considered to provide protection during drought [37]. Earlier research on selenium-fed plants exposed to drought stress showed greater activity of antioxidative enzymes, i.e., catalase (CAT) and oxidoreductase (POX) [7] and an increase in the total carbohydrate content [8]. Our research showed that selenium alleviated short-term drought stress. Selenium supplementation may increase the fresh weight and the water content in broad bean cells [38]. The element regulated the plants' water management by increasing the efficiency of the uptake of water by the roots and limiting transpiration. In this study, the transpiration level in the selenium-treated plants was higher than in the control combination. Selenium applied to rapeseed leaves could improve the yield quality and the quantity in arid and semi-arid regions [39]. The selenium treatment method significantly affects the effectiveness of nutrition [40]. The authors found that spraying plants with selenium increased their tolerance to drought stress.

Our experiment showed that when selenium was applied to the plants' root zone, it effectively alleviated shortterm drought stress. Selenium supply favored the accumulation of biomass of wheat seedlings when the plants were well watered, but it did not significantly affect the biomass accumulation under drought stress although it increased the root activity and some antioxidative indicators [41]. Selenium treatment significantly increased the size of water-deficient wheat plants [42] and increase the yield of plants [43]. As this element is responsible for the activation of the antioxidative system in plant cells, its use has been extended for other stress such as drought, UV radiation, and low and high temperature. Plants treated with selenium exhibit higher tolerance to stress factors, e.g. induced by heavy metals. The use of selenium for nutritional purposes reduces oxidative damage caused by the production of compatible substances. Their compensation prevents cell dehydration. Our study showed that silicon tended to modify changes in the plants' habit, especially in those that were not exposed to stress. This effect was also observed earlier [44] on pansy plants sprayed with a silicon solution. Silicon nutrition increased the number of lateral shoots in French marigolds [45]. A similar effect was observed in our experiment. Similar effect was found earlier [46] in plants sprayed with a preparation containing silicon produced more buds and flowers or inflorescences with greater diameters than the control plants. Our study showed that selenium nutrition stimulated the plants' habit. By contrast [6], it was observed that the foliar application of selenium had no effect on the growth of wheat exposed to drought.

3.2 Gas exchange parameters

The intercellular CO₂ concentration increased in the plants supplemented with silicon and selenium (Table 3). The values of all the parameters under analysis decreased in all the combinations, which might indicate a natural physiological process in plants. However, there were differences between the combinations. The P_N of the control plants exposed to the drought stress was lower than in the control plants that were not exposed to the stress. It is particularly important that the $P_{\rm N}$ level in all the plants treated with silicon or selenium in any combination (except for the plants treated with ch-OSA, which were not exposed to the water stress) was higher than in the control plants, with or without water deficit. The P_N level in the water-deficient plants supplemented with ch-OSA II was significantly higher than in the plants treated with the same supplement but not exposed to the water stress. Measurements of the second gas exchange parameters revealed the highest level in the plants irrigated with silica sol II. However, an elevated level was also noted in the plants with the water deficit. Moreover, the P_N level also increased in the plants supplemented with selenium. Higher P_N levels usually coincided with higher g_s and Elevels. However, the E level in the plants treated with silicon (ch-OSA I and silica sol II) under the drought stress was comparable to the E level of the plants with the highest P_N . The C_i level was the highest in the waterdeficient plants supplemented with ch-OSA I. The C_i level was also elevated in the well-irrigated plants supplemented with silicon (silica sol II) (Table 3).

The silicon and selenium treatments had positive effect. This particularly applied to an elevated silicon level, which is in agreement with the findings of our previous study [12]. The treatment with selenium at both concentrations positively influenced the photosynthetic process. Our findings are in line with the results of research conducted on other plant species [17]. Proline is an amino acid synthesized by plants as a result of stress. It protects cellular structures (proteins and cytoplasmic membranes) from damage. Silicon has been found to reduce proline levels in plant cells, thus alleviating environmental stress and inhibiting the degradation of the cytoplasmic membrane [32,34]. Our study revealed the protective effect of silicon during drought stress. It was particularly noticeable when higher amounts of silicon were applied. The plants also exhibited the highest transpiration rate and stomatal conductance. Silicon improved the water uptake [13]. However, it increased the hydraulic properties of the roots rather than reducing the water loss. Silicon may enhance water capacity and uptake by the roots [17]. A higher water loss through transpiration was caused by an increase in the relative water content and indicated an enhanced water uptake by the roots. The researchers also noted the elevated stomatal conductance due to improved CO2 fixation capacity during water stress. This also ensured an optimum CO2 concentration for carbon reactions and prevented photoinhibition; the transcriptional rate of some photosynthesis-related genes in plants increased and regulated the photochemical process and thus promoted photosynthesis [11]. During drought stress, the intercellular CO_2 concentration (C_i) tended to increase, but silicon reduced it [15]. In this study, this parameter also decreased in all water-stressed plants supplemented with silicon, except ch-OSA I. Higher stomatal conductance was observed in sorghum treated with silicon [14].

Table 3: Gas exchange parameters of *Tagetes patula* L. "Pascal" of applied combinations (P_N – net photosynthesis activity (μ mol (CO_2) m⁻² s⁻¹); g_s – stomatal conductance (μ mol m⁻² s⁻¹), E – transpiration rate (μ mol (EO_2) mol⁻¹) and EO_2 concentration (EO_2) mol⁻¹)

Treatments			Before stress				After stress			
		P _N	gs	E	Ci	P _N	g _s	E	C _i	
Standard watering	Control	24.1a	118.4a	1.9a	138.8a	13.3c	49.4a	0.9ab	140.8c	
	Silica sol-II	32.4f	279.0e	5.3g	296.1ef	19.7i	98.8e	1.6e	149.3c	
	ch-OSA-II	29.8d	235.8cd	4.1d	276.2d	12.7b	47.9a	0.9b	138.1c	
Water stress	Control	25.3b	129.8a	2.2a	212.6b	11.8a	48.4a	0.9ab	85.7a	
	Se-I	29.7d	249.3d	4.5f	301.7ef	14.7e	60.0b	1.1c	103.0b	
	Se-II	30.4e	247.1d	4.5f	299.5ef	17.6h	80.4cd	1.4d	94.3ab	
	Silica sol I	25.6b	184.4b	3.2b	240.2b	13.8cd	61.4b	0.9ab	104.7b	
	Silica sol II	32.8f	286.8e	3.8c	275.8d	16.9g	73.2c	1.4d	89.0ab	
	ch-OSA I	28.3c	285.6e	4.3e	306.8ef	14.3de	83.9d	1.5e	195.1d	
	ch-OSA II	30.8e	226.4c	4.0d	252.3c	15.5f	59.2b	1.0bc	114.6b	

Different letters denote significant differences between combination within certain parameters at level $\alpha \le 0.05$, separately before and after the drought stress.

In this study, there was a similar effect observed in the plants grown under standard conditions. During the drought stress, stomatal conductance depended on the source and the concentration of silicon. Our investigations confirmed the protective role of selenium during the drought stress. Treatment with higher amounts of selenium caused higher P_N , g_s , and E levels. Similar effects of selenium and silicon on the uptake of water by roots during drought stress were also observed [47]. Selenium prevented drought stress by improving cellular enzymatic and nonenzymatic antioxidative mechanisms [7]. Moreover, selenium improves the cellular water balance [37], which is crucial for the photosynthetic process.

3.3 Chemical composition of plants

When silicon was applied under the standard water regime, it significantly reduced the nitrogen content in the shoots. However, the opposite trend could be observed under the water stress (Table 4). The nitrogen and phosphorus content in the plants treated with selenium was lower than that in the control samples. The phosphorus and calcium content in the plants grown under optimal conditions depended on the carrier. The silicon treatment had positive effect on the phosphorus content in the plants exposed to the water stress. The application of silicon to the plants grown under the optimal water regime significantly reduced the potassium content. In general, there were similar trends observed in the plants exposed to the stress. As the increasing amounts of silicon were applied through both sources

to the plants with the water deficit, their calcium content increased significantly compared with the control plants. The silicon treatment of the plants grown without the water deficit had a diversified, source-dependent effect on the calcium content. The sodium content in the shoots of the plants exposed to the drought stress was generally similar to the sodium content in the optimally irrigated control plants. In contrast to the selenium treatment, the silicon treatment had generally positive effect on the sodium content, but it did not affect the magnesium content in the optimally irrigated control plants.

However, the magnesium content tended to increase in the plants exposed to the stress when they were treated with selenium. The treatments also significantly differentiated the content of chemical components in inflorescences. For example, the silicate sol increased the nitrogen content in the plants exposed to the stress, whereas the selenium treatment significantly reduced the phosphorus content. On average, the nitrogen and potassium content in green parts of the plants was about 35–40% greater than that in the inflorescences. The calcium content was as much as about 80% greater. The phosphorus content was similar. Interestingly, the average sodium content in green parts of the plants was about 15% lower, whereas the magnesium content was almost 60% higher than in the inflorescences.

The quantitative relations between the macronutrients and the sodium in the shoots also changed. For example, the N:P ratio in the selenium-treated plants exposed to the stress was higher than that in the plants treated with silicon (Table 5). The N:K ratio in the silicontreated plants exposed to the stress was noticeably higher

Table 4: The influence of the factors under study on the content of macroelement and sodium in shoots and inflorescences of *Tagetes patula* L. "Pascal" (in % of D.M.)

Treatments		N	P	K	Ca	Na	Mg
Shoots							
Standard watering	Control	5.79d	0.78cd	5.69d	1.82bc	0.0083a	0.86ab
	Silica sol-II	5.30c	0.53a	5.10a	1.65a	0.0143cd	0.82a
	ch-OSA-II	5.38c	0.70bc	5.19ab	1.95cde	0.0075a	0.88ab
Water stress	Control	5.10bc	0.69b	5.59cd	1.77ab	0.0138bcd	0.90bc
	Se-I	4.86ab	0.51a	5.67cd	1.86bcd	0.0130abcd	0.95cd
	Se-II	4.71a	0.53a	5.48bcd	1.87bcd	0.0158d	0.97d
	Silica sol I	4.95ab	0.83de	5.16ab	1.96cde	0.0093abc	0.94cd
	Silica sol II	6.21e	0.76bcd	5.61cd	2.05e	0.0080a	0.88ab
	ch-OSA I	5.81d	0.79d	5.57cd	1.96cde	0.0085ab	0.99d
	ch-OSA II	6.05de	0.88e	5.30abc	2.03de	0.0080a	0.91bc
Inflorescences							
Standard watering	Control	3.29c	0.75c	3.71bc	0.38b	0.0090a	0.37b
	Silica sol-II	3.28c	0.66a	3.47ab	0.24a	0.0088a	0.32a
	ch-OSA-II	3.25c	0.73bc	3.48ab	0.30a	0.0130a	0.32a
Water stress	Control	2.84a	0.73bc	3.57abc	0.39b	0.0103a	0.33ab
	Se-I	3.15bc	0.67a	3.51ab	0.39b	0.0230b	0.35ab
	Se-II	2.93ab	0.67a	3.38a	0.36b	0.0170ab	0.35ab
	Silica sol I	3.10abc	0.72b	3.33a	0.38b	0.0095a	0.34ab
	Silica sol II	3.41cd	0.74bc	3.69bc	0.42b	0.0125a	0.35ab
	ch-OSA I	3.60d	0.75bc	3.78c	0.38b	0.0128a	0.35ab
	ch-OSA II	3.34cd	0.79d	3.58abc	0.40b	0.0100a	0.33a

Values within columns (separately for shoots and inflorescences) described with the same letter do not differ significantly at $\alpha = 0.05$.

than that in the plants treated with selenium. Changes in the quantitative relations between the components were also observed in the inflorescences.

In the silicon-treated plants grown under the optimal water regime and the silicon-treated (except ch-OSA II) and selenium-treated plants exposed to the drought stress, there was a relative increase in the nitrogen content in the inflorescences compared with its content in the shoots (Table 6). There were similar relations concerning the phosphorus content in the plants grown under the optimal water regime and the selenium-treated plants exposed to the stress. In general, the drought stress did not cause changes in the flowerto-shoot potassium content ratio. The silicon-treated plants grown under the optimal water regime were characterized by a smaller flower-to-shoot calcium content ratio (0.15 each) than the other combinations. This trend was not so noticeable in the plants exposed to the stress. In all the combinations exposed to the drought stress, the flower-to-shoot magnesium content ratio was smaller than in the control plants grown under the optimal water regime. In comparison with the control plants, both the selenium and silicon treatment of the plants exposed to the water stress caused noticeable changes in the sodium ratio.

The research showed that in general, the factors tested in the experiment had significant influence on the content of micronutrients and silicon in the shoots. For example, the iron content in the control plants exposed to the drought stress was significantly lower than in the plants treated with silicon, regardless of the carrier and the concentration (Table 7). In general, there were inverse relations concerning the manganese content. The silicon treatment of the plants grown under the optimal water regime significantly increased the manganese content in their shoots. There was a similar upward trend observed in the plants exposed to the drought stress compared with the optimally irrigated control plants.

The iron and zinc content in the inflorescences was more stable than in the shoots. In general, there were no differences in the content of these nutrients between the combinations. The silicon treatment of the plants exposed to the drought stress reduced the copper content in their inflorescences. There was a similar tendency concerning the manganese content. Both silicon carriers caused generally significant differences. In all the combinations, the silicon content in the plants exposed to the drought stress was significantly higher than in the optimally irrigated control plants.

Table 5: The quantitative relations between macroelements and sodium in shoots and inflorescences of *Tagetes patula* L. "Pascal." For each combination the N content was taken as 1.00

Treatmen	nts	N	Р	K	Ca	Na	Mg
Shoots							
Standard watering	Control	1.00	0.13	0.98	0.31	0.001	0.15
	Silica sol-II	1.00	0.10	0.96	0.31	0.003	0.15
	ch-OSA-II	1.00	0.13	0.96	0.36	0.001	0.16
Water stress	Control	1.00	0.14	1.10	0.35	0.003	0.18
	Se-I	1.00	0.10	1.17	0.38	0.003	0.20
	Se-II	1.00	0.11	1.16	0.40	0.003	0.21
	Silica sol I	1.00	0.17	1.04	0.40	0.002	0.19
	Silica sol II	1.00	0.12	0.90	0.33	0.001	0.14
	ch-OSA I	1.00	0.14	0.96	0.34	0.001	0.17
	ch-OSA II	1.00	0.15	0.88	0.34	0.001	0.15
Inflorescences							
Standard watering	Control	1.00	0.23	1.13	0.12	0.003	0.11
	Silica sol-II	1.00	0.20	1.06	0.07	0.003	0.10
	ch-OSA-II	1.00	0.22	1.07	0.09	0.004	0.10
Water stress	Control	1.00	0.26	1.26	0.14	0.004	0.12
	Se-I	1.00	0.21	1.11	0.12	0.007	0.11
	Se-II	1.00	0.23	1.15	0.12	0.006	0.12
	Silica sol I	1.00	0.23	1.07	0.12	0.003	0.11
	Silica sol II	1.00	0.22	1.08	0.12	0.004	0.10
	ch-OSA I	1.00	0.21	1.05	0.11	0.004	0.10
	ch-OSA II	1.00	0.24	1.07	0.12	0.003	0.10

Table 6: The inflorescence-to-shoot ratio of the content of macronutrients and sodium

Treatme	ents	N	P	K	Ca	Mg	Na
Ctondord	Control	0.57	0.06	0.65	0.42	1.00	1.00
Standard	Control	0.57	0.96	0.65	0.43	1.08	1.08
watering	Silica sol-II	0.62	1.25	0.68	0.39	0.62	0.62
	ch-	0.60	1.04	0.67	0.36	1.73	1.73
	OSA-II						
Water stress	Control	0.56	1.06	0.64	0.37	0.75	0.75
	Se-I	0.65	1.31	0.62	0.37	1.77	1.77
	Se-II	0.62	1.26	0.62	0.36	1.08	1.08
	Silica	0.63	0.87	0.65	0.36	1.02	1.02
	sol I						
	Silica	0.55	0.97	0.66	0.40	1.56	1.56
	sol II						
	ch-OSA I	0.62	0.95	0.68	0.35	1.51	1.51
	ch-	0.55	0.90	0.68	0.36	1.25	1.25
	OSA II						

There were noticeable differences in the content of microelements and silicon in the inflorescences in relation to the shoots (Table 8). For example, when silicon was applied to the plants grown under the optimal water regime as well as those exposed to the water stress (except ch-OSA I), the relative iron content increased.

Both silicon carriers modified the inflorescence-to-shoot iron content ratio.

The factors analyzed in this study caused changes in the quantitative relations between micronutrients in both the shoots and inflorescences (Table 9). For example, the Fe:Zn ratio in the silicon-treated plants exposed to the drought stress was noticeably higher than in the control plants, regardless of the sources. There was a similar relation observed for the copper and manganese content. The silicon treatment narrowed the Fe:Zn ratio in both the plants grown under the optimal water regime and those exposed to the short-term drought stress. However, the selenium treatment resulted in different Fe:Zn and Fe:Cu ratios.

The average iron content in the shoots was 8% higher than in the inflorescences. Most of the combinations also had higher zinc and copper content in the green parts than in the inflorescences. There was a similar relation observed for the manganese content – in all the combinations, it was higher in the green parts than in the inflorescences.

Changes in the uptake of components and consequently changes in the chemical composition of plants should be treated as their adaptation to specific environmental factors. For example, the uptake and the translocation of nutrients in plants can be modified by silicon

Table 7: The influence of the factors under study on the content of microelement and silicon in shoots and inflorescences of *Tagetes patula* L. "Pascal" (in $mg kg^{-1}$ of D.M.)

Treatments		Fe	Zn	Cu	Mn	Si
Shoots						
Standard watering	Control	108.16c	70.32f	16.98b	272.26f	1 103.5a
	Silica sol-II	97.67bc	60.21cde	12.47a	174.66c	1 483.0b
	ch-OSA-II	106.37c	48.73ab	12.14a	169.75c	1 439.0b
Water stress	Control	81.03a	48.35a	11.54a	201.85de	1 327.0b
	Se-I	87.64ab	59.75cde	13.37a	180.20cd	1 324.0b
	Se-II	77.35a	61.56de	11.32a	217.92e	1 410.5b
	Silica sol I	130.30d	65.90ef	11.09a	136.23b	1 619.5b
	Silica sol II	131.04d	58.97cd	13.69a	140.07b	1 635.0b
	ch-OSA I	97.34bc	54.72bc	11.84a	205.67e	1 576.0b
	ch-OSA II	99.13bc	52.41ab	10.59a	107.43a	1 633.0b
Inflorescences						
Standard watering	Control	105.37a	51.81a	22.04cd	48.58g	853.0a
	Silica sol-II	84.93a	34.08a	21.24bcd	31.28cde	1 205.0b
	ch-OSA-II	96.61a	30.96a	20.85bcd	28.49bcd	1 135.0b
Water stress	Control	78.96a	38.31a	22.57d	38.99f	1 460.0b
	Se-I	78.56a	44.10a	17.42abcd	35.74ef	1 307.5b
	Se-II	79.11a	44.04a	21.62cd	35.36ef	1 301.0b
	Silica sol I	96.83a	40.68a	16.41ab	24.01ab	1 220.0b
	Silica sol II	92.09a	43.33a	16.95abc	24.47abc	1 161.5b
	ch-OSA I	136.98b	110.33b	17.77abcd	30.92cde	1 424.0b
	ch-OSA II	85.06a	53.92a	14.17a	21.31a	1 510.5b

Values within columns (separately for shoots and inflorescences) described with the same letter do not differ significantly at $\alpha = 0.05$.

Table 8: The inflorescence-to-shoot ratio of the content of micronutrients and silicon

Treatn	nents	Fe	Zn	Cu	Mn	Si
Standard	Control	1.03	1.36	0.77	5.60	1.29
watering	Silica sol-II	1.15	1.77	0.59	5.58	1.23
	ch-OSA-II	1.10	1.57	0.58	5.96	1.27
Water stress	Control	1.03	1.26	0.51	5.18	0.91
	Se-I	1.12	1.35	0.77	5.04	1.01
	Se-II	0.98	1.40	0.52	6.16	1.08
	Silica sol I	1.35	1.62	0.68	5.67	1.33
	Silica sol II	1.42	1.36	0.81	5.72	1.41
	ch-OSA I	0.71	0.50	0.67	6.65	1.11
	ch-OSA II	1.17	0.97	0.75	5.04	1.08

[48]. Also drought stress significantly increased the content of K, Na, Ca, Mg, and Fe in plants, whereas silicon treatment reduced the content of these elements [4]. In this study, the opposite tendency was observed for the N, P, K, and Ca content in the shoots, but a similar tendency for the Na content. Similar to this study, it was observed that silicon affected the chemical composition of water-stressed plants [34]. The silicon concentration in the plants was correlated with its content in the nutrient solution. In this study, the silicon content in the shoots

of the plants grown under the optimal watering regime was significantly higher than in the control plants. There were increasing but insignificant tendencies observed in the plants exposed to the water stress. Water stress decreased the calcium and potassium concentrations in plants, but silicon treatment increased their levels [34]. In this study, a similar effect was observed only for the calcium content (at higher silicon concentrations in the nutrient solution). It was found that the silicon treatment may improve the growth of plants and increase its yield during drought, but it could not be a full substitute for an adequate water supply. The silicon treatment caused an upward trend (without significant differences) in the yield of plants exposed to the short-term drought stress. However, the selenium treatment caused significant differences. Silicon treatment reduced the sodium content in rice plants exposed to salinity stress [10]. In contrast to the findings of the other scientist [49], in this study, the simultaneous silicon treatment of the plants exposed to the drought stress caused an upward trend in the sodium content (without statistically significant differences). Selenium supplementation disorders the metabolism of amino acids and thus increases the content of soluble proteins and the nitrate reductase activity in plants exposed to drought stress [50]. When wheat plants

Table 9: The quantitative relations between microelements and silicon in shoots and inflorescences of *Tagetes patula* L. "Pascal." For each combination the Fe content was taken as 1.00

Treatme	nts	Fe	Zn	Cu	Mn	Si
Shoots						
Standard	Control	1.00	0.65	0.16	2.52	10.202
watering	Silica sol-II	1.00	0.62	0.13	1.79	15.184
	ch-OSA-II	1.00	0.46	0.11	1.60	13.528
Water stress	Control	1.00	0.60	0.14	2.49	16.377
	Se-I	1.00	0.68	0.15	2.06	15.107
	Se-II	1.00	0.80	0.15	2.82	18.235
	Silica sol I	1.00	0.51	0.09	1.05	12.429
	Silica sol II	1.00	0.45	0.10	1.07	12.477
	ch-OSA I	1.00	0.56	0.12	2.11	16.191
	ch-OSA II	1.00	0.53	0.11	1.08	16.473
Inflorescences						
Standard	Control	1.00	0.49	0.21	0.46	8.095
watering	Silica sol-II	1.00	0.40	0.25	0.37	14.188
	ch-OSA-II	1.00	0.32	0.22	0.29	11.748
Water stress	Control	1.00	0.49	0.29	0.49	18.490
	Se-I	1.00	0.56	0.22	0.45	16.643
	Se-II	1.00	0.56	0.27	0.45	16.445
	Silica sol I	1.00	0.42	0.17	0.25	12.599
	Silica sol II	1.00	0.47	0.18	0.27	12.613
	ch-OSA I	1.00	0.81	0.13	0.23	10.396
	ch-OSA II	1.00	0.63	0.17	0.25	17.758

were supplemented with selenium in an exogenous form, their uptake of potassium, iron, and zinc improved [18]. Selenium supplementation also improves turgor and regulates transpiration and the accumulation of soluble sugars and free amino acids. Selenium supplementation also improved the wheat grain yield by 24% and simultaneously increased the straw calcium content [18]. In this study, the selenium treatment had positive effect on the calcium content in the shoots. The effect of selenium on the accumulation of potassium and phosphorus was found in the studies on rapeseed [39] and rice [8]. The studies conducted on spinach showed that selenium significantly affected the calcium content [19]. In this study, the nitrogen and phosphorus content in the shoots of the plants exposed to the short-term drought stress exhibited a downward trend, whereas the calcium and magnesium content exhibited an upward trend. Selenium may limit the uptake of metals such as Mn, Zn, Cu, Cd, and Pb [20,21,51]. In this study, when the plants exposed to the drought stress were treated with selenium, the manganese content tended to decrease (insignificant differences), whereas the zinc content tended to increase.

4 Conclusion

- The silicon treatment stimulated the biometric parameters of the control plants grown under the optimal water regime. When the plants were exposed to the drought stress, the values of the biometric parameters after the selenium treatment were higher than that after the silicon treatment.
- 2. Both silicon and selenium significantly modified the gas exchange parameters. During the growing season, the net photosynthesis activity (P_N) , stomatal conductance (g_s) , and transpiration rate (E) deteriorated, but the selenium and silicon treatment increased these parameters significantly.
- 3. In general, the factors under analysis significantly modified the plants' nutrition with both macronutrients and micronutrients, as well as the proportions between them.
- 4. Both silicon and selenium alleviated the short-term drought stress in French marigolds. When silicon is applied, this reaction can be modified by the type of source and its concentration.

Author contributions: T. K.: research concept, co-conducting experiments, data analysis, statistical analyses, authorship of a fragment of the manuscript, made formal analysis, edited the whole manuscript, and proofreading this manuscript; K. B.: measurement of gas exchange parameters, statistical analyses, and authorship of a manuscript fragment; T. K.: analysis of plant material for silicon content and took part in the discussion on the research results; W. B.: took part in the discussion on the research results; B. Ł.: co-conducting experiments, biometric measurements of plants, chemical plant analyses, and statistical analyses

Funding: A study financed from the program "Maintaining research potential" financed by Ministry of Science and Higher Education.

Conflict of interest: The authors declare no conflicts of interest. The founding sponsors had no role in the design of the study; the collection, analyses, or interpretation of data; and the writing of the manuscript or the decision to publish the research results.

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