EFFECTS OF ZEOLITE AND SELENIUM APPLICATIONS ON SOME AGRONOMIC TRAITS OF THREE CANOLA CULTIVARS UNDER DROUGHT STRESS¹

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RESUMO

EFEITOS DA APLICAÇÃO DE ZEÓLITA E SELÊNIO EM TRÊS CULTIVARES DE CANOLA SOB ESTRESSE HÍDRICO

Para estudar os efeitos da aplicação de zeólita e selênio, nas características agronômicas de três cultivares de canola, um experimento foi realizado em duas safras (2006 e 2007). O estudo foi conduzido em área experimental do Instituto de Sementes e Melhoramento de Plantas, em Karaj, Irã. O delineamento experimental utilizado foi o de blocos casualizados, em esquema de parcelas subdivididas, com três repetições. O fator irrigação foi empregado em dois níveis: irrigação normal e estresse hídrico no estádio de alongamento das hastes. A zeólita foi empregada em dois níveis: sem aplicação e aplicação de 10 t ha⁻¹. O selênio foi aplicado em três concentrações: 0 g L-1, 15 g L-1 e 30 g L-1 de seleneto de sódio. Estes tratamentos foram casualizados nas parcelas principais, enquanto as três cultivares de canola (Zarfam, Sarigol e Okapi) foram casualizadas nas subparcelas. Com base nos resultados, é possível inferir que os efeitos isolados foram significativos, enquanto as interações não foram significativas. O fator irrigação foi significativo para todas as variáveis agronômicas estudadas, com o estresse hídrico reduzindo os parâmetros avaliados. A aplicação de selênio aumentou o diâmetro da haste, o número de sementes e síliquas e a produção de sementes, mas não alterou significativamente o comprimento da síliqua. A aplicação de zeólita aumentou significativamente a produção de sementes e os componentes de produção. As cultivares apresentaram diferentes números de síliquas e de sementes. De modo geral, a aplicação de zeólita e selênio apresentaram efeito positivo e significativo no crescimento, nas síliquas e na produção de sementes. Como resultado, pode-se sugerir que a aplicação de zeólita e selênio podem ser benéficas para melhorar o crescimento das plantas sob estresse hídrico.

PALAVRAS-CHAVE: *Brassica napus* L.; diâmetro da haste; número de síliquas; produção de sementes.

INTRODUCTION

In many regions of the world, including Iran, drought stress is one of the most important factors responsible for decreasing agricultural crop yield. Canola (*Brassica napus* L.) is one of the most

ABSTRACT

In order to study the effects of zeolite and selenium application on the agronomic traits of three canola cultivars under drought stress conditions, an experiment was conducted in two growing seasons (2006 and 2007). The study site was the Seed and Plant Improvement Institute, in Karaj, Iran. The experimental design was a randomized complete block arrangement, in a split-plot factorial scheme, with three replications. The irrigation factor was applied at two levels: normal irrigation and water holding at the stem elongation stage. Zeolite was used at two levels: non-application and application of 10 t ha⁻¹. Selenium was sprayed at three concentrations: 0 g L⁻¹, 15 g L⁻¹, and 30 g L⁻¹ of sodium selenate. These treatments were randomized in main plots, while three canola cultivars (Zarfam, Sarigol, and Okapi) were randomized as subplots. The results showed that the main effects were significant, while interaction effects were not significant. Irrigation had significant effect on all traits as drought stress lead to a decrease of those traits. Selenium application increased stem diameter, silique and seed number, and seed yield, but it had no significant effect on silique length. Zeolite application provided a significant increase in seed yield and yield components. The cultivars were different in silique and seed number. In general, selenium and zeolite application had significant and positive effect on growth and silique and seed yield. As a result, it can be suggested that the zeolite and selenium application may improve plant growth under drought stress.

KEY-WORDS: *Brassica napus* L.; stem diameter; silique number; seed number; seed yield.

important oilseed plants in the world. Drought stress and high temperatures during the flowering and seed-filling stages decrease canola yield. Researchers have shown that, in many flowering plants, the pollination and seed-filling stages are sensitive to drought stress (Liu et al. 2004).

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Zahedi et al. (2009) reported that the zeolite and selenium application may improve plant growth under drought stress. Kavoosi (2007) showed that the application of 10 t ha⁻¹ of zeolite significantly increased rice grain yield. It has also been reported that the flowering and primary stages of pod-setting are the most sensitive stages for canola growth (Hashem et al. 1998). The responses of two canola cultivars to drought stress were studied by Wright et al. (1995). They concluded that seed yield and oil percentages were strongly affected by drought stress. According to published results, decreases in canola seed yield are due to decreases in yield components, including number of pods, number of seeds per pod, and 1,000-seeds weight, in response to drought stress. Jensen et al. (1996) showed that normal irrigation, during the flowering and primary stages of pod development, increases the number of pods and number of seeds per pod, but average 1.000-seeds weight was less affected than the number of seeds per pod. The flowering and pod-setting stages are critical and affected by drought stress in canola (Rao & Mendham 1991).

The effects of different irrigation regimes on the flowering stage of canola have been studied by Deepak & Wattal (1995). Their results showed that water stress significantly decreased seed and biological yields. A decrease in biological yield and pod dry matter have been observed in two canola cultivars, under water deficit conditions (Thomas et al. 2004). Porous ceramics, diatomaceous earth, and zeolites are just a few of the more commonly used inorganic soil amendments. Some characteristics of these products, that make them desirable for improving soils properties, are a large internal porosity, which results in water retention; a uniform particle size distribution, that allows them to be easily incorporated; and a high cation exchange capacity, which retains nutrients (Ok et al. 2003). The unique physical and chemical properties of natural zeolites, in combination with their abundance in sedimentary deposits and in rocks derived from volcanic parent materials, have made them useful for many industrial applications. These properties have spurred their use in agronomic and horticultural applications (Dwairi 1998).

Zeolites are hydrated aluminosilicates, characterized by three-dimensional networks of SiO_4 and AlO_4 tetrahedral and linked by shared oxygen atoms. Clinoptilolite is not the most well-known

zeolite, but it is one of the most useful. The chemical formula of clinoptilolite is (Na, K,)(Al, Si, O₇₂).24H,O. Extensive deposits of clinoptilolite are found in the Western United States, Bulgaria, Hungary, Japan, Australia, and Iran. Amendment of clinoptilolite zeolite to sandy soils has been reported to lower nitrogen concentrations in leachate and to increase moisture and nutrients in the soil, due to increased soil surface area and cation exchange capacity (He et al. 2002). Selenium (Se) is an essential trace element for animals and humans (Tapiero et al. 2003), but its role in plants is still unclear (Hartikainen et al. 2000). Most cereal crops and fodder plants are relatively weakly able to absorb selenium, even when grown on soils with higher selenium content. There are indications that it can also play a positive biological role in higher plants (Germ et al. 2005).

MATERIAL AND METHODS

This study was conducted at the experimental field of the Seed and Plant Improvement Institute (SPII), in Karaj, Iran (35°59′N, 50°75′E and altitude of 1,313 m), with three canola cultivars (*Brassica napus* L. c. v. Zarfam, Sarigol, and Okapi), in the 2006 and 2007 growing seasons. The yearly average precipitation (over a 30-years period), which is mostly concentrated in the autumn and winter months, was 244 mm.

Before the experiment began, soil samples were taken to determine their physical and chemical properties. Composite soil samples were collected from a depth of 0-30 cm and 30-60 cm. They were air dried, crushed, and tested for physical and chemical properties. The soil classification was clay loam (Table 1). After plowing and disking, plots were prepared.

The experimental design was a randomized complete block, with a factorial split-plot arrangement of treatments, in three replications. The treatments included normal irrigation (I_1) and water holding (I_2), at the stem elongation stage. Zeolite applications were performed at 0 t ha⁻¹ (Z_1) and 10 t ha⁻¹ (Z_2), and selenium applications were performed at the concentrations of 0 g L⁻¹ (S_1), 15 g L⁻¹ (S_2), and 30 g L⁻¹ (S_3), as sodium selenate, at the initial silique stage. These treatments were applied to the three canola cultivars.

According to the soil analysis, zeolite was distributed on the soil surface and incorporated

| Year | Depth (cm) | EC (ds m ⁻¹) | pН | Organic carbon (%) | Saturated percentage (%) | N (%) | P (ppm) | K (ppm) | Ca (ppm) | Mg (ppm) | T.N.V (%) | Texture |
|------|------------|--------------------------|-----|--------------------|--------------------------|----------|------------|------------|-------------|-------------|--------------|---------|
| 2006 | 0-30 | 1.36 | 7.8 | 0.47 | 30.58 | 0.05 | 4.45 | 171 | 45 | 13 | 8.25 | clay |
| 2006 | 30-60 | 1.76 | 7.7 | 0.35 | 30.84 | 0.04 | 4.92 | 132 | 53 | 21 | 10.69 | loam |
| 2007 | 0-30 | 1.42 | 7.8 | 0.51 | 36.01 | 0.06 | 3.02 | 205 | 64 | 18 | 9.81 | clay |
| | 30-60 | 1.44 | 7.9 | 0.40 | 37.12 | 0.05 | 3.12 | 150 | 51 | 33 | 10.53 | loam |

Table 1. Physical and chemical properties of soil collected from the study site (Karaj, Iran, 2006/2007).

into the soil at a depth of 30 cm. The plots were 5 m long and consisted of six rows, 0.3 m apart. Between blocks and main plots, 6 m and 2.4 m alleys, respectively, were set to eliminate the influence of lateral water movement. The canola seeds were disinfected and sown in early October. Irrigation was carried out similarly in all plots, until the reproductive stage. Two water treatments (normal irrigation and water holding) were used at the stem elongation stage. Weeds were effectively controlled by hand. Selenium applications as sodium selenate were carried out at three concentrations (0 g L⁻¹, 15 g L⁻¹, and 30 g L⁻¹), by using motorized backpack sprayers, at the initial silique stage. At the end of the growing season, plant height, number of branches per plant, number of siliques per plant, number of seeds per silique, 1,000-seeds weight, and seed yield were measured. To measure the number of siliques per plant, after eliminating the margin effect, 10 plants were randomly harvested from the middle of each plot and then the means of the traits were measured. Siliques from these plants were separated, and 30 siliques were randomly selected to calculate the number of seeds per silique and the 1,000-seeds weight.

All data were analyzed by variance analysis (ANOVA), using the SAS GLM procedure (SAS Institute 2002). The assumptions of variance analysis were tested by ensuring that the residuals were random and homogenous, with a normal distribution around a mean of zero. The LSMEANS command was used to compare means, at a p < 0.05 probability.

RESULTS AND DISCUSSION

When the combined analysis of two years was performed, results demonstrated that the effect of the year as a treatment had a significant effect on all traits, except for main silique length (Table 2). Different irrigation levels (complete irrigation and water withholding at the stem growth stage) had

a significant effect on all growth traits (p < 0.01). Selenium application showed a significant effect just on the number of siliques on main and sub-stems and number of seeds on main silique (Table 2), while all the assayed traits were affected by zeolite application. Meanwhile, there was a significant difference among cultivars, concerning stem diameter, main silique length, average of silique length and number of seeds on main silique.

According to the variance analysis table, the main effects of year, irrigation, selenium zeolite, and cultivar were significant on traits, but interactions were not significant, thus the main effects will be explained in this study. Interaction effects among irrigation, selenium, zeolite, and cultivars were significant only for seed yield. There are many reports of a decrease of vegetative growth and plant height under drought stress conditions (Wright et al. 1995). An increase in plant height is related to two phenomena (an increase in node number and inter-node length), and these are strongly affected by drought stress (Wright et al. 1988).

Plant reproductive stages, including stem elongation, flowering, pollination, and seed-filling, are sensitive to water stress (Thomas et al. 2004). The flowering and silique-setting stages are particularly susceptible to drought stress in canola (Wright et al. 1995). Comparison of means of main effects shows that there is a significant difference between first and second year on all traits, except for silique length. In general, silique growth and yield were higher in the second year than in the first one, and this could be due to climate conditions and improvement of management schedules. The results confirm that the flowering and silique-setting stages are critical periods of water supply, and that water stress, in those periods, strongly decreases silique number and seed yield. A decrease in the 1,000-seeds weight is due to a decline in water and nutrient absorption by plants and a decrease in assimilation and assimilates transported to seeds.

Table 2. Combined variance analysis on some agronomic factors of canola treated with water stress, zeolite, and selenium (Karaj, Iran, 2006/2007).

| S.O.V | d.f | Stem diameter | Number of silique on main stem | Number of silique on sub-stem | Length of main silique | Length of subsilique | Length of silique | Number of seeds on main silique | Number of seeds on sub-silique | 1,000-seeds weight | Seed yield |
|-------------|-----|------------------|--------------------------------|-------------------------------|------------------------|----------------------|-------------------|---------------------------------|--------------------------------|-----------------------|---------------|
| Y | 1 | * | * | * | ns | ** | * | ** | * | * | ** |
| R(Y) | 4 | ** | * | ns | ** | ns | ** | ns | ns | ns | ns |
| I ' | 1 | ** | ** | ** | ** | ** | ** | ** | ** | ** | ** |
| S | 2 | ns | * | ** | ns | ns | ns | ** | * | ns | ** |
| \tilde{Z} | 1 | ** | ** | ** | ** | ** | ** | ** | ** | ** | ** |
| I*S | 2 | * | ns | ** | ns | ns | * | * | ns | ns | ** |
| I*Z | 1 | ns | ns | * | ns | ns | ns | * | ns | ns | ** |
| I*Y | 1 | ns | ns | ns | ns | ns | ns | ns | ns | ns | ** |
| S*Y | 2 | ns | ns | ns | ns | ns | ns | ns | ns | ns | ns |
| Z*Y | 1 | ns | ns | ns | ns | ns | ns | ns | ns | ns | ns |
| S*Z | 2 | ns | ns | ns | ns | ns | ns | ns | ns | ns | ** |
| I*S*Z | 2 | ns | ns | * | ns | ns | ns | ns | ns | ns | ** |
| I*S*Y | 2 | ns | ns | ns | ns | ns | ns | ns | ns | ns | ns |
| I*Z*Y | 1 | ns | ns | ns | ns | ns | ns | ns | ns | ns | ns |
| S*Z*Y | 2 | ns | ns | ns | ns | ns | ns | ns | ns | ns | ns |
| I*S*Z*Y | 2 | ns | ns | ns | ns | ns | ns | ns | ns | ns | ns |
| R*I*S*Z(Y) | 44 | ns | ns | ns | ns | ns | ns | ns | ns | ns | ns |
| V | 2 | ** | ns | ns | ** | ns | ** | ** | ** | ** | ** |
| I*V | 2 | ** | ns | ns | ns | ns | ns | * | ns | * | ns |
| S*V | 4 | ns | ns | ns | ns | ns | ns | ns | ns | ns | ** |
| Z*V | 2 | ns | ns | ns | ns | ns | ns | ns | ns | ns | ** |
| V*Y | 2 | ns | ns | ns | ns | ns | ns | ns | ns | ns | ns |
| I*V*Y | 2 | ns | ns | ns | ns | ns | ns | ns | ns | ns | ns |
| S*V*Y | 4 | ns | ns | ns | ns | ns | ns | ns | ns | ns | ns |
| Z*V*Y | 2 | ns | ns | ns | ns | ns | ns | ns | ns | ns | ns |
| I*S*V | 4 | ns | ns | ns | ns | ns | ns | ns | ns | ns | ns |
| S*Z*V | 4 | ns | ns | ns | ns | ns | ns | ns | ns | ns | ns |
| I*Z*V | 2 | ns | ns | ns | ns | ns | ns | ns | ns | ns | * |
| I*S*V*Y | 4 | ns | ns | ns | ns | ns | ns | ns | ns | ns | ns |
| I*S*Z*V | 4 | ns | ns | ns | ns | ns | ns | ns | ns | ns | ** |
| S*Z*V*Y | 4 | ns | ns | ns | ns | ns | ns | ns | ns | ns | ns |
| [*Z*V*Y | 2 | ns | ns | ns | ns | ns | ns | ns | ns | ns | ns |
| I*S*Z*V*Y | 4 | ns | ns | ns | ns | ns | ns | ns | ns | ns | ns |
| C. V. | | 13.67 | 20.04 | 18.10 | 8.23 | 8.01 | 5.51 | 13.70 | 15.33 | 4.8 | 11.4 |

Y: year; R: replication; I: irrigation; S: selenium; Z: zeolite; V: cultivar; * and **: Significant at 0.05 and 0.01, respectively; ns: No significant.

Water stress or withholding irrigation at the stem growth stage decreased stem diameter, number of silique on main stem, number of silique on substems, main silique length, sub-silique length, silique length, number of seeds on main silique and on subsilique, 1,000-seeds weight, and seed yield (Table 3). The effect of drought stress on winter canola has been studied by Andersen et al. (1996). They observed that the number of pods per plant, number of seeds per pod, and seed yield were significantly decreased by drought stress, while 1,000-seeds weight was decreased by drought stress at the flowering and seed-filling stages (Andersen et al. 1996). Water stress usually decreases photosynthesis, also decreasing growth and dry matter production (He et al. 2002). The stem elongation stage in canola is sensitive to water stress and any water stress in this stage decreases final yield.

Previous results showed that flowering and silique development stages are critical stages for

water requirement, and lack of water, in those stages, decreases the number of silique in unit of the cultivated area. Bouchereau et al. (1996) reported that water stress decreases the number of silique in canola, and also that water stress, at the flowering stage, significantly decreases the number of silique in each plant (Triboi-Blondel & Renard 1999), while postponed stress leads to a decrease in the number of seeds in silique. Supplementary irrigation increases the number of silique and the number of seeds in silique, due to the increase of the flowering period. The number of seeds in silique is affected by assimilates production and limitation of that by environmental factors, especially water stress. Irrigation withholding, at the stem elongation stage, significantly decreased growth, number and length of silique, and number of seeds in silique (Table 3). In general, supplying enough water at the flowering and silique-setting stages increases the number of seed in

| Treatments | Level | Stem diameter (mm) | Number of silique on main stem | Number of silique on sub-stem | Length of main silique (cm) | Length of sub-silique (cm) | Length of silique (cm) | Number of seeds on main silique | Number of seeds on sub-silique | 1,000-seeds weight (g) | Seed yield (kg/ha) |
|------------|-----------------------|--------------------------|--------------------------------|-------------------------------|-----------------------------|----------------------------------|------------------------------|---------------------------------|--------------------------------------|------------------------------|-----------------------|
| Vaan | First | 6.73 a | 26.51 b | 44.08 b | 5.82 a | 5.52 b | 5.67 b | 23.28 b | 24.50 b | 5.46 b | 3765.29 b |
| Year | Second | 7.05 a | 31.00 a | 50.25 a | 6.47 a | 6.28 a | 6.38 a | 25.85 a | 22.69 a | 5.68 a | 4518.29 a |
| Irrigation | Complete | 7.67 a | 32.95 a | 57.78 a | 6.39 a | 6.09 a | 6.24 a | 26.27 a | 25.97 a | 5.75 a | 5456.33 a |
| | Stress | 6.11 b | 24.57 b | 36.54 b | 5.90 b | 5.71 b | 5.81 b | 22.86 b | 21.22 b | 5.39 b | 2827.24 b |
| Zeolite | 0 t ha-1 | 6.51 b | 26.09 b | 41.21 b | 6.01 b | 5.78 b | 5.90 b | 2364 b | 22.44 b | 5.48 b | 3563.13 b |
| | 10 t ha ⁻¹ | 7.28 a | 31.42 a | 53.12 a | 6.28 a | 6.02 a | 6.15 a | 25.49 a | 24.75 a | 5.66 a | 4720.44 a |
| Selenium | 0 g L ⁻¹ | 6.69 b | 27.10 b | 42.89 b | 6.05 a | 5.85 a | 5.95 a | 23.80 b | 2266 b | 5.51 a | 3704.92 с |
| | 15 g L ⁻¹ | 6.96 ab | 30.10 a | 48.84 a | 6.18 a | 5.93 a | 6.06 a | 25.27 a | 24.01 a | 5.61 a | 4263.17 b |
| | 30 g L ⁻¹ | 7.02 a | 29.08 ab | 49.75 a | 6.20 a | 5.92 a | 6.06 a | 24.63 a | 24.12 a | 5.58 a | 4457.28 a |
| | Sarigol | 6.45 b | 28.88 a | 65.39 a | 5.84 c | 5.81 b | 5.83 c | 21.29 с | 21.12 b | 5.60 b | 3941.68 b |
| Cultivar | Zarfam | 6.98 a | 28.91 a | 42.39 b | 6.38 a | 5.99 a | 6.18 a | 25.41 b | 24.57 a | 5.77 a | 4275.25 a |

5.90 ab

6.06 a

27.00 a

Table 3. Main effects of year, irrigation, zeolite, selenium, and cultivar on some agronomic factors (Karaj, Iran, 2006/2007).

Within each column followed by the same letter, there are no significant differences (p < 0.05).

33.70 c

6.21 b

28.48 a

7.25 a

Okapi

silique and finally seed yield (Sing & Bhargava 1994, Mendham & Salisbury 1995). Mendham & Salisbury (1995) emphasized that supplying water at first, at the silique-setting stage, is very important and that the number of silique is affected by water stress, while water stress, after this stage, is effective on number of seeds in silique. Mendham & Salisbury (1995) showed that water stress, at the flowering stage, decreased the number of seeds in silique and this reduction was fixed after three or four weeks before ripening. Champolivier & Merrien (1996) reported that the number of silique is more sensitive to water stress than other yield components. Moreover, Mendham et al. (1992) pointed out that complete irrigation at first, at the silique-setting stage, had an important role in final yield, and that water stress, at this stage, decreased the length of silique and seed yield. Xiubin & Zhanbin (2001) showed that zeolite improved water retention capacity and cation exchange capacity in arable soils.

Stem diameter significantly decreased due to water stress, while zeolite and selenium application increased stem diameter (Table 3). Increase of stem diameter can be due to improved water and nutrition availability by zeolite. There was no significant difference between Zarfam and Okapi, for stem diameter, while Sarigol presented the lowest stem diameter (Table 3). Irrigation had a significant effect on the number of silique on the main stem (Table 3). The main effect of zeolite was also significant on the stem diameter (Table 3). It was observed that selenium application increased the number of silique on the main stem, but there was no significant difference between the concentrations of 15 g L⁻¹ and 30 g L⁻¹. Comparison

of means showed that there was no significant difference among cultivars, concerning the number of silique on main stem (Table 3). Selenium application had a significant effect on stem diameter, number of silique on main stem and sub-stem, and number of seeds on main stem and sub-stem. In contrast, selenium had no significant effect on silique length (Table 2). Leggo (2000) studied the response of wheat to poultry manure amended by zeolite and found out that crop faced a better growth rate when zeolite was applied in poultry manure, and reported that the increase of growth and yield is due to nitrogen availability by zeolite. One of the most important roles of selenium is increasing growth (Hartikainen et al. 2000).

25.10 a

5.33 c

4208.43 a

Selenium has been known as an antioxidant molecule. It seems that selenium improves plant growth conditions and that the increase of stem diameter is due to the increase of vegetative growth, while the increase of zygosis, due to selenium, was reported by Quanchang & Cheng (2008). In sum, we can conclude that the selenium and zeolite application had a positive and significant effect on growth and seed yield in canola.

CONCLUSIONS

- 1. Zeolite application, in soils exposed to drought stress, can maintain soil water content and improve plant growth and yield.
- Selenium application can improve yield and yield components under drought stress conditions, and it may be recommended for soils in arid and semiarid regions.

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