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ALLEVIATION OF SALT INJURY BY ADDITION SOIL AND FOLIAR FERTILIZERS IN TERMS OF SOME BIOCHEMICAL AND ANATOMICAL PARAMETERS ON *VICIA FABA* L. PLANTS.

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Abstract

A field experiment was conducted during 2014/2015 growth season to study the effect of three soil fertilizers: control(soil without fertilizers), 200 kg/ha compound fertilizer NPK 18-18-18 and organic (10 ton/ha of sheep manure) and three foliar fertilizers (control without fertilizers, silicon and high potash) singularly and with the combination between them, on alleviation of salt stress injury of broad bean cell membranes which grown in silt-clay soil with 7.8 acidity and 9.4 dS.m^{-1} salinity by estimating biochemical parameters (protein, carbohydrate (CHO), Malondialdehyde (MDA), protease, K^+/Na^+ ratio, Indole acetic acid (IAA), gibberellines (GA3), abscisic acid (ABA)) and anatomical parameters (midrib thickness, xylem vessels diameter and bundle thickness). The results showed that the best fertilizers were (high potash) and the combination (organic + silicon) which improve all parameters mentioned above. In addition the other fertilizers have the ability to eliminate membrane injury partially. .

Key words: Broad bean, Salinity, organic fertilizer, silicon, plant hormones.

Introduction

Salt stress is the major problem on plant product in many countries all over the world ((Munns and Tester 2008)). The fundamental factor for increasing soil salinity is irrigation of plant with saline water, bad cultural practices, and low precipitation. It causes various effects on plant physiology such as increasing respiration rate, ion toxicity, changes in plant growth, mineral distribution, and membrane instability resulting from calcium displacement by sodium, and decreased photosynthetic rate (Hasegawa *et al*., 2000.).

Faba bean (*Vicia faba* L.) is the most important leguminous crops used for human. Exogenous application of fertilizers have been successfully employed to mitigate the salt-induced losses (Ashraf *et al*., 2008). Fertilizers offer the best means of increasing yield and of maintaining soil fertility at a level sufficiently high to ensure that good yields can be obtained consistently, year after year. It has been observed that supplemental silicon improves yield and reduce the plant biotic and abiotic stresses (Epstein, 1999). Possible mechanisms of the Si-mediated alleviation of salt stress in higher plants include: 1. the stimulation of enzymatic and non-enzymatic anti-oxidative defense systems (Hashemi *et al*., 2010), and the reduction of oxidative membrane damage (Gunes *et al*., 2007), 2. the improvement of water uptake via the increasing volume and weight of roots, the prevention of water loss via the reduction of both cuticular and stomatal transpiration, and 3. the reduction of Na^+ uptake (Yin, 2013) with an increasing of K: Na ratio (Tahir *et al*., 2012) and/or the alteration of Na^+ distribution and other ions within plants. The aim of this experiment was to determine the effect of soil and foliar fertilizers on alleviation of salt injury on broad bean plants in terms of enzymatic and non-enzymatic antioxidants.

Materials and Methods

A field experiment was conducted during 2014/2015 growth season to study three treatments of soil fertilizers (control, 200 kg/ha compound fertilizer (NPK, 18-18-18) and organic (10 ton/ha of sheep manure) with three treatments of foliar fertilizers (control, high potash, silicon) in silt-clay soil in which the soil acidity was 7.8 and the salinity was 9.4 dS/m on broad bean (*Vicia faba* L.). Randomized complete block design with three replicates was used. The experimental unit contained 3 ridges (2.4 x 3 m^2) seeded on two sides (25 cm apart) with broad bean seeds (local variety) after soaked in water for 24 hours at 6/10/2014. At seeding time, the organic fertilizer was added according to the treatments as line down the planting line. Two weeks after germination, complete fertilizer (NPK) was added according to the treatment in line 10 cm down the plant line. Foliar fertilizer was added two times, first at one month after germination, and the second at flowering stage. The data were registered during the flowering stage , which included protein (Bishop *et al.,* 1985), carbohydrate **(**Dubois *et al*., 1956), MDA (Heath & Packer, 1968), protease (Donald and Chen, 1965), potassium and sodium concentration (Walsh,1971). phytohormones (Horgan and Smith, 1991) and anatomical parameters (Gerlach, 1977) . The results were analyzed and the means were compared according to Least Significant Difference (LSD$_{0.05}$) (Steel and Torrie, 1981).

Results

Figure (1) demonstrated that almost treatments with soil and foliar fertilizers showed an increase in protein content compared to untreated plants (6.43mg/g F.W.).Whereas, almost treatments showed no significance in its protease activity (Figure 2) except for high pothash which decreased significantly to (71.33U) compared to control plants (116.5 U).

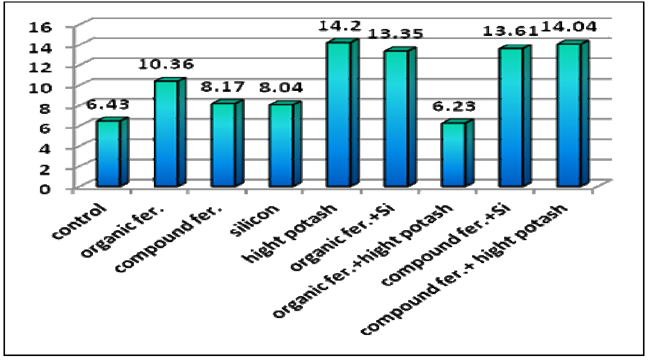

Fig. 1: Effect of soil and foliar fertilizers on protein content (mg/g F.W.). LSD $_{0.05}$= 3.21

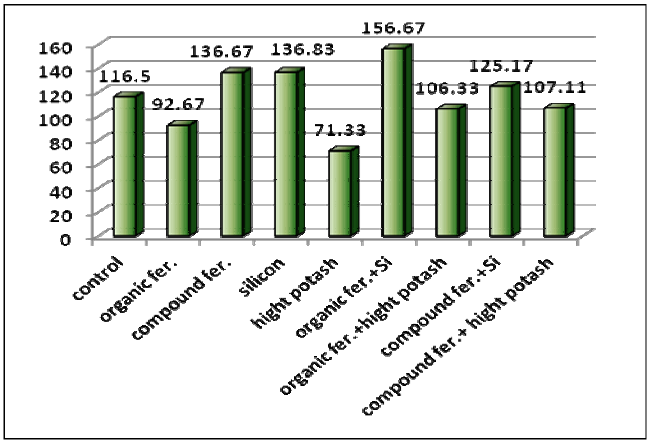

Fig. 2: Effect of soil and foliar fertilizers on protease activity (U) . LSD $_{0.05}$= 26.97

Figure (3) explained that MDA content decreased considerably when the plants treated by all fertilizers which was used. Also, almost treatments caused a significant decrease in carbohydrates content (Figure 4).

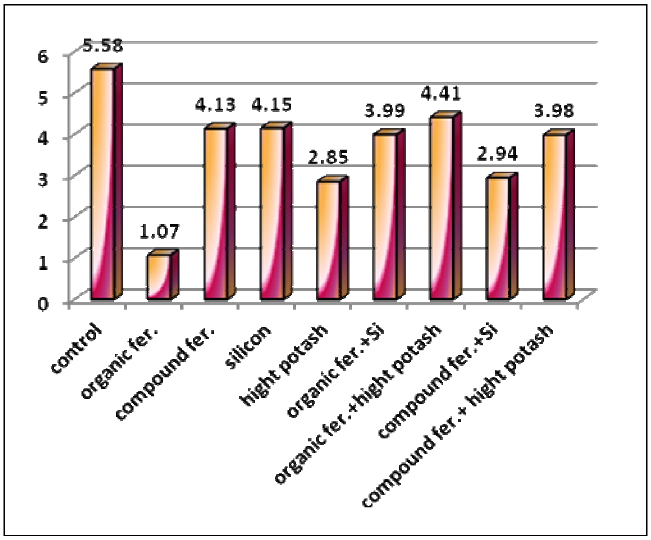

Fig. 3: Effect of soil and foliar fertilizers on MDA (µg/g.F.W). LSD $_{0.05}$= 1.02

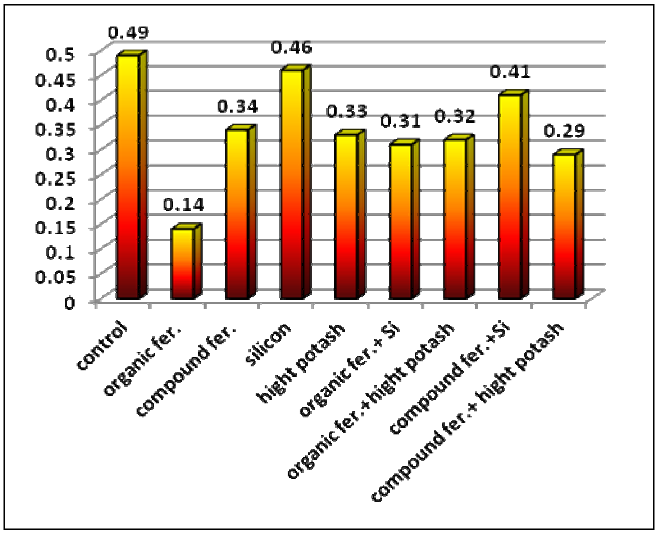

Fig. 4: Effect of soil and foliar fertilizers on CHO (mg/g D.W). LSD $_{0.05}$= 0.11

The influence of soil and foliar fertilizers on phytohrmones (IAA, GA, and ABA) concentrations was observed in figure (5). It has been showed that IAA concentration increased significantly when the plants treated with compound and silicon fertilizers, while, it was decreased significantly in plants treated with the others fertilizers in comparable with the control treatment (3.6 m mM). On the other hand, GA concentration was increased in plants treated with silicon and silicon combined with organic fertilizer, whereas the remaining treatments decreased significantly in comparable with the control treatment (2.6 m mM). in addition, ABA concentration has been decreased significantly in the plants treat with all types of fertilizers compared to control treatment (0.9 m mM).

Figure (6) showed that all fertilizers caused a significant increase in Na^+ concentration in comparable with the control plants (4.13) ppm. On the other hand, almost types of fertilizers showed a significant and non significant decrease of K^+ concentration with exception for compound, silicon and high potash fertilizers which caused a significant increase in K^+ concentration (Figure 7).

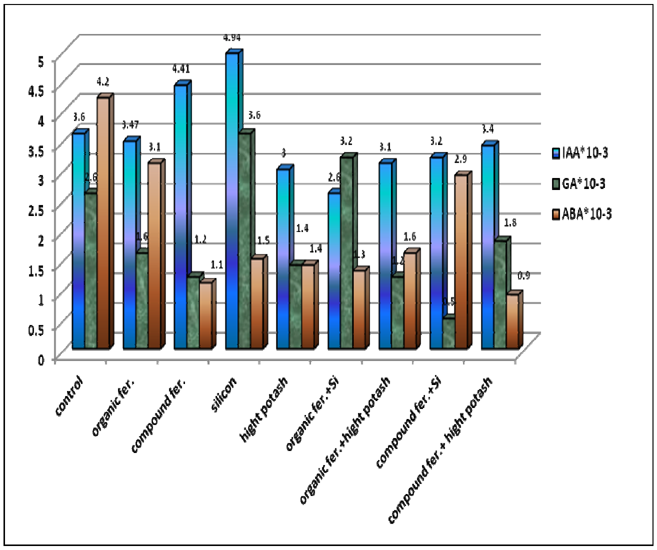

Fig. 5: Effect of soil and foliar fertilizers on IAA,GA and ABA concentrations(mM).

LSD $_{0.05}$ IAA= $0.91*10^{-3}$, LSD $_{0.05}$ GA= $0.066*10^{-3}$, LSD $_{0.05}$ ABA = $0.042*10^{-3}$

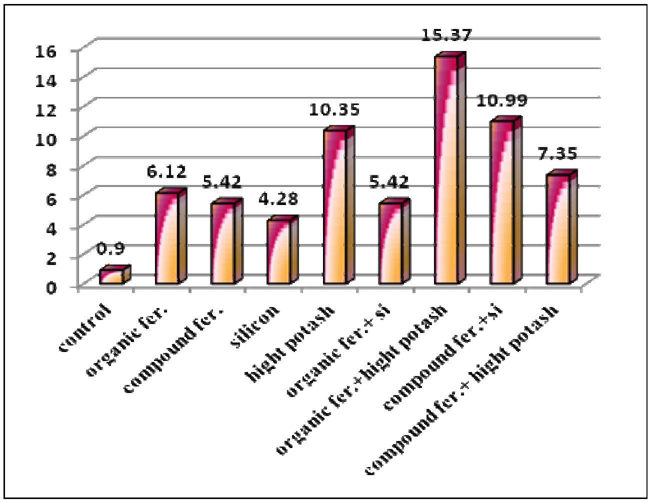

Fig. 6: Effect of soil and foliar fertilizers on K^+/Na^+ ratio. LSD $_{0.05}$= 3.43

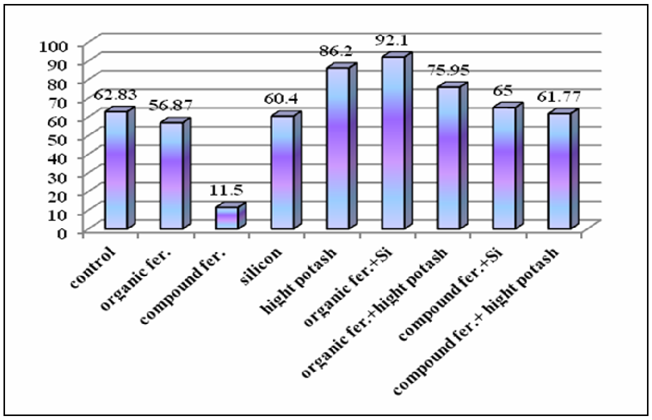

Fig. 7: Effect of soil and foliar fertilizers on bundle thickness (µm). LSD $_{0.05}$= 22.59

Figure (7) showed that compound fertilizer, high potash and organic fertilizer combined with silicon caused a significant increase in bundle thickness. While, midrib thickness increased significantly when the plant treated with high pothash and organic fertilizer combined with silicon there, and no significant effect was observed for the other treatments Figure (8). In addition, xylem vessels diameter (Figure 9) decreased significantly when the plants treated with high potash, organic fertilizer combined with high potash and compound fertilizer combined with silicon.

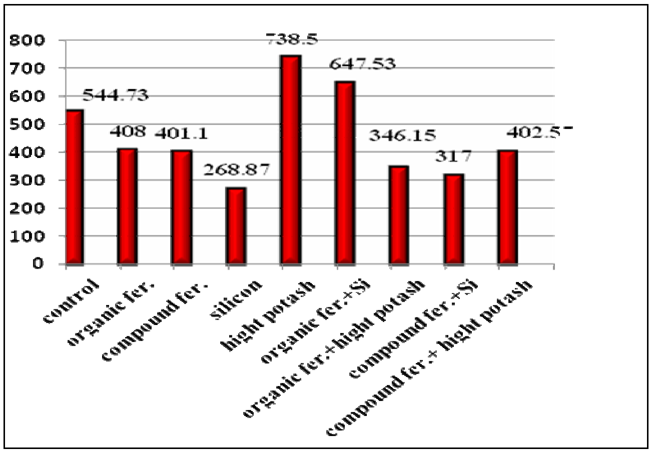

Fig. 8: Effect of soil and foliar fertilizers on midrib thickness (µm) LSD $_{0.05}$= 100.86

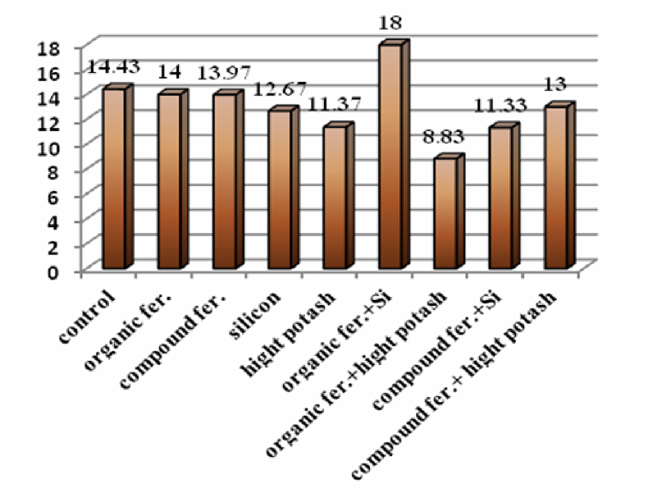

Fig. 9: Effect of soil and foliar fertilizers on xylem vessel diameter (µm) LSD $_{0.05}$= 2.595

Discussion

Many lands around the world have become unfit to agriculture because of essential nutrients lacking, or increase their salinity. So it has been necessary to use the fertilizers to improve plant productivity and increase plants resistant to unfavorable circumstances.

Eight types of fertilizers were used in this research, some of them were soil fertilizers and the others were foliar fertilizers. We found that high potash and organic + silicon fertilizers were the best fertilizers maintaining cell membrane viability during salt stress, by increasing protein content to be 14.2 mg/ g fresh weight (Fig. 1). This finding is compatible with (Jasim *et al*., 2016; Sarwar *et al*., 2018), may be due to the role of K^+ to act as a synergestic effect in N uptake and facilitate protein biosynthesis in addition to its role in increasing ROS scavinger enzyme activity.(Das, 1999; El Sabbagh *et al.,* 2002) causing increase plant growth indicators (Mali *et al*., 2000). Atab *et al.* (2019) showed that *Triticum aestivum* L. treated with high potash had an increased vegetative parameters caused by the effect of K^+ on biochemical and physiological processes and metabolism, by improving membrane stability osmotic adjustment ability (Zhao *et al*., 2016), in addition to K^+ ability to decrease protease activity and the content of MDA (fig. 2,3) respectively. These results may be related to the role of K^+ in decreasing free radicals produced during stress (Johnson *et al*., 2003). Also, both fertilizers have an adequate amount of N which play an important role in increasing cell membrane stability (Premachandra *et al*., 1990). As well as, plants treated with both fertilizers showed a decrease in CHO that used as a potential energy in converting supplied nitrogen to protein as a natural response of the plant (Taiz & Zeiger, 2002). Or, its may be related to the role of K^+ in the regulation pf photoassimilation and translocation processes (Zahoor *et al*., 2017) allowing the plant to do all its vital activity to the fullest by decreasing ABA and IAA concentrations (Fig. 5), its may be related to the role of Si of the second fertilizer on affecting phytohormone homeostasis (Van Bockhaven *et al*., 2013) which lead to decrease ABA concentration (Lee *et al.,*2010). Or, it related to the role of N in decreasing IAA concentration. This is compatible with the finding of (Arigita *et al*., 2005; Wang, 2009). All of these conclusions may leading to a improve stomatal conductance and gas exchange (Borel and Simonneau, 2002; Hejnák, 2011), give rise to increase the efficiency of plants to absorb nutrients, through increasing bundle thickness, midrib thickness and xylem vessels diameter(fig. 7,8,9) respectively. This result is agree with the finding of Fleck et al. (2011) have reported that Si supply enhanced the lignification of sclerenchyma in rice. Therefore it may be affect the stability of higher plants by an inert deposition in lignified cell walls and changing lignin biosynthesis pathway (Marschner, 1995). On the other hand, He *et al*. (2013, 2015) have showed that Si was one of the constituent of the cell walls of suspension-cultured rice cells which can be found as a hemicelluloses bound form, providing mechanical support properties of the cell walls during stresses. Nitrogen as same as silicon caused a thicker, flatter and more turgid leaves, which enhances light absorption while stimulating near-infrared reflection(Yang *et al*., 2011) lead to an increase in photosynthesis efficiency which in turn caused an increase in nutrient uptake , especially K^+ that enhance K/Na ratio (fig. 6) in spite of high Na^+ level. Therefore, high K^+ level accumulation in the cell above a certain threshold due to plant ability to withstand salt stress either by retaining potassium ions or preventing the accumulating of sodium ions because the presence of HKT transporter (high-affinity K^+ transporter) which controlling the transport of Na^+ or Na^+ -K^+ co-transport play a key role in plant Na^+ tolerance mechanisms (Platten *et al*., 2006; Mian *et al*., 2011). HKT represents a basic mechanism in the regulation of Na^+ and K^+ homeostasis, as well as Na^+ exclusion (Byrt *et al*., 2007; Horie *et al*., 2009).

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