Soil supplementation with silicon nanoparticles to alleviate toxicity signs of salinity in strawberry

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Abstract

The current study investigated the efficiency of soil supplementation with Silicon Oxide (SiO2) nanoparticle product (nSi; 20-30 nm; 0, 0.75, and 1.5 gKg-1) or Potassium Silicate (BSi; K2SiO3 as a bulk counterpart) to improve strawberry protection against salinity (NaCl of 2.5 gkg-1). The BSi or nSi utilization not only increased fresh root mass (23%), but also mitigated the inhibitory effects of salinity. The salinity, BSi, or nSi treatments made changes in secondary metabolites confirmed by the differential HPLC chromatograms. Soil supplementation with BSi or nSi induced activity of phenylalanine ammonia-lyase. Likewise, the BSi or nSi treatments enhanced concentrations of phenylpropanoid derivatives, including salicylic acid, ascorbic acid, quercetin, apigenin, caffeic acid, catechin, and chlorogenic acid. The individual salinity treatment caused a severe H_2O_2 accumulation by two folds. However, the BSi or nSi supplementation alleviated the salinity-associated risk of H_2O_2 accumulation. Salt stress caused a drastic increase in lipid peroxidation levels. However, BSi or nSi applications partially relieved the salinity toxicity on membrane integrity. With a similar trend, the BSi or nSi utilization improved the nutritional status of K^+, Na^+, and Ca^{+2} in both leaves and roots. Exposure to BSi, nSi, and/or salinity also enhanced proline concentrations. The BSi or nSi treatments mitigated the salinity-mediated downregulations in photosynthesis performance. Our findings showed that silicon supplements increased salicylic acid (a signaling compound), ascorbate, and quercetin (two vital antioxidants) as a fundamental mechanism.

Keywords: nanoparticle, salicylic acid, silicon, salt stress, secondary metabolites

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Introduction

Silicon (Si) as an essential mineral nutrient is required for different plant species and its classification is a controversial issue. Lsi1 and Lsi2 are respectively the Si-influx and efflux transporters that permit effective transcellular transport of Si in plants (Ma et al., 2007). The Lsi1 gene, a Si-transporter, has a constitutive expression modulated by Si availability (Currie and Perry, 2007). Moreover, Si bioaccumulation in cell wall enhances their physical strength (Epstein, 1999) and is a key mechanism contributing to

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plant resistance against stress condition. Interestingly, at suitable levels, the exogenous Si was shown to enhance growth indexes of *Phragmites australis* (Schaller et al., 2012). Besides, exposure to Si exhibited considerable mitigating effects on the arsenic accumulation in *Solanum lycopersicum* (Marmiroli et al., 2014). Furthermore, the current evidence highlights this hypothesis that Si can trigger mechanisms through which plant growth, anatomy, metabolism, and protection are modified (Moghanloo et al., 2019b; Moghanloo et al., 2019a; Asgari et al., 2018). Furthermore, it has been stated that the manipulation of culture medium by Si may influence cell, tissue, or organ performances (Moghanloo et al., 2019b; Moghanloo et al., 2019a; Sivanesan and Park, 2014). The application of Si-derived fertilizer is considered an eco-friendly way to improve plant growth and protection against different biotic and physicochemical stress conditions (Etesami and Jeong, 2018). Owing to the exclusive physicochemical traits of nanoparticles, the utilization of various nano-based products in diverse industries is rapidly growing (Asgari-Targhi et al., 2018; Babajani et al., 2019). Among these materials are silicon nanoparticles, which are widely used (Moghanloo et al., 2019b; Moghanloo et al., 2019a; Rui et al., 2014). This nano composition is used in various industrial fields such as biosensors, environmental correction, wastewater, agriculture, food processing, and pharmaceutical performance. (Rui et al., 2014). The mesoporous nature of silicon nanoparticles also makes them good candidates as suitable nano carriers for different molecules that may help agriculture. Several studies have shown the importance of silicon nanoparticles in agriculture (Rastogi et al., 2019). Therefore, industrial use of nSi may increase the likelihood of entering plant cells as a food chain primer. It should be noted that its applications in agricultural activities as an insecticide, transfer of specific drugs to the cell, plant nutrition, etc., have received much attention (Moghanloo et al., 2019b; Moghanloo et al., 2019a; Rui et al., 2014). In addition, there are reports of pesticide loss to human health and increased insect resistance (Babajani et al., 2019; Debnath et al., 2011). Interestingly, the nSi application efficiently controlled Sitophilus oryzae, rice weevil (Debnath et al., 2011). Recently, nSi has been shown to mediate changes in growth, biochemistry, gene expression, and anatomy of Astragalus fridae grown under *in vitro* conditions (Moghanloo et al., 2019b; Moghanloo et al., 2019a). Recent findings also indicate that the nSi treatment may efficiently influence nutritional status, enzyme activities, phytohormone balances (Rui et al., 2014), plant structure, tissue differentiation, and cellular ultrastructure (Moghanloo et al., 2019b; Asgari et al., 2018). In fact, various research attempts have been made to exploit the nSi-driven compounds in agricultural activities. However, convincing comparative evidence on the possible benefits or phytotoxicity of nSi on plant growth, physiology, and protection is rare and should be focused further.

Plants encounter diverse environmental stresses, among which salinity is a major limiting factor that severely restricts crop productivity. Salinity stress, commonly provoked by soil high Na^+ ions, causes osmotic stress, disrupts water and nutrient uptake rates and intracellular ionic toxicity, impairs metabolism, and causes oxidative stress to vital intracellular structures and biomolecules (Yang and Guo, 2018). The role of nano-silicon dioxide (nano-SiO2) in plant resistance to salt stress through the improvement of the antioxidant system of squash (*Cucurbita pepo* L. cv. white bush marrow) was studied by Siddiqui et al. (2014), who found that seeds treated with NaCl showed reduced germination percentage, vigor, length, and fresh and dry weights of the roots and shoots. Salinity factor is perceived through the cell wall and increased cytosolic free calcium levels by which specific signaling routes are triggered (Yang and Guo, 2018). Plant acclimation to salt stress leads to activation of multiple signaling cascades and various stress-responsive physiological and molecular traits, especially genes (Gupta and Huang, 2014). Nowadays, scientific efforts have been made to find efficient eco-friendly strategies to reinforce crop productivity and acclimation to salt stress, like different seed priming techniques and utilization of diverse exogenous elicitors, supplements, and chemicals, taking in to account sustainable agriculture (Iranbakhsh et al., 2018; Sheteiwy et al., 2018).

As highlighted above, convincing comparative evidence on the possible roles of nSi on plant growth, physiology, and development is low. Therefore, further studies are required to clarify the exact mechanism through which it may influence plant performance. Moreover, it is well accepted that exogenously applied salicylic acid (SA) may improve plant growth and resistance against unfavorable conditions (Sheteiwy et al., 2018). SA is a multifunctional signaling substance contributing to the activation of stress-responsive genes and defense machinery (Sheteiwy et al., 2018).

In this experiment, we aimed to monitor the possible nSi-mediated differences in the endogenous SA concentration. Taking nanotechnology into account, this study was carried out to explore the potential benefits or phytotoxicity of soil supplementations with bulk Si or nSi on plant growth, physiology, and protection against salinity stress in strawberries.

Materials and Methods

Materials, growth condition, and treatments

Strawberry (*Fragaria × ananassa* Duch. cv. 'Gaviota') plants were grown in a greenhouse condition (Temperature: 20-30/17-19 °C; Relative Humidity: 55-60 %; Light Intensity: 350-400 $\mu Mm^{-2}s^{-1}$). The plants were cultivated in pots containing sandy loam soil supplemented with NH_4NO_3 and KH_2PO_4 at the range of 200 and 62.5 $mgkg^{-1}$ soil, respectively (EC 1.07 dS m^{-1}; pH 7.1). Silicon Oxide (SiO_2) nanoparticles (nSi) were purchased from Iranian Nanomaterials Pioneers, Mashhad, Iran. The physicochemical traits of the nano-product are as follows: purity: 99 %; particle size: 20-30 nm; special surface area: 180-600 m^2/g; color: white; bulk density: <0.10 g/cm^3; true density: 2.4 g/cm^3.

Potassium silicate (K_2SiO_3) was utilized as a bulk control. The strawberry plants were grown in the soil supplemented with the different doses of nSi (0, 0.75, and 1.5 gKg^{-1} soil) or the corresponding concentrations of bulk Si. The NaCl treatment was applied gradually (to avoid an osmotic shock) with the final EC of 5 dS m^{-1}. The effects of different levels of NaCl (ranging from 0, 1, 2, 2.5, 3, 3.5, 4, 5, 6, 7, and 8 gkg^{-1}soil) were evaluated at the pretest stage, among which NaCl of 2.5 g kg^{-1} having about 50 % inhibition was chosen for the main experiment. After 45 days, the plants were exposed to two salinity levels (0 and 2.5 g kg^{-1}). To avoid osmotic shock, NaCl was gradually applied during four days. The treated plants were harvested two weeks after the salinity treatment.

Quantifications of different phenols using HPLC analysis

The leaf extract was prepared using methanol as a solvent and filtered before HPLC analysis. To determine the concentrations of different phenolic metabolites, leaf methanolic extract was subjected to HPLC analysis (Knauer liquid chromatography apparatus; Knauer HPLC instrument; a 1000 Smartline Pump, a 5000 Smartline Manager Solvent Organizer, and a 2800 Smartline Photodiode Array Detector). The separation process was conducted on a 25 cm×4.6 mm Eurospher 100-5 C18 column with a pre-column provided by Knauer (Berlin, Germany). Data acquisition and integration were performed with EZchrom Elite software. The injection of leaf extract into an HPLC column was carried out through a 3900 Smart-line auto sampler injector equipped with a 100 µL loop. Separation was carried out using 0.02 % trifluoroacetic acid in water (elution A) and methanol (elution D) at a flow rate of 0.5 ml min^{-1} and the oven temperature of 20 ℃ under the running time of 55 min. Finally, the concentrations of different phenolic compounds were quantified based on the standard curves of standard compounds. Phenolic content was measured using Folin-Ciocalteu reagent. Gallic acid was used as the standard to draw the calibration curve.

Measurement of Ca, Na, and K contents

The dry-ash method (550 ℃; 8 h) was applied for the determination of minerals, and 0.5 M HCl was used as a solvent to dissolve the ash. The concentrations of Ca, Na, and K were quantified using Inductively-Coupled Plasma-Atomic Emission Spectrometry (ICP-AES; INTEGRA XL2, GBC; Australia).

Enzyme extraction and phenylalanine ammonia-lyase (PAL) activity

The leaves (50 mM) were homogenized in the extraction buffer sodium phosphate (pH 7.0) containing 18 mM β-mercaptoethanol, 2 % (w/v) polyvinyl poly pyrrolidone (PVPP), 2 mM EDTA, and 0.1 % (v/v) Triton X-100. PAL activity was spectrophotometrically estimated at 290 nm based on the formation rate of cinnamic acid according to the method previously explained by (Zucker, 1965).

Hydrogen peroxide (H2O2), Lipid peroxidation, and proline

The homogenized samples in 0.1 % (w/v) TCA were centrifuged. Next, the reaction mixture containing 0.5 ml of the supernatant, 0.5 ml of potassium phosphate buffer (10 mM; pH 7.0), and 1 ml of KI (1 M) were incubated for one h in the dark condition. Then, the H_2O_2 concentrations were spectrophotometrically measured at 390 nm and standard curve (Velikova et al., 2000). The lipid peroxidation rate in the membrane was estimated according to the production levels of malondialdehyde (MDA) in a reaction mixture containing thiobarbituric acid. In addition, proline was extracted by sulfosalicylic acid and its concentration was determined based on the protocol presented elsewhere (Bates et al., 1973).

Photosynthesis efficiency

Fv/Fm (the maximum photosystem II (PSII) photochemical efficiency) and PIabs (the performance index) were quantified using a Packet-PEA chlorophyll fluorimeter (Plant Efficiency Analyzer, Hansatech Instruments Ltd., King's Lynn, Norfolk, PE 32 1JL, England).

Statistical Analysis

The data were subjected to statistical analysis using SPSS (version 16) software. The presented data are the mean and standard error (SE) of three independent replications. Mean separations were done according to Duncan's multiple range test ($P \leq 0.05$).

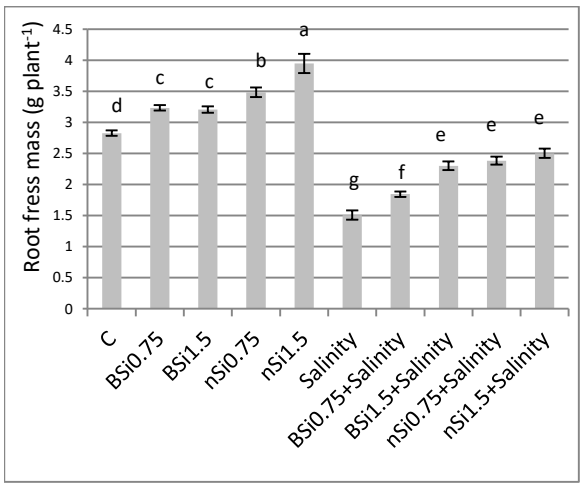

Fig. I. The effects of soil supplementations with different concentrations of BSi or nSi on biomass accumulation in root and post reactions of the supplemented plants to the salinity stress; treatments included C: Control, BSi0.75: Bulk Si of 0.75 gKg^{-1} soil, BSi1.5: Bulk Si of 1.5 g Kg^{-1} soil, nSi 0.75: nano Si of 0.75 gKg^{-1} soil, nSi1.5: nano Si of 1.5 gKg^{-1} soil, Salinity: NaCl of 2.5 g kg^{-1} soil, BSi0.75 + Salinity: Bulk Si of 0.75 g Kg^{-1} soil + NaCl of 2.5 gkg^{-1} soil, BSi1.5 + Salinity: Bulk Si of 1.5 g Kg^{-1} soil + NaCl of 2.5 gkg^{-1} soil, nSi0.75 + Salinity-nano Si of 0.75 g Kg^{-1} soil + NaCl of 2.5 gkg^{-1} soil, nSi1.5 + Salinity: nano Si of 1.5 gKg^{-1} soil + NaCl of 2.5 g kg^{-1} soil

Results

Biomass production

The soil supplementations with different concentrations of BSi or nSi led to a significant increase in fresh root mass by an average of 22.6 % (Fig. I). However, the individual salinity treatment significantly decreased the fresh mass of the root (Fig. I). The BSi/nSi supplements considerably mitigated the salinity-associated risk in the root biomass (Fig. I). Moreover, the nano-form was more effective than the bulk type to reinforce the root system. The same results were also obtained for shoots.

Phenolics

The salinity or Si treatments made changes in secondary metabolites as confirmed by the differential HPLC chromatogram. In non-saline conditions, the applications of Si in both bulk and nano-forms, especially the latter, increased leaf ascorbate content (Fig, IIA). However, individual salinity treatment reduced this property, which was alleviated by the application of Silicon. In both saline and non-saline conditions, the BSi/nSi-

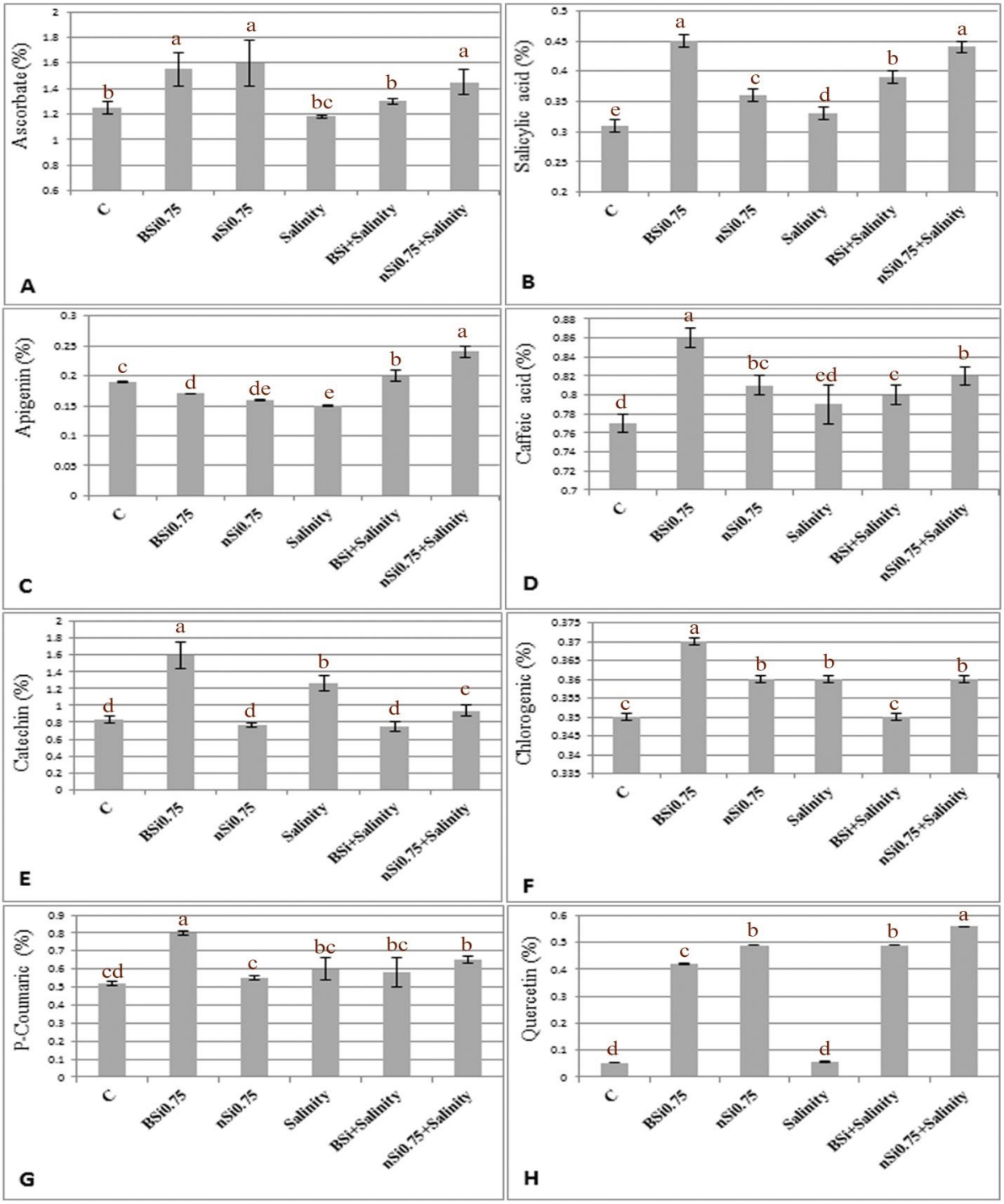

Fig. II. The BSi, nSi, and/or salinity-mediated changes in the concentrations of the various secondary metabolites expressed in percentage (mg per 100 mg fresh mass); Treatments: C: Control, BSi0.75: Bulk Si of 0.75 g Kg^{-1} soil, nSi 0.75: nano Si of 0.75 g Kg^{-1} soil, salinity: NaCl of 2.5 g kg^{-1} soil, BSi0.75 + salinity: bulk Si of 0.75 g Kg^{-1} soil + NaCl of 2.5 g kg^{-1} soil, nSi0.75 + salinity: nano Si of 0.75 g Kg^{-1} soil + NaCl of 2.5 g kg^{-1} soil

treated plants had much more SA by an average of 32% more than the control (Fig. IIB). The nano form was more effective than the bulk form to enhance the apigenin content (Fig. IIC). The applied supplements partly increased the caffeic acid in comparison to the control (Fig. IID). The highest amounts of catechin concentrations were observed in the BSi0.75 and salinity groups (Fig.

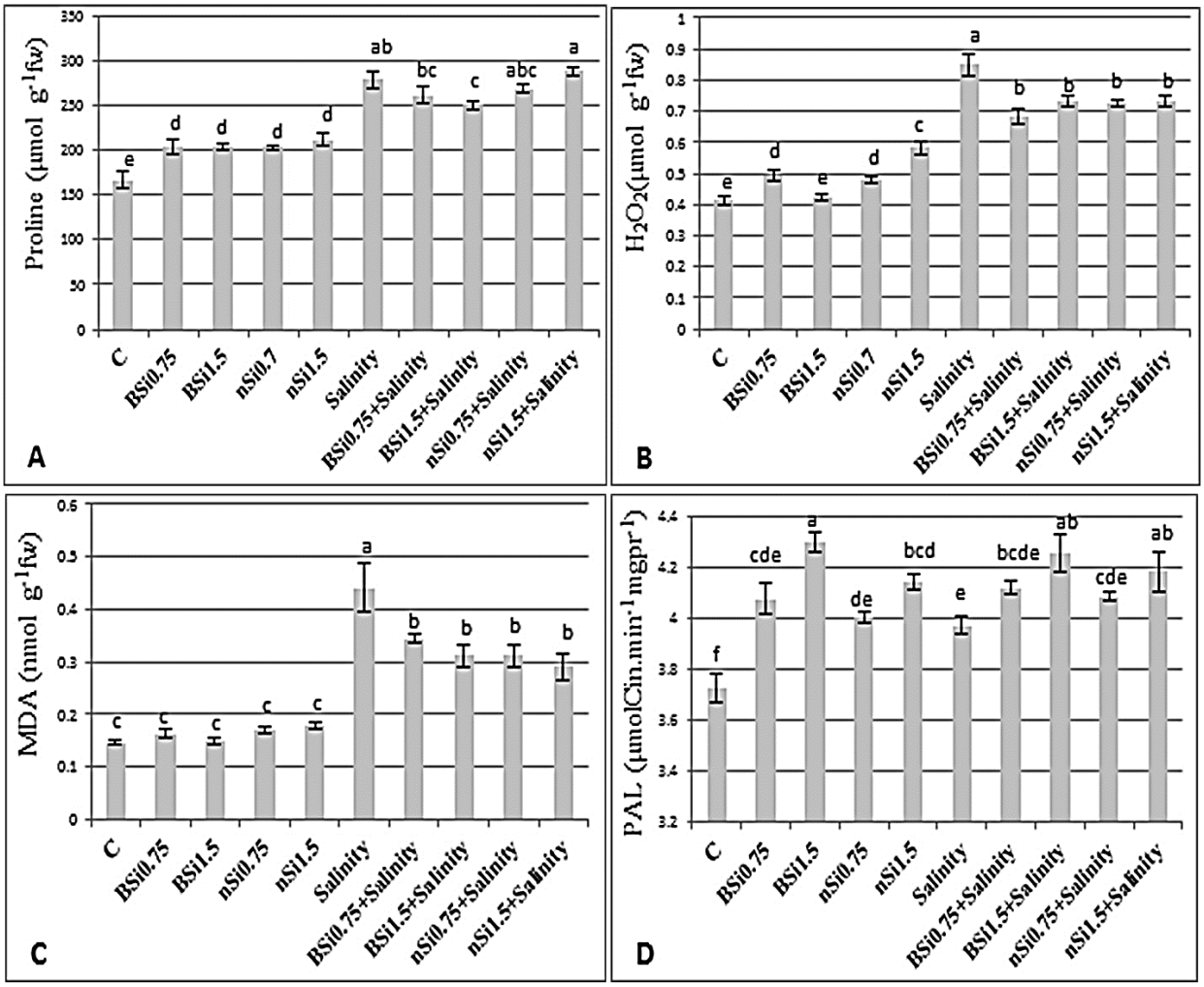

Fig. III. The effects of BSi, nSi, and/or salinity on various biochemical traits of strawberry plants; A: proline contents in leaves, B: H_2O_2 concentrations in leaves, C: Malondialdehyde (MDA) levels in leaves, D: leaf Phenylalanine ammonia lyase (PAL) activity expressed in micro mole cinnamate (Cin) per minute per mg protein; treatments included C: control, BSi0.75: Bulk Si of 0.75 g Kg^{-1} soil, BSi1.5: Bulk Si of 1.5 g Kg^{-1} soil, nSi 0.75: nano Si of 0.75 g Kg^{-1} soil; nSi1.5: nano Si of 1.5 g Kg^{-1} soil, salinity: NaCl of 2.5 g Kg^{-1} soil, BSi0.75 + Salinity: Bulk Si of 0.75 g Kg^{-1} soil + NaCl of 2.5 gKg^{-1} soil, BSi1.5 + salinity: Bulk Si of 1.5 g Kg^{-1} soil + NaCl of 2.5 g Kg^{-1} soil, nSi0.75 + Salinity: nano Si of 0.75 g Kg^{-1} soil + NaCl of 2.5 g Kg^{-1} soil, nSi1.5 + Salinity: nano Si of 1.5 g Kg^{-1} soil + NaCl of 2.5 g Kg^{-1} soil.

IIE). Moreover, slight differences in chlorogenic acid contents were found among the control and the other treatment groups (Fig. IIF). All treatment groups displayed higher concentrations of coumaric acid than the control (Fig. IIG). The BSi or nSi applications in both EC conditions dramatically increased quercetin concentrations by a mean of 9 folds compared to the untreated control groups (Fig. IIH).

Proline, H_2O_2, MDA, and PAL

Under non-saline condition, the proline contents in the BSi or nSi-treated plants were slightly higher than in the control (Fig. IIIA) while the nSi1.5 + salinity group had the highest proline content. The individual salinity treatment caused a severe augmentation in H_2O_2 concentration by two folds. However, the utilization of BSi or nSi significantly alleviated the salinity-associated H_2O_2 accumulations (Fig. IIIB). The individual salinity treatment was also associated with a significant increase in MDA as a membrane damage indicator. Interestingly, the BSi or nSi partially mitigated the salinity-mediated increase in MDA (Fig. IIIC). However, the soil supplementation with BSi or nSi did not significantly differ in MDA levels (Fig. IIIC). All treated groups resulted in a slightly higher PAL activity by an average of 10.7 % (Fig. IIID).

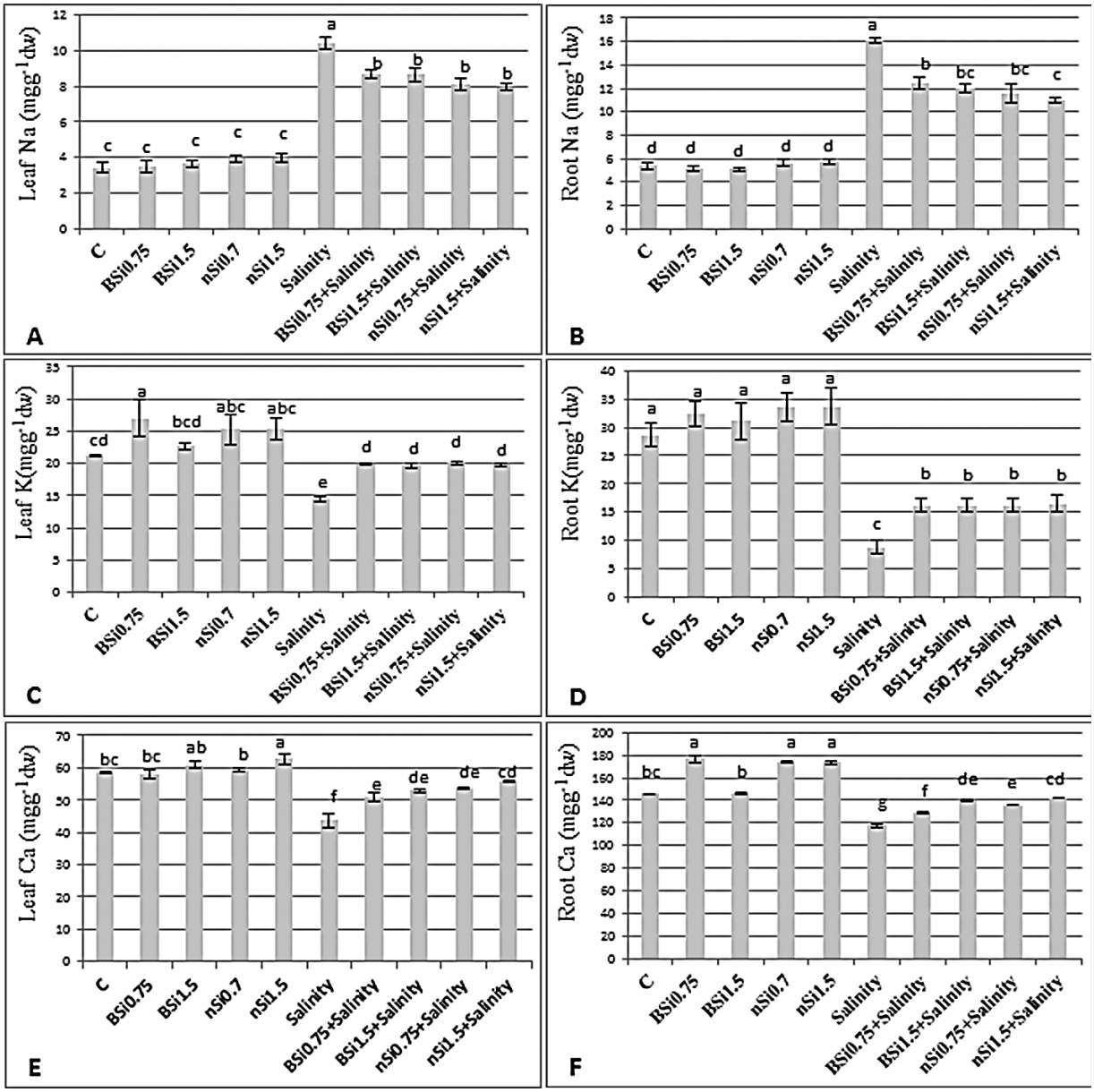

Fig. IV. The differential nutritional status in response to the soil supplementations with bulk Si, nSi, and/or salinity treatments; A: Na^+ concentrations in leaves, B: Na^+ concentrations in roots, C: K^+ concentrations in leaves, D: K^+ concentrations in roots, E: Ca^{+2} concentrations in leaves, F: Ca^{+2} concentrations in roots; Treatments included C: control, BSi0.75: bulk Si of 0.75 gKg^{-1} soil, BSi1.5: Bulk Si of 1.5 gKg^{-1} soil, nSi 0.75: nano Si of 0.75 g Kg^{-1} soil, nSi1.5: nano Si of 1.5 g Kg^{-1} soil, salinity: NaCl of 2.5 g Kg^{-1} soil, BSi0.75 + Salinity: Bulk Si of 0.75 g Kg^{-1} soil + NaCl of 2.5 g Kg^{-1} soil; BSi1.5 + Salinity: Bulk Si of 1.5 g Kg^{-1} soil + NaCl of 2.5 g Kg^{-1} soil, nSi0.75 + Salinity: nano Si of 0.75 g Kg^{-1} soil + NaCl of 2.5 g Kg^{-1} soil, nSi1.5 + salinity: nano Si of 1.5 g Kg^{-1} soil + NaCl of 2.5 gKg^{-1} soil.

Nutritional status

Salinity treatments enhanced Na levels in both roots and leaves (Fig. IV A, B). However, applying BSi or nSi partially declined Na accumulations (Fig. IV A, B). Similarly, supplementation of the soil with BSi or nSi improved K contents compared to the control (Fig. IVC). In saline conditions, the BSi or nSi- treated plants had higher K, and Ca contents compared with the salinity control in both roots and leaves (Fig. IV C, D, E, F).

Photosynthesis performance

Maximum photochemical efficiency of PSII (Fv/Fm) was evaluated to estimate the extent of photo-inhibition phenomenon (Table 1). The NaCl treatment was associated with a slight decrease in

Table 1
The effects of different treatments on the two traits related to photosynthesis performance, including Fv/Fm (the maximum photosystem II (PSII) photochemical efficiency) and PIabs (the performance index)

Treatments	Fv/Fm	PIabs
C	0.8±0.04[a]	2.2±0.5[c]
BSi 0.75	0.79±0.03[a]	2±0.41[c]
BSi 1.5	0.8±0.03[a]	4.6±1.02[b]
nSi 0.75	0.81±0.02[a]	6.5±1.5[a]
nSi 1.5	0.81±0.03[a]	5.2±0.96[b]
Salinity	0.76±0.01[a]	0.84±0.1[d]
BSi 0.75+Salinity	0.77±0.05[a]	1.7±0.2[cd]
BSi 1.5+Salinity	0.76±0.04[a]	1.9±0.32[cd]
nSi 0.75+Salinity	0.78±.0.03[a]	2.3±0.6[c]
nSi 1.5+Salinity	0.77±0.05[a]	2.5±0.51[c]

Mean values of the same category followed by different letters are significant at p≤0.05 level.

the maximal quantum efficiency of PSII (Fv/Fm). The performance index (PIabs) parameter showed a significant reduction under salt stress. However, the salinity-mediated decrease in the performance index was mitigated by the applying Si (Table 1). Interestingly, BSi and nSi-supplemented plants showed higher PIabs under non-saline conditions (Table 1).

Discussion

Biomass and nutritional status

Salinity inhibits growth, elongation, and division of cells and results in cell death, through changes in water status in plants. Although Na concentration increased in roots as a result of salinity stress, applying silica nanoparticles reduced Na concentration in plant tissues. Salinity stress interferes with plant growth due to the reduction of osmotic potential and toxicity of Na ion. Silica nanoparticles reduced Na toxicity by reducing Na absorption, resulting in improved plant growth (Kalteh et al., 2014). The soil supplementation with BSi or nSi not only improved biomass accumulation in roots, but also mitigated the inhibitory effects of salinity stress. Moreover, the nano form was more efficient than the bulk one to increase biomass accumulation. This finding is consistent with some other recent reports (Moghanloo et al., 2019b; Moghanloo et al., 2019a). It has been stated that nanoparticles can cause drastic changes in plant cells differently from their bulk counterparts owing to their unique traits (Moghanloo et al., 2019b; Moghanloo et al., 2019a; Asgari-Targhi et al., 2018; Babajani et al., 2019). Also, nSi treatments were associated with no signs of toxicity. The results are consistent with the findings of Kim et al. (2014) in rice. In line with our findings, the Si supplementation exhibited a protecting role in cucumber under salinity via mediating modifications of the root system, stimulations in water uptake, and reductions in ion toxicity (Wang et al., 2015). The Si-induced stimulation in the root system has been attributed to the enhanced root elongation due to the increased cell wall extensibility (Etesami and Jeong, 2018). Contrary to these findings, the nSi exposure in cotton exhibited phytotoxicity and diminished biomass accumulation in both roots and shoots (Rui et al., 2014). In this experiment, the plants' nutritional status was also modified by the applied supplements. The nSi-modified nutrition may be regarded as a critical mechanism through which nSi may affect plant growth, physiology, and protection. NSi treatment altered Cu, Mg, and Na concentrations in cotton (Rui et al., 2014). In addition, Si supplementation changed the arsenic accumulation in tomato fruits (Marmiroli et al., 2014). Moreover, nSi presence in the culture medium also reinforced the differentiation pattern of xylem and phloem conducting tissues, thereby stimulating growth rates in Astragalus fridae (Moghanloo et al., 2019b; Moghanloo et al., 2019a). Furthermore, Si treatments improved photosynthetic performance. Therefore, nSi-mediated increases in concentrations of certain essential nutrients and photosynthesis performance are critical

mechanisms by which nSi may modify both growth and metabolism. Besides, the nSi-mediated alteration in SA may be responsible for the observed differences. In plants, SA is involved in regulating growth, nutrition, metabolism, and defense system (Sheteiwy et al., 2018). Furthermore, nSi in a concentration-dependent manner changed levels of abscisic acid and auxin as two critical phytohormones (Rui et al., 2014).

PAL and phenylpropanoid derivatives

Applying BSi and nSi in the dose and type-dependent manners induced PAL activity and accordingly led to the differential phenylpropanoid derivatives, especially SA and quercetin, and ascorbate. The involvements of SA (a signaling and hormone-like agent) and ascorbate (an essential vital non-enzymatic antioxidant) in plant protection against various biotic and abiotic stress conditions are well documented (Babajani et al., 2018; Iranbakhsh et al., 2018; Sheteiwy et al., 2018). Modifications in secondary metabolites may efficiently contribute to the mitigation of the risks associated with the salinity stress. In line with our findings, exposure to Si in soybean resulted in increases in SA level while reducing Jasmonate contents (Hamayun et al., 2010). It is well illustrated that SA-triggered signaling contributes to the regulation of stress-responsive genes (Sheteiwy et al., 2018). Moreover, nSi application in culture medium also upregulated expressions of PAL and universal stress protein (USP) genes in Astragalus fridae (Moghanloo et al., 2019b). Furthermore, Si treatment enhanced abscisic acid concentration in wheat under drought (Pei et al., 2010). It has been hypothesized that long-term exposure to Si can modulate hormonal status and antioxidant system (Kim et al., 2014).

Proline, H_2O_2, and lipid peroxidation

The individual salinity treatment intensively increased H_2O_2 accumulation and accordingly reduced membrane integrity while the BSi or nSi supplementations considerably mitigated these signs of salinity toxicity. It seems that these supplements mediated upregulation in the antioxidant system. Our results showed that Si application was associated with stimulation of secondary metabolism and increased production of secondary metabolites, especially ascorbate and quercetin antioxidants, the key mechanisms through which the Si utilization may improve plant protection against stress conditions. In line with our finding, Si in rice improved protection against salt stress by inducing antioxidant enzymes, diminishing H_2O_2, and decreasing salt ion accumulation, protecting photosynthetic pigments (Lekklar et al., 2018). Proline content increased under salinity stress which was a response to stress. Moreover, proline increased by silicon nanoparticles which were due to tolerance induction in plant; Treatments with silicon nanoparticles reduced the pollution effects originated from salinity in strawberries. In plant cells, proline plays multi-taxed roles through which this amino acid contributes to the regulations of cellular growth, nitrogen metabolism, osmotic pressure, protein structures, and dissipation of excess energy during unfavorable environmental conditions (Nazerieh et al., 2018).

Photosynthesis performance

The PIabs severely declined compared with Fv/Fm in response to the salinity. It could be concluded that PIabs were much more sensitive to stress than the Fv/Fm ratio. Applying nSi alleviated the inhibiting role of salinity on photosynthesis performance. These results exhibited that downregulation of photochemical activity during exposure to salinity is functionally modified by the nSi application. Our results are in line with Wang et al. (2012) who reported that drought stress in *Vitis amurensis* has relatively little effect on Fv/Fm while PIABS was more sensitive to stress in drought conditions.

Conclusion

Taken collectively, the soil supplementations with BSi or nSi exhibited a significant efficiency to improve plant protection against salinity conditions. Comparative data on nSi and its bulk counterpart suggested that the protection efficiency of nano-form was partially more than bulk type, mainly due to its unique physicochemical characteristics. Moreover, the BSi or nSi considerably mitigated salinity-

associated risks of H_2O_2 accumulation and lipid peroxidation, implying the improved function of antioxidant machinery. Furthermore, the applied supplements relieved the salinity-mediated downregulation of photosynthesis performance and nutritional status. Both Si types stimulated the production of phenolic secondary metabolites, among which SA, ascorbate, and quercetin are of critical importance. Our findings underline the BSi/nSi-mediated increases in endogenous SA (a signaling compound), ascorbate, and quercetin (two vital antioxidants) as underlying mechanisms.

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