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EFFECT OF EXOGENOUS APPLICATION OF SALICYLIC ACID AND SODIUM NITROPRUSSIDE ON MAIZE UNDER SELENIUM STRESS

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Selenium (Se) is a naturally occurring substance that is toxic at high concentration for all life forms. It enters into environment through natural and anthropogenic sources. Therefore, this experiment was conducted to study the effect of foliar application of salicylic acid (SA) and sodium nitroprusside (SNP) on maize growing in selenium contaminated soil. Salicylic acid and sodium nitroprusside are important growth controller molecules which regulate physiological functions in plants and protect plants from environmental stresses. Two maize varieties (Pioneer 33H15 and Cargill 6142) were used in this experiment. Two concentrations of SA and SNP (50µM and 100µM) were used as a foliar spray and selenium was applied @ 20 mg kg^{-1}. Results showed that selenium stress caused a reduction in plant growth, chlorophyll content, gas exchange parameters and activities of enzymes such as SOD, POD and CAT. Though, foliar application of SA and SNP considerably reduces the negative impacts of selenium by enhancing the activities of antioxidant enzymes as they play an important role in modulating the cell redox balance, thereby protecting plants against oxidative damage. There was significant improvement in membrane stability index (68.33%), relative water contents (78.47%) along with increase in activities of SOD (23%), POD (16.37%) and CAT (70%) in Pioneer 33H15 where SA was applied @ 100µM. These results suggested that foliar application of SA might be more useful for improving maize growth by alleviating harmful effects of Se.

Keywords: Heavy metals, mechanical waste, abiotic stress, selenium, maize, enzymes.

INTRODUCTION

With the development of industry, there has been an impressive increment in the release of mechanical waste to the environment, mostly soil and water, which has prompted the accretion of heavy metals, especially in urban regions (Dixit *et al.*, 2015). These metals also initiate severe health problems due to transference of these pollutants into the food chain. Due to extreme utilization of chemicals and varying environmental conditions; heavy metals are being accrued in the soil and cause serious health problems and danger to human life (Jeelani *et al.*, 2017; Ahsan *et al*., 2018a,b; Sattar *et al*., 2019).
Selenium (Se) is normally present in nature that is randomly dispersed in the outer layer of the earth (0.05 mg kg^{-1}) (Kabata-Pendias and Szteke, 2015). In agriculture, Se is utilized as an expansion, mostly as sodium selenite to bug sprays, composts and foliar showers. Moreover, several livestock foodstuffs are encouraged with this element. It is comparatively communal of different makeups and medicines, as a tonic (Kabata-Pendias, 2011). Due to its semi-conductive possessions, selenium is extensively used in manufacturing electrical things. It is also released from metallurgical processes and largely used in glass manufacturing, dyes, greasing oil, dyestuffs, food supplements, farmed products etc. (Bodnar *et al.*, 2012; Mehdi *et al.*, 2013).
Metal stress is one of the serious problems throughout the world, affecting the metabolic responses of crops, human health and the environment (Deinlein *et al*., 2014). Selenium at higher levels is viewed as harmful to the plants (Germ *et al*., 2007). Hence, in the plants developing in soils having poisonous convergences of Se, different impacts, for example, chlorosis, senescence, diminished development, and potential yield (Zhang *et al*., 2007) have been accounted for.
A range of chemicals is presently utilized on plants existing in stress environments, and in many cases, remarkable results have appeared (Farooq *et al.*, 2009). Utilization of plant growth regulators (PGRs) is a favourable method to expand the resistance against abiotic stresses (Ali *et al*., 2013). Without a doubt, plant weight increases due to various PGRs (El-Tayeb, 2005).
Salicylic acid (SA) is a growth controller of phenolic nature, which play a role in plant protection from environmental hassle (Karlidag *et al.*, 2009). It enhances germination, development, gas exchange parameters and particle take-up (He *et al*., 2010). Moreover, it is currently certain that the SA gives security against various abiotic stresses (Karlidag *et al*., 2009, El-Tayeb, 2005). SA has been accounted for to

give assurance against metal pressure, for example, against mercury, cadmium (Zhou *et al.,* 2009; Noriega *et al.,* 2012) and copper (El-Tayeb, 2006).

Nitric oxide (NO) which is assumed as a vital part in numerous physiological procedures in plants, for example, development, improvement, senescence and versatile reactions to different stresses (Kazemi *et al.,* 2010). Exogenously provided NO has been shown to give a protection in response to metal stress (Sun *et al.,* 2014; Srivastava and Dubey, 2012). Nitric oxide role in plants is extremely varied due to its contribution in various processes (Ruan *et al.,* 2004). Foliar application of nitric oxide has been accounted to enhance development. In addition, it improves the chlorophyll contents (Ruan *et al.,* 2004) and when it applies exogenously prevent plants from oxidative destruction by upgrading the ability of enzymes (Tuncz-Ozdemir *et al.,* 2009).

The higher concentration of Se in ground water may affect the food chain through the mechanism of soil–plant water relations (Hartikainen, 2005; Dhillon *et al*., 2010; Guerrero *et al*., 2014). Se transport in maize resulted though many transporters like Sulphur and affect the plant growth (Garousi *et al.,* 2016). Selenium is known as micronutrient with vital physical functions. It takes part in the growth of plants at lower concentrations but harmful at higher concentration. Maize (*Zea mays* L.) has been selected for this research work because it is top-ranked food crops in the world it is important for human diet and is also a rich source of raw material for industry (Jiang *et al.,* 2017). This experiment was conducted with the objective to study the possible role of exogenous Salicylic Acid (SA) and Sodium nitropruside (SNP) in the modulation of the antioxidant defense system against Se stress in maize.

MATERIALS AND METHODS

***Growth conditions and treatments*:** An experiment was led in the wirehouse of the ISES, SARC, UAF. Two maize (*Zea mays* L.) varieties including Pioneer 33H15 and Cargill 6142 were selected for this experiment. The pots were lined with a plastic sheet to avoid leakage and filled with soil @ 12 kg soil per pot and spiked with Se (20 mg kg^{-1}). Five seeds of both maize varieties were sown in each pot. Two plants from each pot were thinned; three plants were maintained in each pot. The salicylic acid (SA) and sodium nitroprusside (SNP-donor of nitric oxide) were applied as foliar spray one month after sowing using common insecticide sprayer. The sample collection was made 15 days after the treatment application. The treatment plan for the both maize varieties was: T_1 (Control, Se @ 20 mg kg^{-1}), T_2 (Se @ 20 mg kg^{-1} + SA 50µM), T_3 (Se @ 20 mg kg^{-1} + SA 100µM), T_4 (Se @ 20 mg kg^{-1} + SNP 50µM) and T_5 (Se @ 20 mg kg^{-1} + SNP 100µM).

***Determination of chlorophyll contents and gas exchange parameters*:** SPAD chlorophyll meter (Minlota, Japan) was used to measure chlorophyll contents of maize leaves. Gas exchange parameters were recorded before harvesting. The data with respect to photosynthetic rate (A), stomatal conductance (gs) and rate of transpiration (E) were measured on a fully expanded younger leaf by utilizing convenient Infrared Gas Analyzer (IRGA).

***Determination of growth parameters*:** After 2 weeks of SA and SNP application, plants were separated into roots and shoots, and fresh weights were recorded promptly after harvest. The length of shoots and roots was also recorded by using a meter rod. Subsequently, taking the fresh weight of shoots and roots the samples were air dried, then kept in an oven at 70 ºC till constant weight and then the dry weight of root and shoot (g plant^{-1}) were measured.

***Determination of relative water content* (%):** Relative water contents measured by weighing fresh leaves (FW), plunged in deionized water for twelve hours then weight fully turgid leaf (TW) and then dry at 65°C temperature in the oven to take the dry weight of leaves for 48 hours (Sairam *et al.,* 2002).

$$\mathrm{RWC} = [(\mathrm{FW} - \mathrm{DW})/(\mathrm{TW} - \mathrm{DW})]$$

***Determination of membrane stability index*:** To measure membrane stability index of maize plants, leaf samples (0.1 g) were retained in 10 ml of distilled water in two sets. One set was placed at 40°C for 30 minutes and its electrical conductivity (C1) was measured electrical conductivity (C2) of second set was also measured by placing it in a boiling water bath (100°C) for 15 minutes (Sairam *et al.*, 2002).

$$\mathrm{MSI} = [1 - (\mathrm{C_1}/\mathrm{C_2})] \times 100$$

***Determination of antioxidant activity*:** The activities of antioxidant enzymes were determined by taking fresh leaf samples (0.5 g). Leaf sample was grounded by utilizing a grinder in 5 ml of 50 mM cold phosphate buffer (pH 7.8) kept in an ice bath. The subsequent homogenate was centrifuged at 15000 x g for 20 minutes at 4 ºC temperature. Antioxidant enzymes were determined by utilizing the supernatant. SOD activity was measured by its capacity to hinder the photo-reduction of nitro blue tetrazolium (Giannopolitis and Ries, 1977). Peroxidase and catalase were also measured by using the method given by Aebi (1984).

***Determination of selenium and micronutrient concentration*:** Dried plant samples were crushed to make powder by using a mechanical grinder. The grounded shoot and root samples were digested with HNO_3:$HClO_4$ with 2:1 proportion (method 54a of U.S. Salinity Lab staff, 1954). The samples were retained overnight after adding 1g of plant sample and 10 ml di-acid mixture in a conical flask. In order to get transparent material flasks were warmed on a hot plate (Ryan *et al.,* 2001). After processing, samples were cooled, and volume was made 25 mL by adding distilled water and stored in airtight bottles. The prepared samples were run on AAS for the detection of selenium, iron, manganese and zinc.

***Statistical analysis*:** Results presented in this experiment were average of 3 replicates. Statistical software package "Statistics 8.1" was used for interpreting the results. A completely randomized design with the factorial arrangement was used following Steel *et al.* (1997).

RESULTS

***Effect of SA and SNP on plant growth under selenium stress*:** Different concentrations of SA and SNP effected plant height, shoot fresh and dry weight, root fresh and dry weight and root length (Table 1). Highest shoot fresh weight (78.3 g) was recorded where SA was applied @ 100µM and the lowest (30.67 g) was recorded in control. Highest shoot fresh weight (47%) in Pioneer 33H15 was obtained where the SA was applied @ 100µM as compared to the control plants where no treatment was applied. Shoot fresh weight was higher in Pioneer 33H15 than Cargill 6142. A Similar trend was also noticed in shoot dry weight where the application of SA @ 100µM showed the highest increase (31%) as compared to control.

In case of shoot length maximum increase in shoot length (33%) of Pioneer 33H15 was recorded where SA was applied @ 100µM as compared to control plants followed by Cargill 6142 which showed (26%) increase in shoot length at the same treatment level. Similarly, maximum root length 36 and 34%, respectively were observed in Pioneer 33H15 and Cargill 6142 at the same treatment level (SA @ 100µM) as compared to control plants where no treatments were applied. The root, fresh and dry weights were also enhanced with the application of SA @ 100µM as compared to control (Table 1). The highest increase in the root, fresh weight was (50%) while the highest increase in root dry weight was (43.5%) over control. Results further presented that use of salicylic acid considerably improves the lenience capability of the plants in all the above-mentioned parameters. Pioneer 33H15 showed the best performance in all treatments under selenium stress as compared to Cargill 6142.

***Effect of SA and SNP on chlorophyll contents and gas exchange parameters in maize under selenium stress*:** All gas exchange parameters reduced under selenium stress, but the application of SA and SNP improves these parameters. Maximum improvement in photosynthetic rate (A), transpiration rate and stomatal conductance (gs) observed in Pioneer 33H15 which were 66, 47 and 54% respectively where SA was applied @ 100µM. Whereas in Cargill 6142 the more reduction in gas exchange parameters was recorded in control plants where only selenium was applied (Fig.1A, B and C). In case of chlorophyll contents results indicated that the highest increase in chlorophyll contents (52%) in Pioneer 33H15 was observed where SA applied @ 100µM as compared to control plants where no treatment was applied (Fig. 1.D).

***Effect of SA and SNP on relative water contents (%) and membrane stability index in maize under selenium stress*:** Reduction in relative water contents and membrane stability index was observed in control plants treated with selenium 20 mg kg^{-1}. Maximum membrane stability index (MSI) (68.33) and relative water contents (RWC) (78.47%) was observed in Pioneer 33H15 where SA was applied @ 100µM (Table 2). Plants growing under selenium stress showed significant improvement in membrane stability index and relative water contents when SA applied as compare to SNP. More improvement was observed at higher level of SA (100µM) and the level of improvement is better in Pioneer 33H15 then Cargill 6142.

***Effect of SA and SNP on selenium concentration and uptake of micronutrients in maize under selenium stress*:** Results revealed that treatments affected shoot and root concentrations of iron (Fe), manganese (Mn) and zinc (Zn). The maximum Fe concentration (1.94 mg kg^{-1} was recorded where SA @ 100 µM was applied in Pioneer 33H15,

Table 1. Effect of SA and SNP on plant growth parameters under selenium stress

Maize Genotypes	Treatments	Shoot fresh weight (g)	Shoot dry weight (g)	Root fresh weight (g)	Root dry weight (g)	Shoot length (cm)	Root length (cm)
Pioneer 33H15	T_1	41.3±0.8e	8.70±0.24e	12.0±0.57cd	1.86±0.10f	69.0±1.15cd	45.0±1.52e
	T_2	57.3±2.3d	10.90±0.18c	17.6±2.60b	2.42±0.04cd	77.3±1.20ab	55.3±2.33bcd
	T_3	78.3±0.8a	12.60±0.07a	24.0±0.57a	3.30±0.01a	81.3±0.88a	70.7±1.20a
	T_4	66.3±1.4bc	10.20±0.37d	15.7±0.66bc	2.27±0.08de	73.67±2.40bc	50.6±0.88cde
	T_5	71.3±3.7b	11.90±0.21b	18.0±0.57ab	2.53±0.06c	75.0±2.08abc	57.7±1.45bc
Cargill 6142	T_1	33.0±0.6g	7.24±0.08h	9.0±0.57e	1.62±0.04g	60.0±1.85e	40.0±0.57f
	T_2	49.3±1.7e	8.00±0.12fg	13.3±0.88cd	2.27±0.08de	65.6±2.60de	53.6±3.28cde
	T_3	58.0±0.6cd	8.48±0.25ef	17.0±0.57ab	2.87±0.04b	69.3±2.02cd	61.0±1.52b
	T_4	37.6±0.3f	7.64±0.17gh	11.0±0.57de	2.13±0.08e	63.3±3.52de	47.0±2.08e
	T_5	48.6±2.7e	8.27±0.01ef	13.0±0.58cd	2.41±0.05cd	65.3±3.53de	54.3±0.67bc

Each value is an average of three replications ± S.E, Different letters (a-g) showed significant difference among treatments was determined by LSD test (P<0.05). T_1 (Control, Se @ 20 mg kg^{-1}), T_2 (Se @ 20 mg kg^{-1} + SA 50µM), T_3 (Se @ 20 mg kg^{-1} + SA 100µM), T_4 (Se @ 20 mg kg^{-1} + SNP 50µM) and T_5 (Se @ 20 mg kg^{-1} + SNP 100µM).

however the minimum Fe concentration (1.34 mg kg^{-1}) was noticed in control plants in Cargill 6142. In case of root Fe concentration minimum Fe concentration (1.64 mg kg^{-1}) was in roots of control plants in Cargill 6142 and maximum Fe concentration (2.64 mg kg^{-1}) was recorded where SA was applied @ 100 µM in Pioneer 33H15 (Fig. 2A).

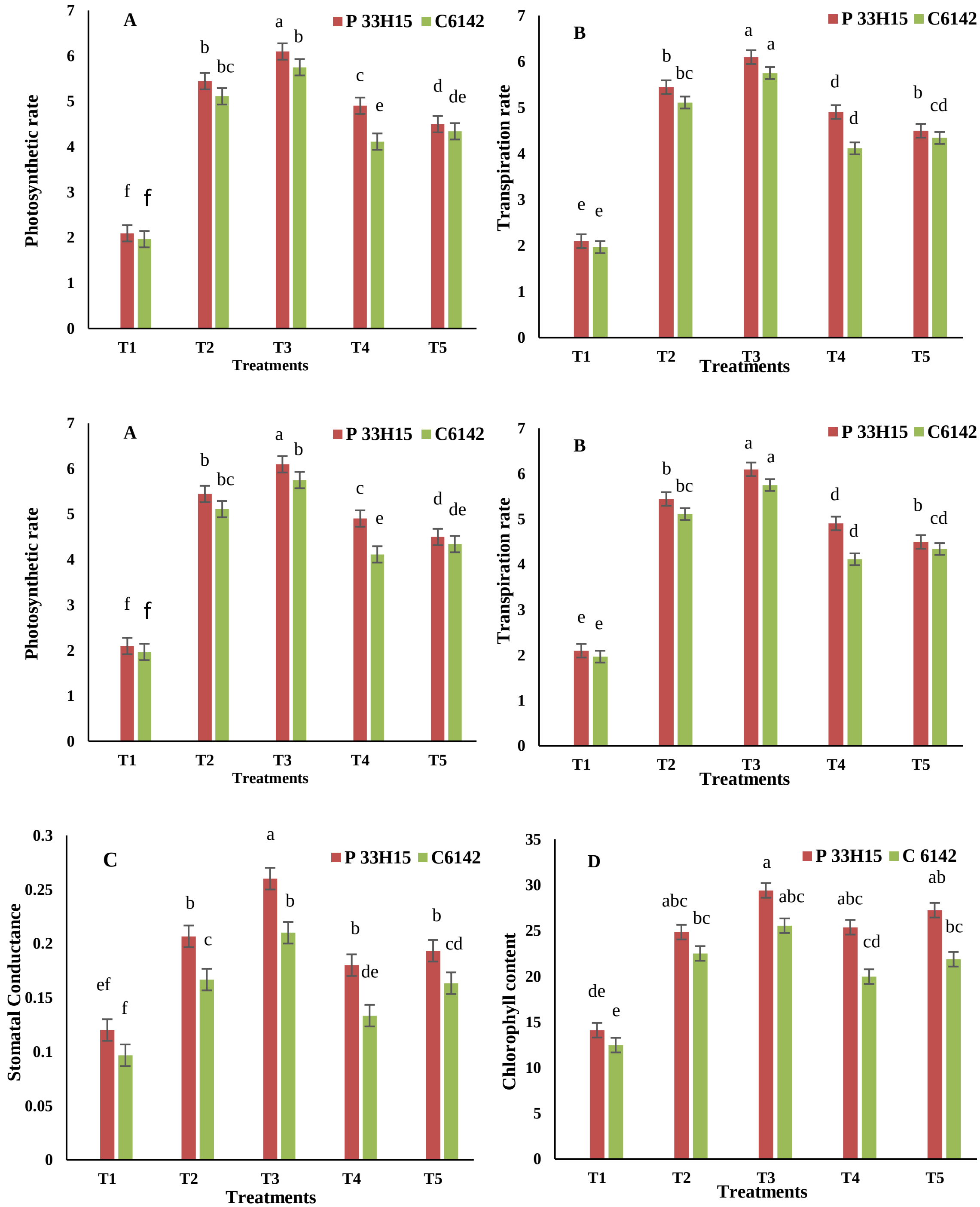

Figure 1. Effect of SA and SNP application on Photosynthetic rate (A), Transpiration rate (B), Stomatal conductance (C) and Chlorophyll contents (D) of maize leaves under selenium stress. T_1 (Control, Se @ 20 mg kg^{-1}), T_2 (Se @ 20 mg kg^{-1} + SA 50µM), T_3 (Se @ 20 mg kg^{-1} + SA 100µM), T_4 (Se @ 20 mg kg^{-1} + SNP 50µM) and T_5 (Se @ 20 mg kg^{-1} + SNP 100µM).

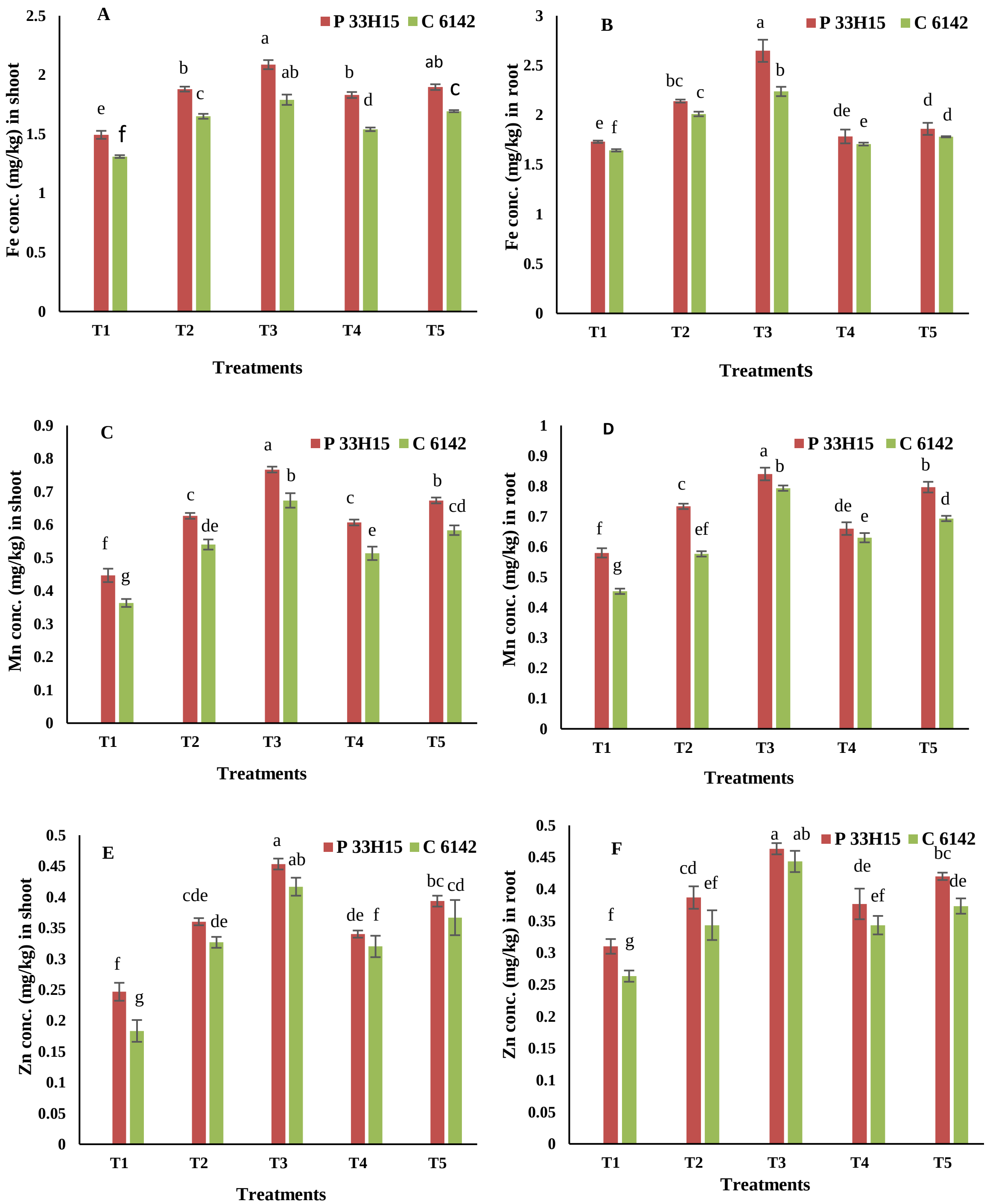

Figure 2. Effect of SA and SNP application on uptake of Fe (A, B), Mn (C, D), and Zn (E, F) in maize shoots and roots under selenium stress. T_1 (Control, Se @ 20 mg kg^{-1}), T_2 (Se @ 20 mg kg^{-1} + SA 50µM), T_3 (Se @ 20 mg kg^{-1} + SA 100µM), T_4 (Se @ 20 mg kg^{-1} + SNP 50µM) and T_5 (Se @ 20 mg kg^{-1} + SNP 100µM).

The results of Mn and Zn analyzed in shoots and roots of both maize genotypes are shown in (Fig. 2B and 2C). Maximum reduction in Mn and Zn concentration of maize shoots were recorded with control plants in Cargill 6142. It is observed in roots that maximum Mn concentration (0.84 mg kg^{-1}) was noted where SA @ 100 µM was applied in Pioneer 33H15, while the minimum Mn concentration (0.45 mg kg^{-1}) was recorded with control plants in Cargill 6142. The highest shoot Zn concentration (0.45 mg kg^{-1}) was also noted where SA @ 100µM was applied in Pioneer 33H15 and the lowest Zn concentration (0.18 mg kg^{-1}) was noted in control plants in Cargill 6142. Highest concentration of Zn was observed in roots then shoots. Data indicated that treatment T_3 was significantly different with control and all the other treatments.

Table 2. Effect of SA and SNP on relative water contents (%) and membrane stability index of maize under selenium stress

Maize Genotypes	Treatments	Relative Water Contents (%)	Membrane Stability Index
Pioneer 33H15	T_1	46.77±1.74g	38.67±1.45g
	T_2	59.76±1.25cd	52.33±3.48cd
	T_3	78.47±1.62a	68.33±1.76a
	T_4	54.68±1.45de	45.00±1.53ef
	T_5	61.14±1.24c	55.67±1.86bc
Cargill 6142	T_1	40.68±1.86h	31.67±1.45h
	T_2	52.90±1.59ef	48.00±1.73de
	T_3	68.56±1.90b	60.00±2.65b
	T_4	48.72±2.14fg	40.67±0.88fg
	T_5	56.90±2.45cde	50.33±1.86cde

Each value is an average of three replications ± S.E, Different letters (a-h) showed significant difference among treatments was determined by LSD test (P<0.05). T_1 (Control, Se@20 mg kg^{-1}), T_2 (Se @ 20 mg kg^{-1} + SA 50µM), T_3 (Se @ 20 mg kg^{-1} + SA 100µM), T_4 (Se @ 20 mg kg^{-1} + SNP 50µM) and T_5 (Se @ 20 mg kg^{-1} + SNP 100µM).

The maximum shoot Se concentration (6.47 mg kg^{-1}) was recorded where SA @ 100µM was applied in Pioneer 33H15 and the minimum Se concentration (6.14 mg kg^{-1}) was recorded with control plants in Cargill 6142. Same trend was also observed in roots of maize genotypes where maximum Se concentration (7.18 mg kg^{-1}) was observed in roots of Pioneer 33H15 where SA was applied @ 100µM while the minimum Se concentration (6.49 mg kg^{-1}) was observed in control plants in Cargill 6142 (Table 3).

Table 3. Effect of SA and SNP on selenium concentration in shoot and root

Maize Genotypes	Treatments	Se in shoot (mg/kg)	Se in root (mg/kg)
Pioneer 33H15	T_1	6.24±0.02def	6.60±0.04g
	T_2	6.34±0.01b	6.98±0.02cd
	T_3	6.47±0.01a	7.18±0.03a
	T_4	6.29±0.03bcd	6.94±0.03de
	T_5	6.41±0.02a	7.09±0.02b
Cargill 6142	T_1	6.14±0.01g	6.49±0.01h
	T_2	6.22±0.01ef	6.92±0.02de
	T_3	6.33±0.02bc	7.04±0.03bc
	T_4	6.18±0.01fg	6.78±0.03f
	T_5	6.27±0.03cde	6.98±0.02cd

Each value is an average of three replications ± S.E, Different letters (a-g) showed significant difference among treatments was determined by LSD test (P<0.05). T_1 (Control, Se @ 20 mg kg^{-1}), T_2 (Se @ 20 mg kg^{-1} + SA 50µM), T_3 (Se @ 20 mg kg^{-1} + SA 100µM), T_4 (Se @ 20 mg kg^{-1} + SNP 50µM) and T_5 (Se @ 20 mg kg^{-1} + SNP 100µM).

***Effect of SA and SNP on antioxidant activity in maize*:** Considerable reduction in the activities of antioxidant enzymes was noticed under selenium stress, but salicylic acid and nitric oxide enhance the antioxidant activity (Table 4). The activity of superoxide dismutase (SOD) was considerably improved in Pioneer 33H15 as compare to Cargill 6142 where salicylic acid was applied @ 100 µM. Peroxidase (POD) and catalase (CAT) activities are also

Table 4. Effect of SA and SNP on superoxide Dismutase (SOD), Peroxidase (POD) and Catalase (CAT) of maize under selenium stress

Maize Genotypes	Treatments	SOD (unit mg^{-1} of protein)	POD (unit mg^{-1} of protein)	CAT (unit mg^{-1} of protein)
Pioneer 33H15	T_1	117.93±2.87gh	113.54±2.14de	12.39±0.25f
	T_2	137.58±2.55cd	121.49±3.15bc	25.41±0.54c
	T_3	153.10±3.93a	135.77±1.55a	40.73±1.56a
	T_4	127.07±1.97ef	117.65±2.74cd	18.80±0.90e
	T_5	133.29±1.14de	127.21±4.49ab	23.33±0.81cd
Cargill 6142	T_1	114.71±1.30h	102.14±1.93f	11.85±0.17f
	T_2	141.78±1.21bc	114.85±1.24cde	22.76±0.73d
	T_3	146.88±2.84ab	121.66±0.64bc	32.62±0.70b
	T_4	124.96±2.53fg	109.71±4.43e	17.94±0.80e
	T_5	130.22±1.65ef	117.70±2.81cd	21.66±1.25d

Each value is an average of three replications ± S.E, Different letters (a-h) showed significant difference among treatments was determined by LSD test (P<0.05). T_1 (Control, Se @ 20 mg kg^{-1}), T_2 (Se @ 20 mg kg^{-1} + SA 50µM), T_3 (Se @ 20 mg kg^{-1} + SA 100µM), T_4 (Se @ 20 mg kg^{-1} + SNP 50µM) and T_5 (Se @ 20 mg kg^{-1} + SNP 100µM).

markedly improved where salicylic acid was used. Activities of SOD, POD and CAT significantly decreased in control plant treated with 20 mg kg^{-1} selenium, in both maize genotypes but more reduction was observed in Cargill 6142. The results revealed that application of salicylic acid (SA) shows more improvement in enzymes activities as compare to sodium nitroprusside (SNP) under selenium stress. The highest improvement in activities of SOD (23%), POD (16.37%), and CAT (70%) was recorded in Pioneer 33H15 where salicylic acid was applied @ 100μM.

DISCUSSION

Harmful metals that cause contamination of the biosphere is a broad environmental issue and among these metals, for example, lead, arsenic, selenium, cadmium, copper, mercury, and nickel cause's severe health problems as they enter the food chain (Jamal *et al.*, 2013). Selenium at greater concentrations is thought to be lethal for plants (Germ *et al.*, 2007). In this experiment excess, selenium deferred the growth of plants and related findings were depicted on various plants (Simojoki, 2003).

Salicylic acid (SA) and Sodium nitroprusside (SNP) are signaling molecule, which assumes an imperative part in numerous biological procedures in plants, for example, development, senescence and versatile reactions to different stresses (Kazemi *et al.*, 2010). In present study application of SA and SNP improved the growth of plant like RL, SL and biomass maximum improvement observed in Pioneer 33H15 especially at 100 μM SA level as compared to control plants. This growth, improvement by SA could be auxin assisted (Shakirova *et al.*, 2003).

Growth improving effects of SA have been reported in wheat (Shakirova *et al.*, 2003), maize (Gunes *et al.*, 2007) and (Gill *et al.*, 2016) in *Brassica napus*. Correspondingly it is also described that sodium nitroprusside is active in improving RDW in maize and enzyme activities in the roots of yellow lupin in severe conditions (Zhang *et al.*, 2006).

In this experiment, selenium stress considerably decreased the photosynthetic rate (A), transpiration rate (E) and stomatal conductance (gs) in both genotypes. But foliar application of SA and SNP improved these photosynthetic parameters as compared to control plants, especially at 100 μM SA level. It has been recommended that the developmental impacts of salicylic acid could be identified with changes in the hormonal status (Abreu and Munne-Bosch, 2009) or due to changes in photosynthetic processes (Stevens *et al.*, 2006). It also enhances the activities of antioxidant enzymes under selenium stress as they play an important role in modulating the cell redox balance, thereby protecting plants against oxidative damage (Song *et al.*, 2017)

Decreasing chlorophyll contents of both maize genotypes leave in control plants was noted might be related to the aggregate action of chlorophyll debasing proteins and the destruction in the structure of the chloroplast. Related findings were described for maize (Abbasi *et al.*, 2014). These outcomes are in accordance with past looks into that an expansion in transpiration rate and stomatal conductance because of foliar use of SA in corn and soybean (Khan *et al.*, 2003) and a foliar splash of SNP upgrades chlorophyll contents (Ruan *et al.*, 2004). In another investigation completed on soybean, foliar utilization of salicylic acid upgraded the water utilize proficiency, transpiration rate and interior CO_2 fixation (Khan *et al.*, 2003). Foliar application of salicylic acid to Se-treated plants, the membrane damage was decreased, which was also improved the relative water content and chlorophyll content.

High selenium concentrations in plants compete with other micronutrients particularly Mn, Zn and Fe and disturb their uptake (Pazurkiewicz-Kocot *et al.*, 2008). In this experiment it is observed that high selenium reduces the uptake of micronutrients whereas improvement was observed in the uptake of micronutrient with foliar application of SA and SNP. Similar result was also presented by Gunes *et al.* (2007) who found that application of SA improves the uptake of micronutrients under stress conditions in maize plants.

In the present experiment, we observed that utilization of sodium nitroprusside (nitric oxide donor) and salicylic acid were found to be active in the reclamation of reduced plant growth by improving antioxidant activity. Additionally, the activities of SOD, POD and CAT reestablished and presented an astounding improvement when plants were treated with SA @ 100 μM under selenium stress. Control plants treated with Se, 20 mg kg^{-1} showed the considerable decrease in enzyme activity, whereas the foliar salicylic acid application, the manifestation of antioxidants was improved. These results are similar to the findings of Simaei *et al.* (2012) who found that salicylic acid and nitric oxide decrease the damaging effects by enhancing the activity of the antioxidative system.

***Conclusion*:** The results of this study demonstrated that the toxic effects of selenium may happen due to intensification in oxidative stress. Exogenous application of salicylic acid and sodium nitroprusside countered these toxic effects by improving photosynthetic parameters, relative water contents, and reducing the oxidative damage. Taken together, it is revealed that salicylic acid and sodium nitroprusside has decreased the selenium initiated oxidative stress and efficiently regulated the antioxidant enzymes. Salicylic acid improves the selenium concentration in shoots and furthermore overwhelmed the deficiency of micronutrients instigated by selenium stress in maize genotypes so the explained investigation of SA signaling might be more useful as compared to SNP in developing the selenium resistant crops and Pioneer 33H15 performed the best towards foliar application of SA and SNP under selenium stress.

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