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ORIGINAL PAPER

Silicon Alleviates Arsenic Toxicity in Maize Seedlings by Regulating Physiological and Antioxidant Defense Mechanisms

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Abstract

Arsenic (As) is a toxic heavy metal, found abundant in the environment, especially in industrial waste and pesticide formulations. The present study was conducted to evaluate the role of silicon (Si) application and its mechanism in the alleviation of toxic effects of As in maize (*Zea mays* L.) seedlings. The treatments consisted of four levels of As (0, 4, 8, and 12 mg kg^{-1} of soil) factorially combined with three levels of Si (0, 50, and 100 mg kg^{-1}) applied in the earthen pots placed in a triplicate completely randomized design. Results have shown a significant correlation of morphological and biochemical traits, which depicts that accumulation of Si-enhanced antioxidant defense mechanism which alleviates the As toxicity. Application of Si improved plant growth and gas exchange attributes by reducing the accumulation of As in plant tissues and reactive oxygen species production and by improving the membrane stability (lower malondialdehyde), synthesis of chlorophyll pigments, antioxidant enzymatic activities (superoxide dismutase, peroxidase, catalase), and proline accumulation. Application of Si at 100 mg kg^{-1} of soil may be proved best for sustainable maize production under As toxicity. This will not only improve plant growth and development but also have positive impact on the environment.

Keywords Arsenic toxicity · Environmental pollution · Superoxide dismutase · Peroxidase · Malondialdehyde · Chlorophyll pigments

1 Introduction

Presence of toxic elements in the soil hinders biological and productive efficiency of different crops. Among these toxic elements, arsenic (As) is non-essential and generally highly toxic for both plants and animals. Increased levels of As can cause various health disorders including, gastric, hepatic, cardiac, respiratory, and reproductive and some time may also cause stillbirth and infant death (Chikkanna et al. 2019; Ali et al. 2021). As is mostly contained in domestic and industrial sewage sludge in much higher quantities than any other toxic elements or metalloids. Arsenic enters soil deliberately or through sewage sludge and fertilizer application (Chikkanna et al. 2019). The bioavailability of arsenic in the soil at any stage depends upon several soil characteristics, soil solution characteristics, and their interactions with other toxic elements (Daiet al. 2018; Suriyagoda et al. 2018a and b). In southern countries and South East Asia mining processes, deliberate use of arsenical pesticides and irrigation with As-contaminated ground water eventually produces residual As accumulation in soils (Rahman and Singh 2019; Mapa 2020). The occurrence of As in soils or its presence in irrigation water leads arsenic accumulation in crop plants. In crop plants, arsenic is mostly accumulated in roots where it hinders root elongation making its proliferation lesser than as compared to upper-ground parts of the plant and produces functional changes and damages of crop plants (Abedi and Mojiri 2020), and produces less crop productivity due to arsenic accumulation (Rosas-Castor et al. 2014).

Maize (*Zea mays* L.) is the 3rd staple food of the world after wheat and rice. Therefore, heavy metal contaminations, especially As contamination, also threaten human health due

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to the possibility of its translocation in the grains which has already been reported in cereal crops (Suriyagoda et al. 2018a and b; Bianucci et al. 2020). Translocation of As in the plant cell occurs through phosphate transporters and aquaporins (Kertulis-Tartar et al. 2009; Farooq et al. 2016) and thus disrupts energy production and nutrient uptake and imbalances the plant metabolism and physiology. There is a lot of evidence that showed an increase in the production of reactive oxygen species (ROS) in response to As toxicity (Tripathi et al. 2016; Suriyagoda et al. 2018a and b) resulting in the damage of photosynthetic apparatus, RUBISCO enzyme, and carbohydrate metabolism (Farooq et al. 2016) and triggers antioxidant defense mechanism. This results in the reduction of cell expansion and biomass accumulation. Most of the plant tissues may lead to death due to interference of arsenic accumulation in critical metabolic processes. Mostly, plants have a mechanism to accumulate higher quantities of arsenic in the roots (Sun et al. 2018; Torres et al. 2020); however, depending upon genotypic variation, some proportion of arsenic is translocated to the shoots and other parts of the plants.

Silicon is the most abundant element of the earth's crust after oxygen and its share is around 28%. Silicon (Si) has been found to have an important role to ameliorate the negative effect of abiotic stresses like drought, salinity, and heavy metals in plants (Malhotra and Kapoor 2019). But its mechanism of action to alleviate the stresses differ from species to species due to change in metal uptake mechanism, translocation, and its binding to cells (Ma et al. 2008; Suriyagoda et al. 2018a and b). Antagonistic effect of Si and As has been reported in literature (Ma et al. 2008; Bogdan and Schenk 2009; Ning et al. 2016). Ma et al. (2008) found that arsenic movement and accumulation in rice roots share the same pathways (OsNIP2;1) as Si. Furthermore, Si has also been found to change the soluble and exchangeable fractions of different heavy metals including As into stable forms by various chemical modifications (Ning et al. 2016). It is found that As concentration in rice decreased when Si concentration in the soil increased (Bogdan and Schenk 2009) which is attributed to the low expression on Si transporter genes (i.e., Lsi1 and Lsi2) which also mediate the As uptake (Bienert et al. 2008). In plants, metal toxicity may be ameliorated through application of Si as Si alters translocation pathway of heavy metals in plants (Ning et al. 2016; Farooq et al. 2016). Recent studies revealed that in wheat and alfalfa, Cd toxicity can be alleviated through Si (1 mM) application (Greger et al. 2016). Therefore, Si emerged as a tonic either in the form of a soil fertilizer or foliar applicant to reduce the heavy metal toxicity in plants and for environmental sustainability.

Physiological and biochemical mechanisms are involved in adaptation of plants to the heavy metal–contaminated soils. At the onset of heavy metal stress, reactive oxygen species produced which triggers antioxidant defense mechanism triggers the production of different enzymes like catalase (CAT), peroxidase (POD), superoxide dismutase (SOD), glutathione reductase (GR), and ascorbate peroxidase (APX) (Suriyagoda et al. 2018a and b; Tripathi et al. 2016). However, a detailed study of the interactive effects of Si during arsenic toxicity conditions and how Si offsets the physiological and biochemical changes under arsenic toxicity is scarce. Furthermore, studies revealing information regarding the regulatory functions of Si in ROS metabolism under arsenic toxicity by activating antioxidant defense mechanism in crop plants, especially in maize, are also limited. Considering the above facts, major objectives of the study were to explore the potential of Si in ameliorating the arsenic toxicity through morphological, physiological, and antioxidant defense mechanisms in maize seedlings grown in arsenic-contaminated soil.

2 Materials and Methods

2.1 Growth Conditions and Treatments

A pot experiment was conducted in a greenhouse at the College of Agriculture, BZU, Sub-Campus Layyah, Pakistan, during autumn season 2017 to evaluate the role of silicon application in mitigating the adverse effect of As toxicity in maize seedlings. Seeds of maize hybrid FMC-2011 were sown in each pot (75 cm in height, 18 cm in diameter) containing 12 kg of well-ground soil passed after sieved. The nitrogen (N), phosphorus (P), and potassium (K) fertilizers were applied at the rates of 100, 75, and 60 mg kg^{-1} of soil, respectively, to sustain the emergence of maize seedlings. Average temperature during the experiment was 35/26 (maximum/minimum) and humidity 60%. The sources of fertilizers applied were urea, di-ammonium phosphate, and potassium sulfate. All the fertilizers were thoroughly mixed. The treatments comprised two factors (a) As concentrations (0, 4, 8, and 12 mg kg^{-1}) and (b) Si levels (0, 50, and 100 mg kg^{-1}), which were used in experimentation. The Ck served as a control treatment. Arsenic was added as $NaAsO_2$ and Si as sodium silicate. After stand establishment (15 days after sowing) of maize seedlings, As concentrations and Si treatments were applied in each pot. Maize seedlings were then grown for 15 days. Each treatment was performed in triplicate using three different replicates and contained five equal-sized seedlings. An experiment was arranged in a complete randomized design (CRD) with three replications where four levels of As were factorially combined with three levels of Si.

2.2 Growth Parameters

After 15 days, the data was calculated for shoot and root lengths and fresh plant biomass. All the samples were collected, and the maize seedlings were cut just above the soil surface and the roots were removed from the soil. The roots and shoots samples were oven-dried at 75 °C until constant weight is gained.

2.3 Chlorophyll Contents

With 5 ml 80% acetone, 0.5 g of fresh leaves was extracted overnight at 4 °C. For 5 min, the leaf extracts were then centrifuged at 10,000×*g*. For calculation of chlorophyll *a* and chlorophyll *b*, the method followed was by Arnon (1949). The supernatant absorbance was noted at 645-nm wavelengths by means of spectrophotometry.

2.4 Physiological Attributes

With the help of an infra-red gas analyzer (IRGA, model Li-6400, Li-Cor), the variables of photosynthetic gas exchange like photosynthetic rate (*Pn*), transpiration rate (*E*), and stomatal conductance (*g*) were measured. For this persistence, from each plant, mature leaves were selected and sited inside the IRGA. During daytime between 10:00 and 12:00 am, all the values of physiological parameters were recorded (Zekri et al. 1991).

2.5 Biochemical Analysis

For the extraction of enzymatic antioxidants from fresh plant samples, 5 ml of 50 mM buffer (pH 7.8) of phosphate solution was used and was then centrifuged for 20 min at 10000×*g*. The superoxide dismutase (SOD) action was determined at 560 nm with the help of the method by Giannopolitis and Ries (1977) and catalase (CAT) action was examined at 240 nm by means of the method practiced by Chance and Maehly (1955).

As a result of photochemical reduction, superoxide dismutase (SOD) action was measured at 560 nm by the inhibition of nitro blue tetrazolium (NBT). The reaction mixture was comprised of 60 μL enzyme extract, 500 μL methionine (14 mM), 1.5 mL NBT (50 mM), 500 μL EDTA (74 mM), 1.5 mL riboflavin (1.4 μM), and 960 μL (50 mM) phosphate buffer. Under illuminations of a 40-W fluorescent lamp, the reaction was started by reaction mixture. The reaction was stopped after passing 5 min of the lamp being turned off. Blue-colored formazan was made through an NBT reaction that was further employed at getting absorbance at 560 nm. The same solution with no enzyme extract was run as a blank. The absorbance of catalase (CAT) action was noted at 240-nm wavelength at a spectrophotometer and the change was measured by peroxide H_2O_2 produced as a consequence of enzyme action. Into the reaction mixture, 1000 μL enzyme extract was added to start the reaction. The superoxide dismutase (SOD) action was estimated by noting the decline at 425-nm absorbance which was because of H_2O_2-dependent oxidation of pyrogallol. The measurement of H_2O_2 and MDA content was determined according to Tripathi et al. (2012). Plant material was digested in nitric and hydrochloric acids as per the 3051A Method described by USEPA (1998). An atomic absorption spectrophotometer coupled with a hydride generator was used to determine the As contents from the digested material. For the determination of Si, the plant material was digested with a mixture of hydrogen peroxide (H_2O_2) and sodium hydroxide (NaOH) in an autoclave by following (Korndörfer et al. 2004).

2.6 Statistical Analysis

All the data of the experiment was analyzed with Fisher's analysis of variance technique using the Statistix 8.1 software (Analytical Software, 2105 Miller Landing Rd., Tallahassee, FL 32312) and the average of treatments was computed by LSD test (Steel et al. 1997). Association between different treatments was measured with the help of correlation analysis function of Microsoft Excel v. 365.

3 Results

3.1 Morphological Attributes

The As stress significantly ($p < 0.05$) reduced the root dry weight, shoot dry weight, root length, and shoot length as compared with the non-stressed controls (Fig. 1). However, under 50 mg kg^{-1} of Si treatment applied in combination with different As stress concentrations (0, 4, 8, and 12 mg kg^{-1}), the plant showed a significant increase in root length (25.0, 43.4, and 54.7%) and root dry weight (30.1, 30.4, and 50.3%) compared with the As-stressed plants without Si application. Plants stressed with different As concentrations along with 100 mg kg^{-1} of Si application also showed a distinct increase in root length (23.0, 37.6, 52.8%) and root dry weight (29.8, 18.1, and 38.8%) in comparison with the As-stressed plants without Si application. Similarly, a significant increase was noted in shoot length (15.8, 27.8, and 34.9%) and shoot dry weight (17.7, 23.1, and 33.7%) under 50 mg kg^{-1} of Si supplementation in combination with different As treatments, as compared with As-stressed plants (Fig. 1). Furthermore, a significant increase was observed in shoot length (15.0, 24.5, and 34.3) and shoot dry weight (14.7, 20, and 29.4%) of plants treated with Si 100 mg kg^{-1} along with various As concentrations, while both Si supplementations in As-stressed and non-stressed plants exhibited an increase in morphological attributes.

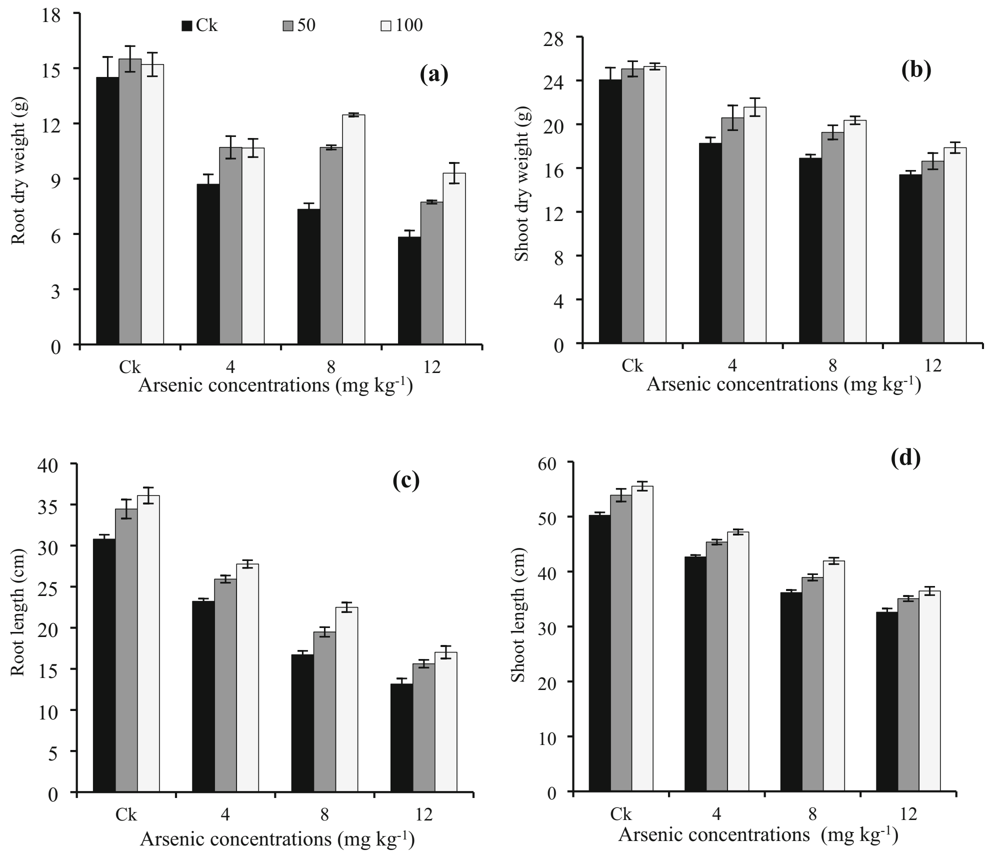

Fig. 1 Influence of silicon supplementation (i.e., Ck; control, 50, 100 mg kg^{-1}) on root dry weight (a), shoot dry weight (b), root length (c), and shoot length (d) of maize seedlings under various level of arsenic treatments (i.e., Ck; control, 4, 8, 12 mg kg^{-1}). Values are the mean SE of four replicates. Bars marked with different letters are significantly different by LSD ($p \leq 0.05$)

3.2 Photosynthetic Pigments

The plants under As stress significantly ($p < 0.05$) reduced the chlorophyll *a* and *b* synthesis as compared with the non-stressed controls (Fig. 2). Both Si treatments in combination with the different As-stressed and non-stressed plants enhanced the chlorophyll *a*, *b*, *a* + *b*, and *a*/*b* attributes. Alleviation of As stress to chlorophyll *a* and *b* varied by Si application according to the concentrations applied but maximum increase was observed in chlorophyll *a* (32.9, 41.2, and 46.3%) and *b* (26.6, 27.3, and 43.6%) under 8 mg kg^{-1} As in combination with Si treatments 50, 100, and 150 mg kg^{-1}.

3.3 Gas Exchanges Attributes

The results showed that Si addition significantly leads to higher contents of photosynthetic rate, transpiration rate, and stomatal conductance for all Si rates applied alone and in combination with As-stressed maize seedlings (Fig. 3). Nevertheless, the highest levels for photosynthetic rate (32.0, 36.3, and 37.6%), transpiration rate (27.6, 43.6, and 50.6%), and stomatal conductance (19.6, 38.1, and 43.7%) were found with 100 mg kg^{-1} of Si application in combination with all As stress–treated maize plants.

3.4 Enzymatic Antioxidant Activities

Significant ($p \leq 0.05$) effects of treatments, i.e., Si and As application, were observed on the enzymatic activities, i.e., POD, CAT, and SOD, where application of Si ameliorates As toxicity by enhancing the activities of the antioxidant enzymes (Fig. 5). Moreover, plants at As stress with 8 mg kg^{-1} of Si application showed a significant increase in SOD (53.9, 97.8, and 84.3%), POD (63.2, 111.2, and 101.8%), and CAT activity (85.4, 70.0, and 144.4%) in comparison with the control. The Si applied to plants grown without As indicated augmented activities of CAT, POD, and SOD enzymes. Similarly, proline contents significantly enhanced under both Si levels with As stress and non-As stress in maize seedlings. Maximum increase was noted in proline contents of maize seedlings treated with 100 mg kg^{-1}Si with all As stress levels.

3.5 Correlation Analysis

Correlation analysis depicted a significant ($p \leq 0.05$) association among shoot traits, biochemical traits, and Si and As shoot contents. Correlation of As shoot contents was negative and significant ($p \leq 0.05$) with shoot traits. Antioxidant enzymatic activities increased under stress conditions and therefore showed negative significant correlation with shoot traits. Accumulation of As increased antioxidant activities, and

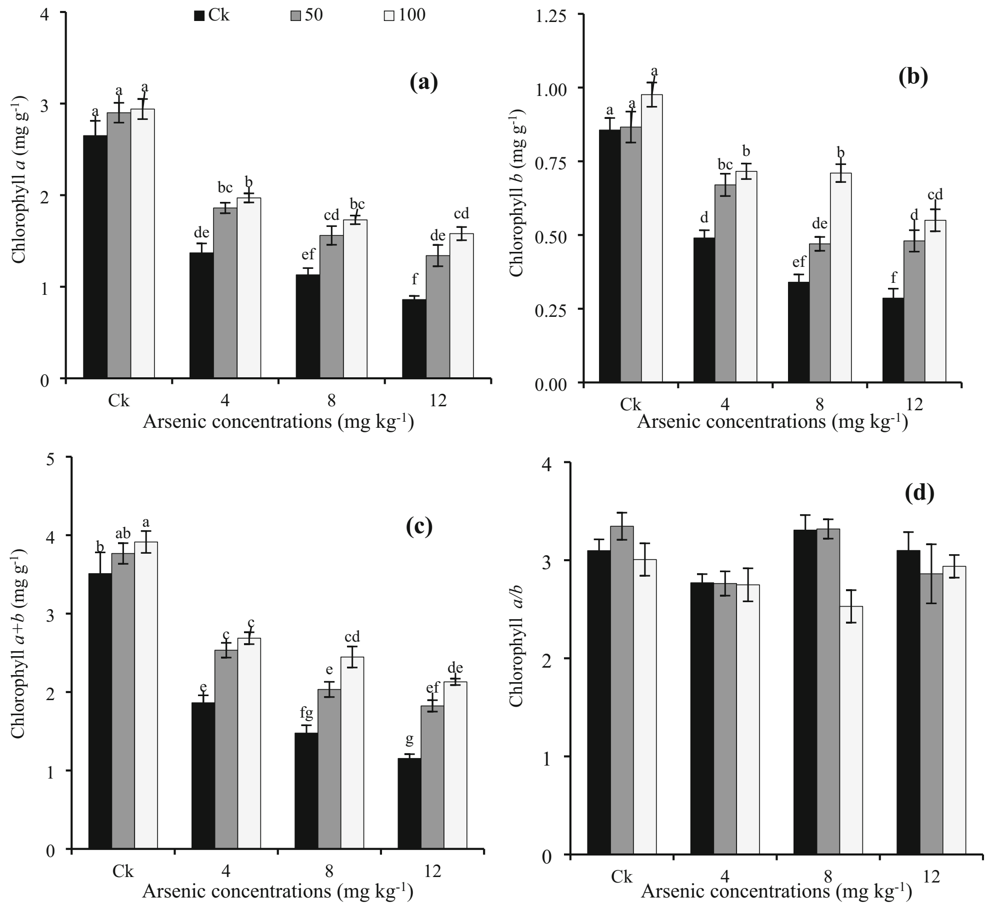

Fig. 2 Influence of silicon supplementation (i.e., Ck; control, 50, 100 mg kg^{-1}) on chlorophyll *a* (a), chlorophyll *b* (b), chlorophyll *a* + *b* (c), and chlorophyll *a/b* (d) of maize seedlings under various levels of arsenic treatments (i.e., Ck; control, 4, 8, 12 mg kg^{-1}). Values are mean SE of four replicates. Bars marked with different letters are significantly different by LSD ($p \leq 0.05$)

especially contents of MDA and H_2O_2 were increased which is an evident strong correlation ($r = 0.96$ and 95 respectively) of these attributes with As shoot contents. But accumulation of Si decreased these attributes which is evident from the negative correlation (non-significant) of these attributes with Si shoot contents. Details of all correlation combinations are presented in Table 1.

4 Discussion

The Si is an essential nutrient which plays an important role in the regulation of physiological mechanism under abiotic stress mechanism of different plant species. In the current study, application of Si improved root-shoot traits of maize seedlings. Application of As severely affected the performance of root-shoot traits, and increasing the toxicity of 40 mg kg^{-1} produces more severe detrimental effects. But application of Si alleviates the As toxicity even at a high concentration of As, due to its ability to provide mechanical strength and boost the growth of plants under a stressed environment (Muneer et al. 2017; Singh et al. 2019). Another reason for the slower performance of root-shoot parameters under As toxicity may be the destruction of chlorophyll pigments under As toxicity. Application of As reduced the chlorophyll pigments by more than 50% comparative to the control, which results in less CO_2 assimilation and ultimately reduced growth of the seedlings. Our results are consistent with that of other researchers who reported a detrimental effect of As toxicity of green pigments of maize (Anjum et al. 2017), mung bean (Srivastava et al. 2017), and wheat (Maglovski et al. 2019). Application of Si has significantly ($p \leq 0.05$) improved the green pigments even under As toxicity and reduced the toxic effect of As, which resulted in better seedling growth. There are two mechanisms behind the improvement in the chlorophyll pigments with Si application under As stress. First is that Si improves synthesis of chlorophyll pigments (Fig. 2; Singh et al. 2019) and second, it reduces the accumulation of As in plant parts (Fig. 4; Singh et al. 2019; Singh et al. 2011) which ultimately results in less destruction of the chlorophyll pigments.

Accumulation of As in roots and shoots directly affects the transport of water and CO_2 assimilation. All gas exchange parameters were reduced due to As toxicity (Fig. 3). Stoeva et al. (2003) reported a 23% reduction in photosynthetic rate, 41–52% reduction in stomatal conductance, and 30–50% in transpiration rate in maize due to As toxicity. They further reported not only that As affects the stomatal apparatus, but also that its

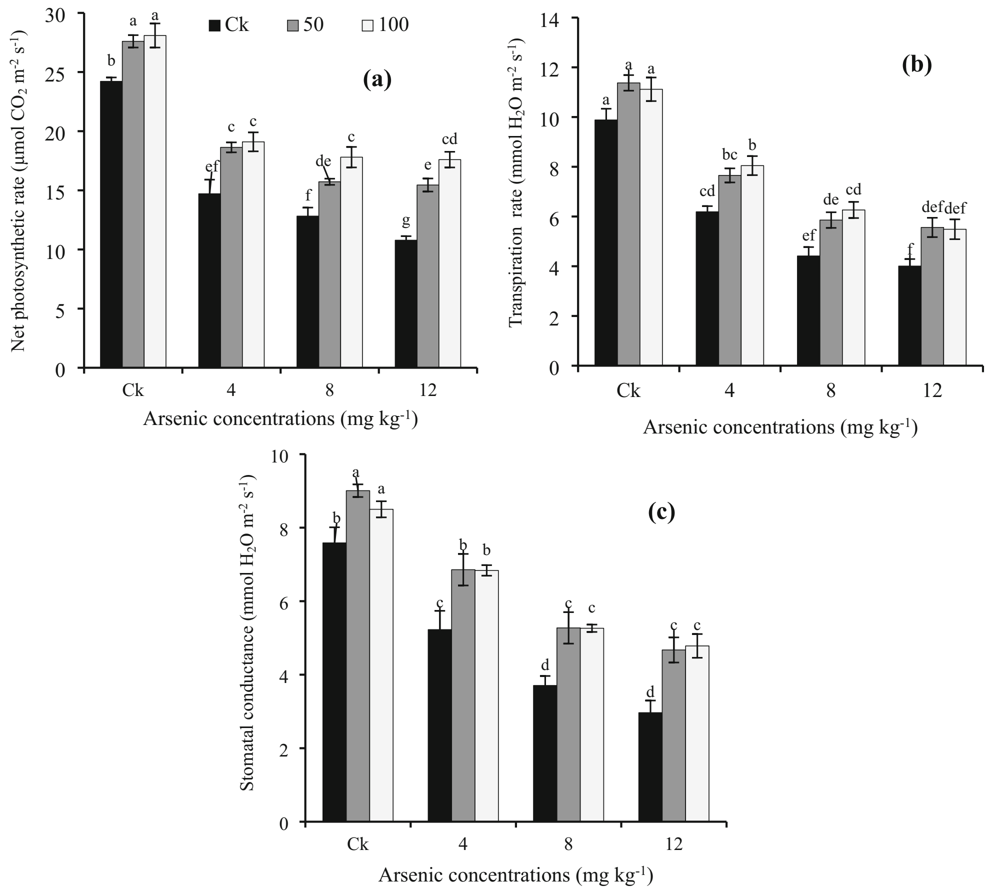

Fig. 3 Influence of silicon supplementation on (i.e., Ck; control, 50, 100 mg kg^{-1}) net photosynthetic rate (a), transpiration rate (b), and stomatal conductance (c) of maize seedlings under various levels of arsenic treatments (i.e., Ck; control, 4, 8, 12 mg kg^{-1}). Values are the mean SE of four replicates. Bars marked with different letters are significantly different by LSD ($p \leq 0.05$)

accumulation causes disruption in uptake of water which ultimately limits the process of photosynthesis and transpiration. Moreover, As toxicity hampers the synthesis of chlorophyll pigments (Stoeva et al. 2003; Srivastava et al. 2017; Fig. 2) which results in less light interception and less photosynthesis. Stoeva et al. (2003) reported that under As stress, the ratio of F_v/F_o and F_v/F_m decreased which indicates the reduction in functional activity of photosystem 2 (PS2). In Fig. 4(c), it is clear that application of Si considerably reduces the As accumulation which results in improvement of gas exchange parameters.

Table 1 Correlation analyses of different biochemical traits and shoot arsenic and silicon concentrations

	SDW	SL	SOD	POD	CAT	PT	PRO	MDA	H_2O_2	As
SL	0.91**	1.00								
SOD	−0.50**	−0.54**	1.00							
POD	−0.77**	−0.86**	0.69**	1.00						
CAT	−0.64**	−0.73**	0.70**	0.91**	1.00					
PT	−0.73**	−0.85**	0.72**	0.96**	0.93**	1.00				
PRO	−0.59**	−0.70**	0.70**	0.88**	0.92**	0.91**	1.00			
MDA	−0.91**	−0.95**	0.53**	0.79**	0.62**	0.75**	0.57**	1.00**		
H_2O_2	−0.89**	−0.87**	0.44**	0.66**	0.45**	0.61**	0.41**	0.94**	1.00	
As	−0.89**	−0.88**	0.49**	0.71**	0.50**	0.65**	0.42**	0.96**	0.95**	1.00
Si	0.06^{NS}	−0.03 ^{NS}	0.26 ^{NS}	0.40*	0.50**	0.45**	0.54**	−0.13 ^{NS}	−0.28 ^{NS}	−0.26 ^{NS}

*p value ≤0.05

**p value ≤0.01

^{NS} p value ≥0.05

SDW, shoot dry weight; *SL*, shoot length; *SOD*, superoxide dismutase; *POD*, peroxidase; *CAT*, catalase; *PT*, protein; *PRO*, proline; *MDA*, malondialdehyde; *H_2O_2*, hydrogen peroxide; *As*, arsenic; *Si*, silicon

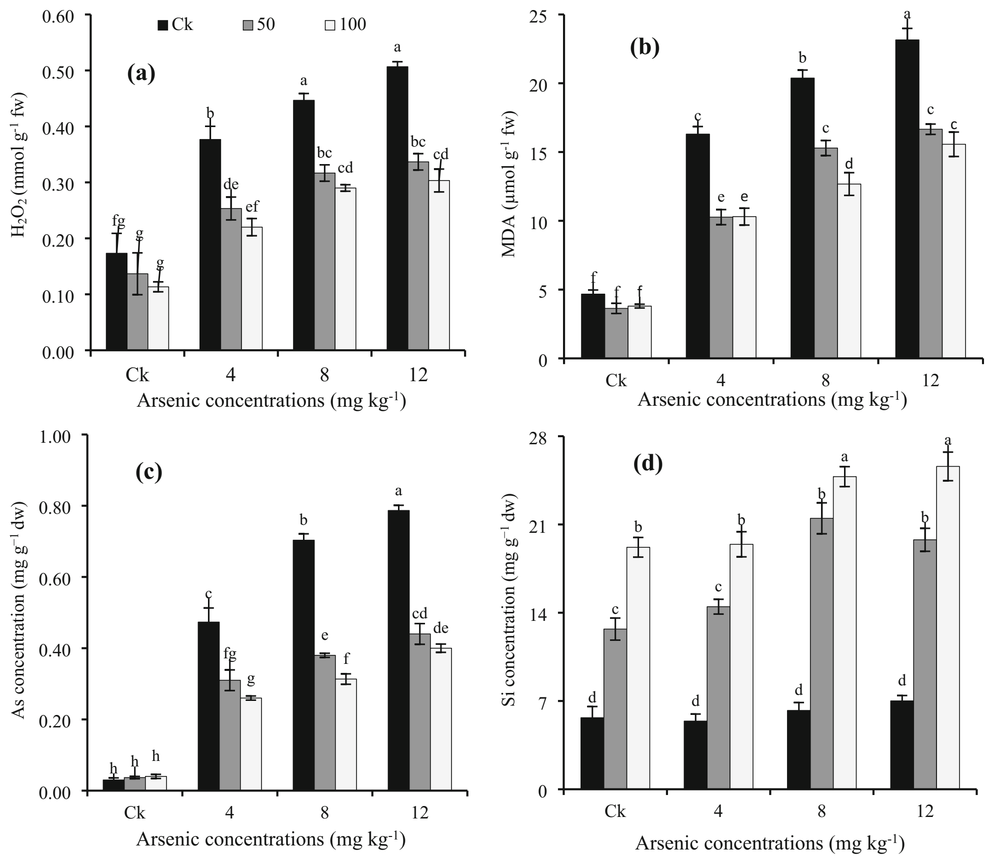

Fig. 4 Influence of silicon supplementation (i.e., Ck; control, 50, 100 mg kg^{-1}) on H_2O_2 (a), MDA (b), As concentration (c), and Si concentration (d) of maize seedlings under various levels of arsenic treatments (i.e., Ck; control, 4, 8, 12 mg kg^{-1}). Values are the mean SE of four replicates. Bars marked with different letters are significantly different by LSD ($p \leq 0.05$)

Metal stress hampers plant growth and development by disturbing the physiological and biochemical mechanisms. An adverse effect of metal stress is the production of reactive oxygen species like H_2O_2, radical oxygen, and hydroxyl ions, which results in oxidative damage to the plant (Tripathi et al. 2017; Anjum et al. 2017; Table 1). With the increase in As concentration, accumulation of As in plant tissue increased which increased oxidative damage. Oxidative damage is evident from more production of H_2O_2 and increases lipid peroxidation in the cell membrane which results in more production of MDA (Fig. 4(a, b); Table 1) which decreased the membrane stability. But application of Si reduced the accumulation of As in plant tissue (Fig. 4(c)) which in turn reduces the ROS production and oxidative damage (Fig. 4(a, b)). This shows that Si can mitigate the adverse effects of As toxicity.

To cope with abiotic stresses which results in the production of ROS, plants have their own defense mechanism which is equipped with the production of antioxidant enzymes like SOD, POD, and CAT and some proteins like proline (Maghsoudi et al. 2019). Proline is a kind of protein which protects plants from oxidative damage. With As damage, plants faced oxidative damage due to membrane damage and disruption in water and nutrient uptake (Tripathi et al. 2017; Maghsoudi et al. 2019; Fig. 4(a, b)); proline production increased and this production is further enhanced with application of Si (Fig. 5(d); Hayat et al. 2012; Kaur and Asthir 2015) and thus regulates the plant growth and development by the carbohydrate metabolism and synthesis of antioxidant enzymes like SOD, POD, APX, and CAT. SOD and POD always act as front-line soldiers in response to ROS production and converts highly toxic ROS like radical oxygen to less toxic ones like H_2O_2 (Zhang et al. 2019). CAT further decomposes H_2O_2 into water which is beneficial (Choudhury et al. 2013; Domazetovic et al. 2017). If we look into Fig. 5, SOD, POD, and CAT activities increased in response to As activity but with application of these activities have enhanced which resulted in membrane stability and regulation of physiological processes.

Correlation analyses revealed that As application plays a dual role in alleviating the As toxicity as it lowers the As concentration in the root, which is evident from a negative association of the two elements (Ning et al. 2016; Table 1), and also improves the antioxidant mechanism which scavenges the ROS and leads to better growth and development of maize seedlings.

5 Conclusion

Arsenic toxicity is a potential threat to global food production but in maize, its effects and alleviation have not been explored well. It is concluded from the study that arsenic toxicity affects

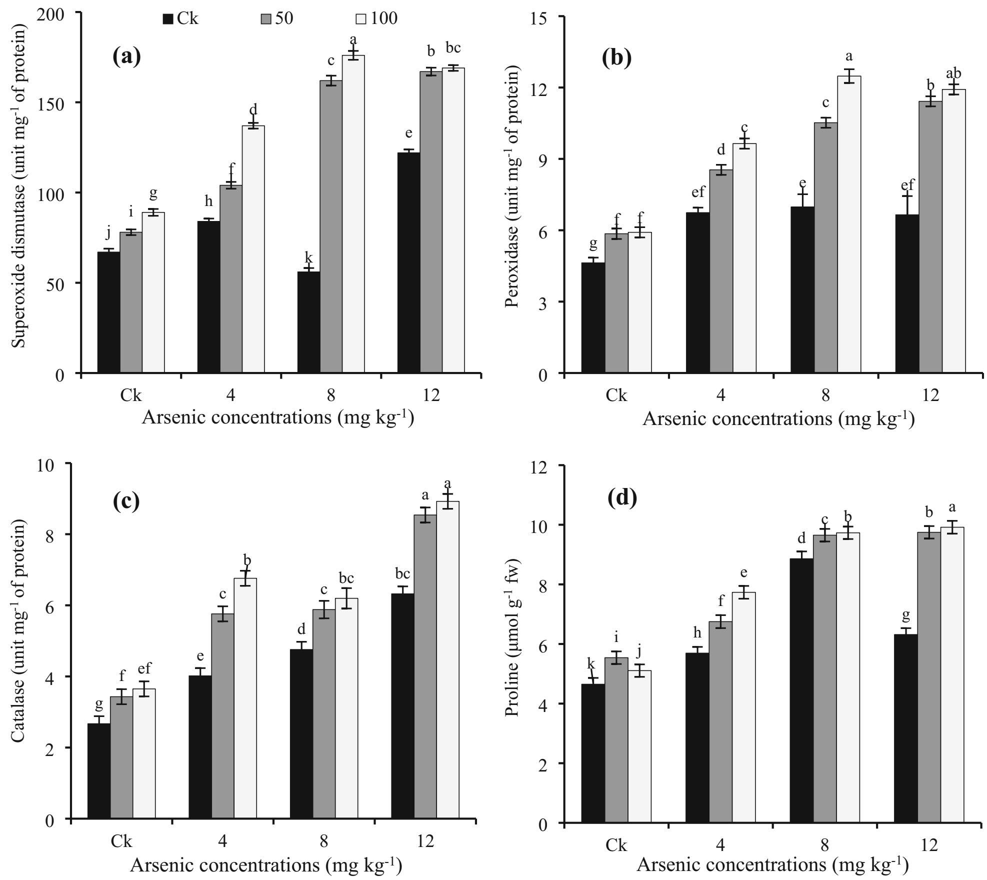

Fig. 5 Influence of silicon supplementation (i.e., Ck; control, 50, 100 mg kg^{-1}) on superoxide dismutase (a), peroxidase (b), catalase (c), and proline (d) of maize seedlings under various levels of arsenic treatments (i.e., Ck; control, 4, 8, 12 mg kg^{-1}). Values are the mean SE of four replicates. Bars marked with different letters are significantly different by LSD ($p \leq 0.05$)

plant growth and development by producing reactive oxygen species and damaging cell membrane and photosynthetic apparatus. Application of silicon alleviates arsenic toxicity by strengthening the antioxidant defense mechanism of plants, improving gas exchange parameters, and reducing damage to the photosynthetic system. 100 mg kg^{-1} of silicon application may be recommended for sustainable maize production under arsenic toxicity. This will not only improve plant growth and development but also have a positive impact on the environment.

Author Contribution Muhammad Kashif—experimentation, data collection, writing of introduction; Abdul Sattar—supervision of the research and initial drafting of the manuscript; Sami Ul-Allah—discussion, review, and editing; Ahmad Sher—methodology, conceptualization, and results; Muhammad Ijaz—introduction and interpretation of data; Madiha Butt—writing of the results; Abdul Qayyum—introduction and conceptualization

Data Availability Data sharing is not applicable to this article as all data has been presented and discussed in Sections 3 and 4.

Declarations

Competing Interest The authors declare no competing interests.

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