Effect of compost addition on arsenic uptake, morphological and physiological attributes of maize plants grown in contrasting soils

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Highlights

- Influence of compost (C) was explored on As uptake, morphological and physiological attributes of maize under As stress
- In Narwala soil, $C_{2.5}$ treatment decreased shoot As and improved shoot dry biomass and physiological attributes
- In Shahkot soil, $C_{2.5}$ treatment increased shoot As, thus reducing shoot dry biomass and physiological attributes
- Compost-mediated soil As immobilization/mobilization and plant As uptake varied with compost level and soil type

ABSTRACT

Contamination of soils with arsenic (As) represents a global environmental and health issue considering the entrance of toxic As in the human food chain. Although partially understood, addition of compost for the remediation of As-contaminated soils may result in distinct effects on plant growth and physiological attributes depending on compost-mediated potential mobility/sequestration of As in soils. This study explores the role of compost addition (C; 0, 1 and 2.5 %) on morphological and gas exchange attributes and photosynthetic pigments (chlorophyll contents) of maize plants under As stress (0, 40, 80, 120 mg kg^{-1}), as well as soil As immobilization/mobilization in a pot experiment, using two contrasting soils. Results revealed that, in Narwala (sandy loam) soil, the addition of compost decreased shoot As concentration of maize plants ($p < 0.05$; 4.01–13.7 mg kg^{-1} dry weight (DW)), notably at $C_{2.5}$ treatment, with significant improvement in shoot dry biomass, gas exchange attributes and chlorophyll (*a* and *b*) contents, i.e., 1.33–1.82, 1.20–2.65 and 1.34–1.66 times higher, respectively, over C_0 at all As levels. Contrastingly, in Shahkot (clay loam) soil, $C_{2.5}$ treatment increased shoot As concentration ($p < 0.05$; 7.02–17.3 mg kg^{-1} DW), and as such reduced the shoot dry biomass, gas exchange attributes and chlorophyll contents, compared to the control – rather C_1 treatment was more effective and exhibited positive effect than $C_{2.5}$. Considerably, at $C_{2.5}$ treatment, phosphate extractable (bioavailable) soil As concentration was also found to be greater in the (post-experiment) Shahkot soil than that of Narwala soil (0.40–3.82 vs. 0.19–1.51 mg kg^{-1}, respectively). This study advanced our understanding to resolve the complex compost-As interactions in As-contaminated soils, which are imperative to understand for developing the effective and soil-specific remediation strategies.

Keywords: Arenic phytotoxicity, Organic amendments, Contamination, Bioavailability, Remediation

1 Introduction

Arsenic (As) contamination of soil, sediment and groundwater systems is a global environmental, agricultural and public health issue due to the toxic and carcinogenic nature of As (Niazi et al., 2012; Shakoor et al., 2015). Both naturally occurring processes and anthropogenic activities, such as coal combustion, mining and smelting, use of arsenical pesticides in agriculture, irrigation with As-laced groundwater and leather tanning operations significantly contributed to soil As contamination (with soil As ranging from 32–3,100 mg kg^{-1}) (Niazi et al., 2016; Niazi et al., 2015; Sheik et al., 2012).

In soil and sediment environments, As mainly exists in two inorganic forms, arsenite (As(III)) and arsenate (As(V)) (Smedley and Kinniburgh, 2002). Arsenic(V) prevails in oxidized conditions, whereas As(III) is prevalent under reduced environments (Niazi and Burton, 2016; Shakoor et al., 2016). Globally, soil contamination with As has posed a potential threat to the humans through contaminating food chain (e.g., Rehman et al., 2016), which has increased interest amongst scientists to explore some sustainable and eco-friendly solutions for remediation and restoration of As-contaminated soils.

In recent years, immobilization of As and heavy metals in contaminated soils, for reducing their accumulation by plants and food chain, has emerged as an attractive and suitable remediation strategy (Arco-Lázaro et al., 2016; Sarkar et al., 2012). Various organic and inorganic waste materials have been used as effective soil amendments due to their reuse potential after recycling as a value added product (Arco-Lázaro et al., 2016; Pardo et al., 2014a). In contrast to inorganic

amendments, organic amendments (e.g., compost) are considered to be essential and eco-friendly option in remediation strategies – as they can supply macro- and micro-nutrients to enhance plant growth and add organic matter in soil to improve soil structure and carbon content (Beesley et al., 2013; Gomez-Eyles et al., 2013; Pardo et al., 2014b). However, As oxyanions could be potentially mobilized in soil by the addition of organic materials, which could possibly increase As bioavailability to plants (Mench et al., 2003).

Although partially investigated, contrasting effects of organic materials have been reported on As dynamics (adsorption and/or potential mobilization) in soil (Arco-Lázaro et al., 2016; Beesley et al., 2014). This depends on different soil properties, e.g., cation exchange capacity, soil texture clay content, as well as the presence of soil minerals, mainly Fe oxides and/or calcium carbonate ($CaCO_3$; primarily in calcareous soils) (Lin et al., 2008; Niazi et al., 2011). Gadepalle et al. (2008) reported that the application of compost (15%) in combination with Fe oxide/zeolite (5%) decreased As concentration in rye grass plants by 2 mg kg^{-1} of dry weight (DW). In another study, compost addition reduced availability of As in soil contaminated by copper-chromium-arsenate (CCA) application (historically used for timber treatment) (Cao and Ma, 2004). Similarly, Wang and Mulligan (2009) exhibited that, in an acidic soil from mine tailings, the organic particles in compost contributed to bind As by making complexes, and as such decreased mobility of As.

Contrastingly, in some other studies, As mobility has been reported to increase in soil following compost or organic amendments. Lin et al. (2008) demonstrated that addition of compost extract to two calcareous soils (compacted in a column bed) led to an increase in bioavailable As concentration in the leachate, although it varied depending on other properties of both soils. Mench et al. (2003) indicated that in soils and sediments (mine spoils), possessing low or no organic matter, compost addition increased the dissolved organic matter in soil solution, thus raised the

leachable amount of As possibly due to competition for adsorption sites on the mineral components. Recently, Arc-Lázaro et al. (2016), based on a sorption-desorption experiment, reported that addition of compost reduced the adsorption of As in mining soils (rich in Fe oxides and having high adsorption capacity), due to increased competition between dissolved organic ligands in compost and As oxyanions. Hence, the knowledge on As immobilization/mobilization in agricultural soils is important for predicting biogeochemical behavior of As in soil-plant systems.

Compost addition can influence soil As bioavailability. After uptake of heavy metal(loid), like As in this study, reactive oxygen species can be produced in plants which could cause oxidative and physiological damage to photosynthetic apparatus and gas exchange attributes of plants followed by plant death (Khalid et al., 2016; Niazi et al., 2016). Therefore, in this study we examined the influence of compost in mediating soil-plant transfer of As, and evaluated the compost-mediated control on plant As toxicity by determining different morphological, physiological and photosynthetic attributes of maize plants.

We hypothesized that, under As stress, the addition of compost in the two (calcareous) soils with contrasting properties (mainly CEC and clay content) may: (1) impact the soil-plant transfer of As, as well as the growth, physiological attributes, including transpiration rate, net photosynthetic rate, stomatal conductance, water use efficiency, and photosynthetic pigments of maize (*Zea mays* L.) plants; and (2) show compost-mediated differences in immobilization/mobilization of As in soils.

2 Materials and methods

2.1 Soil sampling

Surface soil samples (at 0-20 cm depth) were collected from Narwala (district Faisalabad; 31°23′0″ N, 72°56′0″ E) and Shahkot (district Nankana Sahib; 31°26′58″ N, 73°42′23″ E) in Punjab,

Pakistan. Soil samples were air-dried, ground, passed through a 2 mm sieve and thoroughly mixed to ensure homogeneity prior to determination of various physicochemical properties, total soil As concentration (Table 1), and for As-spiking to conduct a pot experiment in the glasshouse. The two soils used in this study mainly differed in CEC and clay content (see Table 1)

2.2 Physicochemical analyses of the soils

Particle size distributions of the two soils were determined using the hydrometer method for soil textural analysis. Soil pH and electrical conductivity (EC) were measured in a 1:5 soil:water suspension; cation exchange capacity (CEC) was determined by sodium acetate method (1 M NaOAc, pH 7); and the soil organic matter (SOM) content was determined by wet digestion employing the modified Walkley-Black method (Rayment and Lyons, 2011). The available soil phosphorus (P) content was extracted using 0.5 M sodium bicarbonate ($NaHCO_3$) (Olsen et al., 1954). Potassium (K) and sodium (Na) contents in the soils were extracted and measured by using a flame photometer (Janway PFP-7, Bibby Scientific Ltd., Staffordshire, UK).

Soil samples were digested in a mixture (2:1) of nitric (HNO_3) and perchloric ($HClO_4$) acids (Miller, 1998). Arsenic concentration in soil digests was measured by using a hydride generation-atomic absorption spectrometer (HG-AAS; Agilent AA240 with VGA 77), as described by Niazi et al. (2011).

2.3 Characterization of compost

Compost used in the pot experiment was obtained from the Soil Microbiology and Biochemistry laboratory of the Institute of Soil and Environmental Sciences (ISES), University of Agriculture

Faisalabad (UAF), Pakistan. Compost was dried in an oven at 50 °C for 24 hours, ground and passed through a 2 mm sieve prior to mixing with the As-contaminated soils.

The compost was characterized for various chemical properties including pH, EC, organic carbon content and total nitrogen (N) (Table 1) following standard methods as described elsewhere (Rayment and Lyons, 2011). Total As, P, iron (Fe), copper (Cu), manganese (Mn) and zinc (Zn) contents of compost were determined after its digestion with HNO_3 and $HClO_4$. Iron (Fe), copper (Cu), manganese (Mn) and zinc (Zn) contents in compost were measured by using an atomic absorption spectrometer (AAS; Thermo Solaar S4AA Spectrometer, Illinois, USA).

2.4 Pot experiment

A pot experiment was carried out in the wire house of the Institute of Soil and Environmental Sciences, University of Agriculture Faisalabad (UAF) in a completely randomized design (CRD) under ambient air and temperature conditions. The air-dried and ground (< 2 mm) soil samples of both soil types were spiked with As ($Na_2HAsO_4 \cdot 7H_2O$) at 0 (As_0), 40 (As_{40}), 80 (As_{80}), and 120 (As_{120}) mg As kg^{-1} dry soil. The As-spiked soils were equilibrated for three months at 50% of field capacity, thus allowing enough redistribution time for As on soil exchange sites.

Each pot was filled with 2 kg of As-contaminated soil and replicated three times. The pots were internally lined with polyethylene sheet to avoid As leaching and moisture loss through the soil over the duration of experiment. Compost as an amendment was thoroughly mixed with As-contaminated soil in each pot at three levels, i.e., 0% (C_0), 1% (C_1) and 2.5% ($C_{2.5}$).

Maize (*Zea mays* L.; cultivar Sahiwal 2003) seeds were obtained from Ayub Agricultural Research Institute, (AARI) Faisalabad. Each pot was sown with four seeds per pot, directly in compost-amended and -unamended soils, and after 7 days of germination only one healthy and uniform

plant was retained in each pot. The uprooted maize plants were mixed well in the soil of the same pot.

Each pot was fertilized with recommended dose of nitrogen (N), phosphorus (P) and potassium (K) at the rate of 120-60-65 mg kg^{-1} dry soil, using urea, di-ammonium phosphate (DAP) and potassium chloride, respectively. The N added by DAP was subtracted from urea to maintain required level of N for each experimental soil in each pot. The P and K were applied in total at the time of sowing while N was supplied in three splits: half of N was applied at the sowing time and remaining half in two equal splits after 20 and 40 days of sowing.

The maize plants in each pot were irrigated regularly to maintain moisture content at about 70% of field water holding capacity, and weeding was done whenever it was required through the duration of pot experiment (8 weeks).

2.5 Measurement of plant growth, photosynthetic and gas exchange attributes of maize plants

2.5.1 Photosynthetic pigments (chlorophyll content) and gas exchange attributes

Prior to plant harvesting, fresh leaves were carefully separated from maize plants using a sharp stainless steel scissor and immediately preserved in an ice-box. The freshly sampled leaves were extracted with 85% (v/v) aqueous acetone in the dark by shaking until the color of leaves was completely disappeared. The assay mixture was centrifuged at 5000 rpm for 10 min at 4°C and the supernatant was collected to measure (mg g^{-1} fresh weight (FW)) photosynthetic pigments (chlorophyll *a*, *b*) at wavelength 663 nm and 644 nm, respectively, using an UV-Vis spectrophotometer (Halo DB-20/ DB 20S, Dynamica Company, London, UK) (Lichtenthaler, 1987). Total chlorophyll content was calculated as the sum of chlorophyll *a* and *b* contents.

The youngest and fully expanded healthy plant leaves were selected to measure various gas

exchange parameters of the maize plants prior to harvesting the plants (after 8 weeks). The gas exchange parameters including stomatal conductance (g_s), transpiration rate (Tr), net photosynthetic rate (P_s) and water use efficiency (WUE) were measured between 10:30 am and 11:30 am during the day using an infrared gas analyzer (IRGA) (Analytical Development Co., Hoddesdon, UK).

2.5.2 Plant growth (morphological) parameters

Various morphological parameters related to maize plant growth were recorded before harvesting the plants. These parameters included plant height, leaf area, number of leaves per plant, and total shoot fresh weight.

2.5.3 Plant harvesting, total shoot dry biomass, digestion and elemental analyses

The above ground parts (total shoot biomass) of all the maize plants were harvested after 8 weeks of vegetative growth. Shoot samples were oven dried at 65^{o}C for 72 h and total shoot dry weight (including leaves and stem of plants) was recorded. Shoot samples were ground (< 1 mm) and digested in a mixture (1:1) of HNO_3 and $HClO_4$.

Total As concentration in the plant shoot was determined using a HG-AAS with a residual standard deviation (RSD) < 2%. Total P content in the digested plant shoot and compost samples was determined following the vanadate-molybdate yellow color method on an UV-Vis spectrophotometer (Chapman and Pratt, 1961).

Arsenic and other elemental analyses were performed in triplicate. Three reagent blanks, one reference plant material (pine needles No. 1575) and one reference soil (Montana 2710) were included to assess the precision and accuracy of the chemical analysis. After every 12 samples, a sample of known As concentration was analyzed to check the precision of the analysis and for quality control.

2.6 Phosphate extractable (bioavailable) soil As

To determine the effect of different compost levels on As mobility and bioavailability in post-experimental soils following compost application, soil samples were carefully taken from post-experimental pots to avoid mixing of plant roots. Soil samples were oven dried and extracted with potassium dihydrogen phosphate (0.2 M KH_2PO_4 solution, in a ratio of 1:5 soil/solution) (Niazi et al., 2012) in three replicates, and As concentration in soil extracts was determined using a HG-AAS as mentioned above.

2.7 Statistical analysis

The differences between individual means were compared by two-way analysis of variance (ANOVA) and the significance of differences between the treatments mean values was determined by Duncan's multiple range (DMR) test at $p \leq 0.05$. The SPSS software package (version 16.0, Chicago, IL) was used for all statistical analyses.

3 Results and discussion

3.1 Soil and compost characterization

The physicochemical properties of both soils and compost used in this study are presented in Table 1. The texture of Narwala and Shahkot soils was sandy loam and clay loam, respectively, thus showing a contrasting soil type. Soil pH was alkaline with values of 8.08 and 8.22 for Narwala and Shahkot soils, respectively. The CEC of Shahkot soil (13.09 $cmol_c\ kg^{-1}$) was ~2 times higher than the Narwala soil (7.08 $cmol_{(+)}\ kg^{-1}$), which could be related to the (~5 times) higher clay content in former soil (Table 1). Organic matter content was < 1 % in both the soils with slightly

higher OM content in Shahkot soil (0.81 %) than the Narwala soil (0.69 %). Chemical analysis of compost revealed Fe in the highest concentration (597 mg kg^{-1}) followed by Mn (53 mg kg^{-1}) and Zn (48 mg kg^{-1}), and As was not detected in the compost.

3.2 Effect of compost on maize plant growth and As phytotoxicity

Arsenic is a toxic element and considered to be non-essential for plant growth (Khalid et al., 2016; Niazi et al., 2016). In this study, As phytotoxicity symptoms appeared in maize plants after 4 weeks of plant growth (qualitative observations), notably at high As levels with no compost (C_0As_{80} and C_0As_{120} treatments), and varied with soil type and applied compost levels. In the high As treatments (As_{80} and As_{120}), plant growth was significantly reduced showing stunted plant growth and appearance of leaf chlorosis (purplish leaf color).

The plant growth (morphological) attributes, including total shoot fresh weight, total shoot dry weight, number of leaves per plant differed significantly ($p < 0.05$) between the two soil types at variable As and compost levels (Table 2). In both type of soils at all As levels with no compost (C_0As_{40}–C_0As_{120}), plant growth attributes tended to decrease significantly ($p < 0.05$) with increasing As concentration compared to their control treatments (C_0As_0).

Plant height, leaf area, number of leaves and shoot fresh weight and shoot dry weight significantly ($p < 0.05$) decreased with increasing levels of As in the absence of compost (C_0As_{40}–C_0As_{120}) in comparison to their respective controls (C_0As_0) (Table 2). In all experimental treatments, plant height, leaf area, number of leaves per plant, shoot fresh biomass and shoot dry biomass of maize plants in Narwala (sandy loam) soil ranged from 34–66 cm, 141–284 cm^2, 2–8, 6.2–25.7 g pot^{-1} and 1.08–4.37 g pot^{-1}, respectively; and spanned 41–68 cm, 110–250 cm^2, 2–6, 9.4–24.8 g pot^{-1} and 1.3–3.8 g pot^{-1} for Shahkot (clay loam) soil, respectively (Table 2).

Shoot dry biomass is a critical parameter for assessing the impact of As stress on plant growth (Niazi et al., 2017). The results revealed that compost application (C_1 and $C_{2.5}$ treatments) in the absence of As (As_0) resulted in significantly greater ($p < 0.05$) percentage increase in shoot dry biomass for maize plants in Narwala soil (22–27 %) than Shahkot soil (8–13 %) with respect to their controls (C_0As_0) (Table 2). This indicated that compost addition in (As_0) soil contributed to increasing nutrient availability for plant uptake, although it appeared to be dependent up on soil properties, with less availability in Shahkot soil (Al-Bataina et al., 2016).

Relatively greater CEC and clay content in Shahkot soil could hold plant mineral nutrients such as K^+, N (NH_4^+) and Zn^{2+} on negatively charged mineral exchange sites, thus reducing their uptake by plants (Caporale et al., 2013). In addition, the possible formation of stable Ca-phosphate precipitates, primarily in Shahkot soil due to its greater Ca content, may reduce phosphate availability to plants, which is required for plant metabolism and growth (this was also evident from shoot P concentration data of maize plants (see description below)). This, at least partly, may suggest that the shoot dry biomass yield was higher for plants grown in Narwala soil compared to those in Shahkot soil following compost application without As (C_1As_0 and $C_{2.5}As_0$).

Under As stress with no compost (C_0As_{40}-C_0As_{120}) treatments, shoot dry biomass decreased with increasing As concentration in both type of soils (Table 2). However, relatively higher percentage reduction in shoot dry biomass was observed for plants in Narwala soil (31–65 %) than that of Shahkot soil (18–54 %) with respect to their control (C_0As_0) (Table 2). It is well-known that As is non-essential for plant growth and causes toxicity even at low and moderate concentrations (Khalid et al., 2016). Arsenic-induced phytotoxicity and reduction in plant growth attributes, mainly shoot dry biomass, in maize plants grown in both soil types could possibly be attributed to malfunctioning of metabolic processes such as respiration and photosynthesis in plants under As

stress. This is in agreement with earlier studies whereby As-induced toxicity in plants was reported to cause stunted and retarded plant growth (Ansari et al., 2013; Gomes et al., 2013; Niazi et al., 2011).

The reduced plant growth was attributed to the hazardous effects of As on the metabolic functions of plant cells. For example, metabolic energy can be exploited for the generation of As stress related compounds such as antioxidases and phytochelatins (Srivastava et al., 2016). The inhibition of shoot growth (plant biomass) in maize plants could be due to increased tissue permeability and tissue loss, reduced enzyme activity and/or As induced oxidative stress (Ansari et al., 2013; Gomes et al., 2013). Also, As can change nutrient balance and their assimilation, protein metabolism and oxidative phosphorylation in plant tissues (Hasanuzzaman et al., 2016; Mirza et al., 2016). It can interfere with photosynthetic activity by affecting uptake of water and essential nutrients, cause lipid peroxidation via alteration in the lipid structure of cell membranes (Flora, 2011).

In Narwala soil, the high compost ($C_{2.5}$) treatment under As stress (As_{40}–As_{120}) resulted in significantly ($p < 0.05$; 1.3–1.8 times) greater improvement in shoot dry biomass yield (reduction percentage in shoot dry biomass ranged from 33–44 %) over control ($C_{2.5}As_0$) (percentage reduction in shoot dry biomass spanned 34–74 %; Table 2). Conversely, in Shahkot soil, C_1 was the most promising compost treatment with significant ($p < 0.05$) improvement in shoot dry biomass at all As levels over the control (C_1As_0) (percentage reduction in shoot dry biomass ranged from 17–34 % at C_1; Table 2). It is worth noting that, at all As levels in Shahkot soil, particularly at As_{80} and As_{120}, a greater reduction in percentage shoot dry biomass was obtained with $C_{2.5}$ (28–50 %) compared to C_1 (17–34 %) (Table 2).

3.3 Plant shoot As concentration

All As (As_{40}–As_{120}) and compost (C_1 and $C_{2.5}$) levels significantly ($p < 0.05$) affected the shoot As concentration in maize plants in both type of soils (Table 3). Plant shoot As concentration significantly ($p < 0.05$) increased under As stress with no compost (C_0As_{40}–C_0As_{120}) treatments in both type of soils compared to their controls (C_0As_0). However, shoot As concentration was found to be relatively higher in Narwala soil (12–18.8 mg kg^{-1} DW) than that of Shahkot soil (7–15.3 mg kg^{-1} DW) for C_0 treatments (Table 3).

In this study, an increasing trend in shoot As concentration was also in agreement with the reduced shoot dry biomass yield obtained for all As treatments with no compost (C_0As_{40}–C_0As_{120}) in both soil types (Tables 2 and 3). Significantly (1.20–1.71 times) higher shoot As concentration in Narwala soil may be linked with relatively greater availability of As for plant uptake than that of the Shahkot soil (Niazi et al., 2011). This could be attributed to the lower clay content and Ca concentration in Narwala soil compared to the Shahkot soil, as observed in this study (Inskeep et al., 2001) (Table 1; see discussion below in phosphate extractable soil As section).

We observed that the shoot As concentration of maize plants concurred with As concentrations in plant shoot reported in earlier studies (Rehman et al., 2016; Rosas-Castor et al., 2014a). For instance, Rosas-Castor et al. (2014b) reported that As concentration in shoot of maize plants varied from 0.365–18.5 mg kg^{-1} DW, where plants were grown in As-spiked sand culture/soils or in As-contaminated aged soils (soil As ranged: 5–586 mg kg^{-1}). In a field survey, Rosas-Castor et al. (2014a) revealed that As concentration in leaves of maize plants ranged from 0.10–3.15 mg kg^{-1} DW, whereby plants were collected from suburban areas of San Luis Potosi, Mexico (soil As ranged: 4.22–43.68 mg kg^{-1}). Rehaman et al., (2016) indicated that shoot As content in 24 different plant species spanned 0.05–1.38 mg kg^{-1} DW from various areas of KP, Pakistan (soil As ranged: 0.89–7.10 mg kg^{-1}).

In our study, shoot As concentration (4.01–18.8 mg kg^{-1} DW) for all As treatments was, although slightly, greater than those reported in different plant species by other researchers. This could possibly be ascribed to comparatively higher As concentration in growth medium (soil As: 40–120 mg kg^{-1}) and more soluble form of As in (artificially) As-contaminated soils (due to less aging time, 3 months) than the earlier studies reported above. Niazi et al. (2011) also demonstrated that shoot As concentration was significantly higher in *Brassica juncea* plants grown in As-spiked soils compared to historically As-contaminated cattle-dip site soils. Notably, shoot As concentration was greater in Narwala soil than the Shahkot soil, which highlights the importance of soil properties controlling the solid-phase partitioning of As in soils, and as such plant availability of As in both soil types, in the present study as discussed earlier.

In Narwala soil, $C_{2.5}$ was the most promising treatment to decrease shoot As concentration and improve shoot dry biomass and other plant growth attributes with respect to control (C_0) under As stress, as mentioned above (Table 2 and 3). Conversely, in Shahkot soil C_1 treatment resulted in a significant decrease in shoot As concentration at all As levels over control, while at high compost ($C_{2.5}$) level, rather a significant ($p < 0.05$) rise was observed in shoot As concentration (Tables 2 and 3).

Compost can influence the adsorption and release of As in soil depending on soil properties (e.g., pH, CEC, clay type and content) and composition of compost (Moreno-Jimenez et al., 2013). In the current study, contrasting soil properties (CEC and clay content) and level of compost application were crucial in controlling As accumulation by maize plants. Significantly, greater clay content and CEC of Shahkot soil (32 % and 13.09 $cmol_c$ kg^{-1}, respectively) than the Narwala soil (6 % and 7.08 $cmol_c$ kg^{-1}) could possibly have substantially increased surface negative charge on soil colloids in the former soil type (Dixon and Weed, 1989). Thus, the addition of compost,

primarily at $C_{2.5}$ level, may cause higher competition between negatively charged dissolved organic groups in compost and As oxyanions in Shahkot soil than that in Narwala soil (Mench et al., 2009; Moreno-Jimenez et al., 2013). This could have potentially led to increase in soil As mobility and As concentration of maize plants in this study (see bioavailable soil As Section below).

3.4 Phosphorus concentration in plant shoot

Plant shoot P concentration ranged from 1690–3690 mg kg^{-1} DW in Narwala soil and spanned 2168–2997 mg kg^{-1} DW for Shahkot soil with the minimum values observed in C_0As_0 treatments (Table 3). Generally, increasing shoot P concentration was ascribed to the presence of P in compost (which may be plant available due to compost mineralization) or it could also be attributed to enhanced release of P in soil solution due to increased microbial activity in compost-rich medium (Caporale et al., 2013). The results revealed that shoot P concentration was greater ($p < 0.05$) for C_1As_0 and $C_{2.5}As_0$ treatments compared to their respective control (C_0As_0), whilst no such significant trend was observed in the case of Shahkot soil (Table 3).

The presence of P in compost may have positive effects on plant growth under As stress, possibly due to P supply for proper functioning of essential metabolic functions, which could occur as: (i) high concentration of P in plant shoot can result in a down regulation of the As/P plasma-lemma transporters; (ii) high amount of P in plant cell can lead to a greater competition with As (As(V)) for different important biochemical processes, where As substitutes for P (Niazi et al., 2017). However, compost addition in both type of soils did not show any significant trend in shoot P concentration under As stress, indicating that the compost-derived P may not have a significant potential impact on As mobility and its uptake by maize plants, in this study.

3.5 Effect of As on chlorophyll contents and gas exchange attributes

For all treatments (C_0As_0–$C_{2.5}As_{120}$), chlorophyll *a*, chlorophyll *b* and total chlorophyll contents in the leaves of plants ranged from 0.27–0.98, 0.14–0.51 and 0.41–1.48 mg g^{-1} FW for Narwala soil, respectively; and spanned 0.10–0.99, 0.05–0.51 and 0.15–1.50 mg g^{-1} FW for Shahkot soil, respectively (Figure 1). The chlorophyll (*a* and *b*) and total chlorophyll contents decreased with increasing soil As levels (As_0–As_{120}) in the absence of compost (C_0) for both Narwala and Shahkot soils (Figure 1).

In Narwala soil, under As stress (As_{40}-As_{120}) the chlorophyll contents in leaves significantly ($p < 0.05$) increased with compost addition, notably at $C_{2.5}$ level, over their respective control (C_0) (Figure 1). Conversely, relatively less increase in chlorophyll concentration was observed for plants in Shahkot soil than the Narwala soil at C_1 level, and chlorophyll concentration tended to decrease primarily at $C_{2.5}$ level, with increasing soil As levels (Figure 1). These results also concur with shoot As concentration data, whereby at $C_{2.5}$ level an increasing trend in shoot As concentration was observed for plants in Narwala soil (Table 3). As described earlier, As-induced toxicity in the plant leaves has been reported to destroy membrane structure, drastically reducing the rate of photosynthesis carried out by plants (Khalid et al., 2016; Niazi et al., 2016), and hinder the biosynthesis of chlorophyll in plants (Hasanuzzaman et al., 2016; Mirza et al., 2016).

Thus, relatively greater chlorophyll content and higher biomass of maize plants in compost amended As treatments compared to their controls (C_0As_{40}–C_0As_{120}), could be an indirect indication on the compost-mediated reduced As uptake by plants. However, a decreasing trend in chlorophyll content for high compost ($C_{2.5}$) treatment, only in the case of Shahkot soil, confirms our shoot As concentration and dry biomass data, whereby the higher shoot As concentration and

lower shoot dry biomass were obtained at $C_{2.5}$ level in Shahkot soil than that of Narwala soil (Tables 2 and 3). Importantly, the level of compost application is a crucial factor while using compost as an amendment for reclamation and restoration of As-contaminated soils possessing contrasting soil properties, as we observed in this study.

Similarly, physiological attributes of maize plants including, net photosynthetic rate, transpiration rate, stomatal conductance, water use efficiency increased significantly ($p < 0.05$) with applied compost under As stress, with respect to their control treatments (C_0As_0) (Table 4). However, similar to chlorophyll contents, physiological attributes showed a decreasing trend with high compost ($C_{2.5}$) level in Shahkot soil at all As levels (Table 4). As discussed earlier, it has been well-identified that As causes growth inhibition, photosynthesis activity diminution and membrane disintegration and affects membrane system of chloroplasts, thereby reducing the physiological functioning and photosynthetic activity of plants (Flora, 2011; Hasanuzzaman et al., 2016). These results further validate our earlier argument (as described above), and as such resolve the important, although partially understood, role of compost in controlling phytoavailability and mobility of As in soils with contrasting properties.

3.6 Effect of compost on phosphate extractable As the in post-experiment soils

For all As treatments, the phosphate extractable (bioavailable) As concentration ranged from 0.22–2.40 mg kg^{-1} for Narwala (sandy loam) soil, and for Shahkot (clay loam) soil, it spanned 0.35–3.8 mg kg^{-1} (Figure 2). In Narwala soil, the bioavailable As concentration significantly ($p < 0.05$) decreased with compost application, considerably with $C_{2.5}$ treatment, over their control (C_0As_{40}-C_0As_{120}); and it followed the order $As_{120} > As_{80} > As_{40} > As_0$ (Figure 2). In Shahkot soil, only C_1 treatment showed a little effect on reducing bioavailable As content at low soil As (As_{40}) level,

whilst $C_{2.5}$ treatment increased phosphate extractable As concentration for all As levels (As_{40}–As_{120}), with respect to their control (C_0As_{40}-C_0As_{120}) (Figure 2).

In soils amended with compost, a decreasing trend in phosphate extractable As concentration, mainly in Narwala soil, was possibly attributed to the adsorption of As with compost, thus reducing plant available As pool by lowering (readily bioavailable) As concentration in soil solution (Caporale et al., 2013; McBride, 2000). Arsenic oxyanions could possibly be bound with protonated biomolecules/ligands present in compost. Several mechanisms could contribute to the adsorption of As oxyanions to compost, which could involve: (i) binding of As with protonated amino groups; (ii) association of As with carboxylate or phenolate functional groups by making covalent bonds; and (iii) ability of As oxyanions to develop relatively insoluble (ternary) complexes with cations (Al^{3+}, Fe^{3+}, Zn^{2+}) present in compost (Mikutta and Kretzschmar, 2011). However, by adding compost at high ($C_{2.5}$) level in the Shahkot soil (having significantly higher CEC and clay content) may have substantially increased the competition between As oxyanions and negatively charged dissolved organic groups or phosphate anions present in compost (Arco-Lázaro et al., 2016), thereby forcing As oxyanions to remain in soil solution in clay and mineral rich (calcareous) soil. Further research is warranted to directly and precisely examine the role of compost in adsorption and desorption of As oxyanions in a wide range of contrasting soils under environmentally-relevant conditions.

4 Conclusions

In stark contrast to the Shahkot (clay loam) soil, the shoot As concentration decreased with a profound improvement in shoot dry biomass, as well as photosynthetic pigments and gas exchange attributes of maize plants in Narwala (sandy loam) soil with $C_{2.5}$ treatment under As stress. This

study highlights that organic material (or phosphate) from compost at $C_{2.5}$ level could partially hinder adsorption sites, and as such decline As adsorption in Shahkot soil. Significantly, the higher CEC and clay content of Shahkot soil (with greater negative charge on the surface of soil colloids) might have resulted in huge competition between As oxyanions and negatively charged groups in compost for adsorption on soil mineral exchange sites, thereby increasing As concentration in soil solution. Overall, this short-term (pilot scale) pot experiment emphasize that the addition of compost or other organic amendments, for restoration and remediation of As-contaminated soils, need a careful optimization in controlled conditions before being applying at the field scale – this depends on soil properties and compost application, as we explored in this study. However, further research is warranted to delineate role of compost on immobilization/mobilization of As and its uptake by different plant species, in a range of historically As-contaminated calcareous soils having differing soil properties and As contents.

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Figures

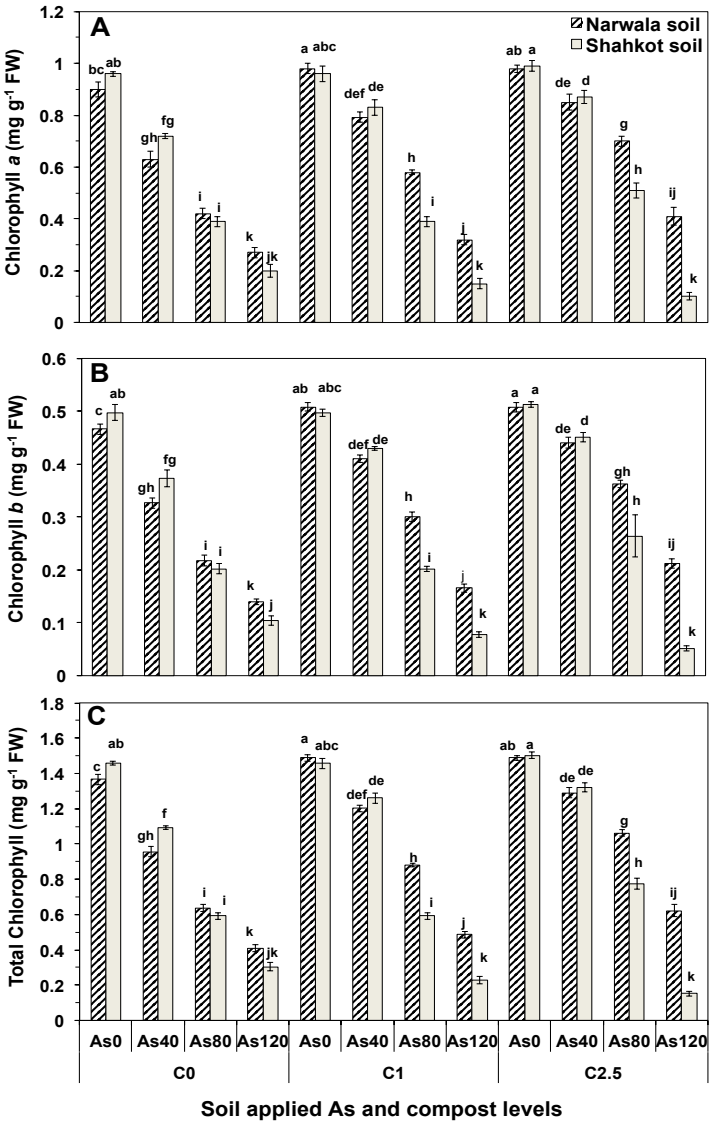


Figure 1: Chlorophyll a, chlorophyll b and total (a + b) chlorophyll contents in leaves of maize plants at four arsenic (As; 0, 40, 80, 120 mg kg^{-1}) and three compost (C; 0, 1, 2.5 %) levels in Narwala soil (sandy loam) and Shahkot soil (clay loam). Data are presented as mean ± standard error of three replicates. Means followed by the same letter are not significantly different (Duncan's multiple-range test, $p < 0.05$).

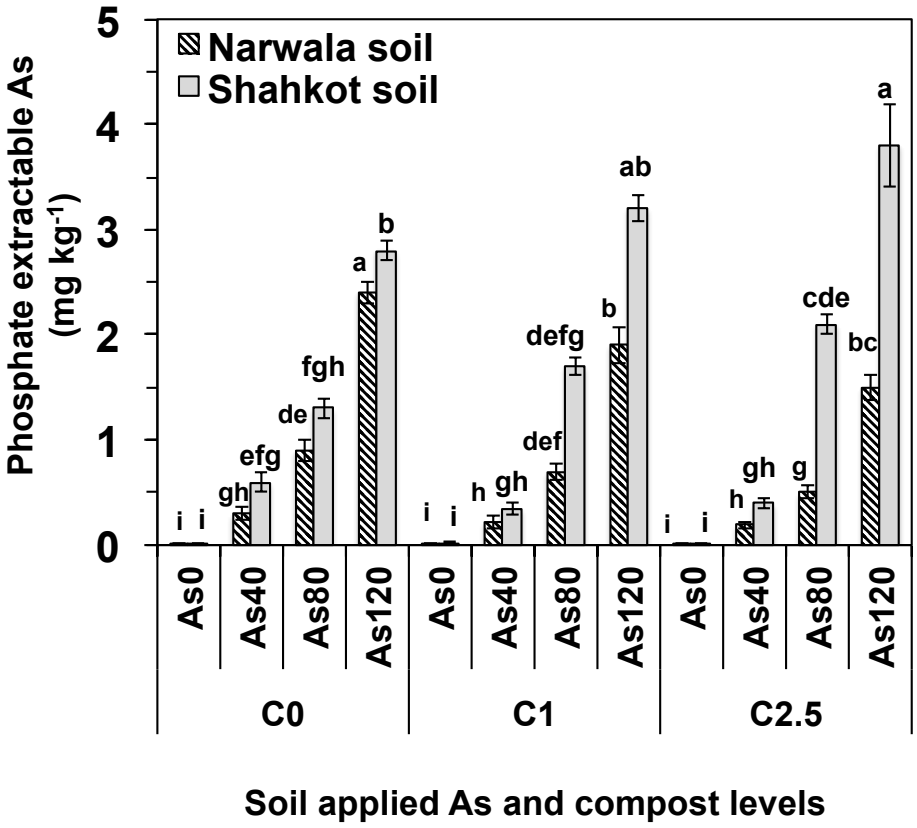


Figure 2: Phosphate extractable (bioavailable) As concentration in the post-experimental Narwala and Shahkot soils under the influence of arsenic (As; 0, 40, 80, 120 mg kg^{-1}) and compost (C; 0, 1, 2.5 %) treatments. Data are presented as mean ± standard error of three replicates. Means followed by the same letter are not significantly different (Duncan's multiple range test, $p < 0.05$).

Tables

Table 1 Physicochemical properties of the Narwala and Shahkot soils and compost used in the pot experiment (data are presented as mean ± standard deviation of three replicates).

Soil properties	Value		Compost properties	Value
	Narwala	Shahkot		
Sand (%)	72±3	45±2	OC (%)	23±2.2
Silt (%)	22±4	23±3	N (%)	2.2±0.16
Clay (%)	6±1	32±2	P (%)	0.31±0.08
Textural class	Sandy loam	Clay loam	K (%)	1.59±1.2
EC (1:5) (dS m^{-1})	0.12±0.08	0.63±0.24	Copper (mg kg^{-1})	1.31±0.06
pH (1:5, soil: water)	8.08±0.06	8.22±0.09	Zinc (mg kg^{-1})	48±2.8
Organic matter (%)	0.69±0.11	0.81±0.19	Manganese (mg kg^{-1})	53±2
CO_3^{2-}($mmol_c$ L^{-1})	1.0±0.06	2.4±0.14	Iron (mg kg^{-1})	597±45
HCO_3^- ($mmol_c$ L^{-1})	2.3±0.54	5.4±0.98	Total As (mg kg^{-1})	ND
Cl^- ($mmol_c$ L^{-1})	10.9±1.01	25.8±2.1	EC (1:5) (dS m^{-1})	7.03±0.18
Ca^{2+} ($mmol_c$ L^{-1})	3.0±0.76	16±1.21	pH (1:5, soil: water)	6.41±0.07
CEC ($cmol_c$ kg^{-1})	7.08±0.98	13.09±1.02		
Extractable P (mg kg^{-1})	7.0±1.2	9±0.97		
Extractable K (mg kg^{-1})	120±4	235±6		
Extractable Na (mg kg^{-1})	11±2	46±4		
Total soil As (mg kg^{-1})	ND	ND		

EC: Electrical conductivity; CEC: Cation exchange capacity; ND: not detected

Table 2 Effect of different soil applied levels of arsenic (As; 0, 40, 80, 120 mg kg^{-1}) and compost (0, 1 and 2.5 %) on growth (morphological) attributes of maize plants.

Soil As and compost levels	Plant height (cm)[a]	Number of leaves per plant[a]	Leaf area (cm^2)[a]	Shoot fresh weight (g)[a]	Shoot dry weight (g)[a]
Narwala soil (sandy loam)					
C_0 As_0	63±3 ab	7±1 a	260±10 a	24.10±1.2 ab	3.20±0.17 b
C_0 As_{40}	47 ± 2 hi	5±2 e	191±12 de	12.83±0.16 def	2.21±0.16 (31%)¶ def
C_0 As_{80}	39 ± 4 lm	3±1 kl	147±9 l	7.66±0.61 lm	1.66±0.12 (48%)¶ l
C_0 As_{120}	34 ± 2 m	2±1 m	141±11 m	6.22±0.69 m	1.11±0.11 (65%)¶ m
C_1 As_0	63 ± 3 ab	8±1 a	272±12 ab	24.31±0.59 ab	4.12±0.09 ab
C_1 As_{40}	50 ± 4 gh	4±2 def	202±9 def	12.73±0.79 def	2.73±0.14 (34%)¶ def
C_1 As_{80}	46 ± 2 hij	5±1 fg	234±9 fg	14.70±0.89 fgh	2.37±0.13 (42%)¶ fgh
C_1 As_{120}	35 ± 3 jklm	3±1 m	128±7 m	7.08±0.56 m	1.08±0.16 (72%)¶ m
$C_{2.5}$ As_0	66 ± 2 a	8±1 a	284±8 a	25.7±1.8 a	4.37±0.14 a
$C_{2.5}$ As_{40}	57 ± 2 abc	6±1 de	240±7 de	17.94±1.4 de	2.94±0.17 (33%)¶ de
$C_{2.5}$ As_{80}	53 ± 3 bcde	6±1 fgh	244±9 fgh	17.01±1.3 fgh	2.47±0.13 (43%)¶ fgh
$C_{2.5}$ As_{120}	40 ±3 defg	5±2 ijk	220±7 ijk	12.1±1.7 ijk	2.02±0.17 (54%)¶ ijk
Shahkot soil (clay loam)					

C_0 As_0	52 ± 2 abcd	6±1 ab	250±11 ab	20.2±1.1 bc	3.30±0.16 abc
C_0 As_{40}	49 ± 4 defg	4±1 ghi	171±9 ghi	13.3±1.6 ghi	2.3±0.15 (30%)[¶] ghi
C_0 As_{80}	47 ± 5 ghi	3±2 jk	137±6 jk	11.2±1.9 jk	1.8±0.12 (45%)[¶] jk
C_0 As_{120}	41 ± 2 ijkl	2±1 lm	131±8 lm	9.4±1.3 lm	1.3±0.18 (53%)[¶] lm
C_1 As_0	68 ± 2 a	7±2 a	259±8 bc	24.8±2.1 a	3.8±0.15 a
C_1 As_{40}	62 ± 4 ab	4±1 d	213±5 d	18.7±2.2 d	3.1±0.15 (17%)[¶] d
C_1 As_{80}	59 ± 3 hijk	4±2 def	219±5 de	17.9±0.9 def	2.7±0.12 (29%)[¶] def
C_1 As_{120}	55 ± 2 m	5±2 fgh	110±4 gh	14.1±0.85 fgh	2.5±0.15 (34%)[¶] fgh
$C_{2.5}$ As_0	61 ± 3 abc	7±1 a	278±8 c	23.9±1.8 ab	3.6±0.17 ab
$C_{2.5}$ As_{40}	55 ± 2 cdef	5±1 efg	220±9 fg	14.6±1.1 efg	2.6±0.16 (28%)[¶] efg
$C_{2.5}$ As_{80}	49 ± 3 efgh	3±1 hij	218±12 hi	12.7±0.8 hij	2.1±0.11 (42%)[¶] hij
$C_{2.5}$ As_{120}	45 ± 4 klm	2±1 jk	189±9 jk	11.8±0.9 jk	1.8±0.20 (50%)[¶] jk
As × C	ns	ns	ns	ns	ns
As × S	*	*	*	*	**
As × S × C	ns	*	ns	*	*

[a] Data are presented as mean ± standard error of three replicates. Means followed by the same letter are not significantly different (Duncan's multiple-range test, $p < 0.05$).

The percent (%) relative change/reduction in shoot dry weight was calculated as 100 – (dry shoot biomass/corresponding control × 100);

C_0, C_1 and $C_{2.5}$: Compost applied in soil at 0, 1 and 2.5 % levels;
As_0, As_{40}, As_{80}, As_{120}: Arsenic applied in soil at 0, 40, 80 and 120 mg kg^{-1} levels;
^{ns}: non-significant; * significant at $p < 0.05$;
As: Soil As level; S: Soil type; C: Compost level

Table 3 Effect of different soil applied levels of arsenic (As; 0, 40, 80, 120 mg kg^{-1}) and compost (0, 1 and 2.5 %) on shoot As and phosphorus (P) concentrations of maize plants.

Soil As and compost levels	Shoot As concentration (mg kg^{-1} DW)[a]	Shoot P concentration (mg kg^{-1} DW)[a]
Narwala soil (sandy loam)		
C_0 As_0	0.02±0.005 jk	1690±22 k
C_0 As_{40}	12.01±1.16 fgh	1872 ±32 jk
C_0 As_{80}	16.29±0.88 cd	2317±34 cd
C_0 As_{120}	18.80±0.88 a	3170±50 ab
C_1 As_0	0.04±0.01j	2238±34 de
C_1 As_{40}	6.01±0.58 hi	2366±44 cd
C_1 As_{80}	14.34±0.88 ef	2122±21 def
C_1 As_{120}	17.04±0.58 ab	2818±11 b
$C_{2.5}$ As_0	0.025±0.005 jk	3690±92 a
$C_{2.5}$ As_{40}	4.01±0.58 hi	2311±40 cd
$C_{2.5}$ As_{80}	11.02±0.58 i	2073±37 g
$C_{2.5}$ As_{120}	13.70±0.33 fg	2574±32 bc
Shahkot soil (clay loam)		
C_0 As_0	0.05±0.01 j	2168±50 j
C_0 As_{40}	7.01±0.58 h	2671±42 ef
C_0 As_{80}	13.3±1.33 fg	2513±37 ghi
C_0 As_{120}	15.3±1.45 cde	2811±24 cd
C_1 As_0	0.04±0.02 jk	2852±27 abc
C_1 As_{40}	5.00±0.56 hi	2726±38 de
C_1 As_{80}	15.0±1.15 de	2793±43 d
C_1 As_{120}	17.0±0.58 ab	2598±32 ef
$C_{2.5}$ As_0	0.04±0.025 jk	2997±38 a
$C_{2.5}$ As_{40}	7.02±0.58 i	2366±37 hij

$C_{2.5}$ As_{80}	16.7±0.88 abc	2500±32 hi
$C_{2.5}$ As_{120}	17.3±0.88 a	2775±34 de
As × C	*	*
As × S	ns	ns
As × S × C	*	ns

[a] Data are presented as mean ± standard error of three replicates. Means followed by the same letter are not significantly different (Duncan's multiple-range test, $p < 0.05$).
C_0, C_1 and $C_{2.5}$: Compost applied in soil at 0, 1 and 2.5 % levels;
As_0, As_{40}, As_{80}, As_{120}: Arsenic applied in soil at 0, 40, 80 and 120 mg kg^{-1} levels;
ns: non-significant; *: significant at $p < 0.05$; nd: not detected; nc: not calculated;
As: Soil As level; S: Soil type; C: Compost level.

Table 4 Effect of different soil applied levels of arsenic (As; 0, 40, 80, 120 mg kg^{-1}) and compost (0, 1 and 2.5 %) on gas exchange attributes of maize plants.

As and compost treatments	Gas exchange parameters			
	Net photosynthetic rate (µmol m^{-2} s^{-1})[a]	Transpiration rate (mmol m^{-2} s^{-1})[a]	Water use efficiency[a]	Stomatal conductance (mmol m^{-2} s^{-1})[a]
Narwala soil (sandy loam)				
C_0 As_0	14.10±1.1 ab	2.34±0.54 abc	4.25±0.14 ab	6.19±0.11 ab
C_0 As_{40}	8.13±0.77 def	1.93±0.26 de	2.89±0.16 de	4.02±0.09 de
C_0 As_{80}	4.66±0.91 k	0.96±0.18 k	1.09±0.07 k	2.34±0.06 k
C_0 As_{120}	3.90±0.59 m	0.45±0.12 m	0.65±0.04 m	0.95±0.03 m
C_1 As_0	17.20±0.97 a	3.28±0.37 a	4.91±0.21 a	6.98±0.08 ab
C_1 As_{40}	9.01±1.89 def	2.11±0.12 f	3.10±0.31 f	4.81±0.07 f
C_1 As_{80}	8.10±0.55 fg	1.90±0.25 fg	2.52±0.15 fg	3.22±0.05 fg
C_1 As_{120}	1.78±0.46 m	1.68±0.20 m	1.56±0.15 m	1.69±0.05 m
$C_{2.5}$ As_0	16.9±0.9 a	2.95±0.33 a	4.45±0.53 a	7.10±0.37 a
$C_{2.5}$ As_{40}	11.04±0.95 de	2.29±0.21 de	3.41±0.28 de	5.21±0.08 de
$C_{2.5}$ As_{80}	10.11±0.54 fgh	1.91±0.29 fgh	2.89±0.13 fgh	3.75±0.07 fgh
$C_{2.5}$ As_{120}	8.10±1.0 i	1.20±0.19 i	1.16±0.11 i	2.06±0.09 i
Shahkot soil (clay loam)				
C_0 As_0	12±0.76 def	2.12±0.53 de	4.02±0.63 d	5.92±0.18 de
C_0 As_{40}	9.3±0.64 ghi	2.01±0.39 gh	2.97±0.29 gh	3.27±0.09 gh
C_0 As_{80}	6.2±1.1 jk	1.07±0.16 jk	1.21±0.14 jk	2.69±0.04 jk
C_0 As_{120}	2.64±0.88 lm	0.54±0.20 m	0.81±0.11 m	1.11±0.04 m
C_1 As_0	19.8±0.98 bc	3.16±0.54 bc	4.98±0.22 a	6.69±0.16 ab
C_1 As_{40}	9.45±1.1 d	2.27±0.21 d	3.29±0.09 d	4.98±0.05 d
C_1 As_{80}	8.76±0.77 de	2.02±0.17 de	1.90±0.20 de	3.09±0.07 de
C_1 As_{120}	2.97±0.85 fgh	1.69±0.15 fg	1.71±0.29 f	1.49±0.09 f
$C_{2.5}$ As_0	18.8±2.1 c	3.08±0.23 c	4.38±0.39 c	6.62±0.19 c
$C_{2.5}$ As_{40}	5.16±0.34 ef	1.99±0.54 ef	2.01±0.29 ef	1.81±0.04 ef
$C_{2.5}$ As_{80}	3.01±0.22 hij	0.93±0.13 hij	0.99±0.26 h	0.91±0.07 h
$C_{2.5}$ As_{120}	1.32±0.32 j	0.39±0.12 j	0.59±0.09 j	0.42±0.03 j
As × C	*	ns	ns	ns
As × S	ns	*	*	*
As × S × C	*	*	*	*

[a] Data are presented as mean ± standard error of three replicates. Means followed by the same letter are not significantly different (Duncan's multiple-range test, $p < 0.05$).

C_0, C_1 and $C_{2.5}$: Compost applied in soil at 0, 1 and 2.5 % levels;

As_0, As_{40}, As_{80}, As_{120}: Arsenic applied in soil at 0, 40, 80 and 120 mg kg^{-1} levels;

[ns]: non-significant; *: significant at $p < 0.05$;

As: Soil As level; S: Soil type; C: Compost level