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# Comparison of grain sorghum and alfalfa for providing heavy metal remediation of sandy soil with different soil amendments and salt stress

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Soil salinity and heavy metal (HM) pollution of soil is an ongoing threat to the plants' survival that adversely affect the crop productivity and global food security. Therefore, an eco-friendly solution is highly desirable for mitigating the adverse affect of toxic pollutants in plants and soils. This study was aimed to explore how municipal solid waste compost (CO) or farmyard manure (M) supplementation regulates biomass yield, mineral nutritions under salinity, and distribution profile of toxic pullutants of (*Medicago sativa* L.) and sorghum [*Sorghum bicolor* (L.) Moench]. The CO and M were supplemented with saline (NaCl) soils, the total experiments were conducted for the three consecutive harvestings (H1, H2 and H3) of sorghum and alfalfa. In this study, the CO supplementation highly enhanced biomas yield (dry weight basis in sorghum during H2, while it showed higher in alfalfa during H3., Interestingly, the M significantly increased nitrogen (N<sub>2</sub>) and potassium (K<sup>+</sup>) but reduced sodium (Na<sup>+</sup>) in alfalfa, while the higher Na<sup>+</sup> and phosphorus (P) were accumulated in sorghum fertilizer. As a consequence of these finding, a positive correlation was observed among the plant biomass yield, N and K<sup>+</sup> content in alfalfa. Conversely, the high Na<sup>+</sup> present in soil declined plant biomass in surghum, indicating that CO supplementaton was not fully effective under high saline soil conditions. However, the N-P-K distribution improved due to CO and/or M supplementation in saline soils, while Cd accumulation was higher in sorghum compared to alfalfa. Therefore, sorghum can be used to clean up contaminated environments. The PCA results showed the same clusters of treatments and amedments were grouped in same plot, which indicated positive correlation between the treatment groups and plants, repectively. These results suggest

that M supplementation is useful to mitigate saline stress compared to CO in alfalfa, while sorghum can be recommended as to clean up heavy metals (HMs) from soils. This study further suggest a correlation of minerals (N-P-K) boosting and salinity stress reduction in plants. Therefore, organic amendment-based ecofriendly approach can be useful to mitigate salinity stress in plants as well as effective for clean environment and smart agriculture.

#### KEYWORDS

sorghum bicolor, medicago sativa, salinity, municipal solid waste compost, farmyard manure, nutrient content, heavy metal phytoextraction

## Introduction

Soil salinization is one of the most common occurrences that significantly declines plant growth and productivity (Isayenkov and Maathuis, 2019). The presence of excess salt ( $\text{Na}^+$ ) in agricultural soils adversely affects crops and the environment (Minhas et al., 2020; Ahmed et al., 2022). According to global projections, it has been predicted that approximately, 50% of arable land will cover with salinity by 2050 (Butcher et al., 2016). Thus, the soil salinity problem will more severe toward agricultural production. Therefore, an ecofriendly approach is highly desirable for plant growth, yield productivity, global food security and sustainable agricultural productions.

Application of organic amendments (OAs) and mineral nutrients substantially mitigate of multiple stress-induced plant damages and yield reduction (Rahman et al., 2018). The organic amendments such as plant based compost, farmyard manure, biofertilizer are cost effective and ecofriendly compared to commercial chemical fertilizers (Kumar et al., 2022). The organic amendments associated plant growth promoting with stress adapting traits have enormous potential to solve these environment challenges (Khan et al., 2022). The more beneficial impact of these OAs contain several mineral nutrients (i.e., N,P,K, Zn, and S), modulates phytohormones, increase plant growth promoting microbes (Kumar et al., 2022; Rahman et al., 2018). These beneficial traits greatly enhance plant growth and fitness to multiple abiotic stress (Ullah et al., 2021). Recent study documented that OAs greatly alleviated soil salinity by influencing microbial activity and nutrient cycling (Wichern et al., 2020). Despite of these advancement of OAs application in plant improvements and abiotic stress adaptation, there has been several mineral-based biostimulant approaches were reported for mitigating salt and heavy metal toxicity in plants and soils (Rahman et al., 2020; Kabir et al., 2021; Choi et al., 2021).

Saline soils generally have low organic matter (OM), and nitrogen (N) content and their physicochemical and biological properties usually need to be improved (El Sabagh et al., 2020). Compost from source-separated household waste (biowaste compost) can be a valuable fertilizer and beneficial as a soil conditioner, supplying nutrients as well as organic matter (Carabassa et al., 2020; Awad et al., 2021) and useful for

bioremediation of salt-affected soil (Kalanaki et al., 2020). This is particularly important for agricultural soils in some Mediterranean regions, which are deficient in OM (1%–3%). Supplying the nutrients N, P, and K, these organic amendments can significantly improve the mineral nutrient status and growth of plants in such soils (Mbarki et al., 2020; Hafez et al., 2021). There is also substantial evidence that applying compost as a soil amendment in areas using saline water for irrigation can decrease soil salinity levels and improve agricultural production (Kalanaki et al., 2020; Khan et al., 2022). Similarly, Farrag et al. (2021) showed that the yield of an important feed crop in Egypt, blue panic grass, irrigated with saline water, is maintained without reducing the already limited freshwater availability. The primary constraint associated with the repetitive use of CO is the contamination with heavy metals in soil which is of concern in agriculture production due to phytotoxicity and environmental health of soil organisms. Increasing its concentration beyond the threshold limit will impose several harmful impacts on plant constituency, thereby adversely influencing soil fertility and development (Arif et al., 2016; Kabasiita et al., 2022; Sahebdehfar et al., 2022). It has been suggested that plant responses to soil amendment are soil-type specific (Mbarki et al., 2008; Guron et al., 2021).

Alfalfa (*Medicago sativa* L.) is an important perennial forage legume species that widely cultivated due to high feeding values (Radović et al., 2009). Alfalfa provides  $\text{N}_2$  benefits to the soils, increases organic matters (OMs), and decrease the reliance on  $\text{N}_2$  fertilizers (Song et al., 2021). This plant exhibits tolerance to multiple stresses including drought, heat, and oxidative stress (Ma et al., 2021). Thus, it can be used as a promising resource for breeding programs with enhanced tolerance to multiple abiotic stresses. Sorghum [*Sorghum bicolor* (L.) Moench] is a fodder crop species. The position of feed quality after maize that is largely cultivated (Ogbaga et al., 2014). Sorghum excellent source of silage, wet forage, dried fodder, and direct grazing as pasture (Galyuon et al., 2019). Sorghum is sensitive to abiotic stress compared to other fodder species. Therefore, the necessity of tolerant forage genotypes or varieties able to cope with complex stress has become a significant challenge for global crop production (Bhardwaj et al., 2010; Tian et al., 2021; Rizvi et al., 2021 and, 2022). Accordingly, the improvement of

abiotic stress tolerance in crops is necessary, along with environmentally friendly approaches are highly demandable by which abiotic stress can be mitigated without influence on yield potentials. Considering these issues, this study focuses on mitigation of drought stresses using municipal solid waste compost and farmyard manure that regulate biomass yield, mineral nutrition under salinity in alfalfa and sorghum, and remediate toxic pollutants from soils.

## Material and methods

### Experimental design, plant cultivation and treatment

A completely randomized design (CRD) was set for these experiments. The viable seeds of alfalfa (*Medicago sativa* L.) and sorghum [*Sorghum bicolor* (L.) Moench] were collected from the Centre of Biotechnology Borj Cedria in Tunisia. The sandy soils were collected from the region of Nabeul (Tunisia) and the soil collecting area was top horizon (0–20 cm). This soil was air-dried for a few days, sieved at 2 mm, and mixed with municipal solid waste compost (CO) with or without livestock farmyard manure (M). The experiments were carried out in controlled environments (air, light, relative humidity and temperature) of a greenhouse. Plant seeds were surface sterilized using 70% ethanol and washed properly with deionized water. The seeds were sown in soil pots filled with 2 kg of dry soil supplemented with the same dose of compost or manure (0 and 40 t livestock farmyard manure ha<sup>-1</sup> Eq. (13.33) g/kg), respectively. The two different concentrations of salt (50 or 100 mM NaCl) were prepared and irrigated daily to the combination of CO or M containing soils. The pot soil field capacity (FC) was maintained at 66%. Total nine treatments were applied: control (C); municipal solid waste compost (CO); livestock farmyard manure (M); control with 50 mM NaCl (C + NaCl1); municipal solid waste compost with 50 mM NaCl (CO + NaCl1); livestock farmyard manure with 50 mM NaCl (M + NaCl1); control with 100 mM NaCl (C + NaCl2); municipal solid waste compost with 100 mM NaCl (CO + NaCl2); livestock farmyard manure with 100 mM NaCl (M + NaCl2). These treatments were considered for three consecutive harvests (H1, H2 and H3) of alfalfa and sorghum. Total thirty-five days were prolonged for each harvest time. At least three individual biological replications were maintained for each treatment.

### Soil and MSW analytical methods

Total C and N content were determined using a Vario micro-cube analyzer (Elementar Analysen Systeme GmbH, Hanau, Germany (Table 1). The soil and compost samples' pH and

EC (electrical conductivity) were determined after 1 h extraction in demineralized water (1/5; w/v) followed by filtration.

The elemental concentration was determined using the protocol (Das et al., 2021). Alfalfa and sorghum plant samples were dried at 80°C in a microwave, and the dried root and shoot samples were completely digested using a ternary solution (HNO<sub>3</sub>, H<sub>2</sub>SO<sub>4</sub>, and HClO<sub>4</sub> in 5:1:1 ratio). However, the same steps were followed for soil sample drying and digestion. The minerals and HMs concentration were determined using an inductively coupled plasma spectroscopy (ICP, Jarell-ash 9000 plasma spectrometer). At least three individual biological replications were considered for each treatment.

### Plant material analysis

The aerial parts and roots were washed, put in paper bags and oven-dried at 65°C for at least 4 days. Once dry, the plant parts were weighed to determine the total dry matter. Plant samples were then ground to a powder and digested with 0.5% nitric acid to determine the content of Na, K and P by flame photometry. Tissue heavy metals were determined using a nitric acid digestion procedure and inductively coupled plasma spectroscopy (ICP, Jarell-ash 9000 plasma spectrometer). Tissue N was determined using the Kjeldahl method (Brookes et al., 1985). Dried plant material (30 mg) was digested in H<sub>2</sub>SO<sub>4</sub>-H<sub>2</sub>O<sub>2</sub>, followed by 0.01 M NaOH, distillation into boric acid and quantitative titration with 10 mM HCl.

### Statistical analysis

Statistical analysis was performed for all the physiological indices using the statistical software, ver. 12.1 (Statsoft Inc. 2012). Data were subjected to one-way ANOVA, and means were compared using Tukey's multiple range test (TMRT) with  $p < 0.05$  used as the value for significance. Canoco 5, was used for PCA (principal component analysis) calculated from centred (not standardized) data.

## Results and discussion

Amendment of agricultural soils with municipal solid wastes (MSW) furnishes a valuable source of plant nutrients and organic matter. The availability of marginal quality water resources, such as drainage water or high salt containing groundwater, is becoming an important issue in Tunisia and other countries with scarce water resources. The CO amendment has been investigated for its effectiveness in soil remediation and plant yield improvement under saline conditions. Such amendment may foster nutrient availability and plant growth (Mbarki et al., 2020). The negative effects of salt can be reduced by choosing

**TABLE 1** The characteristics of clay soil, farmyard manure (M), and municipal solid waste compost (CO). Data are means of three repeats  $\pm$ SE. nd = not determined.

| Characteristics               | Unit                     | Sandy soil        | Compost                        | Manure                         |
|-------------------------------|--------------------------|-------------------|--------------------------------|--------------------------------|
| pH                            |                          | 7.9 $\pm$ 0.37    | 8.12. $\pm$ 0.01               | 8.3 $\pm$ 0.35                 |
| CE                            | $\mu$ S cm <sup>-1</sup> | 115.25 $\pm$ 7.71 | (3.43 $\pm$ 0.44) $\times$ 103 | (8.18 $\pm$ 0.22) $\times$ 103 |
| Total nitrogen                | g.kg <sup>-1</sup> DW    | 0.37 $\pm$ 0.03   | 25 $\pm$ 2.03                  | 1.15 $\pm$ 0.04                |
| Organic matter                | W/W %                    | 0.35              | 35.7                           | 1.03                           |
| C/N                           |                          | nd                | 11.2                           | nd                             |
| NH <sub>4</sub> <sup>+</sup>  | mg.kg <sup>-1</sup> DW   | 0.028 $\pm$ 0.007 | 0.02 $\pm$ 0.004               | 0.12 $\pm$ 0.03                |
| NO <sub>3</sub> <sup>-</sup>  | mg.kg <sup>-1</sup> DW   | 11.2 $\pm$ 2.03   | 350 $\pm$ 20.4                 | 42 $\pm$ 3.2                   |
| P <sub>2</sub> O <sub>5</sub> | g.kg <sup>-1</sup> DW    | 0.24 $\pm$ 0.025  | 26.4 $\pm$ 2.16                | 11.2 $\pm$ 8.24                |
| Potassium                     | g.kg <sup>-1</sup> DW    | 0.14 $\pm$ 0.02   | 7.1 $\pm$ 1.2                  | 0.52 $\pm$ 0.05                |
| Zinc                          | mg.kg <sup>-1</sup> DW   | 44.24 $\pm$ 4.7   | 307.09 $\pm$ 7.51              | 133.68 $\pm$ 7.76              |
| Copper                        | mg.kg <sup>-1</sup> DW   | 12.59 $\pm$ 2.19  | 107.94 $\pm$ 12.3              | 47.53 $\pm$ 4.39               |
| Cadmium                       | mg.kg <sup>-1</sup> DW   | 0.53 $\pm$ 0.001  | 7.78 $\pm$ 0.17                | 4.7 $\pm$ 0.056                |
| Lead                          | mg.kg <sup>-1</sup> DW   | 18.66 $\pm$ 1.87  | 118 $\pm$ 11.23                | 52.81.0 $\pm$ 65               |
| Clay                          | %                        | 5.0               | nd                             | nd                             |
| Limon                         | %                        | 2.0               | nd                             | nd                             |
| Sand                          | %                        | 93.0              | nd                             | nd                             |

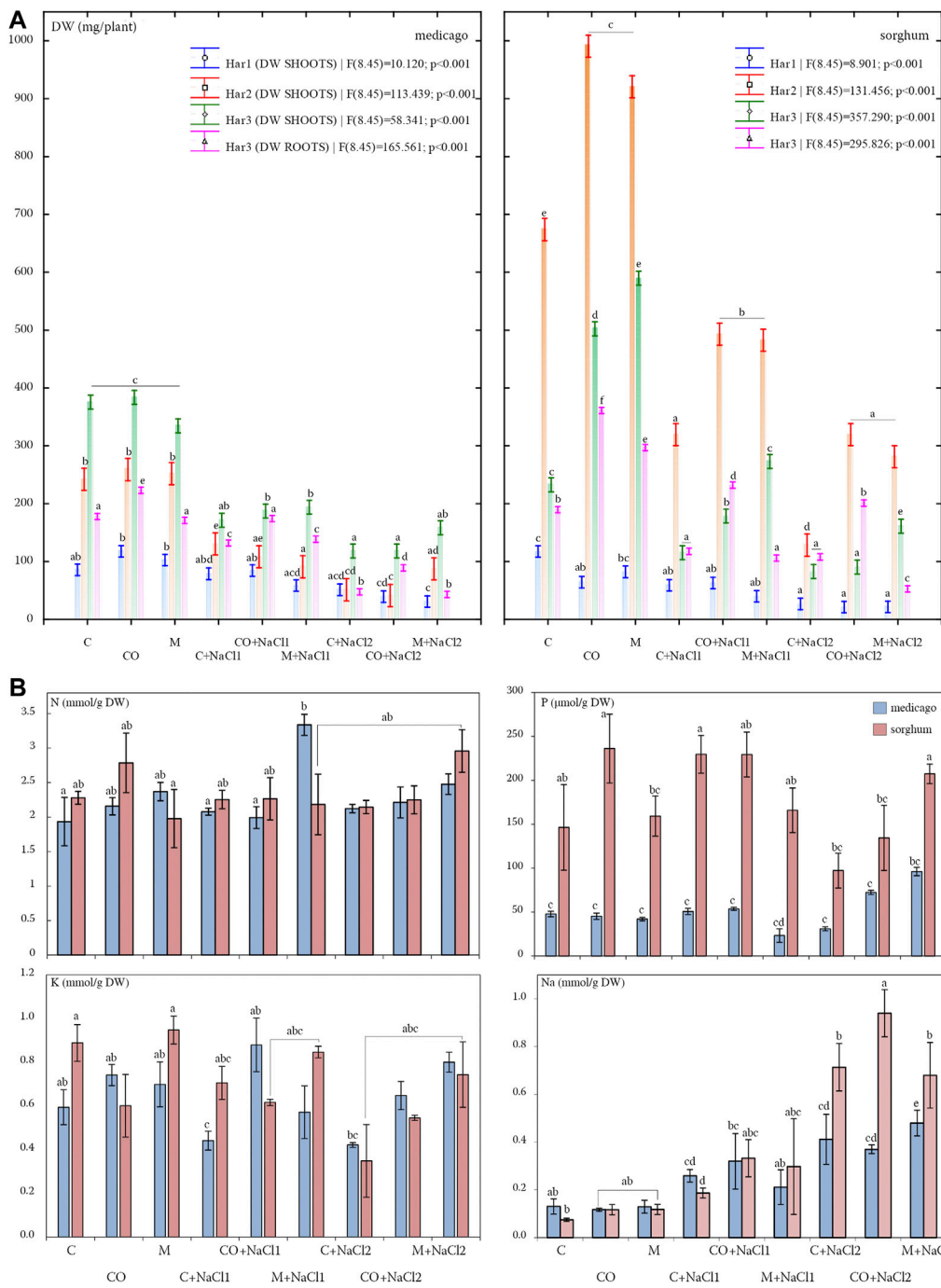
tolerant plants and good agricultural management. Abdrabouet al. (2022) reported that adding organic fertilizers to coarse soils increases the ability of quinoa plants to resist saline water and allows using lower quality water to irrigate these valuable plants. The interaction between two factors, compost and salinity of irrigated water, was studied in this research by using two different forage crops, one legumes plant (alfalfa) and the second cereals plant (sorghum) cultivated in sandy soil. Total dry matter, nutrient content and heavy metals phytoextraction have been studied.

## Effect of municipal solid waste compost and farmyard manure on plant biomass

Alfalfa growth was increased in H2 and H3 after supplementation of the highest CO and M in alfalfa. The H2 harvest showed the highest plant biomass yield in alfalfa (Figure 1) in H harvests. The shoot dry weight of sorghum with CO or manure amendment was lower than control at harvest H1 but increased at harvest H2 and H3 (Figure 1A). The interaction of plant species and fertilizer treatment in the absence of salt stress significantly affected plant growth at all three harvests ( $p = 0.003$  for H1 and  $p < 0.001$  for H2 and H3). Under irrigation with 50 mM and 100 mM NaCl, however, this interaction was not significant at H1 ( $p > 0.05$ ) but highly significant at H2 and H3 ( $p < 0.0001$ ) where the shoot growth was decreased by saline treatment. This effect was more pronounced in plants grown with manure and NaCl-2 watering (Figure 1A). For both crops, the most negligible root dry weight (DW) was recorded at salt condition NaCl<sup>-2</sup> (with and

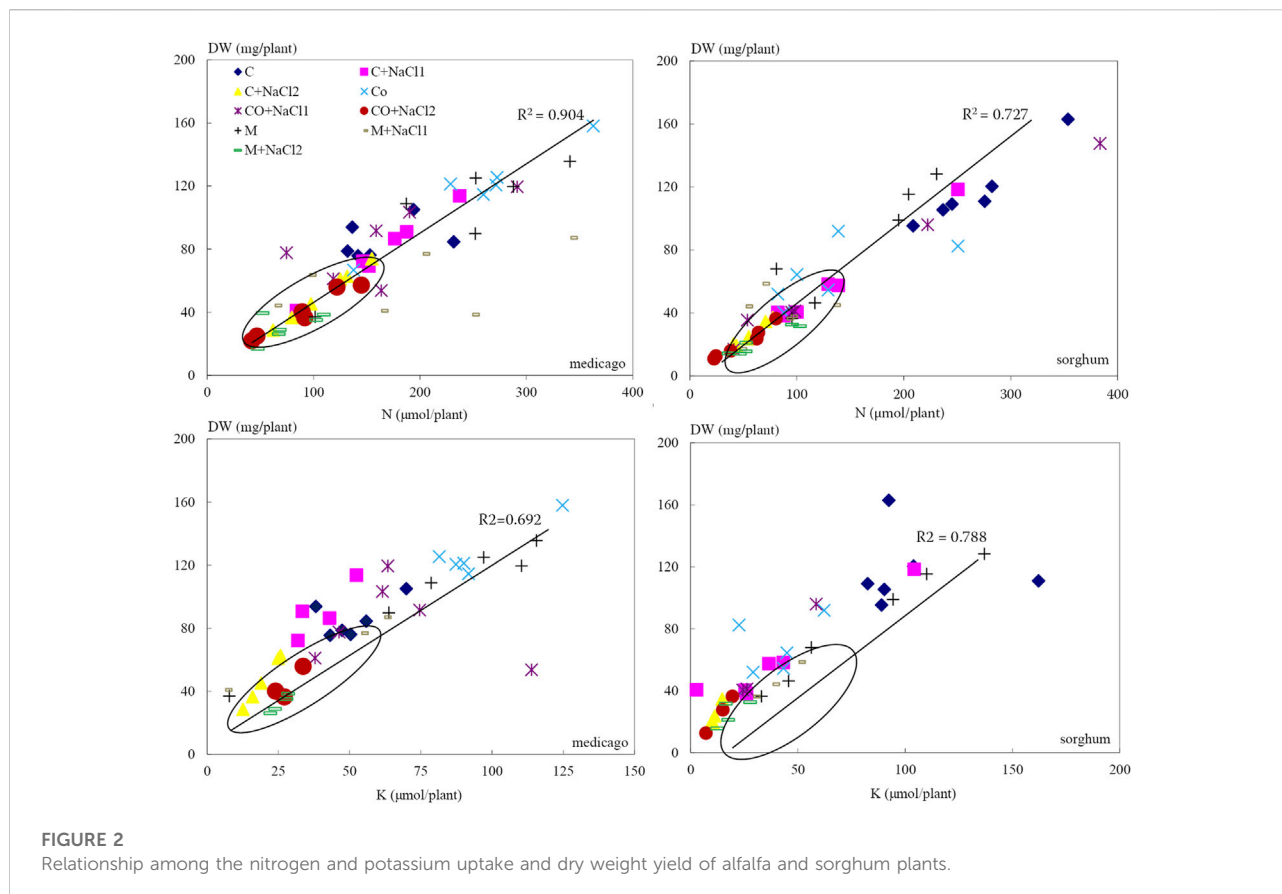
without manure), while the highest root DW was recorded with added CO (Figure 1A). The cumulative DW of sorghum shoots in CO-amended soil was higher than in control plants, and we noted a beneficial effect of the compost on salt stress. In contrast, there was no significant effect of CO on the cumulative DW of alfalfa under salt stress. The soil type was also crucial in determining a crop's response to compost (Mbarki et al., 2008). Similar effects of compost were reported based on experiments in which the soil amendment acted as a slow-release nitrogen source (Mahdy 2011; Singh et al., 2022). Guindo et al. (2022) demonstrated that applying a microdose of 2.5 t ha<sup>-1</sup> of compost, regardless of the kind, increased growth rates, plant height, grain yield, and biomass yield by 15%, 18%, 47%, and 27%, respectively, above controls. Mbarki et al. (2008) reported that MSW Compost, the shoot biomass cumulated over four cuts, was 20%–25% augmented upon CO addition in clay soil, independently of MSW Compost dose.

At 100 mM NaCl concentration, the aerial parts tended to be more reduced (50%) compared with control plants. The shoot growth was decreased by saline treatment (45.88% and 68.80% at 50 mM and 100 mM NaCl for alfalfa; 51.75 and 76.77 at 50 mM and 100 mM NaCl for sorghum). Salt accumulation in the root zone leads to the development of osmotic stress by the accumulation of toxic levels of Na<sup>+</sup> and Cl<sup>-</sup>. It disrupts cellular ion homeostasis by inhibiting the uptake of essential nutrients such as K<sup>+</sup>, Ca<sup>2+</sup>, and NO<sub>3</sub><sup>-</sup> and accumulating toxic levels of Na<sup>+</sup> and Cl<sup>-</sup>. At the whole-plant level, the effects of salinity are reflected through declines in growth, reductions in yield and adverse effects on gas exchange, photosynthesis, and protein synthesis (Ullah et al., 2019). The root decline was (25.77 and 73.27 for alfalfa; 37.93 and 42.93 for sorghum, respectively, for the two salt



**FIGURE 1**

(A) Effect of municipal solid waste compost (CO) and farmyard manure (M) supplementation on biomass yield in alfalfa and sorghum. The treatments indicate, control (C); municipal solid waste compost (CO); livestock farmyard manure (M); control with 50 mM NaCl (C + NaCl1); municipal solid waste compost with 50 mM NaCl (CO + NaCl1); livestock farmyard manure with 50 mM NaCl (M + NaCl1); control with 100 mM NaCl (C + NaCl2); municipal solid waste compost with 100 mM NaCl (CO + NaCl2); livestock farmyard manure with 100 mM NaCl (M + NaCl2). The H1, H2, and H3 presents the consecutive harvests. The different letters above the bar columns indicate significant differences of mean  $\pm$  SE at  $p < 0.05$ . Figure 1. (B) Accumulation of nitrogen, potassium, phosphorus and sodium in alfalfa and sorghum under saline soil with municipal solid waste compost (CO) and farmyard manure (M) supplementation on biomass yield in alfalfa and sorghum. The treatments indicate, control (C); municipal solid waste compost (CO); livestock farmyard manure (M); control with 50 mM NaCl (C + NaCl1); municipal solid waste compost with 50 mM NaCl (CO + NaCl1); livestock farmyard manure with 50 mM NaCl (M + NaCl1); control with 100 mM NaCl (C + NaCl2); municipal solid waste compost with 100 mM NaCl (CO + NaCl2); livestock farmyard manure with 100 mM NaCl (M + NaCl2). The different letters above the bar columns indicate significant differences of mean  $\pm$  SE at  $p < 0.05$ .

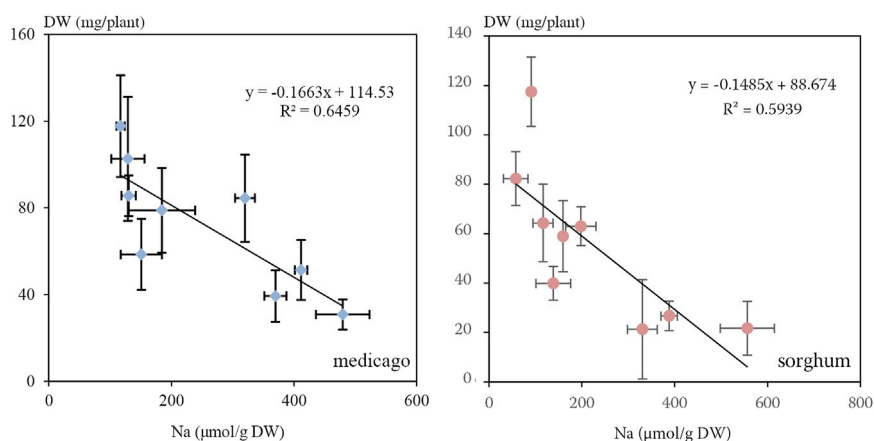


doses. Our results demonstrated that *M. sativa* cultivar Gabes could maintain growth and minimize the influence of salt stress. In particular, experiments with salt-tolerant varieties revealed that they experienced much smaller decreases in DW of aerial parts. Humic acids may have a hormone-like effect on plant development or they may have plant growth regulators adsorbed onto them that affect growth, as seen by the growth responses to salt stress (Ouni et al., 2014).

## Plant nutritional status

Accumulation of  $N_2$ ,  $K^+$ ,  $P$  and  $Na^+$  ANOVA analysis of  $N$  and  $K$  content showed significant effects of treatments ( $p < 0.05$ ). However, the impact of species was non-significant ( $p > 0.05$ ), while the interaction of treatment  $\times$  species was significant ( $p < 0.05$ ). The interaction between compost treatment and salt stress significantly affected  $N$  and  $K$  content in aerial parts (Figure 1B). We also found statistically significant effects of fertilizer treatment, species, and their combined effect on  $P$  content (Figure 1B). The uptake of potassium through fertilization was improved by compost amendment, which offset the impact of salt on the competition for ions at the level of the adsorption sites on root hairs that

reduced potassium uptake. An increase in  $Na^+$  led to a significant reduction in  $K^+$  levels in the aerial parts of both plant species. Similar results were reported (Khan et al., 2022) in the radicals and plumules of maize. Soil amendment with CO or manure increased  $K$  content in plants grown with saline irrigation. There was a highly significant effect of fertilizer treatment, species, and their interaction ( $p < 0.0001$ ) on the  $P$  content of shoots. The presence of  $NaCl$  in the nutrient solution resulted in an increased concentration of  $P$  content in leaves (Lacerda et al., 2006; Giuffrida et al., 2009). The mineralization of the MSW compost provided another source of slow-release organic phosphorus, which significantly improved the continuous  $P$  nutrition of the plants (Horta 2019). Concerning the relationship between plant DW and the accumulation of  $K$  and  $N$  in the shoots of both plants cultivated on sandy soil (Figure 2), there was a positive correlation between shoot growth and  $N$  content for *M. sativa* ( $R^2 = 0.90$ ) and *S. bicolor* ( $R^2 = 0.72$ ) and  $K$  content for *M. sativa* ( $R^2 = 0.69$ ) and *S. bicolor* ( $R^2 = 0.78$ ) (Figure 2). These results suggested that the growth rate was closely tied to the supply of essential nutrients for both crops. It is also probable that the symbiotic fixation of  $N$  would have contributed to the nutritional status of *M. sativa* at later stages in its development. The linear relationship between  $N$ ,  $P$ , and  $K$  soil nutrients and



**FIGURE 3**  
Relationship between sodium uptake and dry weight yield of alfalfa and sorghum plants.

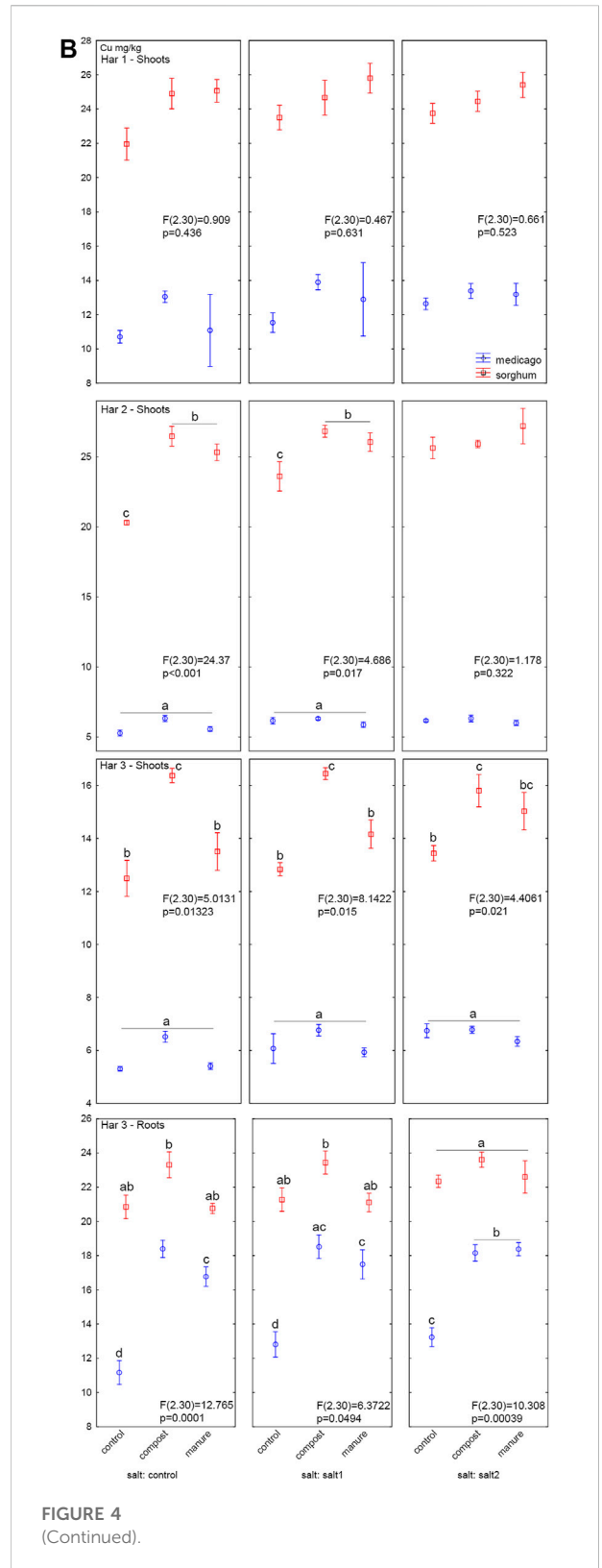
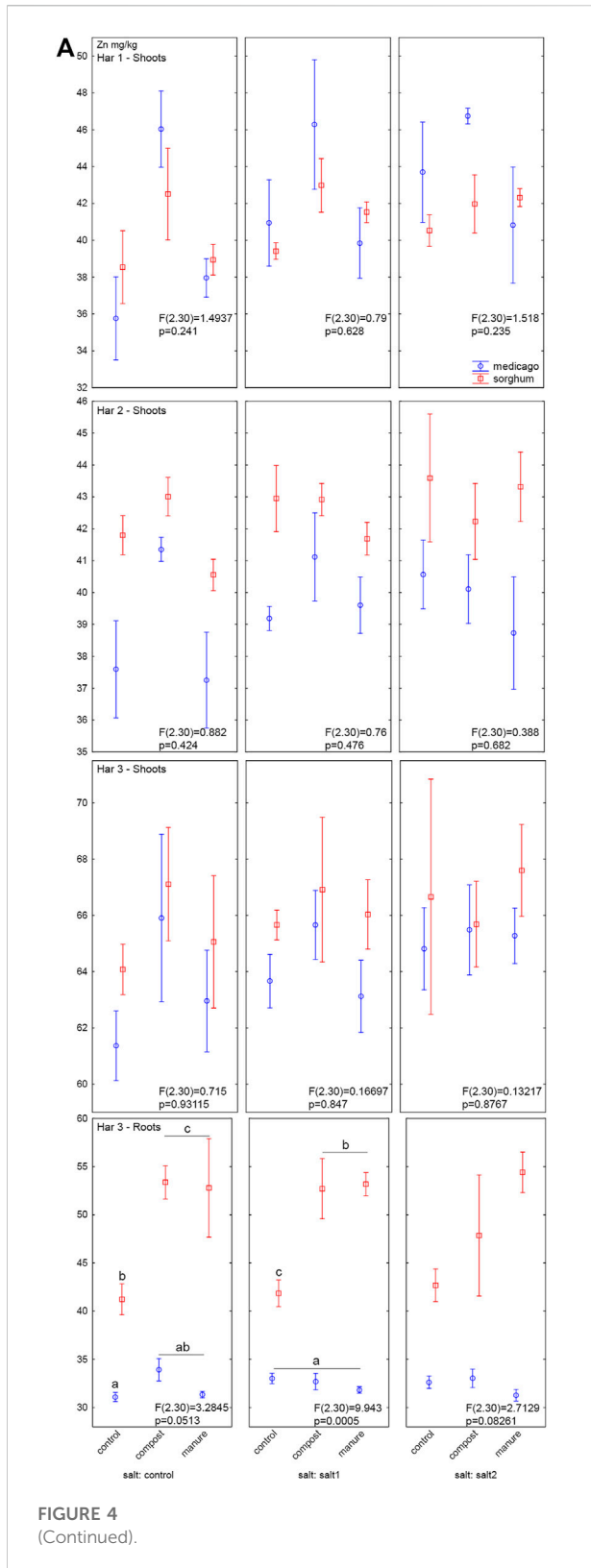
the photosynthesis parameters,  $P_n$ , and chl F, showed how increasing soil fertility improved the physiological performance of sorghum (Ahmed et al., 2020). Our results follow a previous study by Oueriemmi et al. (2021), demonstrated that adding organic amendments improved soil organic matter, cation exchange capacity, and the amount of P, Ca, Mg, and K that was accessible for use, as well as grain production (up to 51%) and nutritional content in the barley plants.

The decreased growth of plants in the presence of salt is thus related to the reduced uptake of N and K at the high dose of salt (Figure 3). The role of K in osmotic regulation and its competitive effect on Na are critical factors for overcoming salt stress (Thu et al., 2017). Potassium contributes to the survival of plants exposed to various abiotic stresses. Its deficiency can significantly affect root morphology, linearly increasing the root DW and the shoot and total DW production (Wang et al., 2013). Soil N and K availability were critical for plant growth and nutrient uptake under salt-water irrigation in sandy soil culture substrates (Mbarki et al., 2018). Grattan and Grieve (1992) indicated that too many ions could interfere with plants' ability to absorb nutrients in solutions, either directly through ionic competition or indirectly through the reduced osmotic potential of the solution, which would reduce the bulk flow of mineral nutrients to the root. The availability and uptake of N, P, and K were impacted by the rate and type of the organic amendments that were studied (van Zwieten et al., 2010). Supplying organic amendment can promote growth and mineral nutrient status under salt stress. However, This is dependent on the saline level and nutrient state of the soil, as well as the plant species' tolerance to salt. The availability of nutrients might not aid growth since the salt stress is more severe than any nutritional shortage (Grieve and Grattan 1999).

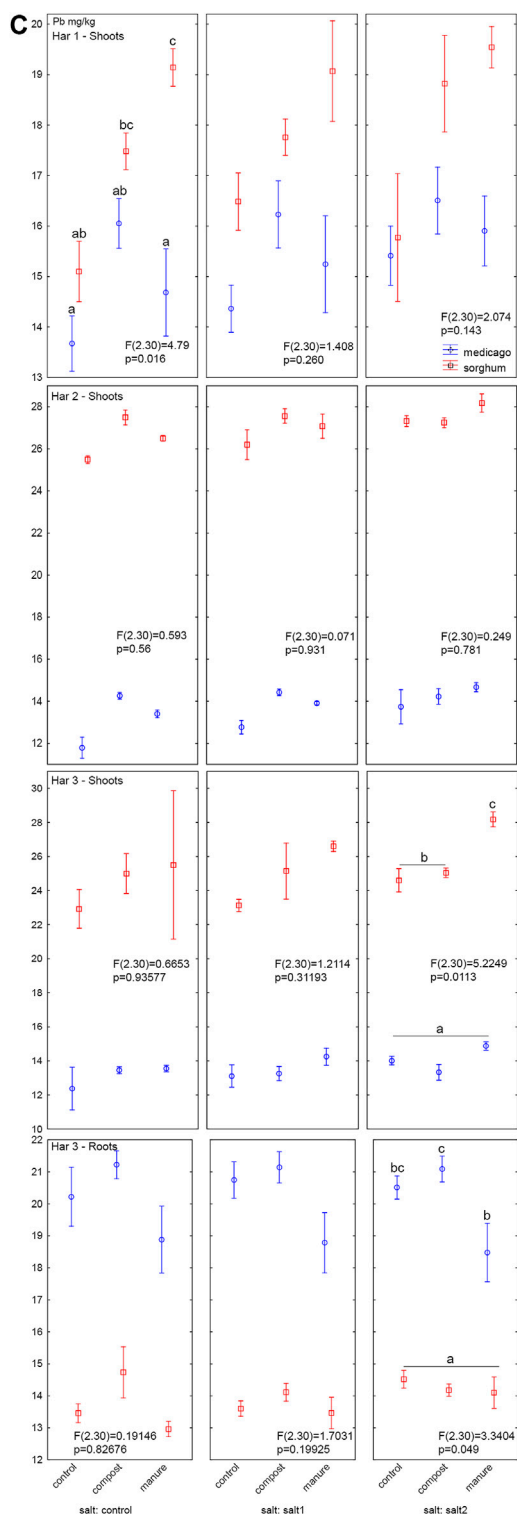
## Sodium concentration

The sodium concentration in shoots was higher in plants cultivated in the presence of salt. The  $\text{Na}^+$  content in this tissue was significantly affected by salt stress, bio-amendment, crop species and their interaction ( $p < 0.001$ ). Na content was higher in the presence of compost or manure, depending on the NaCl level applied. Shoot Na content was higher in sorghum than alfalfa at the same salt concentration in soil (NaCl-2) but strongly depended on the type of bio-amendment applied (Figure 1B). The maximum level of Na attained was about  $1 \mu\text{mol g}^{-1}$  of dry matter at 100 mM NaCl. Salt stress can directly affect nutrient uptake because  $\text{Na}^+$  reduces  $\text{K}^+$  uptake and  $\text{Cl}^-$  can reduce  $\text{NO}_3^-$  uptake. These effects can lead to complex interactions that affect plant metabolism, susceptibility to injury, and internal nutrient requirements. The negative influences of salts on crop plants can reduce their nutrient use efficiency and consequently inhibit growth (Fageria et al., 2011).

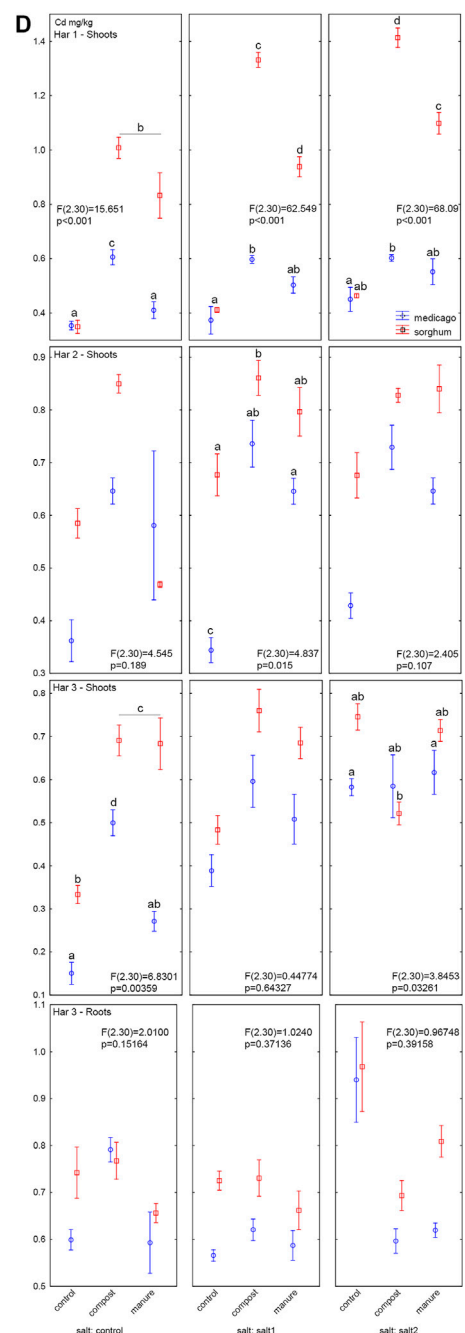
We found a negative correlation between shoot dry matter and Na uptake ( $R^2 = 0.65$ ,  $R^2 = 0.57$  for *M. sativa* and *S. bicolor*, respectively) (Figure 3). However,  $\text{Na}^+$  was not retained in the exchange sites in sandy soil, and the sodium adsorption ratio remained low. In order to the plant defend itself from salt stress, it must either restrict  $\text{Na}^+$  entry through the roots or regulate  $\text{Na}^+$  concentration and distribution once it has moved in (Hanin et al., 2016). Negrão et al. (2017) reported that the most significant plant adaptation to salinity was the ability to restrict the transportation of  $\text{Na}^+$  to leaves and its accumulation there. Rekaby et al. (2020) showed that adding organic soil amendments improved soil fertility by increasing the availability and uptake of the primary macronutrients and enhancing chlorophyll biosynthesis in barley plant tissues. This may be the reason for barley's increased ability to tolerate salinity, which has a more significant impact on sandy soil because this soil contains low levels of minerals and organic colloids (Papafilippaki et al., 2015). Das et al. (2013) reported that



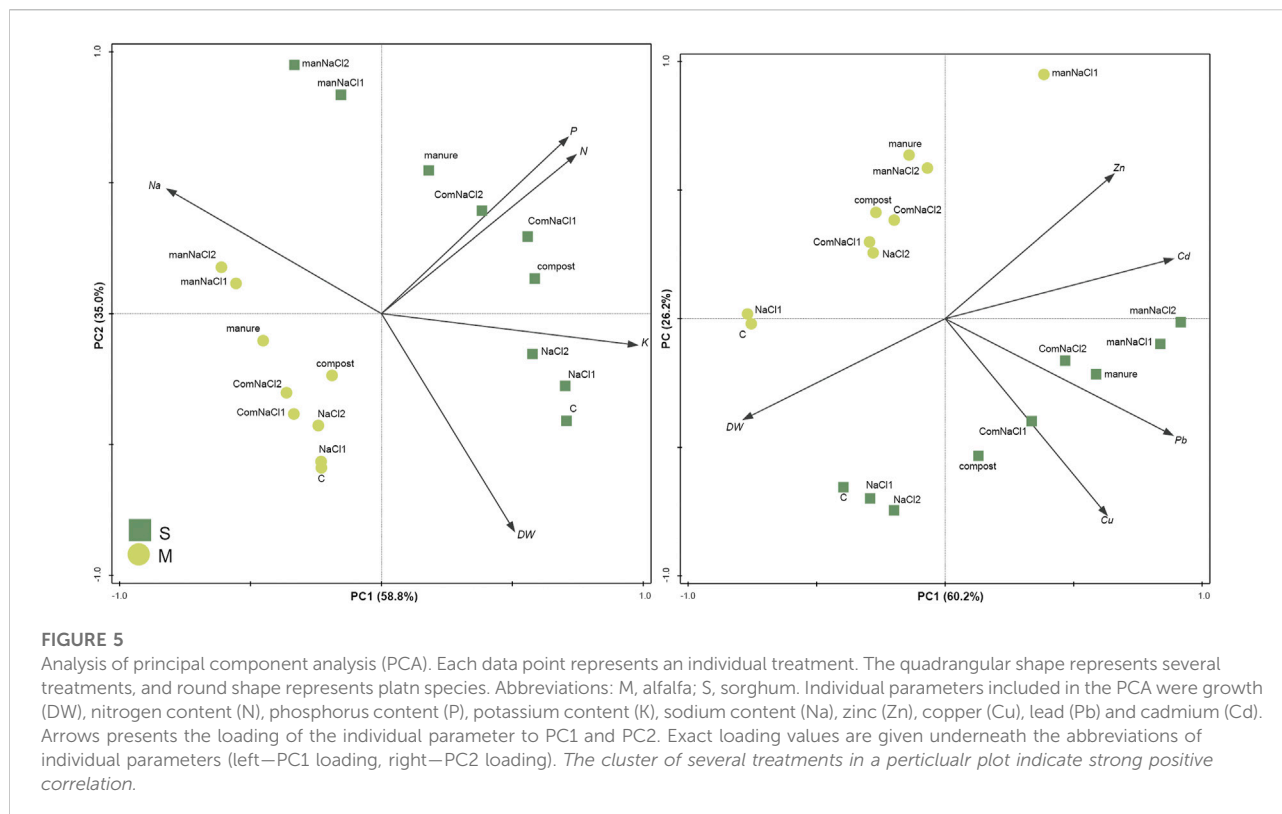




**FIGURE 4**  
(Continued).



**FIGURE 4**  
(Continued). Zinc (Zn-A), copper (Cu-B) lead (Pb-C) and cadmium (Cd-D) concentration (mg/kg DW) in alfalfa and sorghum shoots grown and harvested at H1, H2, H3. Shoots and roots from plants grown on sandy soils with or without MSW compost (CO) or farmyard manure (M) addition and irrigated with water containing 0 mM, 50 mM (NaCl-1, Salt 1), or 100 mM NaCl (NaCl-2, Salt 2). Data are means of six repeats  $\pm$ SE. The different letters represent significant differences between treated samples at  $p < 0.05$ , by Tukey's post hoc test.



soil amendment with farmyard manure conferred tolerance to salinity in maize by increasing chlorophyll content and  $K^+/Na^+$  ratios. Similarly, a study on sorghum plants found that the application of manure mitigated the deleterious impact of saline water on the growth of sorghum by increasing the nutrient content (Murtaza et al., 2020).

## Effect of fertilizer treatment on heavy metal content in shoots

Zn and Cu, essential plant micronutrients, were increased in the presence of compost and with increasing NaCl concentration of soil (level NaCl-1 to NaCl-2). Irrigation with 100 mM NaCl increased the phytoextraction of Zn and Cu by both species (Figure 4), and the concentration of Zn and Cu was higher in sorghum than in alfalfa ( $p < 0.05$ ). The distribution of Cu, Zn, Pb and Cd (Figure 4) in shoots and roots depended on the type of heavy metal element, the plant species and the harvest time (H1, H2, and H3). However, ANOVA revealed a significant effect of treatment  $\times$  species ( $p < 0.05$ ) on Cu, Zn, Pb and Cd at some harvest periods, the data have very high standard errors and standard deviations (95%), and the  $p$ -value is close to 1. The Cu concentration in the shoots of both crops was lower than in roots, and ANOVA showed no significant effect of treatment or species on Cu content in shoots. Sorghum accumulated more Cd than alfalfa ( $p < 0.05$ ). Even though ANOVA didn't reveal a significant

effect of treatment ( $p = 0.054$ ), the Cd concentration was higher in the shoots of plants grown in the presence of an organic soil amendment. The plant garners the essential heavy metals (Zn, Cu, Mg) from the soil solution due to concentration gradients and selective uptake of these metals (Peralta-Videa et al., 2009). Adding bio-solids and cow manure improved the ability of ornamental sunflowers grown in salty soil to accumulate significant amounts of heavy metals (Yazdanbakhsh et al., 2020). However, soil amendment did not increase the concentration of metals in the harvestable tissue of *S. bicolor* during the crop cycle. Sorghum has been proven to have the potential for the phytoremediation of heavy metals. Still, in some cases, the organic matter appeared to bind these elements making them less available for plant uptake (Marchiol et al., 2007). Shoot analysis showed no heavy metal toxicity as the concentration was less than the phototoxic level. Because of their low buffering capacity and inability to adsorb metals, sandy soils are considered poor candidates for the remediation of metal-contaminated waste used as an organic amendment (McBride 2003).

## Principal component analysis for traits

PCA was performed to reveal the potential relationships among salinity, soil bio-amendment, heavy metal phytoextraction, and nutrient content (PC1). This analysis explained 58.8% of the total variability between traits and

plant species. It was associated with elevated loading of the macronutrients N, P and K, high positive P and N content and high negative loading of Na<sup>+</sup> content (Figure 5A).

The species at the lower end of PC1 was *M. sativa* which retained high levels of Na<sup>+</sup> in leaves, while at the higher end was *S. bicolor*, with higher levels of N, P and K content. PC2 accounted for an additional 35% of the total variability among plant traits, which appeared to be related to the ion content, the N-P level, and the growth traits (Figure 5A). The accessions with high growth parameters and higher accumulations of Na<sup>+</sup> in the leaves were located at the lower end of PC2 (Treatment: Man NaCl-1, Man NaCl-2). In contrast, those with high leaf N and P content and high content of Na<sup>+</sup> in the leaves were located at the higher end of PC2. Vectors at right angles and opposites for Cu indicated no correlation and negative correlation, respectively.

In contrast, the second component showed that plant growth and leaf K content were negatively correlated with the other representative characteristics. Even though Na<sup>+</sup> reduced the availability of K binding sites for critical metabolic processes in the cytoplasm (Wei et al., 2017), Salinity tolerance may also be indicated by the accumulation of Na<sup>+</sup> in relation to biomass. Similar patterns in the two species point to the conservation of salinity tolerance-related features between them. Growth maintenance has been deeply recognized as a reasonable evaluate of salinity tolerance (Negrão et al., 2017). In the second graph (Figure 5B), DW is negatively influenced by plants' Pb and Cd content. Overall treatments, sorghum took up more Zn, Pb, Cu, and Cd than alfalfa with or without salt stress; therefore, sorghum could be used for the remediation of these metals. In general, both species were tolerant to low salinity (50 mM NaCl), but growth was most adversely affected with manure treatment under salt stress at 100 mM NaCl.

## Conclusion

The results of this study explore municipal solid waste compost (CO) or farmyard manure (M) mitigate salinity stress along with improved mineral nutrients and biomass yield in alfalfa and sorghum plants. These findings suggest that the two soil amendments had significant impact on soil. The alfalfa alleviates high Na<sup>+</sup> toxicity by enhancing several mineral nutrients including N<sub>2</sub>, K, P compared to sorghum. The sorghum crop accumulated higher Cd related to alfalfa after supplementation of two amendments. These findings suggest that M supplementation is effective to alleviate salt stress compared to CO in alfalfa, while sorghum can be recommended as to clean up heavy metals from the soils. This study also opens a link between minerals (N, P, and K) enhancing and ability to mitigate salinity stress in plants. Therefore, organic amendment-based ecofriendly approach could be

important to alleviate salinity stress in plants, and can be a promising approach for green environment and smart agriculture.

## Data availability statement

The datasets presented in this study can be found in online repositories. The names of the repository/repositories and accession number(s) can be found in the article/supplementary material.

## Author contributions

Conceptualization, SM, CA; Methodology, SM; Resources, OT; Investigation, SM and MS; Data curation, SM and PV; Formal analysis, PV and MS; Writing of the original draft, SM; Funding acquisition, I-A, VH, and FH; Manuscript revision and editing, I-A, AE, MAR; Supervision, PT and MS.

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## Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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