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Organic amendments affects soil concentration, accumulation and availability of copper in chilli (*Capsicum annuum* L.)"

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Metals are dangerous to both human health and the environment since they are poisonous elements with a density greater than 5 g cm^{-3}. The abundant and common heavy metals contaminating Agricultural lands are lead (Pb), cadmium (Cd) and chromium (Cr). Different organic amendments have been used to degrade and immobilize the metals with different efficiency. The present study was conducted to determine the impact of organic amendments on the accumulation, availability, and concentration of copper in chili (*Capsicum annum* L.). The experiment was run at the wirehouse at the University of Agriculture, Faisalabad Institute of Soil and Environmental Sciences. Different organic amendments, such as compost, farm manure, and chicken manure, were applied at 20 and 40 t ha^{-1} in an experiment that was set up in a completely randomized design (CRD). The Institute of Soil and Environmental Sciences soil farm provided surface soil (0–15 cm), which was collected. The soil was air dried and then spiked with 20 ppm $CuSO_4$ salt and kept for acclimatization for one month. Spiked soil was sieved through a 2 mm sieve. Twenty-one pots were filled with 14 kg of soil. Results revealed that maximum Plant height (cm), Total number of chili peppers per plant, Chilli peppers fresh weight per plant (g), Chilli peppers dry weight per plant (g), Chili peppers length per plant (cm), Shoot fresh and dry weight per plant (g), Root fresh and root weight per plant (g), Root length per plant (cm), No. of branches/plant, Number of leaves/plant, and Chlorophyll contents (SPAD value) were maximum where Compost 40 t ha^{-1} was applied while minimum in the control treatment. However, the lowest copper concentration in roots, shoots, leaves and branches was recorded with Compost @40 t ha^{-1} treatment. In addition, copper concentration in soil was observed at minimum in the control treatment and Maximum in Compost @40 t ha^{-1}. From this study, it was concluded that the application of Compost @40 t ha^{-1} amendment significantly improved the growth of chili by reducing the uptake of copper from the soil**.**

Keywords: Organic Amendments, Availability of Copper, Chili Crop

INTRODUCTION

Heavymetalsare **t**oxic elements having **a** densitygreater than cm^{-3} and are considered a danger to human health and the environment (Tchounwou et al. 2012). Theabundant and common heavy metals contaminating agricultural lands are lead (Pb), cadmium (Cd) and chromium (Cr) (Khan et al. 2018a). Heavy metal contamination of soil has turned into a serious issue in agriculture production throughout the world in the previous few years because of anthropogenic activities. For example, the removal of industrial waste and inappropriate utilization of heavy metal-improved resources in agriculture such as synthetic pesticides and fertilizers, sewage waste, industrial wastes and wastewater (Kuo et al. 2006; Ramadan and Al-Ashkar, 2007). While a few of them such as cadmium, chromium, lead and arsenic are similar to iron, zinc, manganese and copper, etc. are basically required in fewer quantities which are harmful throughout the food chain and might represent an extraordinary danger to animals, plants and human (Costa, 2000). A higher number of heavy metals in the soil would increase the take-up of these metals by plants. Along these lines, hazard evaluation of heavy metal collection in agricultural land is increased by the utilization of synthetic fertilizer, pesticides and natural wastes to soil (Papafilippaki et al. 2007; Zeng et al. 2011; Alengebawy et al. 2021).

The most environmentally friendly way of extracting metals is to accumulate heavy metals with rapid growth and high biomass yield. Extracting the heavy metals from the decomposed granulate is a safe way to reuse sewage sludge in agriculture and its reintroduction in the environment (Nunes et al. 2021).

There are only some plants that can grow for a short time in heavy metal-contaminated soils and produce sufficient biomass for economical and feasible extraction of the metals (Chaney et al. 2007; Baniet al. 2015). Unfortunately, most of the local metal-extracting plants in Europe did not produce enough biomass for the economical phytoextraction of the metal as reported by Simmons et al. (2015). But still, even the transgenic and exotic plants are not the guarantee for the Phyto-exaction of the metals because bio granulate might be itself a limiting factor in metal uptake and plant growth (van der Entet al. 2013; Hassan, 2013). It is critical to understand the properties of the heavy metals to check the application ability of phytoextraction (Baker et al. 2000). Because metals are bound to the soil and organic content which affect the redox potential of the metal existence in ionic and plant-available form (Tangahu et al. 2011). Plants can also lower the pH of the sediments and soils to increase the bioavailability of the metal (Tang and Rengel, 2003).

Plants grown on heavily metals contaminated soils showed stunted growth, less biomass production, abnormal metabolism and metal accumulation into plants which have adverse effects on human health (Khan et al. 2016; Kalaivanan and Ganeshamurthy, 2016). Organic fertilizers showed positive effects in relation to crop productivity and are an important source of plant micronutrients (Ahmad et al. 2019; Wajid et al. 2020).

Hot pepper (Capsicum spp.) evolved from South and Central America and domesticated from Africa to Asia via Europe (Tsaballa et al. 2015). It is a member of the Nightshade family (Solanaceae) and belongs to the Capsicum genus (Yadeta et al. 2011). This genus has 25 species of which only five are cultivated (Kraft et al. 2014). Other cultivated species of the genus Capsicum are *Capsicum frutescens* Mill, *Capsicum chinense*, *Capsicum baccatum* L. and *Capsicum pubescens* (Csilléry, 2006).

Due to its high economic and nutritional value, it is considered a cash crop of Pakistan (Howard et al. 2000) and shares 1.5% of Pakistan's GDP (GOP, 2017). At the national level, chilies occupy an area of 0.058 million hectares producing 0.037 million tonnes of chilies annually (Anonymous, 2020). Punjab and Sindh are major chilli-producing provinces contributing 96% of total chilli production in Pakistan (GOP, 2017). Area under chilies has decreased by 2.5% due to abiotic and biotic factors (Khaitovet al. 2019).

Hot Pepper has high nutritious value and is a rich source of antioxidants and vitamins such as A, C and E (Olatunji and Afolayanet al. 2018). It provides carotenoids, flavonoid and mineral elements and this quality make this the most important natural colorant of the food (Arimboor et al. 2015). It is an important condiment having many commercial and therapeutic values. The pungent flavor of hot pepper is due to volatile compounds called Capscinoides. Capsaicin is the most common capsaicin with marvelous pharmaceutical properties. The essential oil of *C. annum* has immunomodulatory and antibiotic properties (Valdivieso-Ugarteet al. 2019). It is helpful in treating certain diseases such as cough, toothache, heart problems, and cancer (Cortés-Ferré et al. 2021). Pepper extracts are used in cosmetics and pharmaceuticals. As well, as used in food as a spice, peppers have also a decorative attraction when grown in gardens or pots (Padilha and Barbieri, 2016).

Plant phenotypic characteristics such as fruit shape, color, anthocyanin content, and plant growth habits were used to differentiate pepper genetics and arrange it into groups (Stommel and Griesbach, 2008). Hot pepper is cultivated in large areas worldwide with different shapes and colors. Pepper shows variation in fruit shape, color, and size thus named as a heterogeneous plant (Hill et al. 2013). For the most part, pepper varieties are classified based on their fruit size, shape, color, and pericarp thickness (Naegele et al. 2016).

Different concentration of poultry manure has a different impact on the yield and biomass of the plants. The higher quantity of poultry manure (10%) produced a high spinach yield as compared to the 5% (Dikinya and Mufwanzala, 2010). Continuous application of poultry manure decreased the accumulation of lead and cadmium in rice grain by 7.2 to 59.4% and 7.8 to 79.3% respectively (Wan et al. 2020). The combination of poultry manure with $CaCO_3$ significantly reduced the bioavailability of zinc, lead and cadmium in acidic mixed chelator (MC)-washed soils (Guo et al. 2019). The objective of this current study was to observe the impact of organic amendments on the accumulation, and bioavailability of copper in chilies and to monitor the impact of organic amendments on copper concentration in soil.

MATERIALS AND METHODS

Experimental site:

The research trail was executed at the wirehouse of the Institute ofSoil and Environmental Sciences (ISES), UniversityofAgriculture Faisalabad. The study was performed to observe the effects of organic amendments on the availability, concentration and accumulation of copper in the chilies (*Capsicum annum* L.).

Soil sampling:

Surface soil (0-15 cm) was collectedfromthe farm of ISES. The soil was air-dried and then spiked with 20 mg kg^{-1} $CuSO_4$ salt and was kept for acclimatization for one month. Spiked soil was sieved through a 2 mm sieve. Twenty-one plastic pots were filled with 14 kg of soil per pot. The treatments consist of compost (Comp.), farm manure (FM) and poultry manure (PM) with 20 and 40 $tonha^{-1}$ (Figure 2). Five plants from chili nursery were transplanted in each pot and tap water was used for irrigation purposes.

Layout and Treatments:

Each pot was contained 14 kg of soil and arranged according to a Completely Randomized Design (CRD) with seven treatments and three replications. The following treatments were used as T1=Control (uncontaminated), T2=Cu spiked with poultry manure (PM) at 20 ton ha^{-1}, T3=Cu spiked with poultry manure (PM)at 40 ton ha^{-1}, T4=Cu spiked with farm manure (FM) at 20 ton ha^{-1}T5=Cu spiked with farm manure (FM) at 40 ton ha^{-1}T6=Cu spiked with compost (COMP) at 20 ton $ha^{-1}$and T7=Cu spiked with compost (COMP)at 40 ton ha^{-1}

The recommended dose (30:60:30 kg ha^{-1} NPK) of fertilizers (N, P and K) was applied for proper growth and development of chili by using the source of single super phosphate (SSP) andmuriateofpotash (MOP) respectively. Irrigation was applied according to the plants' requirements. Before conducting the experiment, a complete soil and water analysis was carried out in the soil and water chemistry laboratory (Tables 1 and 2).

Table 1: Physio-chemical properties of soil used for the experiment

Characteristics	Value	Unit
Ph	7.11	
Ece	1.71	dS m^{-1}
TSS	17.10	meq L^{-1}
SAR	7.86	$(mmol\ L^{-1})^{1/2}$
Na^+	10.00	mmol L^{-1}
CO_3^{2-}	0.50	mmol L^{-1}
HCO_3^-	8.00	mmol L^{-1}
Ca^{2+} + Mg^{2+}	2.00	mmol L^{-1}
Textural class	Sandy loam	
Sand	53.50	%
Silt	31.20	%
Clay	15.30	%
Saturation percentage	32.00	%
Bioavailable Cu	0.68	mg kg^{-1}

Electrical Conductivity (ECe) of Soil Extracts:

For this purpose, 250 g of soil sample was thoroughly saturated by using distilled water until the soil paste reflected the characteristics of standard soil paste. The paste was allowed to stand for 30 minutes and then rechecked the paste to add more water if needed. For soil extract, 100g of saturated soil paste was filtered by using a vacuum filtration system having Buchner funnels containing well-fitted What man No. 42 filter paper. Lovibondsen direct EC meter was used to test electrical conductivity (Model con.200). A 0.01 N 0 KC l0 solution 0 was 0 used to calibrate the electrical conductivity0meter. The Cell0constant (k) was derived using the following formula K=1.4118dSm^{-1}/Observed ECof 00.010NKCl(dS m^{-1}) whereas, Sample EC meter reading x Cell constant (k).

pHof Saturated Soil Paste:

For soil pH, air-dried soil was taken, and sieved from <2mm wire mesh to remove gravels or stones. Weighed 50 g of sieved soil on digital balance into a 100 ml beaker. Then 50ml, distilled water was added to that beaker with the help of a cylinder and assorted fine-through glass rods. The solution was allowed 30 minutes to settle down. At this time, the solution was stirred again. The PH meter was calibrated before using buffer solutions. After calibration, the pH meter combines electrodes about 3 cm deep into the solution for a total time of 30 seconds. Took the reading with 1 decimal. The combine electrode was removed after taking readings and washed cleanly in another beaker then dried with tissue paper for the purpose of removing extra water.

Soil bulk density:

The bulk density of the soil was measured by following the clod method. The volume of a clod is measured using this technique's foundation, the buoyancy principle, as follows; When a solid object with a density higher than water is submerged in water, a force equal to the weight of the water displaced is applied in an upward direction. So, if a solid item is weighed first in the presence of air (where the buoyant force can be regarded as minimal), then in the presence of water, The weight of the displaced water is represented by the difference between the two weights. The volume in cm^3 is equal to the weight in grams of the displaced water. A soil clod is covered with paraffin before being measured in order to prevent water from getting into the pores. The amount of water that the clod displaces is equal to the volume of the paraffin-coated object minus the amount of paraffin. Paraffin wax (melted wax) was used as a reagent.

Methodology:

Firstly, collect soil clod from all pots. If roots are present, carefully remove them with scissors before using a tiny copper wire to knot and secure the clod as you carefully weight it. The clod should be dipped in melted wax and dried for 30 minutes. To make the clod more water-resistant, additional melted wax was added. Next, coated clod and wire were weighted in the air. The clod was weighted while submerged in water using a balance that could accommodate the clod hanging from the balance beam by a thin copper wire. The weight loss is equal to the volume of the clod times the weight of the water that has been displaced. If there is no such balance available, totally submerge the clod in a graduated cylinder halfway full of water, then measure the water volume change in the cylinder (v).

Soil Texture:

The hydrometer method (Bouyoucos, 1962) was used to determine the particle size. In a 400 mL beaker, 40 mL of a 2 percent sodium hexametaphosphate $[(NaPO_3)_6]$ solution was added along with 40 grams of air-dried soil. The mixture was then shifted to a dispersion cup and swirled for 10 minutes. The dispersion cup's contentsweretransferredinto a 1000 mL graduated cylinder with a 1Lcapacity that was 36+2 cm tall. The cylinder was filled with distilled water until the hydrometer's volume reached 1000 mL. The hydrometer was taken out, and the cylinder's contents were manually shaken with a metal plunger. After achieving uniform suspension, the plunger was removed, and a hydrometer reading (HR1) was recorded after 4 minutes. After minimally disturbing the shaking process, the hydrometer was removed, and the second hydrometer reading (HR2) was collected after two hours. Since the hydrometer's calibration temperature is 68ºF (20ºC), the HR1 and HR2 were created as CHR1 and CHR2, respectively, to account for temperature variation

Figure 1: Soil texture analysis

For each degree above 20ºC, the reading was increased by 0.3, and for each degree below 20ºC, it was decreased by 0.3. The texture of the soil is represented in Figure 1Calculations involved are; Silt+Clay (%) = [(CHR1)100] / (weightofsoil), Clay (%) = [(CHR2)100] / (weightofsoil), Silt(%) = % (silt+clay) - (%)clay, Sand(%) = 100- [% silt+ %clay] and USDA textural triangle was used to determined soil textural class.

Preparation of saturated soil paste:

Soil (250 g) was taken in a 500ml beaker. Distilled water was added gradually while mixing carefully with the spatula. It was allowed for overnight soaking. On the next day, the saturated soil paste was prepared ensuring its criteria. At saturation, the paste flowed freely from the spatula when it was tilted, reflected light and there was no water standing in the beaker.

Soil saturated extract:

To evaluate the EC of saturated paste extract and soluble ions or salts, an extract of saturated soil paste was obtained. It was obtained by using an air pump to apply pressure. To avoid salt precipitation during storage, one drop of Sodium hexametaphosphate (2%) solution was added to each 25ml extract following the methods described by (Richards,1954).

Total soluble salts (TSS):

TSS ($mmolL^{-1}$) was created from the ECe using the relationship between ECe and TSS (U.S. Salinity Lab. Staff, 1954).

Soluble Ca^{2+} and Mg^{2+}:

10 ml of the extract was combined in a China dish with 1-2 drops of the Eriochrome Black T (EBT) indicator and 10 drops of the buffer solution (NH4OH + NH4Cl). This sample was titrated to a blue endpoint in comparison to a 0.01 N EDTA (disodium) solution. The following formula was used to compute the Ca2+ and Mg2+ which is Ca^{2+} and Mg^{2+} ($mmolL^{-1}$) = (mL of EDTA used x 0.01/mL of extract taken)x 1000

Soluble $CO_3{}^{2-}$:

Carbonates were measured using phenolphthalein as an indicator and titrating10 mlof saturationextractagainst0.010*N* H_2SO_4toacolorless endpoint. The carbonate concentration in the sample was determined using the formula: $CO_3{}^{2-}$(mmol L^{-1}) = [(2 x ml of H_2S0_4 used) × 0.01/ml0of extract0taken] x 1000

Soluble $HCO_3{}^-$:

Bicarbonateswere measured in the sample after CO_3 2- was calculated by titrating aliquot 0.01 *N* H_2SO_4 with methyl orange as an indicator to apinkish-yellow endpoint. Using methyl0orange0as0an0indicator.

HCO_3^- ($mmol_cL^{-1}$) = (ml of H_2SO_4 $_{used}$ for HCO_3) - (ml of H_2SO_4 used for $CO_3{}^{2-}$) × 0.01/mL of extract taken

Soluble Cl^- :

Following measurements of CO3 2- and HCO3-, the amount of soluble Cl- was determined by titrating the aliquot with 0.010 N0AgNO3 solutions to a brick-red endpoint's$^-$ (mmol $_c$ L^{-1}) = mL of $AgNO_3$ used x 0.01/ mL of extract taken

Sodium Adsorption Ration (SAR):

The SARwas determined using the followingformula: SAR (mmol L^{-1}) = $Na^+ / [(Ca^{2+} + Mg^{2+}) / 2]^{1/2}$ whereas all ion concentrations are measured in mmol L^{-1}.

Table 2: Analysis of irrigation water used for the experiment

Characteristics	Value	Unit
EC	0.46	dS m^{-1}
TSS	4.6	meL^{-1}
SAR	3.41	(mmol $L^{-1})^{1/2}$
RSC	1.06	me L^{-1}
Cu	0.32	mg L^{-1}
Cu	0.32	mg L^{-1}

Data Collection:

Data was collected for the following parameters at crop maturity (Figure 2).

Figure 2: Effect of organic amendments on plant growth of *Capsicum annum*.

Plant0height (cm):

At theend of the crop season, height was planted with the helpof measuring tape.

Total number of chili peppers per plant:

The total number of chili peppers was calculated by counting the number of fruits present in the plant.

Chilli peppers fresh weight per plant (g):

The weight of all chili peppers was determined by the electrical balance in grams.

Chilli peppers dry weight per plant (g):

After recording the fresh weight of chili peppers, these were sliced, air drying and then placed. Then, dry weight was recorded with the help of weighing balance.

Chilli peppers length (cm):

Five random chili peppers from each plant were selected. The length of these fruits was measured with measuring tape in centimeters. The average length of fruit was calculated by using the following formula Average length of fruit=sum of fruit length (cm)/5

Shoot fresh weight per plant (g)

The shoot of three plants was collected and weighed on digital balance in grams and the average value was computed.

Shoot dry weight per plant (g):

Shoots of three plants were collected, spread in a dry place and exposed to sunlight for 15 days. After that, the dry weight of the leaves was weighed on a digital weighing balance and the average value was computed.

Root fresh weight per plant (g):

The root of three plants was collected and weighed on digital balance in grams and the average value was computed.

Root dry weight per plant (g):

The roots of three plants were collected, spread in a dry place and exposed to sunlight for 15 days. After that, the dry weight of the leaves was weighed on a digital weighing balance and the average value was computed.

Root length per plant (cm):

Root lengths of three selected plants were recorded in centimeters from the emergence of roots to the end tip of the longest root by using the measuring tape and the average length was computed.

Number of branches per plant:

The number of branches per plant was counted manually from each plant. To count the branches, three plants were selected randomly at maturity and the total number of branches were counted and mentioned on average.

Number of leaves per plant:

The numberofleaves per plantwas quantitatively measured by counting leaves per plant. For this purpose, three random plants were collected from each experimental unit at maturity total leaves were counted and the average was computed.

Chlorophyll contents (SPAD value):

Chlorophyll content was determined from the upper leaves of plants of all treatments during daylight by using a chlorophyll meter (SPAD-502) and then the average was taken.

Table 3: Operation conditions employed in the determination of heavy metals by atomic absorption spectrophotometer

Elements	Wave length (nm)	Slit Width (nm)	Lamp Current (mA)	Burner Head	Flame	Burner Height (mm)	Oxidant gas pressure (Flow rate) (kpa)	Fuel gas Pressure (Flow rate) (kpa)
Copper	283.3	1.3	7.5	Standard type	Air-C_2H_2	7.5	160	7

Preparation of plant samples:
The following procedure was used to determine the copper in different plant parts: The plant samples (shoots, roots, leaves and branches) were firstly dried in two steps; the first step was sun-drying followed by the second step was oven-drying of the sample at about 65ºC. After drying, the dried samples were ground properly through a grinder. The samples which ground were put into a plastic zipper bag.

Plant Sample Digestion:
Di-acid (HNO_3+$HClO_4$) was used for plant sample digestion. To digest oven-dry samples 0.5g dried plant sample and 6mL concentrated HNO_3 was taken in flask and kept overnight. After that 3ml $HC1O_4$ was added to it and placed on a hot plate. It was heated slightly the first time and then strongly until white fumes were observed. These make the solution colorless. After cooling of digested plant sample was crossed from the filter paper. The extract was collected in a volumetric flask of 50ml and made the volume of 50mL by adding distilled water in it.

Soil sample digestion:
AB-DTPA method was used for the basic analysis and bioavailability of heavy metals. It is done by using atomic absorption spectrophotometer. For this method, DTPA (1.97g) was dissolved in pure water (800ml). To enable the dissolution 2ml of NH_4OH was added in it. DPTA became in solution form, and NH_4HCO_3 (79.06g) was added to the solution and stirred slightly. After that final volume was made 1liter.

Determination of heavy metals :
An Atomic Absorption Spectrophotometer (Hitachi Polarized Zeeman AAS, Z-8200, Japan) was used to analyze the elements in the samples in accordance with AOAC (1990) procedure for a specific element, copper. Table 3 lists the instrumental operating conditions for the needed elements.

Standards Preparation:
From the commercially accessible stock arrangement calibrated arrangements were arranged (Applichem®) within the form of a fluid arrangement (1000 ppm). Profoundly filtered de-ionized water was used for working standards preparation. The glass apparatus that was utilized all throughout the method of expository work was submerged in 8N HNO_3 overnight and washed with a few changes of de-ionized water earlier to utilize.
Statistical Analysis:
Data were analysed statistically by using the software "Statistics 8.1". The Analysis of variance (ANOVA) technique was used to determine the overall significance of data and compared by using the LSD test at 5% level of probability (Steel et al. 1997).
RESULTS AND DISCUSSION

Plant height (cm):
Maximum plant height (45.12 cm) was observed where we have applied compost @40 tha^{-1}. Minimum plant height was recorded in control treatment (Figure 3).Maximum plant height in case of compost was due to the better moisture holding capacity, aeration,supplyof nutrient contentsand balancing of soil density (Huang et al. 2013). Ma et al. (2019) also concluded that fertilizer application increased the plant height of celery by changing the enzyme properties of the soil.

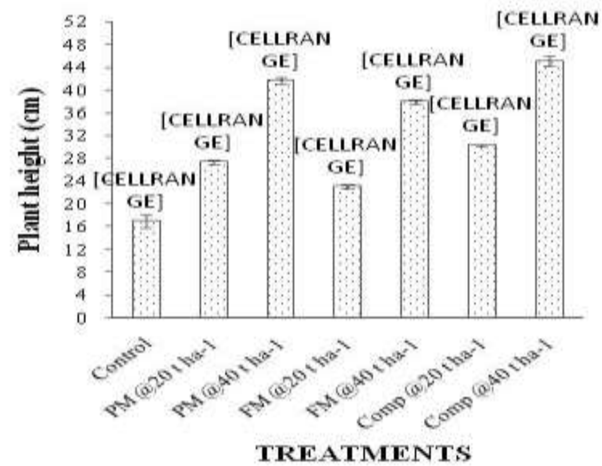

Figure 3: Effect of organic amendmentson plant height of *Capsicum annuum*L.

Total number of chilli peppers per plant:
Maximum total number of chilli peppers per plant (29.0) was recorded in that treatment where compost @40 tha^{-1} was applied while, minimum total number of chilli peppers per plant (5) was observed in control treatment as shown in Figure 4.These results were similar with the findings of the Villena et al. (2018) that compost significantly increased the number of fruits per plant from 2.48 to 2.66 by adding 7 and 13 tha^{-1} as compared to control treatment (2.20) in melon.

Application of copper significantly increased the number of flowers in plants. Maximum flowering is responsible for the highest number of fruits per plant under copper availability (Strelin and Aizen, 2018; Sheng et al. 2021).

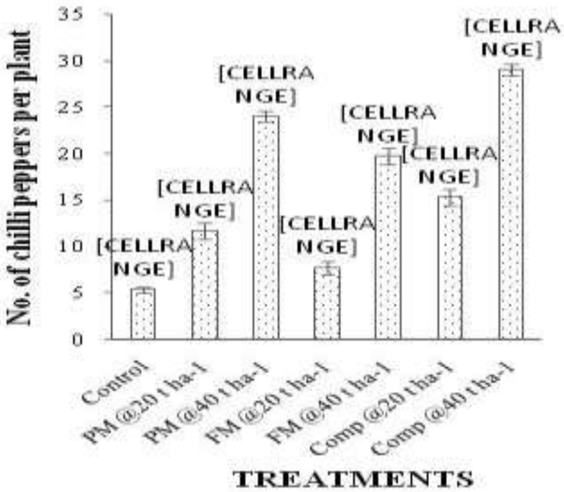

Figure 4: Effect of organic amendments on total number of chilli peppers per plant of *Capsicum annuum*L.

Chilli peppers fresh weight per plant(g):

Compost treatment@40 t ha^{-1}produced maximum chilli peppers fresh weight pr plant (172 g). Whereas, minimum chilli peppers fresh weight per plant (61.32 g) was observed in control treatment where not any organic amendment was applied as shown in Figure 5.The increase in chilli peppers fresh weight per plant might be due to increase in availability of Cu in contaminated soil with compost application (Hernández-Hernández et al. 2019;Sanmanee and Nubdee, 2020). Similar results were also recorded by Imran et al. (2021), they reported that addition of bioactive compost increased the yield of chilli per plant from 266.66 g to 1058.6 g due to increase in availability of other nutrients.

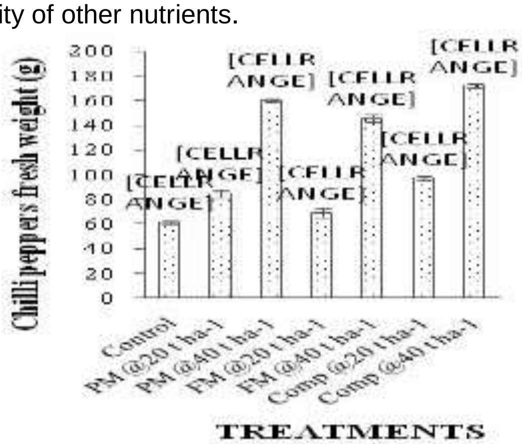

Figure 5: Effect of organic amendments on chilli peppers fresh weight of *Capsicum annuum*L.

Chilli peppers dry weight per plant (g):

Plants which received @40 tha^{-1}compost produced maximum chilli peppers dry weight per plant (86 g). While, those plants which did not received any amendment (Control treatment) produced minimum chilli peppers dry weight per plant (30.65 g) as shown in Figure 6.Chilli peppers dry weight with compost application might be due to enormous effects of compost on soil nitrogen balance through presence of additional nutrients that might be maintained continuous nitrogen supply and improved plant growth (Setyowati et al. 2017).

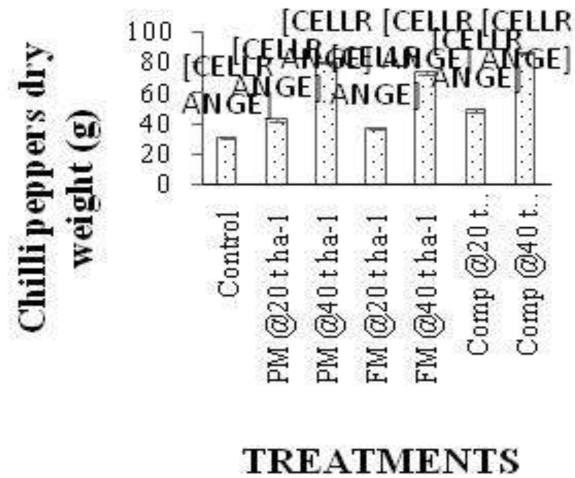

Figure 6: Effect of organic amendments on chilli peppers dry weight of *Capsicum annuum*L.

Chilli peppers length (cm):

Those plants which received compost @40 t ha^{-1}producedmaximum fruit length (6.2 cm). While, minimum chilli peppers length (0.97 cm) was recorded in control treatment as shown in Figure 7.Uddin et al. (2022) reported that increasing the copper dose from 1 ml/L to 3 ml/L significantly increased the chilli fruit length from 4.2 cm to 4.7 cm as compared to control treatment (4.0 cm). Because copper improved the photosynthesis by enhancing the phosphorylation and electron transport chain in light reaction which inevitably increase enzymatic activity during dark reaction and activate nitrogen and carbon metabolism (Pradhanet al. 2015).

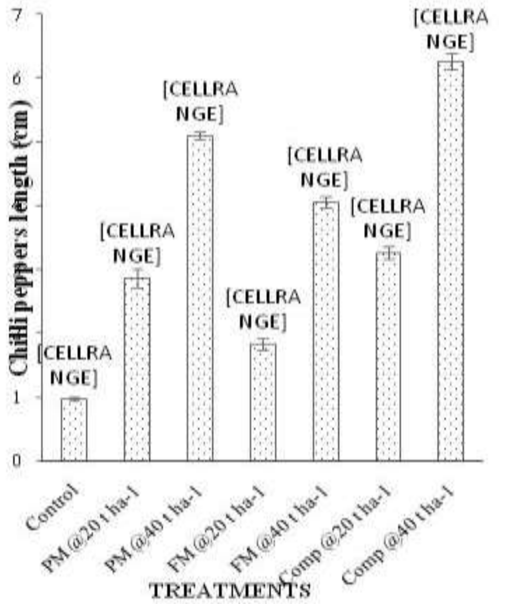

Figure 7: Effect of organic amendments on chilli peppers length of *Capsicum annuum*L.

Shoot fresh weight per plant (g):

Maximum shoot fresh weight (187.64 g) was observed where we have applied compost @40 tha^{-1}. While, minimum shoot fresh weight per plant(95.32 g) was recorded in control treatment as shown in Figure 8.Application of compost improve the soil properties by slowly releasing of plant nutrient like potassium, phosphorus, magnesium, calcium and nitrogen etc., (Setyowati et al. 2017). Increase in shoot fresh weight might be due to compost because additional nutrient increased the vegetative and reproductive growth of chilli (Khaitov et al. 2019).

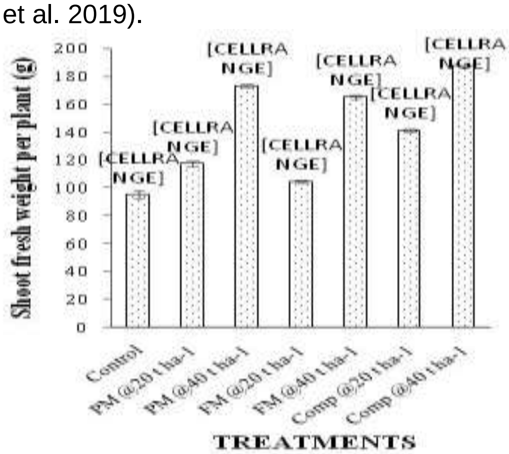

Figure 8: Effect of organic amendments on shoot fresh weight per plant of *Capsicum annuum*L.

Shoot dry weight per plant (g):

Compost treatment @40 t ha^{-1} produced maximum shoot dry weight per plant (106.95 g). Whereas, minimum shoot dry weight per plant (51.33 g) was observed in control treatment as shown in Figure 9.Compost amended soil have balanced soil nitrogen due to presence of additional nutrients by compost (Setyowati et al. 2017). Additional nutrition significantly increased the shoot dry weight of chilli as compared to control treatment as reported by Khaitov et al. (2019).

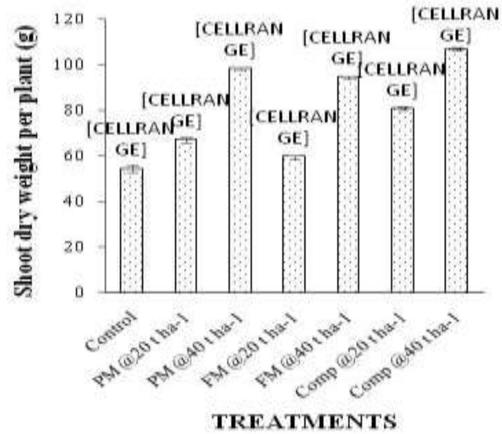

Figure 9: Effect of organic amendments on shoot dry weight per plant of *Capsicum annuum*L.

Root fresh weight per plant (g):

Those plants which received@40 t ha^{-1}compost produced maximum root fresh weight per plant (17.1 g). While, those plants which did not received any amendment (Control treatment) produced minimum root fresh weight per plant (3.2 g) as shown in Figure 10.Our findings were supported by the results of Ahmad et al. (2021), who reported that addition of compost to the soil increased the 19 % root fresh weight because compost additions in the soil alters biological, physical and chemical properties of soil and plant growth.

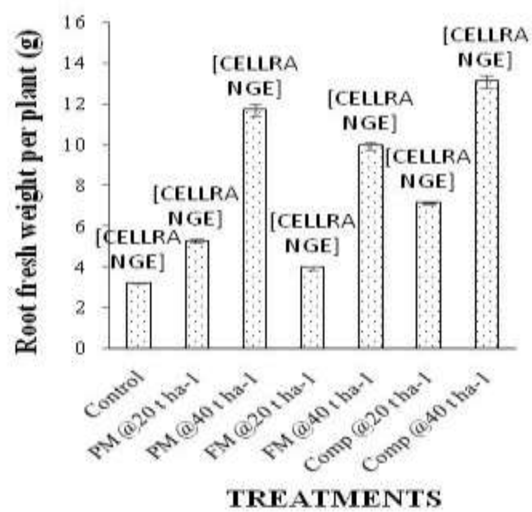

Figure 10: Effect of organic amendments on root fresh weight per plant of *Capsicum annuum*L.

Root dry weight per plant

(compost treatment @40 t ha^{-1} produced maximum root dry weight per plant (8.51 g). Whereas, minimum root dry weight per plant (1.8 g) was observed in control treatment as shown in Figure 11.Our results were parallel with Khaitov et al. (2019), who reported that compost significantly increased the root dry weight of chilli from 46 gplant^{-1} to 55 gplant^{-1} as compared to control treatment (42 gplant^{-1}) which might be due to enormous effects of compost on soil nitrogen balance through presence of additional nutrients (Setyowatiet al. 2017).

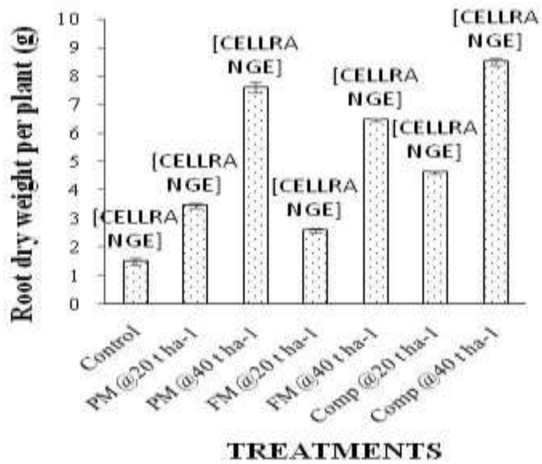

Figure 11: Effect of organic amendments on root dry weight per plantof*Capsicum annuum*L.

Root length per plant (cm):

Those plants which received @40 t ha^{-1}compost produced longest root (7.21 cm). While, those plants which did not received any amendment (Control treatment) produced minimum root length per plant (1.01 cm) as shown in Figure 12. Zhang et al. (2020) evidenced the increased root length with compost application by improving the soil structure and microbial community. Compost removes the compaction of the soil by increasing the organic matter and water conservation which favoured the root growth (Shahzad et al. 2014).

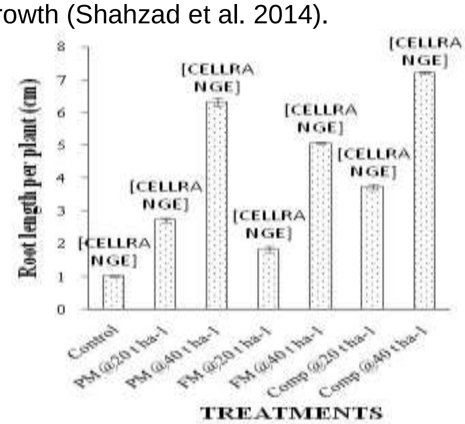

Figure 12: Effect of organic amendments on root length per plant of *Capsicum annuum*L.

Number of branches per plant:

Maximum number of branches per plant (26.35) was observed where we have applied compost @40 t ha^{-1}. While, minimum number of branches per plant (1.02) was recorded in control treatment as shown in Figure 13.Tella et al. (2016) testified that application of compost to the copper contaminated soil improved the copper availability.

Maximum number of branches in compost treatment may be due to copper availability in plant. Uddinet al. (2022) supported our results that 3 mlL^{-1} treatment of copper showed the maximum number of branches per plant (15.3) in chilli as compared to control treatment (9.0)

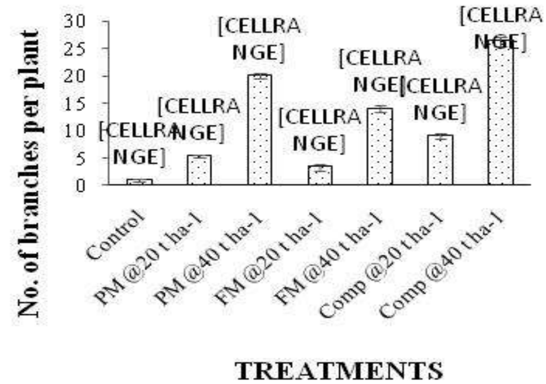

Figure 13: Effect of organic amendments on number of branches per plant of *Capsicum annuum*L.

Number of leaves per plant:

Compost treatment @40 t ha^{-1} produced maximum number of leaves per plant (107.33). While, minimum number of leaves per plant (34.67) was observed in control treatment as shown in Figure 14.Compost treatment improved copper availability in the plant which increased the plant growth as well number of leaves (Tellaet al. 2016).Similar effect of compost was confirmed by the Uddinet al. (2022) that application 3 mlL^{-1} treatment of copper showed the maximum number of leaves (96.7) in chilli at 90 days of transplanting.

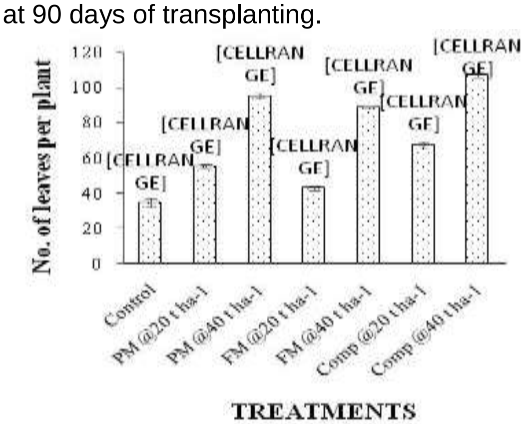

Figure 14: Effect of organic amendments on number of leaves per plant of *Capsicum annuum*L.

Chlorophyll contents (SPAD value):

Those plants which received @40 t ha^{-1}compost showed maximum chlorophyll contents (60.23 SPAD). While, those plants which did not received any amendment (Control treatment) produced minimum chlorophyll contents (39.17 SPAD) as shown in Figure 15.Chlorophyll content in chilli leaves is the critical parameter which closely related with vegetative parameters and yield because its play an important role in photosynthetic accumulation and transportation of photosynthetic along with crop performance (Li et al. 2018).Uddinet al. (2022) reported that copper significantly improved the chlorophyll content in chillileaves. They noted maximum chlorophyll contents (52.7 SPAD) in highest copper dose (3 mlL^{-1}).

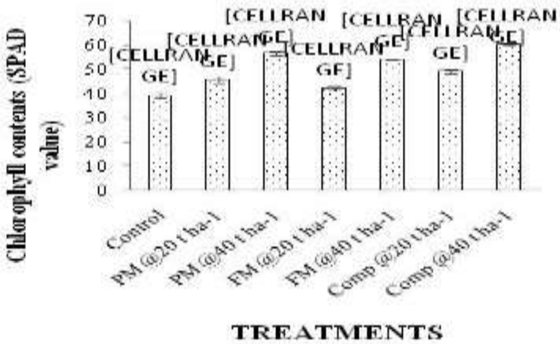

Figure 15: Effect of organic amendments on chlorophyll contents of *Capsicum annuum*L.

Copper concentration in rootsof chillies(mg kg-1):

Maximum copper concentration in roots (69 mg kg^{-1})was recorded where have applied no organic amendment (control treatment). While, minimum copper concentration in roots (50.33mg kg^{-1}) was observed in compost @40 t ha^{-1}treatment as shown in Figure 16.Decrease in copper concentration with the application of compost at the rate of 40tha^{-1}might be due to formation of different complex with the carbon phase of compost or might be due to the formation of copper hydroxides at the surface of compost (Tsang et al. 2014).

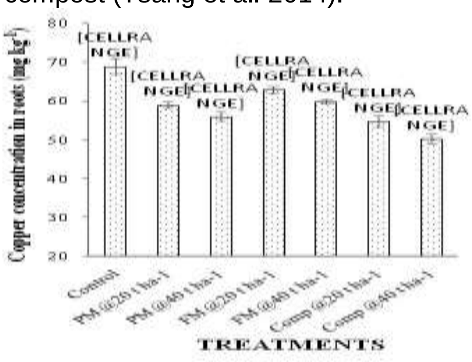

Figure 16: Effect of organic amendments oncopper concentration in roots of *Capsicum annuum*L.

Copper concentration in shoots of chillies (mg kg^{-1}):

Maximum copper concentration in shoots (42.33 mg kg^{-1}) was recorded in control treatment while, those plants which received compost @40 t ha^{-1}showed minimum copper concentration in shoots (30 mg kg^{-1}) as shown in Figure 17.

The decrease in copper bioavailability by the application of compost might be due to adsorption and complex formation of copper on compost as previously studied by Shaheen et al. (2015).

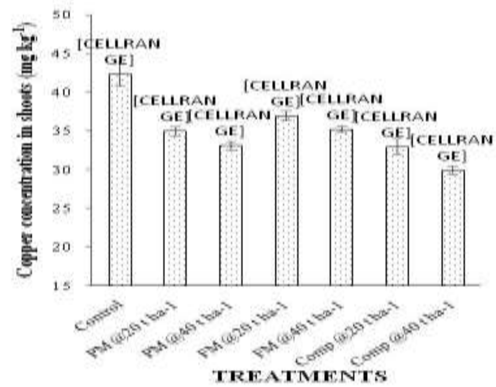

Figure 17: Effect of organic amendments on copper concentration in shoots of *Capsicum annuum*L.

Copper concentration in leaves of chillies(mg kg^{-1}):

Maximum copper content in leaves of chillies (27 mg kg^{-1}) was noted in control treatment while, minimum copper concentration in leaves (18.17 mg kg^{-1}) was observed in compost @40 t ha^{-1} treatment as shown in Figure 18.The decrease in copper bioavailability by compost application might be due to the improvement in cation exchange capacity (CEC) and soil organic carbon (SOC)of soil by predominantly addition of organic matter through compost which decreased the concentration of copper in leaves (Marković et al. 2019).

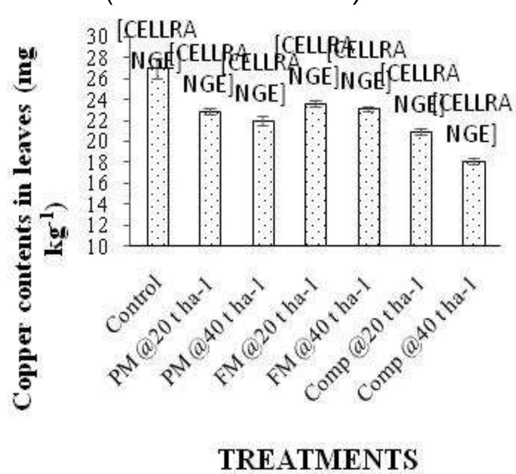

Figure 18: Effect of organic amendments on copper concentration in leaves of *Capsicum annuum*L.

Copper concentration in branches of chillies (mg kg^{-1}):

Maximum copper concentration in branches (30mg kg^{-1}) was observed in control treatment. While, Minimum copper concentration in branches (19.87) were recorded where we have applied compost @40 t ha^{-1}(Figure 19).Minimum copper concentration in branches in case of compost was due to increasing the complexion ability and CEC of soil by organic matter which significantly decreased the bioavailability of Cd Cu, Zn, and Pb (Malik et al. 2021).

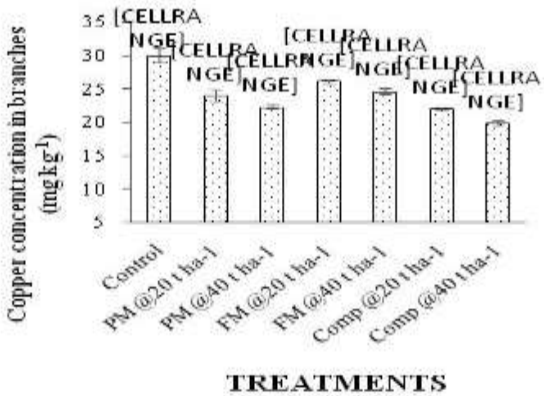

Figure 19: Effect of organic amendments on copper concentration in branches of *Capsicum annuum*L.

Copper concentration in soil (mg kg^{-1}):

Maximum copper content in (41.26mg kg^{-1}) was recorded in compost @40 t ha^{-1}treatments. While, minimum copper concentration in soil (15.17 mg kg^{-1}) was noted in control treatment as shown in Figure 20.High concentration of copper in soil in compost treatment might be due to presence of organic compound which decreased uptake of heavy metals (Cd, Cu, Zn andPb) in plants tissue by making insoluble complex with heavy metals and retain these metals in soil (Rosenand Chen, 2014; Li et al. 2021).

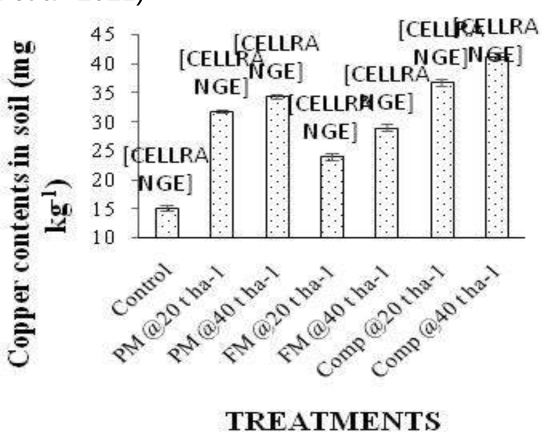

Figure 20: Effect of organic amendments on copper concentration in post soil

CONCLUSION

The results of the experiment showed that organic amendments affected all the vegetative parameters of chili plants. Data regarding plant height showed significant differences among the treatments under Cu contamination. Plant height was noted lowest in Cu-contaminated soil while increasing under organic amendments. Maximum plant height was observed in plants where compost was applied @40 t ha-1. In terms of average shoot length, a significant decrease in shoot length was seen in the Cu-contaminated control group. The treatments that applied organic amendments improved the length of the shoots. There was a maximum increase in branch lengthseen in plants receiving @40 t ha-1 of compost.

CONFLICT OF INTEREST

The authors declared that the present study was performed in the absence of any conflict of interest.

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AUTHOR CONTRIBUTIONS

Muhammad Afzan and Muhammad Usman designed the research work and performed it among all other Co-Authors Muhammad JamilTehseen Ali Jilani,Memoona Bashir helped out in performing the research workMuhammad Salman Hameedand Bismillah Khan helped in statistics, Muhammad Gulsher,Shamim Akhtar and Esha Niaz proofread the manuscript

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