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Research Article

Effects of Bio-Slurry and Chemical Fertilizer Application on Soil Properties and Food Safety of Tomato (*Solanum lycopersicum* Mill.)

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This study evaluated the effects of bio-slurry (BS) and chemical fertilizer (CF) application on soil properties and food safety of tomato (*Solanum lycopersicum* Mill.). A field experiment consisting of 100% BS (5 ton BS ha^{-1}), 100% CF (90 kg N·ha^{-1} + 30 kg P·ha^{-1} + 13 kg S·ha^{-1}), and control was conducted. Soil samples from all the treatments were collected for their physico-chemical characteristics. The level of ten heavy metals in experimental soil and tomato fruit samples was also determined. Compared to CF and control, the application of BS improved soil physico-chemical characteristics. The BC significantly reduced the mean concentrations of Cd and Mn in the tomato fruit samples. The mean concentration of Ni (18.24 ± 0.61, 23.9 ± 0.3, and 9.66 ± 1.2 mg kg^{-1}) and Mn (15.4 ± 2.4, 38 ± 3.3 and 21.8 ± 0.99 mg kg^{-1}) in tomato fruit samples of BS-treated, CF-treated, and control soil, respectively, was above the safety limit set by the Food and Agriculture Organization/World Health Organization for human consumption. Similarly, the mean concentration of Cd (7.98 ± 0.72 and 3.29 ± 0.37 mg kg^{-1}) in tomato fruit samples of CF-treated and control soil was above the safety limit. From this perspective, the consumption of these tomato fruits could be unsafe for human health with respect to Ni, Mn, and Cd toxicities. The application of BS could remediate the Cd toxicities, yet other scenarios of phytoremediation would be praiseworthy to address Ni, Cd, and Ni toxicities.

1. Introduction

Vegetables constitute a major source of vitamins, crude fiber, protein, antioxidant, and minerals [1]. Specifically, tomato (*Solanum lycopersicum* Mill.) is one of the most important vegetable crops for its special nutritive value and widespread production [2]. It is the world's largest vegetable crop after potato and sweet potato [3]. In accordance, the Central Statistical Agency (CSA) of Ethiopia pointed out that Ethiopia is the world's 84th largest producer of tomato where its national mean yield is 6.2 ton ha^{-1} [4].

Injudicious application of chemical fertilizer causes environmental pollution, damages soil physico-chemical characteristics, and causes various problems to human health [5–9]. This has initiated researchers around the globe to search for eco-friendly alternative fertilizers that would ensure agricultural biosafety and environmental and human health [6]. Bio-slurry (BS) is one of such alternatives, which is an aerobically digested organic material obtained from biogas plants. It is environmentally friendly with only few harmful effects when compared with chemical fertilizers. It contains appreciable amounts of organic matter (20 to 30%) and is much needed for poor soils [10]. Its use can reduce the quantity of chemical fertilizer up to 50% [11]. Specifically, it is most suitable for farming horticultural crops [12].

The quality of soil determines environmental health [13] and food safety and quality [14], which could be expressed in terms of physico-chemical indicators [15, 16]. Soil organic matter is considered as one of the most important indicators of soil quality as it controls many soil properties, such as nutrient cycling, soil structure maintenance, and pH buffering [9, 17]. However, soil contamination by heavy metals is one of the factors that cause soil health deterioration and plant health problems [18]. Among the main sources of heavy metals in the soil are agrochemicals (fertilizers and pesticides) [19]. The uptake of heavy metals by plants depends on different factors, including solubility of heavy metals, soil pH, soil type, and plant species [20]. Vegetables can take up heavy metals (Cu, Zn, Fe, Pb, Cd, Mn, and Cr) from the soil on which they grow [21–23], which upon consumption could cause human health problems [24].

Increasing crop productivity by applying chemical fertilizers has been the main objective in most agricultural production systems, whereas product quality and environmentally friendly approaches are given little attention [25]. Increased productivity of cultivated land and higher input use efficiency with no harm to the soil and product quality are among the development strategies in vegetable production [25, 26]. Particularly, the use of chemical fertilizer is not the appropriate solution to overcome these restraints, exclusively for vegetables that have steadily short time and are consumed fresh. Moreover, the use of chemical fertilizer is not only a threat to human health [27] but also its continuous use may lead to the accumulation of heavy metals in plant tissues, which compromises the nutritional value and food safety [5, 8, 28]. Therefore, considering environmental and human health risks, sole dependency on chemical fertilizer is not recommended at least for small-scale farmers who can have options of using organic sources of fertilizer [29]. Replacing chemical fertilizer with BS reduces soil pollution and food safety problems [11, 30].

In most areas of Ethiopia, the use of BS as a source of fertilizer is uncommon, even among farmers who have access to bio-slurry. In this regard, although a considerable number of farmers around the study area own biogas plants and have access to BS, the tradition of using it as organic fertilizer for tomato production is missing primarily due to lack of awareness and proper agricultural extension services. In spite of this, tomato is widely grown, primarily with the use of chemical fertilizers. In the study area, previous studies exploring how soil qualities and tomato food safety are affected by the application of BS and chemical fertilizer are lacking. Therefore, determining the effects of BS and chemical fertilizer application on (1) soil physico-chemical properties, (2) heavy metal concentration levels in soil and tomato fruit samples, and (3) the food safety of tomato fruits for human consumption were the objectives of the current study.

2. Materials and Methods

2.1. Description of the Study Area. Hawassa University main campus agricultural research farm was used for the experimentation. Hawassa University is located at 275 km south of Addis Ababa, the capital city of Ethiopia. It is situated at an elevation of 1768 meter above sea level (Figure 1).

2.2. Agro-Climatic Conditions. Hawassa receives a bimodal rainfall, where March to June is the main cropping season for planting late and mid-maturing maize varieties. The months of June to October are used for growing early maturing crops, such as maize and pulses [31]. The average mean monthly rainfall during the cropping period was 146.78 mm, while the maximum and minimum temperature during the same period was 30.8°C and 14.3°C, respectively. In addition, the monthly rainfall and mean monthly maximum and minimum temperature recorded during 1990–2019 are presented in Figure 2.

2.3. Agriculture and Soils. Agriculture is the dominant means of livelihood for the majority of people living around the study areas. Vegetables (head cabbage, tomato, onion, and carrot), perennial crops such as ensete (*Ensete ventricosum*), chat (*Catha edulis* L.), coffee, and avocado, and annual crops such as maize, sweet potato, and haricot bean are widely grown [31]. The soils are tropical Andosols with textural class ranging mostly from sandy loam to silty loam [29].

2.4. Experimental Design, Treatments, and Procedure. The experimental field was ploughed, and plots were leveled manually. Tomato variety Venes was used as a test crop. The tomato seeds were planted in a germination box in a mesh house. After 42 days, healthy and vigorous seedlings with four true leaves were transplanted to the experimental plots. Seedlings failed to establish were replaced within a week of transplanting to maintain the appropriate plant population.

The experiment was laid out in a randomized complete block design with three replications. CF and BS were used as fertilizers. There were three treatments, including 100% BS (5 ton ha^{-1}), 100% CF (92 kg $N{\cdot}ha^{-1}$ + 30 kg $P{\cdot}ha^{-1}$ + 13 kg $S{\cdot}ha^{-1}$), and control (Table 1). The full dose of CF was applied at transplanting following Ethiopian Agricultural Research Organization (EARO) recommendation [33]. The bio-slurry was obtained from a model farmer who resided close to the study area and had a biogas plant. Cow dung was the main feedstock.

There were 9 plots (Table 2), each measuring 3 m $*$ 6 m (18 m^2). The spacing between each block and plot was 1.5 and 1 m, respectively. The spacing between each row and plant was 0.5 and 0.3 m, respectively. There were six rows, each with six plants, comprising 36 plants in each plot. All agronomic practices (weeding, cultivation, supplementary irrigation, etc.) were employed [34]. Weeding was done manually with a hand hoe four times between transplanting and harvesting. Tomato fruit samples were harvested at four-day interval, the first harvest was done on the 85th day after transplanting, and the final harvest was done on the 100th day after transplanting.

2.5. Plant Protection Measure. During the experimental period, the treatments were regularly observed for the occurrence of disease, pest, or any other kinds of disorders, and data were recorded. Ridomil gold MZ 68 WP 400 g/200 L fungicide was applied at a ten-day interval to control late blight and leaf blight.

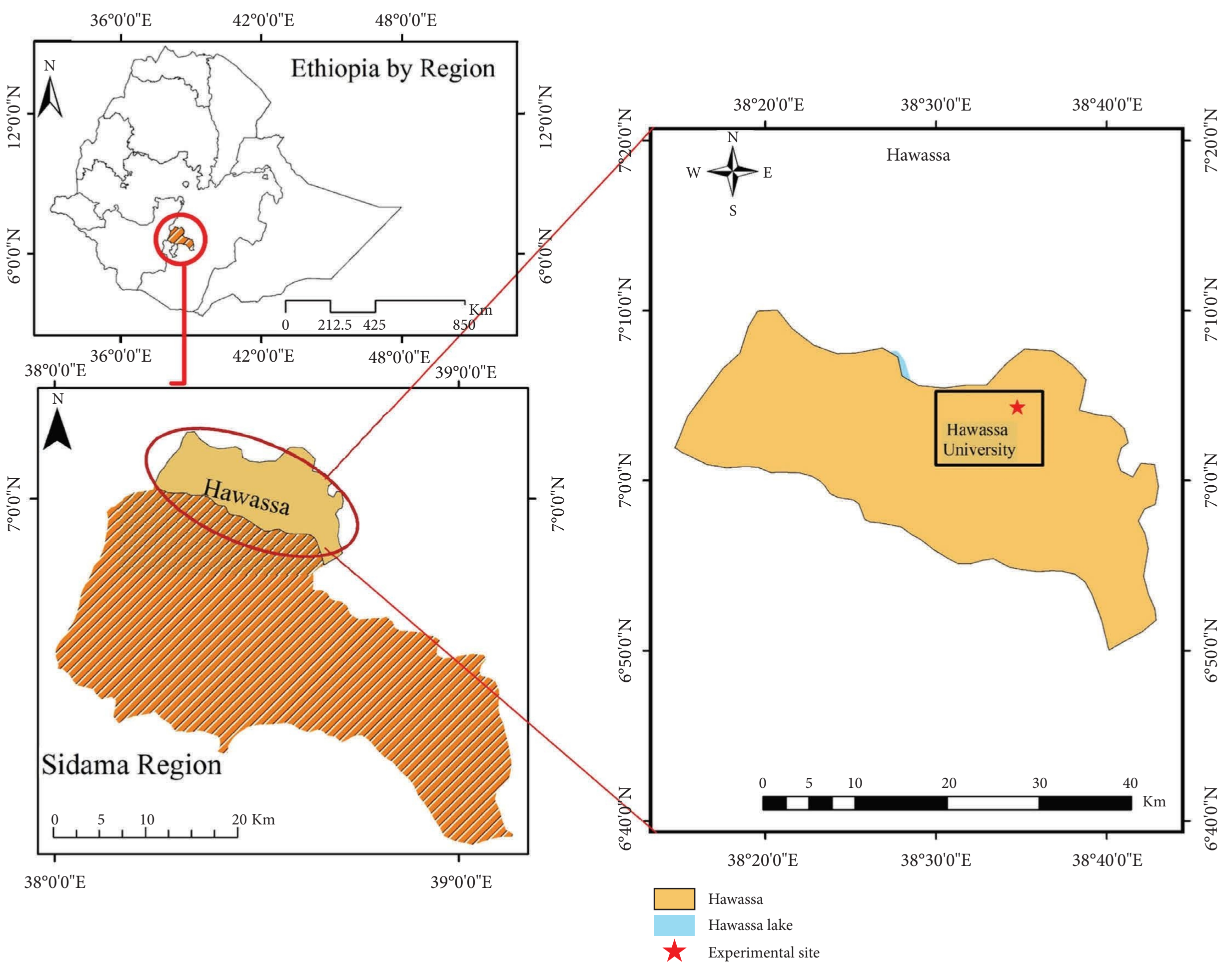
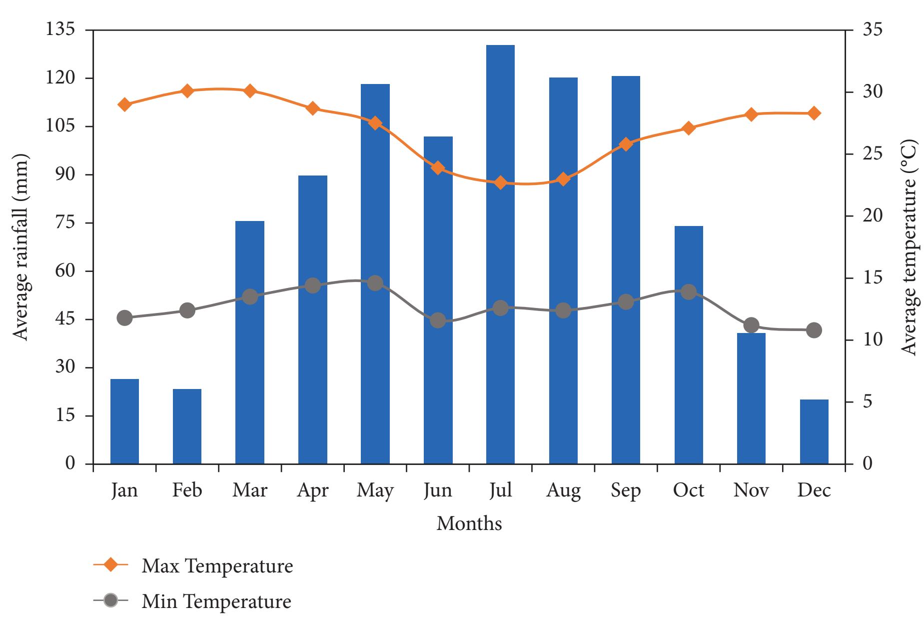
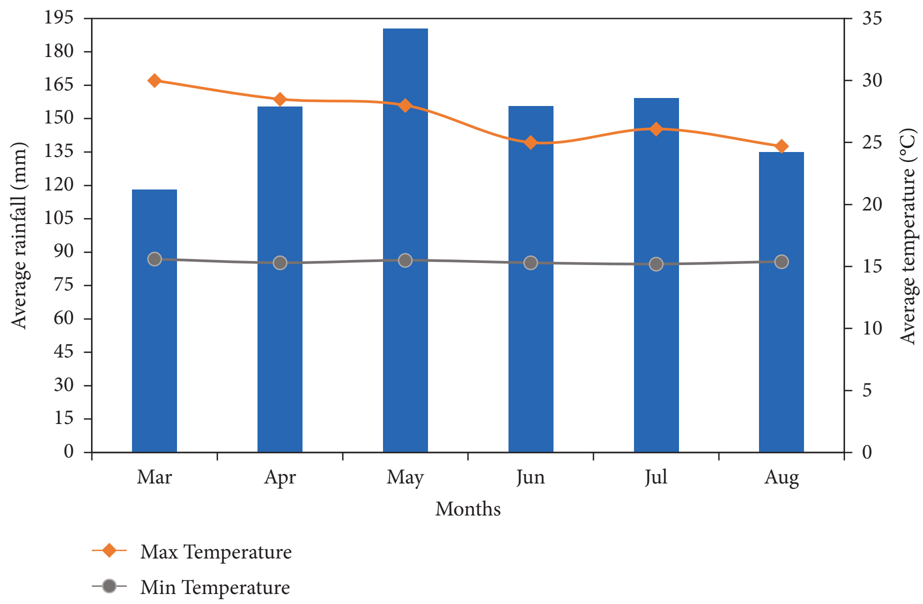


Figure 1: Location map of the study area.



(a)

Figure 2: Continued.



(b)

Figure 2: Monthly average rainfall and maximum and minimum temperature of Hawassa recorded during 1990–2019 (a) and during the experimental season (b) (data source: National Metrology Service Agency [32], Hawassa Branch, 2020).

2.6. Soil Sampling and Analysis for Physico-Chemical Properties. Soil samples were collected from the top 20 cm depth in a zigzag manner using soil auger prior to transplanting. The samples were thoroughly mixed to form one composite sample and were analyzed for physico-chemical properties. At harvest, a composite sample was taken from each of these plots at the same soil depth for physico-chemical analysis.

Organic carbon (OC), pH, cation exchange capacity (CEC), total nitrogen (TN), available phosphorus (P), and exchangeable bases (Ca, Na, and K) were analyzed following the established procedures and methods.

Pre- and post-experiment soil bulk density, porosity, and moisture content were determined following established and standard procedures and methods.

2.7. Bio-Slurry Sample for Physico-Chemical Analysis. A sample of BS of 0.5 kg was analyzed for pH, OC, TN, CEC, and Av. P using standard methods.

2.8. Soil Sample Collection, Preparation, and Analysis for Heavy Metals. A 0.5 kg composite soil sample from each treatment was prepared and stored in plastic bags. The sample was properly labeled and placed inside plastic bags and transported to HortiCoop Laboratory, Bishoftu, Ethiopia, for heavy metal analysis. The soil sample was oven dried at 25°C for 2 days until constant weights were reached. The sample was then crushed into powder using mortar and pestle, sieved through 2 mm sieve, and then stored in plastic bags until analysis. The dried 0.5 g soil sample was transferred into digestion vessel containing HCl, HNO_3, and H_2O_2 mixture and digested at 100°C for 2 hours. Then, the sample was removed from block digesters and allowed to cool down, and 40 mL of distilled water was added and mixed well. After cooling to room temperature, the digested sample was filtered using Whatman No. 42 filter paper. Finally, the concentrations of heavy metals (As, Zn, Cd, Pb, Fe, Cu, Mn, Ni, Cr, and Co) were determined using inductively coupled plasma optical emission spectrometry (ICP-OES).

2.9. Bio-Slurry Samples for Heavy Metal Analysis. A 0.5 kg BS sample was analyzed for heavy metals (As, Zn, Cd, Pb, Fe, Cu, Mn, Ni, Cr, and Co) using ICP-OES.

2.10. Tomato Fruit Samples for Heavy Metal Analysis. Six ripe and healthy tomato fruit samples were collected randomly from each treatment and washed with distilled water to remove any adhered contaminants. The samples were then cut into small pieces using clean sterilized plastic knife and dried in oven at 60°C for 24 hours to remove moisture and maintain constant mass. The samples were then properly labeled and placed in clean plastic bags and transported to HortiCoop Laboratory, Bishoftu, Ethiopia, for further processing and analysis. The dried samples were crushed into powder using mortar and pestle and then screened to pass through a sieve of 2 mm mesh size. The sieved samples were carefully labeled and stored in polyethylene bags and kept in desiccators.

A 0.5 g of homogenized powdered sample was added into a digestion flask and then 10 mL aqua regia (with a 3 : 1 ratio of HCL to HNO_3) and 3 mL H_2O_2 were added. The mixture was heated at 300°C for 1 h on block digester. After digestion has been completed, the final mixture was filtered out using Whatman No. 42 filter paper and then the clear and colorless solution was transferred to a 50 mL volumetric

Table 1: Treatment, fertilizer type, rate, and application time.

Treatment code	Treatment (%)	Fertilizer type and rate				
		BS (g/planting hill)	Urea (g/planting hill)	NPS (g/planting hill)	Amount added per hectare (ha)	Application time
T1	100% BS	250	0	0	5 ton ha^{-1}	Applied at transplanting
T2	100% CF	0	10	7.9	92 kg $N \cdot ha^{-1}$ + 30 kg $P \cdot ha^{-1}$ + 13 kg $S \cdot ha^{-1}$	Applied at transplanting
T3	Control	0	0	0	0	No fertilizer was applied

Table 2: Field layout of the experiments.

Plot	Rep 1	PN	Rep 2	PN	Rep 3
1	T1	1	T1	1	T2
2	T3	2	T2	2	T3
3	T2	3	T3	3	T1

T1 = treatment with 100% BS, T2 = treatment with 100% CF, T3 = control, Rep = replication, and PN = plot number.

flask. The samples were diluted with distilled water, and then the concentrations of heavy metals (As, Zn, Cd, Pb, Fe, Cu, Mn, Ni, Cr, and Co) were determined using ICP-OES.

After analysis, the concentration levels of heavy metals in the soil, BS, and tomato fruit samples were compared with the maximum permissible limit (MPL) for the respective parameters according to FAO/WHO [35].

2.11. Assessment of Ecological Risk of Heavy Metals in the Soils. Contamination factor (CF) and pollution load index (PLI) were used to ascertain the potential ecological risk of heavy metals in the soil.

2.12. Contamination Factor (CF). The CF was used to express the level of contamination of soil by heavy metals [36]. This method has been developed further in soil analysis [37, 38]. It was calculated as a ratio between the measured concentration of the heavy metal in soil and the pre-industrial reference value of the same metal as follows:

$$CF = \frac{\mathbf{Cs}}{\mathbf{Cb}}, \tag{1}$$

where CF is the contamination factor, *Cs* is the concentration of metal in the soil (mg kg^{-1}), and *Cb* is the baseline concentration.

There is lack of baseline concentration level for the heavy metals in Ethiopia; therefore, the concentrations of heavy metals in the Earth's crust were used as a reference value [39]. Therefore, the reference value (mg kg^{-1}) for the different meals was considered: Cd (0.15), Pb (12.5), As (15), Co (29), Cr (100), Ni (75), Cu (55), Zn (70), Fe (5000), and Mn (1060). Håkanson [36] categorized the CF value as follows: low contamination factor ($CF < 1$), moderate contamination factor ($1 < CF < 3$), considerable contamination factor ($3 < CF < 6$), and very high contamination factor ($CF > 6$).

2.13. Pollution Load Index (PLI). The level of soil contamination by heavy metals was evaluated using PLI that provides a simple and comparative means of assessing the soil quality. This parameter allows assessing the level of environmental contamination in order to undertake monitoring or repair activities aimed at improving soil quality [40]. Each sampling site can be evaluated for the extent of heavy metal pollution, employing the PLI method developed by Tomlinson et al. [41] and later applied by Bhutiani et al. [42] as below:

$$\mathrm{PLI} = (CF1 \times CF2 \times CF3 \times \cdots\cdots CFn)\frac{\mathbf{1}}{\mathbf{n}}, \tag{2}$$

where *CF* is the contamination factor, *n* is the number of metals studied, and CF^n is contamination factor for n^{th} element as mentioned above.

According to Tomlinson et al. [41], $PLI < 1$ *denotes* no heavy pollution and $PLI > 1$ denotes heavy metal pollution.

2.14. Statistical Analysis. Statistical Analysis System (SAS) software version 9.4 [43] was used for data analysis. Wherever there was significant difference, mean separation was carried out using the least significant difference (LSD). Significant difference between means of treatments was determined at the 5% significance level ($p < 0.05$).

3. Results and Discussion

3.1. Pre-Experiment Soil Physico-Chemical Properties. The soil samples consisted of silt (42%), sand (28.5%), and clay (22.7%), which is categorized into silty loam textural class. The soil pH, OC, TN, available P, CEC, and exchangeable Ca, Na, and K were 5.64, 2.73, 0.24%, 49.6 mg·kg^{-1}, 24.6 cmol kg^{-1}, and 46.15 cmol kg^{-1}, 0.21 cmol kg^{-1}, and 2.29 cmol kg^{-1}, respectively (Table 3). The soil is moderately acidic. Moderately acidic soils have a pH ranging from 5.6 to 6.0 [44]. The OC is in a medium range [45] where soil OC content ranging from 1-2%, 2–4%, and 4–6% is rated as low, medium, and high, respectively. The TN is within the range of medium [46] where the TN content rated as <0.1, 0.1–0.15, 0.15–0.25, and >0.25% is categorized as very low, low, medium, and high, respectively. The available P is categorized in the high range (>10 mg·kg^{-1}) [47]. The CEC is in the medium range where CEC value ranging from 5–15, 15–25, and 25–40 cmol·kg^{-1} is rated as low, medium, and high, respectively [45]. The concentration of exchangeable bases of Ca, Na, and K is high, low, and very high, respectively [48]. Most of the soil properties appear to be favorable for the growth of vegetables.

The mean concentrations of heavy metals (mg kg^{-1}) in pre-experiment soil were As (16.5), Pb (27.2), Zn (15.75), Cd (8.62), Cu (0.88), Ni (2.45), Co (0.3), Fe (120.9), Mn (142.1), and Cr (4.53). Except for Cd, these concentrations were below the maximum tolerable limit of FAO/WHO [35] of heavy metal concentrations in the soil. This shows that the soil is slightly contaminated with Cd (Table 4). Pesticides and phosphate chemical fertilizers that had previously been used for conducting numerous tests on the same experimental field could be the most likely sources of the Cd contamination. Cd can enter agricultural soil through various pathways which include application of agrochemicals (synthetic phosphate fertilizers and pesticides) [49, 50] and contaminated animal manure [50].

3.2. Chemical Properties of BS. The BS had a pH of 7.52, 6.24% OC, 0.54% TN, 262.2 ppm available P, 11.56 C: N, 10.33 ppm available K, and 39 cmol·kg^{-1}CEC as well as 10.3, 0.39, and 52.34 exchangeable K, Na, and Ca, respectively (Table 4). The high pH value of the bio-slurry reduced the acidity of the experimental soil and made it favorable for the growth of tomato. Previous studies reported similar findings: application of BS reduces the acidity of soils [51] and improves the quality of agricultural soil by neutralizing acid condition [52].

The high organic carbon in BS could be important to maintain nutrient balance by suppressing the mobility of heavy metals and facilitating the decomposition of organic matter [53]. The high OC in BS could also imply that it can be a good source of plant nutrients. This is because increasing OC levels increases overall soil CEC and increases the ability of soils to store NH_4^+, Ca^{+2}, Mg^{+2}, and K^+. Such soil conditions would in turn make fertilization more efficient [52, 54, 55]. Organic materials are slightly alkaline and could improve soil suitability for plant growth [48]. Besides, the higher TN and available P in BS implies that its application can supply the soil with high amounts of TN and P. BS sourcing from livestock contains high concentrations of available nutrients, especially N and P [56, 57].

Apart from that, the mean concentrations of heavy metals in BS were within the acceptable range of FAO/WHO [35] except for Cd (Table 4). The source for higher concentration of Cd in the BS might attribute to the cow dung used as feedstock. The cow feed was prepared from diverse sources including plant residues and additives. Significant amount of heavy metals including Cd can be found in animal manures based on the types of animal feed and the additive used [58, 59].

4. Effects of BS and CF Application on Soil Physico-Chemical Properties

4.1. Soil Physical Properties. The application of BS slightly reduced bulk density (0.83 g/cm^{-3}) and increased porosity (70%) compared with the application of CF (Table 5). It reduced bulk density by 17% and increased porosity by 11% compared with the CF, featuring positive improvements in soil physical properties (Table 5). The effect of the application of CF on porosity of soils is minimal. This exhibits the advantage of applying organic fertilizer over CF in managing the physical properties of soils over short term. The decrease in bulk density after the application of BS may be related to the increase in OC, which modifies the porosity and bulk density of soils. White [60] stated that the value of bulk density for soils having high OC ranges from <1 g/cm^3, that for well-aggregated soils ranges from 1 to 1.4 g/cm^3, and that for sandy soils ranges from 1.4 to 1.8 g/cm^3. In accordance, the application of BS yielded higher OC, which might be the reason for the increased porosity and decreased bulk density of the respective soils (Table 5).

Apart from that, the application of BS increased the moisture content of the soil compared with the application of CF. It increased the moisture content by 58.3% and 58.1% over CF-treated soils and control soils, respectively (Table 5). Findings from previous studies reported that the application of farmyard manure, BS, and other organic fertilizers increases porosity and decreases bulk density, which are associated with increasing water-holding capacity of soils [61–64].

4.2. Soil Chemical Properties. The application of BS and CF influenced the status of soil pH, OC, TN, available P, CEC, and exchangeable bases (Ca, Na, and K) (Table 5). The application of BS increased soil pH from moderately acidic (5.6) to faintly alkaline (7.4). In contrast, the application of CF slightly reduced soil pH. It is supported by findings from previous studies: increment in pH after BS amendment could be explained by decomposition of organic materials that release basic cations K+, Ca^2+, Mg^2+, and OH^- to the soil and substitute acid cations (H^+, Al^{3+}, and Fe^{3+}), which would in turn result in a slight increase in soil pH [65]. Application of compost releases alkaline substances and cations such as Ca^{2+}, Mg^{2+}, and K^+, which increase CEC and pH level and counteract soil acidification [66]. The application of organic fertilizers increases soil pH and reduces exchangeable acidity while the application of chemical fertilizer slightly influences soil pH [62, 67].

The application of BS and CF yielded the highest (2.98%) and lowest (2.5%) soil OC, respectively (Table 5). The application of BS increased the OC content by 8.4% over the control soil as well as 16.1% over the CF-treated soil. Nevertheless, the application of CF reduced the OC content of the control soil by 8.42%. This shows that the application of BS increases OC relative to the application of CF. This might be attributed by high amount of organic matter in BS, which enhances the OC content of the soil; organic materials have a major impact on mineralization rates by increasing soil organic carbon directly [68]. Other previous studies also demonstrated significantly increasing OC of the soil by the application of organic fertilizers compared to CF [69–72].

The application of BS increased the TN (0.26%) contents compared to the control soil (0.22%) and CF-treated soil (0.24%). The application of BS and CF increased TN by 15.38% and 8.3% over the control soil, respectively (Table 5). BS contains a high concentration of organic nitrogen and contributes to the direct addition of nitrogen from nitrogen fertilizers [56]. Organic fertilizers increase the TN content of soils compared to pre-application [73].

The available P in the soil treated with BS (59.6 mg·kg^{-1}) and CF (42.5 mg·kg^{-1}) was higher compared to the same in the control soil (42.3 mg·kg^{-1}) (Table 5). The soil with available P (mg·kg^{-1}) content <3 is very low, 4–7 is low, 8–11 is medium, and >11 is high [47]. Accordingly, the available P contents of all treatments are higher except the soil treated with BS, which is higher than that of the soil treated with CF and control (Table 5). Organic amendments can increase the P recovery in the soil by increasing the P mobility in the soil [74]. It could also be due to the fact that organic fertilizers, on decomposition, solubilize insoluble organic P fractions through the release of various organic acids, thus resulting in a significant improvement in soil available P content [57, 75].

The CEC value of the control soil (24.9 cmol·kg^{-1}) is lower than the CEC value of BS-treated soil (29 cmol·kg^{-1}). This was, however, higher than the CEC value of CF treated soil (23.6 cmol·kg^{-1}) (Table 5). Ranges in CEC of 5–15, 15–25, and 25–40 cmol·kg^{-1} are rated as low, medium, and high, respectively [45]. Accordingly, the CEC of the soil is rated as high. The application of BS increased the CEC from the medium in control soil to high; however, the application of CF decreased the CEC of the soil, which could attribute to the decrease in soil pH and exchangeable bases.

Table 3: Physico-chemical properties of pre-experimental soil.

Para	pH	OC	OM	TN	C : N	P	CEC	Ca	K	Na	Sa	Si	Cl	TC	BD	Po
Value	5.64	2.73	4.74	0.24	11 : 01	49.6	24.6	46.15	2.29	0.21	35.28	42	22.72	Silt loam	1	62.26

Para: parameter, OM: organic matter, Sa: sand, Si: silt, Cl: clay, TC: textural class, BD: bulk density, and Po: porosity. pH (H_2O), OC (%), OM (%), TN (%), P (ppm), CEC (cmol/kg), Ca (cmol/kg), K (cmol/kg), Na (cmol/kg), Sa (%), Si (%), Cl (%), BD (g/m^3), and Po (%).

The application of CF decreased the OC content of the soil and consequently lowered the CEC value. This result agrees with findings from previous studies: application of organic fertilizers increases CEC while chemical fertilizers decrease CEC [64, 76]. There is a direct association between OC and CEC; soils with low CEC are often low in OC [77, 78]. An increase in soil OC and CEC content increases the buffering capacity of soil and soil fertility through retaining nutrients against leaching and enhancing their availability.

Unlike CF ($22.2\,cmol{\cdot}kg^{-1}$), the application of BS increased the amount of exchangeable Ca ($28\,cmol{\cdot}kg^{-1}$) compared to the control soil ($24.6\,cmol{\cdot}kg^{-1}$) (Table 5). This indicates that the application of BS yields at higher amount of Ca over the application of CF. Similarly, the application of BS increased the amount of exchangeable Na ($0.23\,cmol{\cdot}kg^{-1}$) over the control soil ($0.21\,cmol{\cdot}kg^{-1}$) and CF ($0.2\,cmol{\cdot}kg^{-1}$) (Table 5). In addition, the application of BS increased the amount of exchangeable K ($2.7\,cmol{\cdot}kg^{-1}$) over the control ($2.29\,cmol{\cdot}kg^{-1}$) and the application of CF ($2.18\,cmol{\cdot}kg^{-1}$). This indicates that in contrast to CF, the application of BS increases the amount of soil K (Table 5). In general, the application of BS increases exchangeable bases and CEC and improves other soil physico-chemical characteristics. This could attribute to the higher CEC and organic matter content in BS and the nature of the organic matter to buffer change in pH. Previous studies reported increasing organic matter content and available nutrients with the application of organic fertilizers, which in turn increases the exchangeable bases and the cation exchange capacity of the soil [17, 69, 79]. Organic matter increases the buffering capacity of soils and prevents acidification through binding cations [80].

5. Levels of Heavy Metals in Soil and Tomato Samples

5.1. Level of Heavy Metals in Soil. Compared with the application of CF and control, the application of BS resulted in the highest mean concentrations of Fe ($213.1 \pm 6.55\,mg{\cdot}kg^{-1}$), Mn ($144.7 \pm 3.5\,mg{\cdot}kg^{-1}$), Cu ($1.88 \pm 0.08\,mg{\cdot}kg^{-1}$), Ni ($4.96 \pm 1.4\,mg{\cdot}kg^{-1}$), Co ($0.33 \pm 0.011\,mg{\cdot}kg^{-1}$), and Zn ($16.43 \pm 1.22\,mg{\cdot}kg^{-1}$) (Table 6). This might attribute to the very nature of the applied BS, which was sourced from cow dung. Luo [81] reported higher concentrations of Cu, Ni, Zn, and Fe in soils from livestock manure application. Similarly, the application of BS resulted in higher concentrations of Fe, Zn, Mn, and Cu [82]. On the contrary, the application of CF yielded the highest concentration of As ($16.6 \pm 2.7\,mg{\cdot}kg^{-1}$), Pb ($28.1 \pm 1.2\,mg{\cdot}kg^{-1}$), Cd ($12.2 \pm 3.27\,mg{\cdot}kg^{-1}$), and Cr ($6.14 \pm 0.66\,mg{\cdot}kg^{-1}$) compared to the BS-treated and control soils. At the same time, the application of CF decreased soil pH, which might have contributed to the high concentration of toxic heavy metal (for instance, Mn, Cd, and Cr) in the soil. Low pH increases the solubility and availability of toxic metals like Mn, Cd, and Al in soils [83]. The application of CF increased the mean concentration of Cd, As, Cr, and Pb in soil [84–86].

The application of BS significantly ($P < 0.05$) increased the mean concentration of Zn, Cu, Fe, and Ni over the application of CF and control soil. In addition, the application of CF in turn significantly ($P < 0.05$) decreased the mean concentration of Zn, Cu, Fe, and Mn over the control. Regardless of treatments, only the mean concentration of Cd was above the maximum permissible limits for agricultural soils recommended by FAO/WHO [35]. The application of the BS reduced the Cd concentration of the control soil; however, this did not bring the level of the metal to the permissible range. In this sense, the application of BS and CF as well as the control soil itself may not be suitable for the tomato cultivation with respect to Cd toxicity, which agrees with findings from earlier studies [87].

5.2. Level of Heavy Metals in Tomato Fruits. The mean concentration of heavy metals in tomato fruits sampled from BS-treated, CF-treated, and control plots followed the order of Fe > Ni > Zn > Mn > Co > Cu > Cr > Cd > Pb > As, Fe > Mn > Ni > Zn > Cd > Co > Cu > Pb > Cr > As and Fe > Mn > Zn > Ni > Co > Cd > Cu > Cr > Pb > As, respectively (Table 7).

The mean concentration of Pb in tomato fruits sampled from CF-treated soil was higher ($0.25 \pm 0.2\,mg\,kg^{-1}$) than those sampled from BS-treated soils ($0.016 \pm 0.011\,mg\,kg^{-1}$), which was statistically insignificant ($P > 0.05$). In fact, the mean concentration of Pb in tomato fruits sampled from all treatments was below the MPL for human diets [35]. These tomatoes could therefore be safe for human consumption with regard to Pb toxicities. In contrast, there were significant differences ($P < 0.05$) among the mean concentrations of Cd in tomato samples collected from all the three treatments. In this regard, the application of CF yielded higher mean concentration of Cd ($7.98 \pm 0.72\,mg\;kg^{-1}$) compared to the application of BS ($0.0246 \pm 0.23\,mg\,kg^{-1}$). This may attribute to the presence of Cd in phosphate fertilizer and its high mobility and bioavailability at the low pH of the soil to plants. Sêkara et al. [88] suggested that Cd is found in CF and it is a mobile element, easily absorbed by the roots and transported to shoots where it is uniformly distributed in plants. Besides, the lowest concentration of Cd in tomato samples collected from BS-treated soil might be due to the ability of organic matter to immobilize the heavy metals like Cd, Pb, and As in the soil [89]. On top of that, the

Table 4: Mean concentration (mean ± SD) of heavy metals in pre-experiment soil and bio-slurry.

Sample	Heavy metal concentration (mg/kg dry weight)									
	As	Pb	Zn	Cd	Cu	Ni	Co	Fe	Mn	Cr
Soil	16.60 ± 2.76	28.17 ± 1.20	11.23 ± 0.93	4.34 ± 0.90	0.54 ± 0.002	3.30 ± 0.32	0.33 ± 0.01	213.13 ± 6.56	129.86 ± 4.47	4.54 ± 0.12
Bio-slurry	10.48 ± 0.63	17.69 ± 0.83	66.13 ± 3.1	8.80 ± 0.22	7.57 ± 0.79	3.59 ± 0.54	13.39 ± 0.33	550.87 ± 15.20	322.90 ± 4.94	0.26 ± 0.03
MPL	20.0	50.0	1000.0	3.0	300.0	50.0	50	50000	2000	75.0

MPL: maximum permissible limit for agricultural soils according to FAO/WHO [35]; SD: standard deviation.

Table 5: Effects of bio-slurry and chemical fertilizer application on the physico-chemical properties of the soils.

Treatment	Soil chemical property	Values
100% bio-slurry	Bulk density (g cm^{-3})	0.83
	Total porosity (%)	70
	Moisture content (%)	37
	pH (H_20)	7.4
	OC%	2.98
	TN	0.26
	Av. P (ppm)	43.2
	CEC (cmol kg^{-1})	29
	Ca (cmol kg^{-1})	28
	Na (cmol kg^{-1})	0.23
	K (cmol kg^{-1})	2.7
100% chemical fertilizer	Bulk density (g cm^{-3})	1
	Total porosity (%)	62.1
	Moisture content (%)	15.4
	pH (H_20)	5.62
	OC%	2.5
	TN (%)	0.24
	Av P (ppm)	42.5
	CEC (cmol kg^{-1})	23.6
	Ca (cmol kg^{-1})	22.2
	Na (cmol kg^{-1})	0.2
	K (cmol kg^{-1})	2.18
Control	Bulk density (g cm^{-3})	1
	Total porosity (%)	62
	Moisture content (%)	15.5
	pH (H_2O)	5.65
	OC (%)	2.73
	TN (%)	0.22
	Av P (ppm)	42.3
	CEC (cmol kg^{-1})	22.4
	Ca (cmol kg^{-1})	21.7
	Na (cmol kg^{-1})	0.21
	K (cmol kg^{-1})	2.29

mean concentration of Cd in tomato sampled from CF-treated soil and control soil was above the MPL standards for human diets [35]. Therefore, the application of CF and the control soil is unsafe for tomato cultivation for human consumption regarding Cd toxicities. Findings from previous studies showed that Cd is a highly mobile metal and is found to accumulate in plants in large amounts without showing phytotoxic symptoms [90, 91]. Similarly, higher mean concentration of Cd due to the application of CF was reported in previous studies [87, 92, 93].

The differences in mean concentrations of Ni in tomato samples among all the treatments were statistically significant ($P < 0.05$), with CF yielding the highest Ni concentration (23.9 ± 0.33 mg·kg^{-1}) and the control soil yielding the lowest Ni concentration (9.66 ± 1.2 mg·kg^{-1}). There are similar reports from previous studies [94, 95]. More importantly, the mean concentrations of Ni in tomato samples from all treatments were above the MPL for human diets [35]. This shows that tomatoes from the study site are unsafe for human consumption with respect to Ni toxicities.

The highest mean concentration (10 ± 0.22 mg·kg^{-1}) and the lowest (2.15 ± 0.34 mg·kg^{-1}) of Co were recorded in tomato samples sampled from BS and CF-treated soils, respectively,

Table 6: Comparison of mean concentration of heavy metals in bio-slurry-treated, chemical fertilizer-treated, and control soil (mg kg^{-1}) (mean ± SD) with MPL.

Metals	Soil treated with bio-slurry	Soil treated with chemical fertilizer	Control	MPL
As	13.8 ± 1^{a}	16.6 ± 2.7^{a}	16.5 ± 1.4^{a}	20
Pb	25.8 ± 1.09^{a}	28.1 ± 1.2^{a}	27.2 ± 1.6^{a}	50
Zn	16.43 ± 1.22^{a}	11.2 ± 0.93^{b}	15.75 ± 0.58^{a}	100
Cd	4.34 ± 0.9^{b}	12.2 ± 3.27^{a}	8.62 ± 1.9^{a}	3
Cu	1.88 ± 0.08^{a}	0.54 ± 0.002^{c}	0.88 ± 0.09^{b}	300
Ni	4.96 ± 1.4^{a}	2.454 ± 0.14^{b}	2.453 ± 0.43^{b}	50
Co	0.33 ± 0.011^{a}	0.29 ± 0.03^{a}	0.3 ± 0.03^{a}	60
Fe	213.1 ± 6.55^{a}	106.8 ± 1.4^{c}	120.9 ± 4.9^{b}	1500
Mn	144.7 ± 3.5^{a}	129.8 ± 4.47^{b}	142.1 ± 2^{a}	600
Cr	3.67 ± 0.76^{b}	6.14 ± 0.66^{a}	4.53 ± 0.12^{a}	75

Mean values with the different superscript letters in a row are significantly different from each other at $\alpha = 0.05$. MPL = maximum permissible limit of agricultural soil [35].

which were statistically significant ($P < 0.05$). There are similar findings from previous studies [96, 97]. Contextually, the mean concentration of Co in tomato samples from all treatments, including the control, is below the MPL for human diets [35]. Therefore, BS and CF application as well as the sole agricultural soils can be used for tomato cultivation, which is safe for human consumption with regard to Co toxicities.

There were significant differences ($P < 0.05$) between the mean concentrations of Zn in tomato fruits sampled from BS-treated (16.87 ± 2.26 mg·kg^{-1}) and CF-treated soil (10 ± 0.74 mg·kg^{-1}) and those from BS-treated and the control soil (9.77 ± 1.15 mg·kg^{-1}). The higher mean concentration of Zn in tomato fruit samples might have stemmed from the BS, which is in line with results from previous studies [92, 98, 99]. However, the mean concentrations of Zn in tomato fruit samples from all treatments, including the control, were below the MPL for human diets [35]. This shows that the application of BS and CF as well as the agricultural soils can be used for tomato cultivation, which is safe for human consumption with respect to Zn toxicities.

There were significant differences ($P < 0.05$) among the mean concentrations of Fe in tomato fruit samples from all the treatments, with the highest mean concentrations of Fe (57.1 ± 1.853 mg kg^{-1}) in BS-treated soil and the lowest (42.1 ± 0.34 mg kg^{-1}) in the control soil. The high Fe concentration in tomato fruits might be sourced from the BS, CF, and the agricultural soil itself. There are similar reports from previous studies [96, 100, 101]. Yet, the mean concentrations of Fe in tomato fruit samples were below the MPL for human diets [35]. This indicates that tomato can be grown with the application of BS and CF in the study site that is safe for human consumption with respect to toxicities of Fe.

There were significant differences ($P < 0.05$) among the mean concentrations of Mn in tomato fruit samples from all treatments, with the highest mean concentrations of Mn (38 ± 3.3 mg·kg^{-1}) in tomato fruit samples from CF-treated

Table 7: Concentrations of heavy metals (mg kg^{-1}) in tomato fruits grown in bio-slurry and chemical fertilizer-treated soil and control soil.

Metal	Bio-slurry-treated soil	Chemical fertilizer-treated soil	Control	MPL
As	0.013 ± 0.011^{a}	0.04 ± 0.01^{a}	0.03 ± 0.02^{a}	0.1
Pb	0.016 ± 0.011^{a}	0.25 ± 0.2^{a}	0.016 ± 0.011^{a}	0.3
Zn	16.87 ± 2.26^{a}	10 ± 0.74^{b}	9.77 ± 1.15^{b}	60
Cd	0.024 ± 0.023^{c}	7.98 ± 0.72^{a}	3.29 ± 0.37^{b}	0.2
Cu	2.63 ± 0.5^{a}	0.31 ± 0.24^{b}	1.0 ± 0.99^{b}	40
Ni	18.24 ± 0.61^{b}	23.9 ± 0.33^{a}	9.66 ± 1.2^{c}	2
Co	10 ± 0.22^{a}	2.15 ± 0.2^{c}	4.95 ± 0.9^{b}	50
Fe	57.15 ± 1.85^{a}	49.33 ± 3.2^{b}	42.1 ± 0.34^{c}	425
Mn	15.4 ± 2.4^{c}	38 ± 3.3^{a}	21.8 ± 0.99^{b}	0.2
Cr	0.093 ± 0.015^{b}	0.173 ± 0.011^{a}	0.031 ± 0.005^{c}	2.3

Mean values with the different superscript letters in a row are significantly different from each other at $\alpha = 0.05$. MPL = maximum permissible limit of agricultural soil [35].

soil and the lowest (15.4 ± 2.4 mg·kg^{-1}) from BS-treated soil. Previous studies reported that the application of CF and agricultural pesticides causes a high concentration of Mn in vegetables [102]. On top of that, the mean concentrations of Mn in tomato fruit samples from all the treatments, including the control, were above the MPL for human diets according to the FAO/WHO [35] standards. This indicates that the application of BS and CF could result in tomato fruits, which are unsafe for human consumption with respect to toxicities of Mn.

There was no significant difference ($P < 0.05$) in the mean concentrations of As in tomato fruit samples from all treatments despite the fact that the CF yielded higher mean concentration of As (0.04 ± 0.01 mg·kg^{-1}) than the BS application (0.013 ± 0.011 mg·kg^{-1}). There are similar reports from previous studies [93, 96]. Furthermore, the mean concentrations of As in all tomato fruit samples were below the MPL for human diets according to the FAO/WHO [35] standards. This shows that tomato grown in all the three soil treatments could be safe for human consumption with respect to As toxicity.

There was a significant difference ($P < 0.05$) in the mean concentrations of Cu between the tomato fruit samples from BS-treated and CF-treated soil as well as between bio-slurry-treated and control soil. The Cu concentration might be sourced from the BS that contains high concentrations of Cu (Table 4). There are similar reports from previous studies [94, 103]. The mean concentration of Cu in the tomato grown in all treatments is below the MPL for human diets according to the FAO/WHO [35] standards. This shows that tomato can be grown in the study site with the application of BS and CF, which is safe for human consumption in relation to Cu toxicity.

The highest mean concentration of Cr (0.173 ± 0.011 mg kg^{-1}) and the lowest (0.03 ± 0.005 mg·kg^{-1}) were recorded in tomato grown with CF-treated soil and the control soil, respectively. There were statistically significant differences ($P < 0.05$) among the mean concentrations of Cr in the tomato fruit samples from all treatments. There are similar reports from previous studies [87, 95, 104, 105]. The mean concentrations of Cr in tomato fruit samples from all treatments were below the MPL for human diets according to the FAO/WHO [35] standards. This shows that the application of BS and CF as well as the agricultural soil of the study site could be used for cultivation of tomato, which is safe for human consumption in association with Cr toxicity.

5.3. Potential Ecological Risks of Heavy Metals in the Soil. Contamination factor (CF) and pollution load index (PLI) were used to assess the potential ecological risks of heavy metals in the soil.

5.4. Contamination Factor (CF). The CF in BS-treated, CF-treated, and control soil followed this decreasing order: As > Pb > Fe > Cd > Zn > Mn > Ni > Cr > Cu > Co, As > Cd > Pb > Zn > Mn > Cu > Cr > Ni > Fe > Co and As > Pb > Cd > Zn > Mn > Cr > Ni > Fe > Cu > Co, respectively (Table 8). It mostly appears that there is a similarity in the distribution of heavy metals regardless of their sources. The CF values indicated contamination ranging from no contamination to moderate contamination of the soils. Accordingly, soil samples from the CF treatment and the control were moderately contaminated with Cd. This might be due to the fact that the agricultural soils used for the present study had been receiving various kinds of chemical fertilizers and agricultural pesticides in the production of various kinds of crops. Said et al. [106] also unveiled similar findings from their previous studies. The remaining heavy metals experienced CF values less than 1. Therefore, there is no contamination with reference to these metals (Table 8). There are similar findings from previous studies [22, 107–109].

5.5. Pollution Load Index (PLI). Table 8 presents the PLI and the combined pollution effect of different metals at different sampling locations. The PLI values of heavy metals were 0.13, 0.28, and 0.25 in soil treated with BS, CF, and control, respectively (Table 8). The highest PLI value was recorded in CF-treated soil, followed by the control soil, with the BS-treated soil registering the lowest PLI value. Despite that, the PLI values of all soil samples were less than 1, indicating no detectable pollution with heavy metals. The agricultural soil in the study site is not polluted (PLI < 1) with heavy metals and is therefore safe for tomato cultivation. There are similar reports from previous studies [22, 110–112].

Table 8: Contamination factor (CF) and pollution load index (PLI) of heavy metals.

Soil type	Contamination factor										PLI	Soil quality
	As	Pb	Zn	Cd	Cu	Ni	Co	Fe	Mn	Cr		
BTS	0.92	0.8	0.23	0.34	0.034	0.066	0.011	0.042	0.13	0.036	0.13	Not polluted
CFTS	1.1	0.88	0.16	0.98	0.098	0.04	0.01	0.02	0.12	0.067	0.28	Not polluted
CS	1.1	0.85	0.22	0.69	0.012	0.04	0.01	0.024	0.134	0.045	0.25	Not polluted

BTS = bio-slurry-treated soil, CFTS = chemical fertilizer-treated soil, and CS = control soil.

6. Conclusions

Compared to chemical fertilizers, the application of BS improved the physico-chemical properties (soil bulk density, porosity, and moisture content) of the soil. The agricultural soil of the study sites experienced Cd concentrations exceeding FAO/WHO standard for agricultural soils. The application of bio-slurry significantly influenced the mean concentrations of Cd, Cr, Cu, Ni, and Fe of the soil. The application of chemical fertilizers significantly increased the Cd concentration level in the soil. The application of bio-slurry significantly reduced the mean concentrations of Cd and Mn in the tomato fruit samples and optimized the Cd concentration to the range of safety limit for human consumption. Regardless of their sources, the concentrations of Ni and Mn in tomato fruit samples were above the safety limit for human consumption. When growing tomatoes, it is important to pay attention to the agricultural soil as well as the levels of heavy metals present in bio-slurry in relation to both food safety and soil health. Thus, monitoring the heavy metal concentrations in fertilizers, agricultural soil, and plant tissues is important to prevent excessive build-up of heavy metals in the human food chain and safeguarding food safety. Particularly, to reduce the accumulation of Mn and Ni in the tomato, other alternative heavy metal remediation methods such as phytoremediation are commendable.

Data Availability

The data used to support the findings of this study are included within the article.

Disclosure

A preprint has previously been published in Research Square in the following link: https://www.researchsquare.com/article/rs-1649597/v1.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

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