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RESEARCH ARTICLE

Heavy metal distribution and uptake by maize in a mudflat soil amended by vermicompost derived from sewage sludge

Wengang Zuo[1,2] · Kaida Xu[1] · Wenjie Zhang[1] · Yao Wang[1] · Chuanhui Gu[3] · Yanchao Bai[1,4] · Yuhua Shan[1,4] · Qigen Dai[2]

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Abstract

Sewage sludge has been regarded as an economic and efficient soil amendment for mudflat soil amendment despite of the concern of heavy metal contamination. Converting sewage sludge into vermicompost by earthworms may be effective to minimize the risk of heavy metal contamination caused by direct application of sewage sludge in mudflat soil. The objective of this study was to assess the feasibility of vermicompost amendment (VA), and its influence on heavy metal contamination compared with sewage sludge amendment (SSA) in mudflat soil. The results showed that VA improved the physicochemical properties of mudflat soil by decreasing soil bulk density, salinity, and pH, increasing soil organic carbon, nitrogen, and phosphorus contents in the soil. Consequently, the maize biomass and yield were significantly elevated by VA. For heavy metals, VA increased total Cd, Cu, Ni, Pb, and Zn concentrations in mudflat soil, and the maximum increments occurred at 250 t ha^{-1}. Available Cd, Cu, Mn, Ni, and Zn concentrations significantly increased with increasing VA rates. VA increased the accumulation of Cd, Cu, Mn, Ni, Pb, and Zn in maize tissues, especially in root of maize. Compared with SSA under the condition of maintaining equal carbon input, VA allowed heavy metals to accumulate in a more stable binding form in the top 20-cm layer of mudflat soil. Thus, the risk of runoff and leaching of heavy metals and their bioavailability to plants reduced in mudflat soil. As a result, VA decreased the accumulation of heavy metals in maize plant compared with SSA in mudflat soil. In summary, vermicompost can be an effective and safe substitute for sewage sludge for mudflat amendment.

Keywords Saline-alkaline soil · Soil amendment · Vermicompost · Heavy metal · Maize

Introduction

The mudflat lands in eastern China are feasible alternative land resources for agricultural purpose. Converting mudflat into cropland is an effective way to solve the shortage of cultivated land resources and meet the growing demand for food. However, high soil salinity, low soil fertility, and poor soil structure are the main factors restricting direct use of the newly reclaimed mudflat for crop cultivation. Applying organic fertilizers is an effective measure to reduce soil salinity and elevate soil fertility in mudflat (Long et al. 2016; Zhang et al. 2015).

Sewage sludge produced by municipal sewage water treatment is an organic resource that can be reused primarily on account of the larger quantities of nutrients (especially phosphorus and nitrogen) and organic matter. Both nutrients and organic carbon in sewage sludge are technically and economically feasible to recycle, which makes sewage sludge a possible economic and efficient soil amendment material (Alvarenga et al. 2015; Antolín et al. 2005; Rigby et al. 2016; Singh and Agrawal 2008). However, amending agricultural soil with sewage sludge is still hampered firstly due to the nature of sewage sludge itself with strong stink, high moisture

Wengang Zuo and Yanchao Bai contributed equally to this work.

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✉ Yuhua Shan
shanyuhua@gmail.com

[1] College of Environmental Science and Engineering, Yangzhou University, Yangzhou 225127, China

[2] Co-Innovation Center for Modern Production Technology of Grain Crop / Jiangsu Key Laboratory of Crop Genetics and Physiology, Yangzhou University, Yangzhou 225127, China

[3] School of Environment, Beijing Normal University, Beijing 100875, China

[4] Jiangsu Collaborative Innovation Center for Solid Organic Waste Resource Utilization, Nanjing 210095, China

content, and compact physical structure, and secondly due to the safety concern of sewage sludge especially heavy metals for its high toxicity, easy accumulation, and biological transfer effect. Many studies have found that applied sewage sludge improved soil physicochemical properties, but led to accumulation of heavy metals in soils and plants (Antonkiewicz et al. 2018; Jalali and Arfania 2011; Kidd et al. 2007), and agricultural use of sewage sludge potentially had adverse effects on human and ecosystem health (Antonkiewicz et al. 2019; McBride 2003). Our previous studies showed that sewage sludge amendment improved mudflat soil physicochemical properties, promoted plant growth, and, inevitably, caused accumulation of heavy metals in mudflat soils and plants grown in mudflat (Bai et al. 2017a, b; Zuo et al. 2018). The amount of sewage sludge input and heavy metal concentration in the initial sewage sludge are main factors determining the concentration of heavy metals in sludge-amended soil (Gu et al. 2013). Therefore, pre-treatments to minimize the risk of heavy metal accumulation in plant and soil caused by direct application of sewage sludge are necessary for its safe use in agriculture. Feeding the earthworms with sewage sludge and converting the sludge into vermicompost are potentially effective measures to reduce heavy metal concentration in sludge. Studies have shown that heavy metal concentrations in sewage sludge were reduced after vermicomposting (He et al. 2016; Azizi et al. 2013). The reduction of heavy metal in the sludge was mainly due to the bioaccumulation of heavy metals by earthworms during vermicomposting (Wang et al. 2013). Additionally, the stability of heavy metal in sewage sludge was enhanced after vermicomposting (Gupta and Garg 2008; Hait and Tare 2012; Suthar 2009), which could reduce the risk of heavy metal contamination.

Furthermore, converting sewage sludge into vermicompost can effectively improve the initial properties of sewage sludge, including deodorization, moisture content reduction, and physical structure improvement. Vermicompost as a stabilized organic material is considered to be an optimum soil amendment and plant growth media due to its excellent physical properties, rich nutrients, and high microbial activity (Edwards 2004). Application of vermicompost could improve soil physical structure, increase soil aggregate stability, and provide vital macronutrient elements and microelements (Arancon et al. 2008; Edwards 2004; Atiyeh et al. 2000a). In addition, application of vermicompost to soil increased crop growth and yields (Atiyeh et al. 2000b; Warman and Anglopez 2010; Zhao et al. 2017).

Previous studies have mainly focused on variations of heavy metals in the vermicomposting process (Gogoi et al. 2015; Liu et al. 2005) and on the changes of soil physicochemical properties and plant growth after vermicompost amendment in farmland and greenhouse (Khan et al. 2015; Wang et al. 2017). As we know, information on mudflat amendment with vermicompost derived from sewage sludge was scarce, and specifically, distribution and uptake of heavy metals in mudflat soil with high salinity, low fertility, and poor soil structure remained unknown. In this study, vermicompost derived from sewage sludge was applied to a newly reclaimed mudflat soil in different application rates, and an initial sewage sludge treatment with equal carbon application rate was set up to compare the differences in heavy metal behaviors in mudflat soil between sewage sludge and sludge-based vermicompost. Maize (*Zea mays* L.) was selected as a trial plant. The objectives of this study were to investigate feasibility and safety of mudflat soil amendment with vermicompost and evaluate the risk of heavy metals in mudflat soil and maize compared with sewage sludge amendment. Specifically, the effects of vermicompost amendment on soil physicochemical properties, accumulation, and distribution of heavy metals in mudflat soil and uptake of heavy metals by maize were studied.

Materials and methods

Site description and experimental materials

The field experiment was conducted in Tiaozini reclamation area (32° 50′ 4.39‴ N, 120° 56′ 34.57″ E) in Dongtai county of Jiangsu Province, China. The experimental site was reclaimed from coastal mudflats in 2015. This area has distinct seasons and marine monsoon climate characteristics. The annual average temperature, relative humidity, and average annual evaporation of this area are 14.6 °C, 81%, and 1417 mm, respectively. Rainfall is mainly concentrated between June and September. The soil in the experimental site is typical muddy beach saline soil and its basic properties are shown in Table 1.

Sewage sludge used was taken from municipal sewage treatment plant of Rudong county in 2015, which complied with the control standards for agricultural use in China (GB 4284-2018). Thereafter, sewage sludge was transported to earthworm breeding farm (Yangzhou Agricultural Environmental Safety Technical Service Center, Jiangsu, China) to complete vermicomposting process of sewage sludge. Firstly, after the moisture content of sewage sludge was air-dried to 75–80% in plastic greenhouse, sewage sludge was piled up into a series of ridges (500 cm in length, 60 cm in height, and 35 cm in height), and the distance between the ridges was about 35 cm. After that, earthworms (*Eisenia fetida*) were inoculated in sewage sludge ridges according to the amount of 20-kg earthworms per ton of sewage sludge. Then, the black shading net was used to cover the top of sewage sludge ridges, and the moisture content of sludge ridges was regularly regulated to maintain a relatively constant temperature (18 ± 2 °C) and humidity. During the vermicomposting period, sewage sludge was turned over

Table 1 Basic properties of mudflat soil, sewage sludge and vermicompost used in this study

Items	Mudflat soil	Sewage sludge	Vermicompost	Municipal wastewater treatment plant-control standards for agricultural use in China (GB 4284-2018)	The control standards of soil environment in China (GB15618-2008)
pH	9.63	5.91	6.34		
Salinity (‰)	4.26	27	22.3		
Organic matter (g kg^{-1})	4.52	501	465		
Total N (N g kg^{-1})	0.41	32.8	24.4		
Total P (P g kg^{-1})	0.56	18.6	15.9		
Alkaline N (N mg kg^{-1})	42.2	1838	2466		
Available P (P mg kg^{-1})	27.0	517	869		
Total Cd (mg kg^{-1})	0.53	3.47	2.94	15	1.0
Total Cu (mg kg^{-1})	15.1	587	436	1500	100
Total Mn (mg kg^{-1})	184	210	168		
Total Ni (mg kg^{-1})	25.1	56.3	45.6	200	100
Total Pb (mg kg^{-1})	21.8	48.2	38.3	1000	80
Total Zn (mg kg^{-1})	63.5	1062	805	3000	300

N, nitrogen; *P*, phosphorus; *Cd*, cadmium; *Cu*, copper; *Mn*, manganese; *Ni*, nickel; *Pb*, plumbum; *Zn*, zinc

every 10 days to ensure that sewage sludge was fully digested by earthworms. When the sludge ridges had no distinct odor, it can be considered that sewage sludge was completely digested by earthworms. About 30–40 days later, the vermicompost was obtained after removal of earthworms. The basic properties of initial sewage sludge and vermicompost are shown in Table 1.

Experimental design

Six vermicompost treatments based on the amount of vermicompost (0, 25, 50, 100, 150, and 250 t ha^{-1} on dry weight basis) were conducted in this experiment, which were defined as V0, V25, V50, V100, V150, and V250, respectively. Meanwhile, an initial sewage sludge treatment (140 t ha^{-1} on dry sewage sludge weight basis) with equal carbon application rate compared with 150 t ha^{-1} vermicompost application rate was set up, which was defined as SS140. In all treatments (V0, V25, V50, V100, V150, V250, and SS140), V0 was defined as a control treatment. Vermicompost and sewage sludge were used through a one-off application. These treatments were laid out in a randomized complete block design and replicated 3 times. The plot area is 10.08 m^2 and the length and width of each experiment plot are 3.6 m and 2.8 m, respectively. Vermicompost and sewage sludge were mixed with the soil in the plow layer by rotary cultivator in April 2015. Maize (*Z. mays* L.) was sown by artificial seeding on June 9, 2016, with the inter-row distance of 50 cm × 25 cm, two seeds per hole. After 20 days of sowing, the seedlings were thinned and one seedling was left in each hole. Weeds were removed by hand at 20 and 48 days after sowing. The maize was rain-fed without irrigation throughout the growing season. Soil and maize plant in each plot were sampled for lab analysis on August 23, 2016.

Soil sampling and analysis

Nine soil samples were randomly collected at each plot throughout the upper 20 cm and then they were mixed up. The visible organic residues and plant debris in the mixed soil samples were removed during naturally air drying process. After soil samples were air-dried, all samples were crushed and sieved (1 mm and 0.150 mm) for further physicochemical analysis. The determination of soil physicochemical properties had duplicates. In addition, we used certified reference materials (CRM) to ensure the accuracy of the results. Soil bulk density was measured by cutting ring method. Soil pH was detected in a 5/1 (water volume/soil weight) suspension by pH meter (Model IQ150, Spectrum, USA). Soil salinity was measured by gravimetric method. For analysis of soil organic carbon (SOC), 0.30-g air-dried soil sample through the 0.150 mm mesh sieve was measured by the potassium dichromate ($K_2Cr_2O_7$) oxidation method. The Kjeldahl method and digestion-Mo-Sb anti-spectrophotometric method were taken for the analysis of soil total nitrogen (N) and total phosphorus (P) contents. Alkaline N and available P contents were analyzed by alkaline hydrolysis diffusion method and $NaHCO_3$ extraction method, respectively. Specific determination methods and steps for available heavy metals, total heavy metals, and sequential extraction for heavy metals geochemical fractions in soil samples were described in our previous studies (Bai et al. 2016; Zuo et al. 2018).

Plant analysis

Maize was harvested on August 23, 2016. The maize cobs in each plot were collected separately and then dried in the sun. When the moisture content of the grain decreased to 13%, the maize cobs were threshed by threshing machine to get maize grain and grain yield was recorded. For the determination of maize biomass, 5 maize plants were excavated completely in each plot, and then the roots of maize were washed with water until there was no soil residue. Maize plant samples were then separated into four parts: root, stem, leaf, and grain. The four fresh parts were killed at 105 °C for 15 min and then oven-dried at 80 °C to reach constant weight. The four parts were then weighed separately to get the biomass of maize plant, which was expressed as grams per plant. Specific method and steps for the determination of heavy metals in maize tissues samples were described in our previous study (Zuo et al. 2018).

Statistical analysis

Translocation factors (TFs) for aerial part (stem, leaf, and grain) and root were calculated using the following equation: $TF_{ap/root} = C_{ap}/C_{root}$. C_{ap} and C_{root} represent the heavy metal concentration in aerial part and root, respectively. Bioaccumulation factors (BAFs) were calculated as the ratio of the heavy metal concentration in maize root to the heavy metal concentration in mudflat soil.

Accumulation ratio (AR) and uptake ratio (UR) were defined for comparison of heavy metal accumulation in soil and heavy metal uptake by maize between vermicompost amendment (VA) and sewage sludge amendment (SSA). AR was calculated using the following equation: $AR = (H_{VA}/I_{VA})/(H_{SSA}/I_{SSA})$. I_{VA} and I_{SSA} represent the average initial total amount of heavy metal in the experimental plot after vermicompost application and sludge application, respectively. H_{VA} and H_{SSA} represent the average total amount of heavy metal in vermicompost-amended mudflat soil and sludge-amended mudflat soil in maize harvesting stage, respectively. AR > 1 indicates greater metal accumulation in VA than that in SSA, and vice versa. UR was calculated using the following equation: $UR = (M_{VA}/M_{SSA})/(I_{VA}/I_{SSA})$. M_{VA} and M_{SSA} represent the average total amount of heavy metal in maize grown in vermicompost-amended mudflat soil and sewage sludge-amended mudflat soil, respectively. UR > 1 indicates greater heavy metal uptake by maize in VA than that in SSA, and vice versa.

The analysis of variance (ANOVA) for the data with the model of randomized complete block (RCB) design was carried out by SPSS 13.0 software (SPSS Inc., USA). Then, multi-comparisons using least significant difference (LSD) method at $p < 0.05$ were conducted to identify significant differences between the treatments.

Results

Physicochemical properties of amended mudflat soil

Vermicompost addition decreased soil bulk density in mudflat (Table 2). Compared with 1.31 g cm^{-3} in the control soil (V0), the decrements of soil bulk density in vermicompost-amended mudflat were 2.3%, 5.3%, 8.4%, 12.2%, and 15.3% in V25, V50, V100, V150, and V250, respectively. In the treatments with ≥ 50 t ha^{-1} VA rates, soil bulk density was significantly higher ($p < 0.05$). With increasing rates of vermicompost, soil pH and salinity decreased in mudflat soil (Table 2). The decrements of salinity in vermicompost-amended mudflat soil in V25, V50, V100, V150, and V250 treatments were 10.3%, 25.9%, 34.7%, 39.4%, and 47.8%, respectively. Soil pH and salinity in vermicompost-amended mudflat at the treatments with ≥ 100 t ha^{-1} and ≥ 25 t ha^{-1} VA rates were significantly lower than those in the control soil, respectively. Soil organic carbon, total N, total P, alkaline N, and available P contents in vermicompost-amended mudflat soil increased with increasing VA rates and were significantly higher than those in the control soil. Compared with SSA under the condition of equal carbon application rate, bulk density and total N were significantly lower and pH and alkaline N were significantly higher in vermicompost-amended mudflat soil.

Biomass and yield of maize

Under the influence of vermicompost addition, the dry matter of root, stem, leaf, and grain of maize grown in vermicompost-amended mudflat soil was significantly higher than that in the control soil (Table 3). The maximum increments in dry matter of root, stem, leaf, and grain of maize were 423.8%, 140.1%, 140.2%, and 249.1% in V250, respectively. Due to the improvement in soil fertility, an increase in maize yield was observed under vermicompost treatments. Grain yields of maize in V25, V50, V100, V150, and V250 were 2.51, 3.38, 3.67, 4.20, and 4.62 t ha^{-1}, with the increments of 71.4%, 137.8%, 158.6%, 196.1%, and 225.6%, respectively, compared with 1.42 t ha^{-1} in the control treatment. Except for root dry weight, the dry matter of stem, leaf, and grain of maize and yield were higher in vermicompost-amended mudflat than that in sewage sludge-amended mudflat under the condition of equal carbon application rate.

Heavy metals in maize

VA significantly affected heavy metal concentrations in different maize plant parts (Fig. 1). Cd concentrations in root, stem, and grain of maize increased with increasing VA rates. Cd concentrations in root, stem, and grain of maize at ≥ 50 t ha^{-1} VA rates were significantly higher than those in the control soil. However, Cd concentrations in the leaf of maize did

Table 2 Selected soil physicochemical properties under vermicompost and sewage sludge amendment in mudflat soil

Parameters	Treatments						
	V0	V25	V50	V100	V150	V250	SS140
Bulk density (g cm^{-3})	1.31 ± 0.03 a	1.28 ± 0.02 ab	1.24 ± 0.05 bc	1.20 ± 0.06 cd	1.15 ± 0.02 de	1.11 ± 0.03 e	1.23 ± 0.03 bc
pH (5:1)	9.34 ± 0.06 a	9.32 ± 0.09 a	9.26 ± 0.07 a	9.03 ± 0.04 b	8.93 ± 0.06 b	8.97 ± 0.04 b	8.73 ± 0.04 c
Salinity (‰)	4.04 ± 0.13 a	3.62 ± 0.17 b	2.99 ± 0.13 c	2.64 ± 0.03 d	2.45 ± 0.05 d	2.11 ± 0.10 e	2.63 ± 0.08 d
Organic carbon (g kg^{-1})	4.86 ± 0.74 f	6.92 ± 0.31 e	8.77 ± 0.51 d	11.76 ± 1.12 c	14.59 ± 0.62 b	17.66 ± 1.16 a	14.25 ± 0.37 b
Total N (N g kg^{-1})	0.46 ± 0.07 g	0.91 ± 0.07 f	1.22 ± 0.09 e	1.43 ± 0.07 d	1.83 ± 0.06 c	2.20 ± 0.09 a	2.04 ± 0.08 b
Total P (P g kg^{-1})	0.64 ± 0.01 d	0.68 ± 0.02 d	0.70 ± 0.01 d	1.05 ± 0.05 c	1.27 ± 0.07 b	1.43 ± 0.01 a	1.22 ± 0.06 b
Alkaline N (N mg kg^{-1})	47.1 ± 2.17 f	55.0 ± 0.83 e	62.7 ± 4.93 d	88.5 ± 5.96 c	104.0 ± 5.20 b	111.6 ± 4.93 a	94.3 ± 2.11 c
Available P (P mg kg^{-1})	34.0 ± 4.04 e	55.6 ± 13.96 d	79.5 ± 4.63 c	90.2 ± 7.56 c	127.1 ± 15.34 b	169.2 ± 11.29 a	113.6 ± 6.25 b

Values are mean ± SD of three replicates. Different letters in each row indicate significant difference at $p < 0.05$

not show significant difference among different VA rates. Cu concentrations in grain of maize showed no significant change ($p > 0.05$). However, Cu concentrations in root, stem, and leaf of maize slightly increased in the VA treatments. Pb concentrations in root, stem, leaf, and grain of maize all increased with increasing VA rates. Compared with those in the control soil, the maximum increments for Pb in root, stem, leaf, and grain of maize were 119.5%, 211.8%, 76.7%, and 263.6% in V250, respectively. Mn, Ni, and Zn concentrations in root, stem, and leaf of maize all increased with increasing VA rates. Compared with those in the control soil, the maximum increments for Mn in root, stem, and leaf of maize were 102.2%, 188.4%, and 156.4% in V250. The maximum increments for Ni in root, stem, and leaf of maize were 238.1%, 106.9%, and 122.8% in V250. The maximum increments for Zn in root, stem, and leaf of maize were 130.7%, 256.0%, and 149.0% in V250, respectively. There were no significant changes ($p > 0.05$) for Mn and Zn concentrations in grain of maize with increasing VA rates. Under the condition of equal carbon application rate, Cd, Cu, Mn, Ni, Pb, and Zn in root, stem, leaf, and grain of maize were all lower in vermicompost-amended mudflat than those in the sewage sludge-amended mudflat.

Heavy metals in mudflat soil

VA affected total metal concentrations in mudflat soil. Total Cd, Cu, Ni, Pb, and Zn concentrations in vermicompost-amended mudflat soil increased with increasing VA rates, and the increments reached significant levels compared with those of the control soil (Fig. 2). The maximum increments of Cd, Cu, Ni, Pb, and Zn concentrations were 66.2%, 183.2%, 28.6%, 48.6%, and 85.8% in V250, respectively. Mn concentrations showed no significant change with increasing VA rates in mudflat soil. Under the condition of equal carbon application rate, VA significantly increased total Cd, Ni, Pb, and Zn concentrations in mudflat soil compared with SSA. Total Cu and Mn concentrations had no significant change in response to VA and SSA.

VA significantly increased the available metal concentrations for Cd, Cu, Ni, and Zn in mudflat soil (Fig. 3). The maximum increments of available Cd, Cu, Mn, Ni, and Zn concentrations in vermicompost-amended mudflat soil were observed in V250 treatment. The threshold VA rate for significant differences between the control and VA treatments for available Cd, Cu, Mn, Ni, and Zn concentrations was found in

Table 3 Biomass and yield of maize in response to vermicompost and sewage sludge amendment in mudflat soil

Treatments	Dry matter (g $plant^{-1}$)				Yield (t ha^{-1})
	Root	Stem	Leaf	Grain	
V0	2.61 ± 0.53 d	35.53 ± 4.02 d	36.29 ± 3.69 d	20.25 ± 3.21 e	1.42 ± 0.33 e
V25	5.94 ± 0.29 c	45.33 ± 4.42 d	45.61 ± 3.82 c	37.39 ± 4.76 d	2.51 ± 0.38 d
V50	10.62 ± 0.49 b	63.05 ± 3.62 bc	64.93 ± 4.26 b	47.97 ± 8.02 c	3.38 ± 0.81 c
V100	9.83 ± 0.63 b	65.08 ± 7.51 bc	68.26 ± 6.50 bc	56.14 ± 3.89 b	3.67 ± 0.30 bc
V150	10.78 ± 0.99 b	71.30 ± 8.89 b	71.58 ± 5.71 b	60.78 ± 4.11 b	4.20 ± 0.18 ab
V250	13.67 ± 2.99 a	85.31 ± 6.42 a	87.18 ± 4.77 a	70.70 ± 2.31 a	4.62 ± 0.12 a
SS140	15.78 ± 1.00 a	58.60 ± 4.55 c	62.36 ± 4.31 c	56.19 ± 3.75 b	3.86 ± 0.06 bc

Values are mean ± SD of three replicates. Different letters in each column indicate significant difference at $p < 0.05$

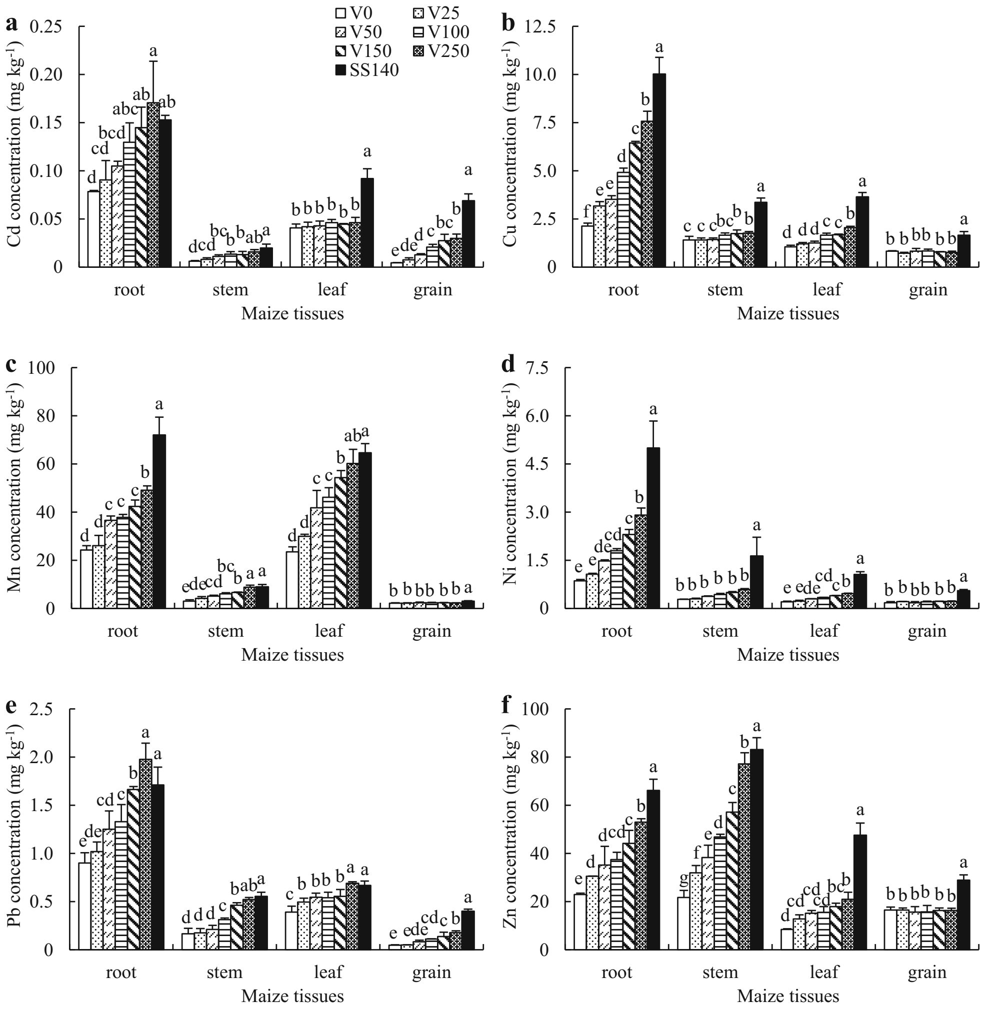

Fig. 1 Heavy metal concentrations for Cd (**a**), Cu (**b**), Mn (**c**), Ni (**d**), Pb (**e**), and Zn (**f**) in different maize plant parts under vermicompost and sewage sludge amendment in mudflat soil. The vertical bars denote standard errors. Different small letters denote significant difference at $p < 0.05$. Cd, cadmium; Cu, copper; Mn, manganese; Ni, nickel; Pb, lead; Zn, zinc

V100, V50, V50, V100, and V100, respectively. The available metal concentrations for Pb showed no significant change with increasing VA rates. Compared with SSA under the condition of equal carbon application rate, VA significantly decreased the concentrations of available heavy metals in mudflat soil. The decrements of Cd, Cu, Mn, Ni, Pb, and Zn were 11.7%, 48.8%, 38.7%, 37.3%, 15.8%, and 41.5%, respectively.

The acid-soluble/exchangeable (EX) fraction is regarded as the labile fraction and the residue (RES) fraction is regarded as nonlabile fraction. The activity of reducible (RG) and oxidizable (OXI) fractions depends on soil physical and chemical properties. Each heavy metal fractions for Cd, Cu, Mn, Ni, Pb, and Zn were presented as a percentage of all fractions (Fig. 4). VA significantly affected the distribution of heavy metal fractions in mudflat soil. With increasing rates of vermicompost applying, the labile fraction (EX) of heavy metals increased. Compared with the control soil, the maximum increments of EX fractions of Cd, Cu, Mn, Ni, Pb, and Zn in vermicompost-amended mudflat soil were 138.3%, 122.2%, 51.2%, 99.3%, 14.0%, and 92.4% in V250, respectively. In contrast, VA decreased the nonlabile fraction (RES) of heavy metals. The sum of RG and OXI fractions also increased with increasing VA rates. VA decreased the labile fraction for Cd, Cu, Mn, Ni, Pb, and Zn in mudflat soil compared with SSA under the condition of equal carbon application rate.

BAF and TF of heavy metals

The bioaccumulation factors (BAFs) indicating the difficulty of metal uptake by maize root from the mudflat soil were all less than 1 (Table 4). VA increased the BAFs of heavy metals in mudflat soil. BAFs for Cd, Cu, Mn, Ni, Pb, and Zn increased with increasing VA rates. Mean values of BAFs indicating the difficulty of plant uptake of heavy metals were: Zn > Cd > Cu > Mn > Pb > Ni. The translocation factors (TFs), ratio of heavy metal concentration in aerial part (stem, leaf, and grain) and root part in maize, for all heavy metals were less than 1 (Table 4). TFs of Cd, Cu, Ni, and Pb showed a decreasing trend with increasing VA rates, while Mn and Zn showed higher TF values in vermicompost-amended mudflat

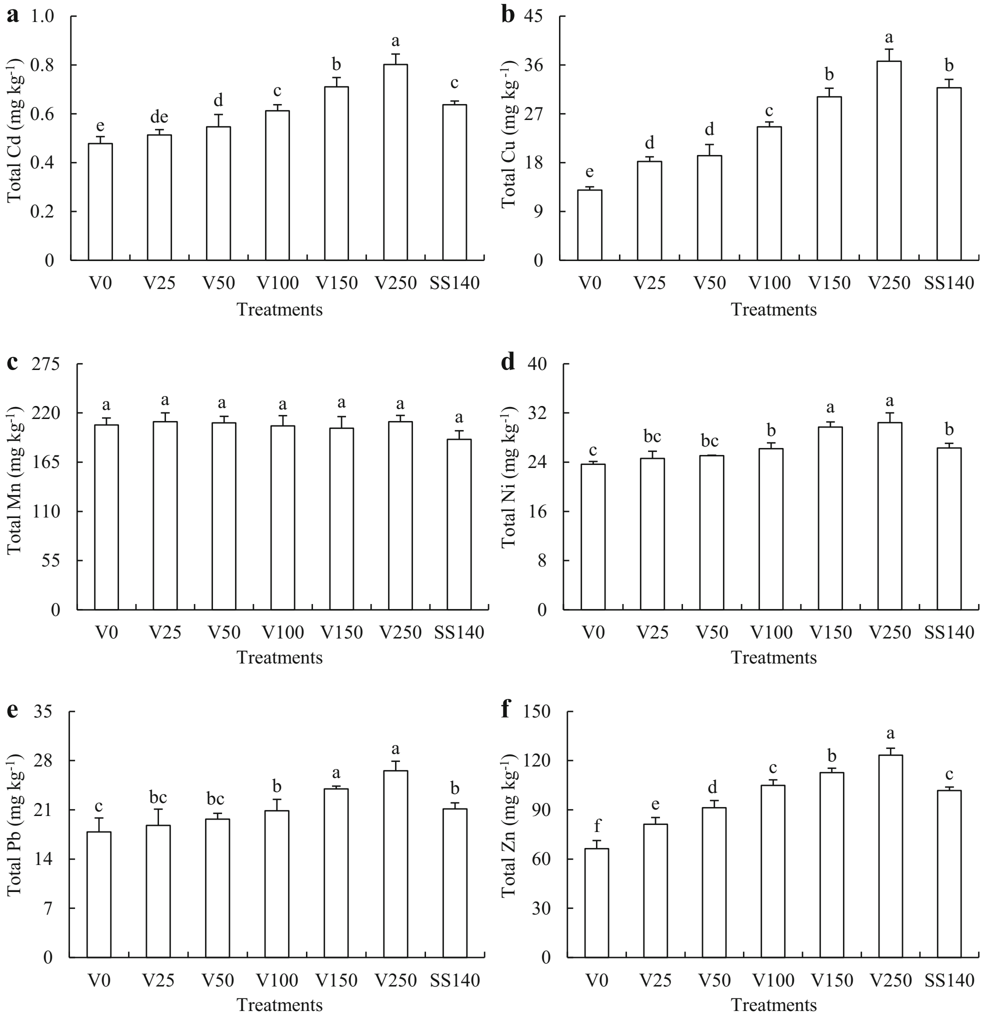

Fig. 2 Total metal concentrations for Cd (**a**), Cu (**b**), Mn (**c**), Ni (**d**), Pb (**e**), and Zn (**f**) in vermicompost-amended and sewage sludge-amended mudflat soil. The vertical bars denote standard errors. Different small letters denote significant difference at $p < 0.05$. Cd, cadmium; Cu, copper; Mn, manganese; Ni, nickel; Pb, lead; Zn, zinc

soils than those in control soils. Mean values of TFs followed the order of Zn > Mn > Cu > Pb > Cd > Ni.

Discussion

VA provided the nutrients needed for maize growth in mudflat soil and promoted the growth of maize. Previous field and greenhouse studies reported that application of vermicompost significantly increased growth and yield of crops including vegetable crops such as tomato, cucumber, and pepper (Arancon et al. 2004a; Wang et al. 2017; Zhao et al. 2017); fruit crops such as strawberry (Arancon et al. 2004b); and field crops such as maize, bean, and wheat (Doan et al. 2013; Kizilkaya et al. 2012; Valdez-Pérez et al. 2011). In this study, we found that maize yield and VA rate were positively and significantly correlated, with the quadratic equations of $y = -5\mathrm{E} - 05x^2 + 0.024x + 1.8002$ ($R^2 = 0.91$, $n = 18$). The increase of yield and dry matter of maize were mainly due to available nutrients and microbial metabolites from vermicompost amendment, which was considered to be high-quality plant growth regulators (Edwards 2004). In addition, the improvement of physicochemical properties of vermicompost-amended mudflat soil also favored maize growth.

High soil salinity and low soil fertility are the main factors restricting the direct use of the newly reclaimed mudflats for crop cultivation. The higher salinity and the long-term repeatability of water-salt movement in mudflat soil are probably due to the high bulk density and dense capillary. In our previous studies (Bai et al. 2013, 2017b), SSA improved soil physical properties of mudflat by reducing bulk density and increasing water-stable aggregate content and thereby decreased the soil salinity. In this study, VA also reduced soil bulk density in mudflat soil. The decrease in soil bulk density and the increase in soil porosity broke the soil dense capillary and thereby prevented salty groundwater from moving up to the ground surface (Bai et al. 2013; Jorenush and Sepaskhah 2003). Consequently, the salinity of mudflat soil dropped. VA decreased soil pH, which was similar to SSA in our previous studies. The reason might be the release of humic acid substances from the biodegradation of organic materials (Moreno et al. 1997). Besides, lower-pH vermicompost

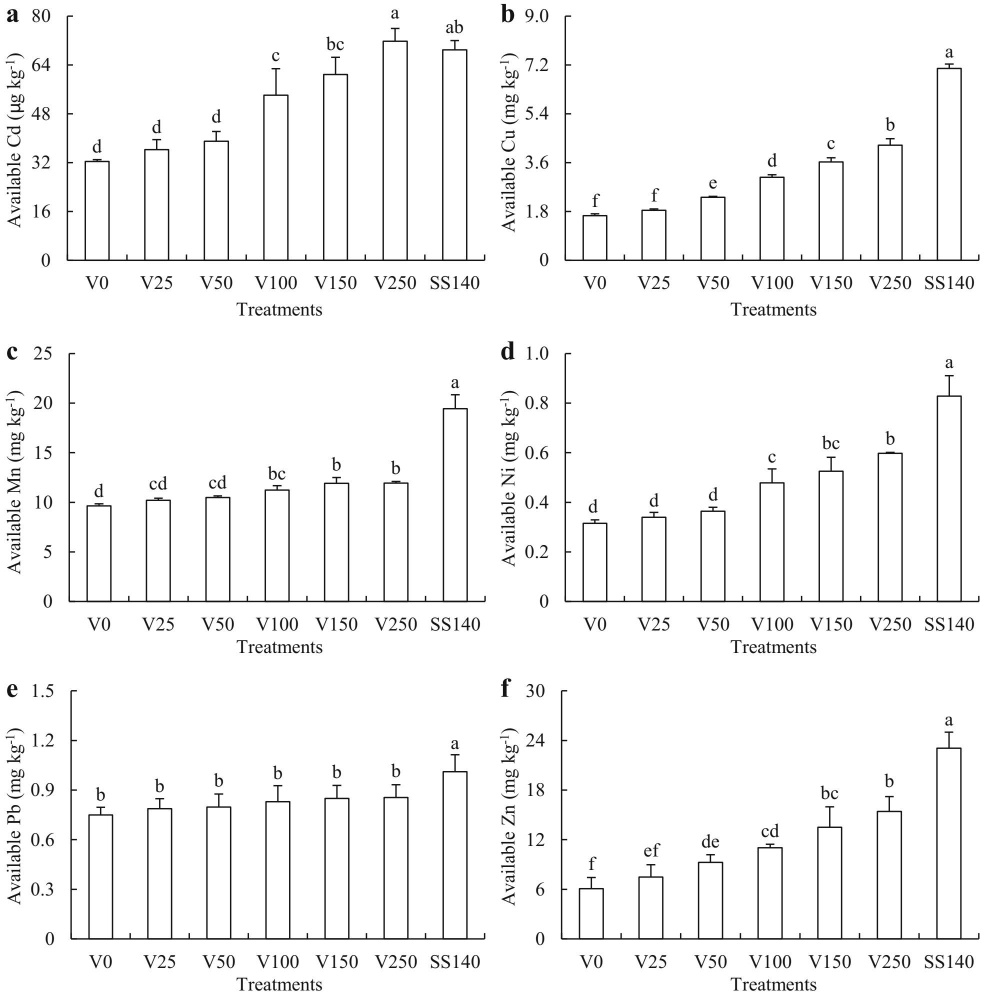

Fig. 3 Available heavy metal concentrations for Cd (**a**), Cu (**b**), Mn (**c**), Ni (**d**), Pb (**e**), and Zn (**f**) in vermicompost-amended and sewage sludge-amended mudflat soil. The vertical bars denote standard errors. Different small letters denote significant difference at $p < 0.05$. Cd, cadmium; Cu, copper; Mn, manganese; Ni, nickel; Pb, lead; Zn, zinc

addition could neutralize alkaline mudflat soil, especially at high vermicompost application rate. Rapid mudflat soil fertility improvement requires the input of a large amount of organic materials because soil fertility improvement is extremely slow under natural conditions. VA increased soil organic carbon, total nitrogen, total phosphorus, alkaline nitrogen, and available phosphorus contents, which is consistent with previous studies in greenhouse and farmland soils amended by vermicompost (Kale et al. 1992; Lim et al. 2015; Yang et al. 2015; Zhao et al. 2017), which found that application of vermicompost improved soil fertility because of the considerable amounts of organic matter, nitrogen, and phosphorus nutrients in vermicompost. Existing researches found that low concentrations of some heavy metals were beneficial to plant growth, and excessive concentrations might show toxicity and inhibit plant growth (Pritchard et al. 2010; Sinha et al. 2005). Although higher vermicompost application rate led to higher uptake of heavy metals in maize in this study, the maize growth was not inhibited even at the highest vermicompost application rate probably because of relatively low heavy metal accumulation in maize. In addition, the improvement of physical properties of mudflat soil and large amounts of nutrients supplied by application of vermicompost provided a favorable condition for maize root growth, which could override heavy metal stress. Therefore, application of vermicompost can also achieve rapid improvement of mudflat soil and promote crop growth compared with the direct application of sewage sludge.

The total heavy metal concentration in organic material-amended soil is mainly determined by the amount of the organic material input plus the heavy metal concentration of organic material (Gu et al. 2013). In this study, application of vermicompost resulted in the increase of soil total heavy metal concentrations except for Mn in mudflat. The concentrations of heavy metals in vermicompost-amended mudflat and VA rates were positively and significantly correlated with the linear equations of $y = 0.001x + 0.4822$ ($R^2 = 0.99$, $p < 0.01$, $n = 18$), $y = 0.0923x + 14.79$ ($R^2 = 0.99$, $p < 0.05$, $n = 18$), $y = 0.029x + 23.826$ ($R^2 = 0.92$, $p < 0.01$, $n = 18$), $y = 0.0357x + 17.881$ ($R^2 = 0.98$, $p < 0.01$, $n = 18$), and $y = 0.2151x + 76.011$ ($R^2 = 0.90$, $p < 0.01$, $n = 18$) for Cd, Cu, Ni, Pb, and Zn, respectively. The increases of total heavy

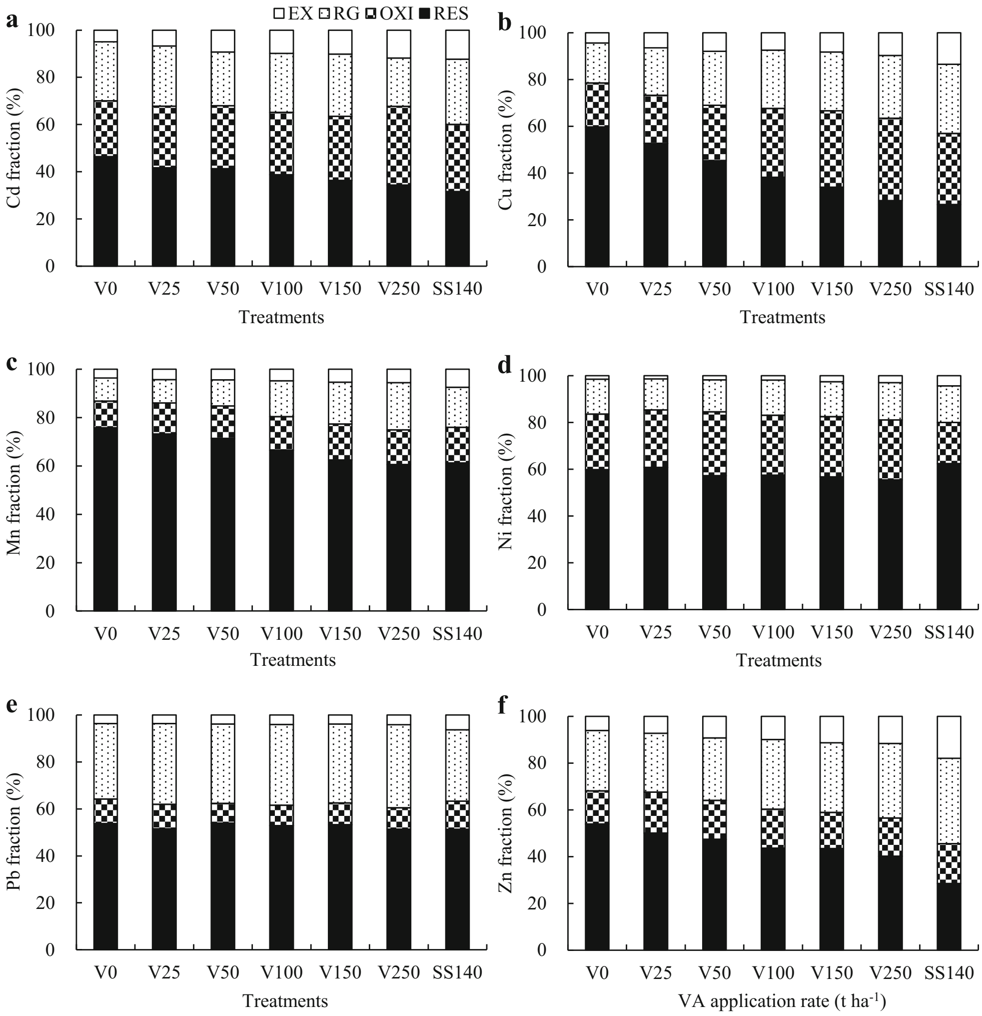

Fig. 4 Effect of vermicompost and sewage sludge amendment on geochemical fractions of total metals for Cd (**a**), Cu (**b**), Mn (**c**), Ni (**d**), Pb (**e**), and Zn (**f**) in mudflat soil. Cd, cadmium; Cu, copper; Mn, manganese; Ni, nickel; Pb, lead; Zn, zinc

metal concentrations were mainly due to much higher Cd, Cu, Ni, Pb, and Zn concentrations in vermicompost compared with those in the mudflat soil. Relatively stable total Mn concentrations in mudflat soil were probably because of relatively low Mn concentration in vermicompost. Although VA increased the concentrations of total heavy metals in mudflat, the metal levels in all treatments were below the maximum permitted concentrations as specified according to the control standards of soil environment in China (GB15618-2008). Furthermore, in this study, all vermicompost was applied once in the initial stage of mudflat amendment and was not applied afterwards. Therefore, the concentration of heavy metals in vermicompost-amended mudflat soil can be further effectively controlled with the progress of amendment. Compared with SSA under the condition of equal carbon application rate, VA caused more heavy metals accumulated in mudflat soil. An experimental study on leaching of heavy metals found that heavy metals tend to move downwards from the top 20-cm layer to the 20-40-cm depth after application of sewage sludge duo to redistribution of heavy metal fractions (Gu and Bai 2018). Therefore, heavy metals introduced by sewage sludge leached deeper into the soil layer due to leaching in mudflat soil. Meanwhile, heavy metals introduced by vermicompost accumulated more in the top 20-cm layer in mudflat soil, mainly because vermicomposting enhanced the stability of heavy metals and thus reduce the risk of heavy metal leaching.

The available heavy metal concentration in organic material-amended soil is mainly determined by soil total metal concentration and chemical properties such as pH and organic matter content (McBride 2003). Previous studies have shown that humic acid substances from biodegradation of organic compounds could decrease soil pH, which increases the solubility and availability of heavy metals (Moreno et al. 1997; Singh and Agrawal 2007; Singh and Agrawal 2009). In this study, the increasing concentrations of total heavy metals and the decreasing pH with increasing VA rates resulted in a significant increase in available Cd, Cu, Mn, Ni, and Zn concentrations and a slight increase in available Pb concentrations. The available heavy metal concentrations of Cd, Cu, Mn, Ni, Pb, and Zn in vermicompost-amended mudflat soil were lower than those in sewage sludge-amended mudflat soil. The reason may be that vermicomposting could reduce the

Table 4 Effect of vermicompost and sewage sludge amendment rate on bioaccumulation factors (BAFs) and translocation factors (TFs) for heavy metals involved in this study

Items	Treatments	Heavy metals					
		Cd	Cu	Mn	Ni	Pb	Zn
BAFs	V0	0.165*	0.165**	0.118**	0.036**	0.050**	0.346**
	V25	0.176*	0.174**	0.124**	0.044**	0.054*	0.376**
	V50	0.193^{ns}	0.182**	0.175**	0.059**	0.064*	0.385**
	V100	0.212^{ns}	0.200**	0.184**	0.069**	0.064*	0.357**
	V150	0.204^{ns}	0.214**	0.209**	0.078**	0.069^{ns}	0.392**
	V250	0.213^{ns}	0.206**	0.233**	0.096**	0.074^{ns}	0.481**
	SS140	0.239	0.315	0.378	0.190	0.081	0.650
TFs	V0	0.248**	0.536**	0.450^{ns}	0.270*	0.255^{ns}	0.667*
	V25	0.221**	0.359*	0.490*	0.234^{ns}	0.248*	0.676*
	V50	0.222**	0.341^{ns}	0.490*	0.200^{ns}	0.239*	0.671^{ns}
	V100	0.209**	0.287^{ns}	0.501*	0.185^{ns}	0.249^{ns}	0.709^{ns}
	V150	0.197**	0.224*	0.523*	0.165*	0.239*	0.706^{ns}
	V250	0.181**	0.208*	0.516*	0.150*	0.244*	0.742^{ns}
	SS140	0.398	0.292	0.370	0.196	0.319	0.808

Superscript symbols indicate significance of the differences between vermicompost treatments and sewage sludge treatment (SS140)

*$p < 0.05$, **$p < 0.01$; *ns*, not significant

concentration of diethylenetriaminepentaacetic acid (DTPA)–extractable heavy metal and thus improved the stability of heavy metal (Suthar 2009). Compared with available heavy metal concentration, the fraction distribution of heavy metal could reflect the bioavailability of metal more effectively (Kidd et al. 2007). In this study, VA increased the labile fraction of heavy metals in mudflat soil probably because decreasing soil pH enhanced metal availability in mudflat soil (Singh and Agrawal 2010). Furthermore, the increase of DOC caused by vermicompost application may also enhance metal solubility and increase the mobile fraction of metals (Bai et al. 2018). In contrast, VA decreased the nonlabile fraction of heavy metals in mudflat soil. The labile fractions of Cd, Cu, Mn, Ni, Pb, and Zn in vermicompost-amended mudflat soil were lower than those in sewage sludge-amended mudflat soil under the condition of equal carbon application rate. Existing research has found that vermicomposting could promote the degradation of sewage sludge and formation of humus (Gupta and Garg 2008), which was conducive to the complexation reaction of heavy metals and fulvic acid and humic acid, forming more stable compounds, thereby increasing the stability of heavy metals (Hait and Tare 2012; Kang et al. 2011).

Application of vermicompost increased the uptake of heavy metals by maize in mudflat soil. The results were well supported by the increase of BAFs with the amount of vermicompost applied. The BAFs could reflect the transferability of metals in plant-soil systems. In this study, VA increased the BAFs values for Cd, Cu, Mn, Ni, Pb, and Zn, which indicated that application of vermicompost enhanced the transferability of heavy metals from mudflat soil to maize. In addition, VA significantly affected heavy metal concentrations in different maize parts. The Cd, Cu, Ni, and Pb more likely accumulate in root than in stem, leaf, and grain of maize, which indicated that root system may have barrier function inhibiting the migration of Cd, Cu, Ni, and Pb from root to aerial plant parts. The results were well supported by the TF values < 1 for Cd, Cu, Ni, and Pb as TF value < 1 indicated less transportation of heavy metals from root to aerial plant parts, and more heavy metals accumulated in maize root. Gondek and Filipek-Mazur (2018) also found that applying vermicompost led to heavy metal accumulation in maize root, and the heavy metal (Cd, Cu, Ni, and Pb) concentrations in maize root were significantly higher than those in maize aerial parts. Cd accumulation in the root probably involved the formation of Cd bonding with sulfhydryl and phytochelatins (Kabata-Pendias and Pendias 1993). Cu and Pb accumulation were probably due to the formation of sparingly soluble forms in root system (Gondek and Filipek-Mazur 2018). Although the TF values were < 1 for Mn and Zn, the Mn concentrations in leaf and the Zn concentrations in the stem were higher than that in the root. The results suggested that Mn was readily transported from root to leaf, and Zn was readily transported from root to stem in maize plant, which was consistent with previous study (Carbonell et al. 2011), who found that Zn in maize has strong mobility from root to different plant parts in a greenhouse experiment. The mean values of TFs (Mn > Zn > Cu > Pb > Cd > Ni) also supported these results. In addition, TFs of Cd, Cu, Ni, and Pb in vermicompost-amended mudflat soil were lower than those in the control soil probably due to heavy metals exclusion effect which prevented the transfer of studied heavy metals from root to aerial parts of maize (Baker 2008). The stronger mobility of Zn and Mn led to the increasing TFs of Zn and Mn with increasing VA rates. Although application of vermicompost increased the concentration of heavy metals in maize, the metal levels in maize grains were below the maximum permitted concentrations as specified in the national control standard in China (GB2715-2016). Compared with SSA under the condition of equal carbon application rate, VA decreased the concentration of heavy metals in maize probably due to the relatively low heavy metal bioavailability in vermicompost-amended mudflat soil.

In our previous research, we only studied the effects of the direct sewage sludge application on soil physicochemical properties and distribution of heavy metals in maize and mudflat soil (Bai et al. 2016). To investigate the safety of mudflat soil amendment with vermicompost and evaluate the safety of heavy metals in mudflat soil and maize compared with SSA, we defined two formulas AR and UR to compare the difference of accumulation of heavy metals in soil and uptake of heavy metals by maize between VA and SSA in this study. The AR could reflect the difference of heavy metal accumulation in mudflat soil amended by vermicompost and sewage sludge. The average AR values for Cd, Cu, Mn, Ni, Pb, and Zn were > 1 (Table 5), which indicated that application of vermicompost was more likely to lead to heavy metals accumulation in mudflat soil compared with application of sewage sludge. Chlopecka (1996) considered that the exchangeable/acid-soluble form was the strongest bioavailable and the most easily migratory form in various heavy metal forms. Liu et al. (2005) found the exchangeable form and the bioavailability of heavy metals decreased over the period of vermicomposting, probably due to the formation of stable metal-humus complexes during vermicomposting progress (Kang et al. 2011). Therefore, in this study, the stronger accumulation ability of heavy metals in vermicompost-amended mudflat soil was mainly due to more stable heavy metal binding form in vermicompost compared with sewage sludge. More stable heavy metal binding form in vermicompost could reduce the risk of heavy metal leaching after vermicompost application in mudflat soil. The UR could reflect the difference of heavy metal uptake by maize grown in vermicompost-amended mudflat soil and sewage sludge-amended mudflat soil. Although VA and SSA led to heavy metal accumulation in maize plant, the average UR values for Cd, Cu, Mn, Ni, Pb, and Zn were all < 1, which indicated that VA decreased the heavy metal uptake by maize compare with SSA. The results were in agreement with the report performed by Gondek and Filipek-Mazur (2018), who found that converting sludge into vermicompost could reduce heavy metals accumulation in winter rape, sunflower, and oat compared with the direct application of sludge. Liu et al. (2005) also found that the application of sewage sludge treated with earthworms for 60 days reduced the accumulation of Cd and Cu in cabbage plants. The reason was probably that relatively low heavy metal bioavailability in vermicompost led to lower heavy metal accumulation in the maize plant compared with sewage sludge.

Table 5 Effect of vermicompost amendment and sewage sludge amendment on accumulation of heavy metals in mudflat soil and uptake of heavy metals by maize

Items	Heavy metals					
	Cd	Cu	Mn	Ni	Pb	Zn
AR	1.137	1.098	1.074	1.069	1.152	1.216
UR	0.572	0.623	0.738	0.412	0.813	0.713

Conclusions

In this study, VA improved mudflat soil physicochemical properties by decreasing soil bulk density, salinity, and pH, increasing soil organic carbon, nitrogen, and phosphorus contents. Consequently, the maize biomass and yield were significantly elevated by VA. VA increased total Cd, Cu, Ni, Pb, and Zn concentrations, while total Mn showed no significant change with VA rates in mudflat soil. Available Cd, Cu, Mn, Ni, and Zn concentrations significantly increased with increasing VA rates. VA resulted in an increase in the labile fraction (EX) of heavy metals and a reduction in the nonlabile fraction (RES) of heavy metals in mudflat soil. VA increased the accumulation of Cd, Cu, Ni, Mn, Pb, and Zn in maize tissues, especially in the root of maize. Compared with SSA under the condition of equal carbon application rate, heavy metals introduced by vermicompost exist in a more stable binding form in the top 20-cm layer in mudflat soil, which could decline their bioavailability to the plants and reduce the risk of runoff and leaching in mudflat soil. As a result, VA decreased the accumulation of heavy metals in maize plant compared with SSA in mudflat soil. Therefore, vermicompost can be an effective substitute for sewage sludge for the rapid improvement of the mudflat soil while reducing the risk of heavy metal contamination.

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