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Full Length Research Paper

Trace metals distribution and uptake in soil and rice grown on a 3-year vermicompost amended soil

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This study was designed to investigate the influence of vermicompost (VC) on trace metals distribution and uptake in soil and rice plant in research field as split plot arrangement based on randomized complete block design with three replications in 2008. Main-plot was VC and chemical fertilizer (CF) that were added to soil in 6 levels (20 and 40 ton/ha VC, 20 and 40 ton/ha VC + 1/2 CF, CF and control). Application years considered as sub-plot comprised 1, 2 and 3 years. The results indicated that fertilizers and application periods treatments influenced micronutrients in soil and rice. Available copper (Cu) had no significant difference under different treatments. The highest available iron (Fe) was found in the 40 ton treatment group. During the 3 years, application of 20 ton and enriched 40 ton gave the most available zinc (Zn) and manganese (Mn). In VC and enriched VC, treatments happened to give the highest Zn uptake by rice. Under the 3 years, application of 40 ton/ha VC, the highest Fe (91.19 ppm) and Cu (13.66 ppm) concentration was seen in flag leaf, while Fe (31.35 ppm) and Mn (27.56 ppm) was seen in grain. With the application of enriched 20 ton VC, the maximum uptake of Mn by flag leaf and Cu by grain was obtained.

Key words: Rice, soil, trace metals, vermicompost.

INTRODUCTION

Trace elements or micronutrients are important because of their association with environmental issues and the health of plants, animals and humans (Swaine, 2000). The availability of these elements in the soil can strongly affect the production and quality of crops such as rice. Different biowastes treatments (municipal solid waste, sewage sludge, manures, etc.) of a long-term field experiment can change soil micro and macronutrients and their available concentrations, which in turn affects soil micronutrient levels (Kaushik et al., 1993; Bole and Bell, 1978; Veeken et al., 2000). Land application of organic fertilizers as vermicompost is both environmentally and economically advisable. It provides organic matter (OM) to soil (Atiyeh et al., 2002) and this addition may represent a good alternative in preventing degradation of soils (Roldan et al., 1996) and improving many physical properties of agricultural soils such as water holding capacity, aeration, porosity and cation exchange capacity (Engelhart et al., 2000). Moreover, the application of this residue offers the possibility of recycling plant nutrients with the beneficial effects on soil fertility and plant nutrition (Cogger et al., 2001, Warman and Termeer, 2005; Stabnikova et al., 2005).

Nutrients in vermicompost are present in readily available forms for plant uptake; for example, nitrates, exchangeable P, K, Ca and Mg (Edwards and Burrows, 1988). Khoshgoftarmanesh and Kalbasi (2002) found that municipal waste leachate markedly increased the amounts of available macro- (N, P, K) and micronutrients (Fe, Mn, Zn, and Cu) in soil and rice plant.

Sainz et al. (1998) reported that addition of vermincompost to soil resulted in increased mineral contents in the substrate and higher concentrations of P, Ca, Mg, Cu

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Abbreviations: VC, Vermicompost; **CF,** chemical fertilizer; **OM,** organic matter; **DTPA,** diethylene triamine pentaacetic acid.

Table 1. Some physicochemical properties of soil and applied organic amendments.

Parameter	Soil	Vermicompost	Unit
Soil texture	Silty-clay	-	-
Sand	7.3	-	%
Silt	44.7	-	"
Clay	48	-	"
OC	1.6	9.63	"
pH	7.63	8.05	-
EC	1.84	2.06	dS/m
Total Fe	1640	5465.35	ppm
Available Fe	3.67	55.56	"
Total Mn	311	351.60	"
Available Mn	1.01	26.66	"
Total Cu	23.07	12.92	"
Available Cu	3.56	3.26	"
Total Zn	68.72	51.71	"
Available Zn	1.23	9.22	"

Mn and Zn in shoot tissues of red clover and cucumber. Chamani et al. (2008) showed that the Zn and Fe concentrations in plant tissues tends to increase with increasing vermicompost in the base media compared to the control. Furthermore, addition of either vermicompost or peat to base media significantly decreased Mn concentrations in Petunia shoot tissues. Since micro-nutrients, Cu, Zn, Fe and Mn, in grain directly affect food quality and are closely related to human and livestock nutrition, deficiency of micronutrients will result in anaemia, decreased immunity, slow growth and nyctalo-pia. However, little attention has been paid to micro-nutrients response to vermicompost and how perennial application of theses fertilizers impacts on the uptake of micronutrients from soil and their transfer from tissues to grains are not well documented. The objectives of this paper are to study the changes of micronutrients accu-mulation in soil, and to investigate the transfer amounts of micronutrients from soil to rice shoot (flag leaf and grain) in response to 3 years application of vermin-compost.

MATERIALS AND METHODS

This experiment was performed in 2008 cropping season in the research field of Sari Agricultural Sciences and Natural Resources University. This area is located in 34° 33´ N; 52°6´ E and 16 m altitude. The experiment was conducted as split plot arrangement based on randomized complete block design with three replications. Main-plots which were fertilizer treatments with 6 levels comprise, 20 and 40 ton ha^{-1} vermicompost (VC) (F1 and F2), 20 and 40 ton ha^{-1} VC + 1/2 chemical fertilizer (CF) (F3 and F4), CF (F5 that based on soil testing comprise 100 kg.ha^{-1} urea, 150 kg.ha^{-1} triple super phosphate and 100 kg.ha^{-1} potassium sulphate) and control (without any fertilizers or F6). Sub-plots were application periods of fertilizers (P1 = 2006, P2 = 2006 + 2007 and P3 = 2006 + 2007 + 2008). This investigation started in 18 plots with size of 3 × 12 m in 2006. In 2007, in 2/3 of the initial plots, (3 × 8 m) and in 2008 in 1/3 of the initial plots, (3 × 4 m), fertilizer treatments were added to soil (sum 54 plots). Prior to planting (in 2006), soil samples were taken for the determination of some of the physical and chemical properties of soil. In addition, some samples were taken from VC in order to determine their chemical properties (Table 1).

Agricultural practices started in late April with plowing of the land and preparing the rice nursery (Tarom cultivar). This was followed by application of respective fertilizers with certain amounts in plots and seedlings transplanted with 25 × 25 cm in May 10. After rice harvesting, soil samples were taken from paddy soil (0 - 30 cm) for the determination of total and available micronutrients (Fe, Mn, Cu and Zn). Total forms of these metals in soil were measured using Varian Spectra A A-10 Atomic Absorption Spectrometer (Baker and Amacher, 1982), and available form was measured using Lindsay and Norvell (1978) method. Two different samples of rice grain and flag leaf were taken at harvest and flowering stages, respectively, and micronutrients were determined according to AOAC (1990). Statistical analysis of data was done using MSTATC and statistical package for the social sciences (SPSS) software and means were compared using Duncan's Multiple Range Tests (DMRT) at 0.05 probability levels.

RESULTS AND DISCUSSION

Trace elements in soil

Fertilizers and application periods treatments influenced accumulation of studied micronutrients in soil. Interaction effects of fertilizers × application periods were significant on available Mn and Zn.

Table 2 shows total and diethylene triamine pentaacetic acid (DTPA) extractable micronutrients concentration in soil under different fertilizers and application periods treatments. Application of 20 ton.ha^{-1} VC accumulated the most total Cu (27.97 ppm) which showed significant difference with the control. Although, this treatment showed the highest available Cu (4.11 ppm), there was

Table 2. Means comparison accumulation of trace elements in soil.

Treatments	T- Cu	A- Cu	T- Fe	A- Fe	T-Mn	T- Zn
Fertilizer						
20 ton.ha^{-1} VC	27.97^{a}	4.11^{a}	1663.11^{b}	5.11^{a}	568.42^{b}	92.39^{a}
40 ton.ha^{-1} VC	26.76^{b}	3.89^{a}	1719.72^{b}	5.35^{a}	587.13^{a}	88.06^{a}
20 ton.ha^{-1} VC +1/2 CF	27.06^{ab}	4.10^{a}	1799.72^{ab}	5.06^{a}	573.54^{b}	91.79^{a}
40 ton.ha^{-1} VC +1/2 CF	27.06^{ab}	4.00^{a}	1930.85^{a}	5.25^{a}	574.68^{b}	88.60^{a}
Chemical fertilizer	26.40^{b}	4.04^{a}	1710.09^{b}	4.54^{b}	572.21^{b}	88.36^{a}
Control	25.08^{c}	3.84^{a}	1651.74^{b}	3.88^{c}	538.95^{c}	86.96^{a}
Application periods						
2006	25.77^{c}	3.66^{c}	1626.48^{b}	4.05^{c}	554.26^{c}	84.51^{a}
2006 + 2007	26.73^{b}	4.01^{b}	1699.56^{b}	4.83^{b}	570.71^{b}	87.43^{a}
2006 + 2007 + 2008	27.66^{a}	4.33^{a}	1911.57^{a}	5.72^{a}	582.50^{a}	87.45^{a}

T = Total; A = available; Means within the same column and each treatment followed by the same letter not significantly difference according to DMRT ($P \leq 0.05$); all values are given in ppm.

Table 3. Interaction effects of fertilizer × application periods on available Mn and Zn in soil (ppm).

Fertilizer application	Available Mn			Available Zn		
	2006	2006 + 2007	2006 + 2007+2008	2006	2006 + 2007	2006 + 2007 + 2008
20 ton.ha^{-1} VC	1.01^{fgh}	1.05^{e-h}	1.53^{abc}	1.10^{def}	1.51^{cde}	2.61^{a}
40 ton.ha^{-1} VC	0.96^{fgh}	1.26^{c-f}	1.67^{ab}	0.76^{f}	1.10^{def}	1.53^{cd}
20 ton.ha^{-1} VC +1/2 CF	1.09^{d-g}	1.35^{cde}	1.52^{abc}	1.01^{def}	1.79^{bc}	2.20^{ab}
40 ton.ha^{-1} VC +1/2 CF	0.76^{hi}	1.27^{c-f}	1.81^{a}	0.91^{ef}	1.09^{def}	1.59^{cd}
Chemical fertilizer	0.89^{gh}	1.33^{cde}	1.40^{bcd}	1.04^{def}	1.26^{c-f}	1.18^{def}
Control	0.55^{i}	0.73^{hi}	0.75^{hi}	0.75^{f}	0.79^{f}	0.81^{f}

Means within the same column and each treatment followed by the same letter was not significantly different according to DMRT ($P \leq 0.05$).

no significant difference with the other treatments. Also, total Fe under enriched 40 ton.ha^{-1} VC was maximum (1930.85 ppm) but had no significant difference with other treatments. The highest available Fe (5.35 ppm) was shown in 40 ton.ha^{-1} VC treatment that is significantly different with chemical and control treatment (Table 2). The maximum amount of total Mn (587.13 ppm) was accumulated in 40 ton.ha^{-1} VC and about 9% enhanced compared to the control (Table 2). The measured total Zn under 20 ton.ha^{-1} treatment was highest (92.39 ppm) but had no significant difference with other treatments. Sainz et al. (1998), Salim Akhter (1990) and Jordao et al. (2005) in their researches obtained similar results.

With increasing application periods from one to three years, accumulation of studied metals in soil were regularly enhanced (Table 2). For instance, total Fe after two and three years application was enhanced with about 4.49 and 12.4%, respectively. Available Fe concentration in two and three years fertilization compared to the first year was enhanced with about 19.2 and 41%, respectively. Total Cu concentration during the 3 years fertilization was enhanced by 3.7 and 3.14%, respectively. Available Cu with increase fertilization periods showed about 9.5 and 7.9% enhancement, respectively (Table 2). Al- Najar et al. (2005) in a similar project stated that Zn and Cu concentration of soil were enhanced in response to the application of sewage sludge.

The amount of total Mn in the third year fertilization compared to the second year enhanced 2.06% while about 3.5% was more than the first year fertilization (Table 2). Although, the amount of total Zn increased with enhancement of application periods, there was no significant difference (Table 2). Mc Grath and Cegara (1992) stated that Cu and Zn accumulation in soil significantly increase in response to 10 years application of sewage sludge. In another study, Gigliotti et al. (1996) in a 6-year field study demonstrated that compared to untreated soils, amended soils with urban waste compost showed a significant increase in Cu and Zn concentrations in the last 2 years.

In our experiment, the maximum concentration of available Mn in soil (1.81 ppm) belong to enriched 40 ton.ha^{-1} treatment when added to soil for three continuous years that was about 87% more than non-fertilized treatment (Table 3). Under the three years, application of enriched 20 ton accumulated the highest available Zn (2.61 ppm) that was about 3.22 times of measured content in the control (Table 3).

Table 4. Means comparison uptake of Zn by rice plant (ppm).

Treatments	Flag leaf	Grain
Fertilizer		
20 ton.ha^{-1} VC	15.45^{a}	24.96^{ab}
40 ton.ha^{-1} VC	13.86^{a}	25.50^{a}
20 ton.ha^{-1} VC +1/2 CF	15.45^{a}	25.81^{a}
40 ton.ha^{-1} VC +1/2 CF	13.92^{a}	25.13^{ab}
chemical fertilizer	11.66^{b}	23.37^{bc}
control	11.94^{b}	22.98^{c}
Application periods		
2006	12.56^{b}	23.74^{b}
2006 + 2007	14.12^{a}	25.49^{a}
2006 + 2007 + 2008	14.46^{a}	24.65^{ab}

Means within the same column and each treatment followed by the same letter was not significantly different according to DMRT ($P \leq 0.05$).

Trace elements in rice plant

Fertilizers and application periods treatments significantly influenced uptake of trace elements by rice plant. Interaction effects of fertilizers × application periods were significant on Fe, Mn and Cu uptake by rice tissues. However, Zn uptake was similar in all treatments.

With the application of 20 ton and enriched 20 ton VC per hectare, maximum uptake of Zn accumulated in flag leaf (15.45 ppm) that only showed significant difference with chemical fertilizer and control treatment (Table 4). In addition, the highest accumulation of Zn in grain (25.81 ppm) belong to enriched 20 ton.ha^{-1} VC treatment that was about 12.3% more than the control (Table 4). These results are consistent with those of Chamani et al. (2008).

With increase application periods, Zn uptake by flag leaf was also enhanced. The second year and third year showed approximate enhancement of 12.4 and 2.4%, respectively (Table 4). Zn accumulation in grain in the second year showed about 7.3% enhancement compared to the first year but gave a decreased of about 3.4% in the third year (Table 4).

In terms of Fe in flag leaf, the highest uptake (91.19 ppm) belongs to 40 ton.ha^{-1} VC treatment when applied for 3 continuous years (Table 5). Under three years application of 40 ton VC ha^{-1} and enriched 20 ton (F3) maximum accumulation of Fe in grain (31.35 ppm) recorded was about 48.7% more than the control. The lowest Fe uptake by flag leaf and grain happened to be in the control (Table 5).

With the application of enriched 20 ton treatment during the 3 continuous years, it was shown that the highest Mn concentration was in flag leaf (257.3 ppm) with about 60% enhancement compared to control. Mn uptake by grain in the three years application of 40 ton.ha^{-1} VC was maximum (27.56 ppm), although, it was only significantly different with the control (Table 5).

The highest concentration of Cu in flag leaf (13.66 ppm) obtained in the 3 continuous years application of 40 ton VC ha^{-1} was about 2.68 times of the content under non fertilized plot (Table 5). Under 3 continuous years' application of enriched 20 ton VC ha^{-1}, (F3) caused the most Cu uptake by grain (10.38 ppm) with about 174% enhancement compared to control treatment which had significant difference when compared to other treatments (Table 5). Khoshgoftarmanesh and Kalbasi (2002) obtained a similar result.

In general, vermicompost treatments with increase application periods from one to three years enhanced the uptake of studied trace elements by rice plant; this has also been shown by Tejada and Gonzalez (2006). In some cases, uptake contents do not increased with enhancement of application periods, regularly, that comprise: Fe uptake by flag leaf under enriched 20 ton VC ha^{-1} (F3), Fe uptake by grain in 20 ton and enriched 40 ton VC ha^{-1} (F4). These results may be correlated to soil properties such as acidity and organic carbon (Gao et al., 2003; Kwiatkowska and Maciejewska, 2006). As a result of the change of basic soil characteristics, such as pH, organic matter and nutrients in response to long-term fertilization field experiments, status and behaviors of micronutrients in soil and crop vary with different fertilization practices (Wei et al., 2006). Soil organic matter exerts a significant and direct impact on the availability of Zn, Fe and Mn but has little influence on the availability of soil Cu (Zhang et al., 2001). In addition, the interaction of other soil macronutrients and micronutrients also affected micro-nutrients uptake by crops (Aulakh and Malhi, 2005).

The results of simple correlation between studied traits showed that there is significant and positive correlation among total Fe with available Fe ($r = +0.33^{*}$), Mn ($r = +0.41^{**}$), Cu ($r = +0.42^{**}$) and Zn ($r = +0.33^{*}$). Also, significant and positive correlation among Fe uptake by grain ($r = +0.36^{**}$), Mn concentration in flag leaf ($r = +0.35^{**}$) and grain ($r = +0.37^{**}$) and Cu uptake by grain ($r = +0.53^{**}$) was recorded. Moreover, negative correlation existed among total Zn with available Zn and Mn, total Cu, Mn, Fe and uptake amounts of studied trace elements by rice plant. The maximum significant and positive correlation belong to available Mn and available Fe ($r = +0.86^{**}$). Although, total Zn showed negative correlation with available Zn and also uptake content of Fe, Mn, Cu and Zn by rice, it was not statistically significant (data not shown). In general, among the available form and uptake contents of mentioned elements, a positive correlation was obtained (Table 6).

Conclusion

Application of vermicompost as organic fertilizer in rice planting system had significant effects on micronutrients accumulation in soil and plant. Also, with increase application periods, enhancement in concentration of micronutrients in soil and rice was very significant. Based on our findings, the accumulation of Fe in soil was

Table 5. Interaction effects of fertilizers × application periods on trace elements uptake by rice (ppm).

Parameters	Treatments	F1	F2	F3	F4	F5	F6
Fe- flag leaf	P1	68.27^{cde}	47.38^{g}	76.93^{bc}	66.23^{c-f}	62.66^{def}	71.83^{cd}
	P2	71.83^{cd}	54.51^{fg}	72.85^{cd}	71.83^{cd}	58.08^{efg}	67.76^{cde}
	P3	86.61^{ab}	91.19^{a}	70.30^{cde}	77.44^{bc}	68.77^{cde}	57.57^{efg}
Fe- grain	P1	28.10^{bcd}	26.48^{de}	24.32^{ef}	27.56^{bcd}	24.32^{ef}	21.08^{g}
	P2	27.02^{cd}	30.27^{ab}	28.64^{a-d}	30.27^{ab}	23.24^{fg}	21.08^{g}
	P3	29.18^{a-d}	31.35^{a}	31.35^{a}	29.73^{abc}	23.78^{ef}	21.08^{g}
Mn- flag leaf	P1	184.9^{e}	173.7^{ef}	227.7^{bc}	184.9^{e}	175.8^{ef}	146.2^{g}
	P2	203.8^{d}	226.2^{bc}	231.8^{bc}	212.5^{cd}	222.6^{cd}	164.1^{fg}
	P3	221.6^{cd}	226.2^{bc}	257.3^{a}	243.5^{ab}	225.2^{bc}	160.5^{fg}
Mn- grain	P1	20.54^{gh}	22.16^{d-g}	21.62^{e-h}	23.24^{cde}	20.54^{gh}	20.00^{h}
	P2	21.08^{fgh}	23.78^{cd}	22.70^{c-e}	24.32^{bc}	23.78^{cd}	19.87^{h}
	P3	25.94^{ab}	27.56^{a}	27.02^{a}	26.48^{a}	25.94^{ab}	21.08^{fgh}
Cu-flag leaf	P1	5.73^{fgh}	5.60^{gh}	6.62^{ef}	5.88^{fgh}	5.03^{h}	5.40^{gh}
	P2	10.19^{c}	7.13^{de}	7.13^{de}	6.10^{fg}	5.40^{gh}	5.03^{h}
	P3	11.20^{b}	13.66^{a}	10.29^{c}	7.13^{de}	7.64^{d}	5.09^{h}
Cu-grain	P1	3.78^{g}	3.78^{g}	5.04^{ef}	5.96^{cde}	2.70^{h}	3.24^{gh}
	P2	5.40^{e}	4.05^{fg}	5.76^{cde}	6.78^{c}	3.78^{g}	2.70^{h}
	P3	6.48^{cd}	5.47^{de}	10.38^{a}	7.92^{b}	4.32^{fg}	3.78^{g}

= 20 ton.ha^{-1} VC; F2 = 40 ton.ha^{-1} VC; F3 = 20 ton.ha^{-1} VC + 1/2 CF; F4 = 40 ton.ha^{-1} VC + 1/2 CF; F5 = CF and F6 = control (or without any fertilizer); P1 = 2006; P2 = 2006 + 2007; P3 = 2006 + 2007 + 2008; Means within the same column and each treatment followed by the same letter was not significantly different according to DMRT (P ≤ 0.05).

Table 6. Simple correlation among available form and uptake contents of studied elements.

Elements	Fe		Zn		Mn		Cu	
	Flag leaf	grain	Flag leaf	grain	Flag leaf	grain	Flag leaf	grain
Available Fe	+0.43**	+0.57**	+0.35**	+0.31**	+0.48**	+0.49**	+0.61**	+0.42**
Available Zn	+0.42**	+0.51**	+0.51**	+0.37**	+0.45**	+0.34*	+0.62**	+0.63**
Available Mn	+0.25ns	+0.58**	+0.30*	+0.31*	+0.50**	+0.57**	+0.57**	+0.48**
Available Cu	+0.40**	+0.31**	+0.32*	+0.11ns	+0.30*	+0.30*	+0.52**	+0.44**

* and ** are significant in 5 and 1% levels, respectively, and ns is not significant (based on Duncan test).

maximum and Cu concentration was lowest. Also, the highest uptake by rice under different treatments belongs to Mn by flag leaf. In addition, the highest amounts of Fe (31.35 ppm) accumulated in rice grain under 3 years application of 40 ton and enriched 20 ton VC per hectare. Attention to the beneficial and effective effects of vermicompost is recommended for long time investigations.

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