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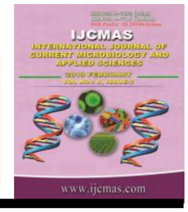

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Long Term Effect of Manure and Fertilizers on Chemical Fractions of Fe and Mn in Surface Soils under Rice-Wheat System

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The present research study has been conducted with prime objective to investigate the effect of manure and fertilizers on chemical fractions of Fe and Mn under rice-wheat system. Laboratory analysis was made on the soil samples collected (October 2013) from an on-going long-term field experiment (in progress since *Kharif*2009-10) at Department of Soil Science, PAU, Ludhiana. The organic manure through bio gas slurry (BGS) @ 6 t ha^{-1} was incorporated along with nitrogen (N @ 80 and 120 kg ha^{-1}), phosphorus (P @ 30 kg ha^{-1}) and potassium fertilizer (K @ 30 kg ha^{-1}) to the rice crop. On the other hand in the wheat crop, nitrogen (N @ 120 kg ha^{-1}), phosphorus (P @ 0, 30 and 60 kg ha^{-1}) and potassium fertilizer (K @ 30 kg ha^{-1}) were applied without addition of organic manure. It was observed that the concentration of micronutrients was found higher in the fractions where organic manure was applied along with chemical fertilizers. It was found that the residual micronutrient fraction is the dominant portion of total Fe and Mn fraction. The WSEX fraction contributed limited in amount as compared to the other fractions. Among chemical fractions viz. WSEX, SpAd, MnOX, AFeOX, CFeOX, OM-bound associated with Zn, Cu, Fe and Mn showed their edge with combined application of manure and chemical fertilizers. However, WSEX, SpAd, CFeOX and OM-bound fractions contributed towards uptake of micronutrients by wheat and rice grains.

Introduction

Rice-wheat cropping system is most vital cropping system of Indian subcontinent. Rice (*Oryza sativa* L.) and wheat (*Triticum aestivum* L.) are the two most important energy giving food globally (Singh *et al*., 2011; Meena *et al*., 2013). Rice and wheat grown sequentially in an annual rotation (Singh and Singh, 2009) constitute a rice-wheat cropping system (RWCS) and in a system occupy nearly 13.5 million hectares area in the Indo-Gangetic Plains (IGP) of South Asia. Integrated nutrient management practices for rice-wheat cropping system are of supreme importance for sustainable crop production in country (Singh and Kumar, 2009).The study of various fractions of Fe and Mn present in soil and conditions under which they become available to plants is pre-requisite in assessing their availability to plants. It is important to know the relationship between chemical fractions of micronutrients in the soil and their uptake by the crop. Under continuous cropping system, micronutrients are generally considered to be present in association with soil solution, organic and inorganic solid phases and this association is often referred to as speciation (Behera *et al.*, 2009), thus, forming their various chemical fractions such as water soluble plus exchangeable, specifically absorbed and those associated with free calcium carbonate, oxide surfaces, soil organic matter and minerals.

The alternate flooding (reduced stage) in rice and upland (oxidized stage) conditions in wheat affects transformation of Zn and Cu from one chemical form to another (Manchanda *et al.*, 2003). Dhaliwal (2008) reported that green manure and soil applied Mn to rice–wheat system increased the DTPA-extractable, water soluble plus exchangeable and Mn specifically adsorbed on the inorganic sites whereas, Mn held on organic sites and oxide bound surfaces decreased. Duhan and Singh (2002) reported that the use of organic manures increased uptake of micronutrients which may be attributed to increase in DTPA-extractable Zn and Fe in soil and to increased yield by these organic materials. Sekhon *et al.*, (2006) reported that application of organic manures resulted in increase and redistribution of Zn from non-available forms to readily available (water-soluble plus exchangeable) and potentially available forms in soil.

Hellal (2007) reported that addition of composted mixtures increased MnOX-Zn in soil as a result Fe availability is increased in calcareous soil by high acidulation effect of compost. Herencia *et al.*, (2008) showed that percentage of Zn in the specific fractions with respect to total content are Zn and addition of OM caused Zn to move from less soluble forms to more plant available fraction which was always favoured by organic amendment. Sekhon *et al.*, (2006) reported that addition of GM to rice increased AFeOX form of Zn under rice-wheat rotation. Hellal (2007) reported that addition of composted mixtures increased amorphous Fe oxide but occluded fractions did not differ significantly due to application of composted mixtures. Consequently, the present research study was conducted with a prime objective to investigate the effect of manure and fertilizers on transformations (distribution) of micronutrients (Fe and Mn) in soil.

Materials and Methods

In order to achieve the objectives mentioned earlier, laboratory studies were made on the soil samples collected from an on-going long-term experiment on role of manure and fertilizers in rice-wheat cropping system (in progress since *Kharif* 2009-10) at Department of Soil Science, Punjab Agricultural University, Ludhiana. The soil of experiment field was classified as *Typic Ustochrept.* The experiment was laid out in a split plot design with four main and three sub treatments. The organic manure through bio gas slurry (BGS) @ 6 t ha^{-1} was incorporated along with nitrogen fertilizer (N @ 80 and 120 kg ha^{-1}), phosphorus fertilizer (P @ 30 kg ha^{-1}) and potassium fertilizer (K @ 30 kg ha^{-1}) were applied to the rice crop. Whereas in wheat crop, nitrogen fertilizer (N @ 120 kg ha^{-1}), different levels of phosphorus fertilizer (P @ 30 and 60 kg ha^{-1}) and potassium fertilizer (K @ 30 kg ha^{-1}) were applied (Table 1).

Laboratory analysis

The soil samples were used to fractionate into following chemical forms as per sequential extraction procedure described below:

Water soluble plus exchangeable fraction (WSEX)

Five grams of soil was shaken with 20 ml of 0.005 M Pb $(NO_3)_2$ in 100 ml centrifuge tubes for fifteen minutes at 25˚C in Orbital shaker and mixture was centrifuged for ten minutes at 6000 rpm the supernatant filtered, separated and stored for analysis (Manchanda *et al*., 2006).

The Reagent 0.005 M $Pb(NO_3)_2$ is prepared by dissolving 1.65gm of lead nitrate in one litre adjusting the pH of solution to 6.8 by 0.5M ammonium acetate (NH_4OAC) which is prepared by dissolving 38.5 gm of ammonium acetate in 1 litre.

Specifically adsorbed (SpAd) fraction

The soil residue from water soluble plus exchangeable fraction was shaken with 20 ml of 0.05M $Pb(NO_3)_2$ for 2 hours at 25˚C in orbital shaker and; the sample was, thereafter, centrifuged ten minutes at 6000 rpm and the supernatant filtered (Iwaski *et al*., 1993).

The sequential extraction continued in the remaining of the soil sample The Reagent 0.05 M $Pb(NO_3)_2$ is prepared by dissolving 16.56gm lead nitrate in one litre adjusting the pH of solution to 6.0 by 0.5M ammonium acetate (NH_4OAC)

Mn-Oxide bound fraction (MnOX)

To the remaining soil sample 20.0 ml of $NH_2OH.HCl$ (hydroxylamine hydrochloride) 0.1 mol l^{-1} at pH 2.0 were added and the mixture was shaken for 30 min, centrifuged and filtered; the separated supernatant was stored for analysis (Chao, 1972). The Reagent 0.1 M $NH_2OH.HCl$ in 0.01M HNO_3 is prepared by dissolving 6.95 gm of $NH_2OH.HCl$ and 0.625 Nitric acid ($HNO_{3)}$ in water and make the volume to one litre.

Amorphous Fe-Oxides bound (AFeOX) fraction

To the Mn-Oxide Bound Fraction free soil sample 20.0 ml of $NH_2OH.HCl$ (hydroxylamine hydrochloride) 0.1 mol l^{-1} plus HCl 0.25 mol l^{-1}, at pH 1.3 were added, and the mixture was shaken for 30 min at 25˚C in orbital shaker, centrifuged and filtered; the separated supernatant was stored for analysis (Maskina *et al*., 1998). The reagent 0.25 M $NH_2OH.HCl$+0.25 M HCl is prepared by dissolving 17.37 gm of $NH_2OH.HCl$ in water and pour 21 ml of Hydrochloric acid (HCl) in it and make the volume of solution to one litre.

Crystalline Fe-Oxides bound (CFeOX) fraction

To the AFeOx free soil sample 20.0 ml of 0.25 M $NH_2OH.HCl$ +0.25 M HCl + ascorbic acid 0.01 mol l^{-1}, at pH 1.21 were added, the mixture was heated with boiling water (100˚C) in a beaker placed on hot plate for 30 minutes, shaking from time to time; there after centrifuged and filtered; the separated supernatant was stored for analysis (Manchanda *et al*., 2006). The sequential extraction continued in the remaining of the soil sample The Reagent 0.25 M $NH_2OH.HCl$ +0.25 M HCl +0.1 M ascorbic acid is prepared by dissolving 17.37 gm of$NH_2OH.HCl$ in water, pour 21 ml of hydrochloric acid (HCl) and 17.61gm of ascorbic acid in it and make the volume of solution to one litre.

Organically bound (OM) fraction

To the CFeOX free soil sample was shaken with 20 ml of 1% $Na_4P_2O_7$ for one hour at 25˚C in Orbital shaker and mixture was centrifuged for ten minutes at 6000 rpm the supernatant filtered, separated and stored for analysis (Raja and Iyengar, 1986). The

Reagent prepared by dissolving 4.46 gm of Sodium-pyrophosphate in one litre.

Residual (RES) fraction

Residual fraction (cation) = Total content (cation) - sum of all fractions (cation). The amount of Zn, Cu, Mn and Fe in different fractions was estimated using atomic absorption spectrophotometer.

Statistical analysis

Critical difference (CD) was used to compare the treatment effects at $P<0.05$. The statistical analysis was done with the help of method described (Panse and Sukhatme, 1985).

Results and Discussion

Effect of manure and fertilizers on chemical fractions of Fe

The data for WSEX-Fe presented in Table 2 of the surface soil samples which were collected after harvesting of rice ranged from 0.11 to 0.16 mg kg^{-1} in all the treatments. Significant increase in WSEX-Fe contents was observed in the treatments where organic manure @ 6 t ha^{-1} was incorporated along with N @ 80 kg ha^{-1} and P_2O_5 @ 30 kg ha^{-1} and in the treatments where organic manure @ 6 t ha^{-1} was applied along with N @ 80 kg ha^{-1} without phosphatic fertilizer as compared to the treatments where only N @ 120 kg ha^{-1} was applied without organic manure and P_2O_5 application to the rice crop and also in the treatments where only N @ 120 kg ha^{-1} and P_2O_5 @ 30 kg ha^{-1} were applied without addition of organic manure to the rice crop. Hellal (2007) reported from his green house experiment that the addition of organic manure increased WSEX-Fe in soil, as a result of application of organic mixture. Similarly, Maskina *et al.*, (1998) observed an increase in WSEX fraction of Fe with addition of organic manure. Long-term application of farmyard manure also increases the organic matter content in soil which also enhances Zn and Fe availability (Rehman *et al.*, 2012). Earlier studies have shown that FYM and single super phosphate contain considerable amount of Fe, which, when applied to the soil, results in higher availability of this micronutrient (Walia *et al.*, 2010), and thus, the crop uptake of this micronutrient significantly increases (Mann *et al*., 2006).

The SPAD-Fe reported significantly higher magnitude in the treatments where organic manure @ 6 t ha^{-1} was added in combination with N @ 80 kg ha^{-1} and P_2O_5 @ 30 kg ha^{-1} and also in the treatments where organic manure @ 6 t ha^{-1} was incorporated along with N @ 80 kg ha^{-1} was applied without incorporation of phosphatic fertilizer in contrast to the treatments where no organic manure was incorporated and only N @ 120 kg ha^{-1} was applied without P_2O_5 application to the rice crop. The SPAD-Fe varied from 0.25 to 0.27 mg kg^{-1} in the treatments where organic manure and inorganic fertilizers were applied in combination and it was ranged from 0.20 to 0.24 mg kg^{-1} and 0.22 to 0.24 mg kg^{-1} in the treatments where no organic manure was incorporated and only inorganic fertilizers were applied. Iu *et al.*, (1981) reported increase in amount of SPAD-Fe with addition of organic manure. These results are also in agreement with the results obtained by Chatterjee *et al.*, (1992) who reported increase in this form with addition of organic manure.

The MnOX-Fe showed significant increase in its fractions with fertilizers and manure. It was reported higher in the treatments where organic and inorganic fertilizers were applied in combination as compared to the treatments where only chemical fertilizers were applied to the rice crop. The MnOX-Fe ranged from

44.77 to 48.73 mg kg^{-1} in the treatments organic manure @ 6 t ha^{-1} was incorporated along with N @ 80 kg ha^{-1} and P_2O_5 @ 30 kg ha^{-1} were applied and 41.58 to 43.25 mg kg^{-1} in the treatments where organic manure @ 6 t ha^{-1} was incorporated along with N @ 80 kg ha^{-1} was applied without incorporation of phosphatic fertilizer. On the other hand, it varied from 36.72 to 38.35 mg kg^{-1} in the treatments where no organic manure was incorporated and only N @ 120 kg ha^{-1} was applied without P_2O_5 application to the rice crop and 39.12 to 41.03 mg kg^{-1} in the treatments where no manure was incorporated and only chemical fertilizers were applied. Sekhon *et al.,* (2006) reported that addition of organic manure to rice increased potentially available fraction of Fe under rice-wheat rotation. Hellal (2007) reported that addition of composted mixtures increased MnOX-Fe in soil, as a result Fe availability is increased in calcareous soil by high acidulation effect of compost.

The AFeOX-Fe ranged from 386.8 to 390.1 mg kg^{-1} in the treatments where organic manure @ 6 t ha^{-1} was incorporated along with N @ 80 kg ha^{-1} and P_2O_5 @ 30 kg ha^{-1} were applied. Similar pattern of increase was observed in the treatments where organic manure @ 6 t ha^{-1} was incorporated along with N @ 80 kg ha^{-1} was applied without incorporation of phosphatic fertilizer where it ranged from 383.4 to 385.9 mg kg^{-1}. Hellal (2007) reported that addition of composted mixtures increased AFeOX fraction but occluded Fe did not differ significantly due to application of composted mixtures. Agbenin (2003) reported a similar increase in AFeOX-Fe and Mn fractions fertilized with NPK, FYM and FYM+NPK. Singh *et al.,* (1988) in a study on 11 soils reported that 9 and 5 per cent of total Fe and Mn was associated with AFeOX fraction. The CFeOX-Fe fraction increased in soil many folds as compared to the other fractions (Table 3). In all the treatments CFeOX-Zn varied from 564.80 to 631.30 mg kg^{-1} where the higher content was noticed in the treatments where organic manure was incorporated along with chemical fertilizers. The significant higher concentration ranged from 616.47 to 631.30 mg kg^{-1} of this fraction was noticed in the treatments where organic manure @ 6 t ha^{-1} was incorporated along with N @ 80 kg ha^{-1} and P_2O_5 @ 30 kg ha^{-1} were applied. Singh *et al.,* (1988) and Randhawa and Singh, (1997) reported that about 52 per cent of the total soil Fe was presented in RES fraction and about 41 per cent of the total Fe was associated with CFeOX fraction. Similarly, Nayyar and Chhibba, (2000) reported that the prevalence of alternative oxidized and reduced conditions under rice-wheat system caused a decline in the content of CFeOX form concomitant with an increase in the easily reducible AFeOX form of these micronutrients leading to their increased availability.

The significant increase was noticed in organically bound fraction (OM-Fe) in the treatments where organic manure @ 6 t ha^{-1} was incorporated along with N @ 80 kg ha^{-1} and P_2O_5 @ 30 kg ha^{-1} were applied where it ranged from 24.60 to 26.28 mg kg^{-1} as compared to the treatment which were treated inorganically and where no organic manure was incorporated. However, the significant difference was also observed in OM-Fe fraction in case of wheat crop where different levels of P_2O_5 (0, 30 and 60 kg ha^{-1}) were applied. The higher concentration of OM-Fe and Mn in the soil solution indicated that the micronutrients associated with the OM bound fraction may play a beneficial role in the uptake of these nutrients by the plants. Sekhon *et al.,* (2006) reported that OM bound fraction of Fe and Mn increased with application of organic manure in rice-wheat system. It was observed that application of P fertilizer and organic manure with incorporation of straw resulted in significant increases in soil total Cu, Zn, Fe and Mn (Li *et al*., 2010).

Effect of manure and fertilizers on chemical fractions of Mn

The concentration of Mn in WSEX fraction ranged from 3.77 to 4.79 mg kg^{-1} in all the treatment combinations in Table 4. Significantly increased concentration of WSEX-Mn contents was observed in the treatments where organic manure @ 6 t ha^{-1} was incorporated along with N @ 80 kg ha^{-1} and P_2O_5 @ 30 kg ha^{-1} were applied where it ranged from 4.41 to 4.79 mg kg^{-1} and it ranged from 4.17 to 4.39 mg kg^{-1} in the treatments where organic manure @ 6 t ha^{-1} was incorporated along with N @ 80 kg ha^{-1} was applied without incorporation of phosphatic fertilizer as compared to the treatments where no organic manure was incorporated and only N @ 120 kg ha^{-1} was applied without P_2O_5 application to the rice crop where it was ranged from 3.77 to 3.81 mg kg^{-1} and in the other treatments it varied from 3.98 to 4.27 mg kg^{-1} where no manure was incorporated and only chemical fertilizers like N @ 120 kg ha^{-1} and P_2O_5 @ 30 kg ha^{-1} were applied to the rice crop. The maximum concentration of WESX-Mn was reported in organically treated plots which may be attributed to the reduction of higher valent forms of Mn (Mn^{4+}) to its available form (Mn^{2+}) accompanied by increase in its solubility under submerged conditions and chelating action of the organic manures. Earlier authors have reported that balanced fertilization not only increases grain yield and maintains soil nutrient balance, but also accelerates rice nutrient uptake (Mann *et al*., 2006; Li *et al*., 2007; Xue *et al*., 2014).

The SPAD-Mn reported significantly higher magnitude in the treatments where organic manure was incorporated in combination with chemical fertilizers in contrast to the treatments where no organic manure was incorporated and only chemical fertilizers were applied. The SPAD-Mn varied from 2.25 to 2.89 mg kg^{-1} in all the treatments. Significant higher concentrations were reported in organically treated plots. Iu *et al.*, (1981) reported increase in amount of SPAD-Mn with the addition of organic manure. These results are also in agreement with the results obtained by Chatterjee *et al.*, (1992) who reported increase in this form with addition of organic manure. Dhaliwal (2008) reported that rice-wheat cropping system increased the levels of Mn in WSEX and SPAD on the inorganic sites, whereas Mn held on organic sites and oxide bound surfaces decreased.

The MnOX-Mn showed significant increase in its fractions with fertilizers and manure. It was reported higher in the treatments where organic and inorganic fertilizers were applied in combination as compared to the treatments where only chemical fertilizers were applied to the rice crop. The MnOX-Mn ranged from 61.43 to 66.20 mg kg^{-1} in the treatments organic manure @ 6 t ha^{-1} was incorporated along with N @ 80 kg ha^{-1} and P_2O_5 @ 30 kg ha^{-1} were applied and 68.20 to 73.90 mg kg^{-1} in the treatments where organic manure @ 6 t ha^{-1} was incorporated along with N @ 80 kg ha^{-1} was applied without incorporation of phosphatic fertilizer. On the other hand, it varied from 52.87 to 56.63 mg kg^{-1} in the treatments where no organic manure was incorporated and only N @ 120 kg ha^{-1} was applied without P_2O_5 application to the rice crop and 50.97 to 55.40 mg kg^{-1} in the treatments where no manure was incorporated and only chemical fertilizers were applied. Sekhon *et al.*, (2006) reported that addition of organic manure to rice increased potentially available fraction of Mn under rice-wheat rotation. Hellal (2007) reported that addition of composted mixtures increased MnOX-Mn in soil, as a result Fe availability is increased in calcareous soil by high acidulation effect of compost. Herencia *et al.,* (2008) showed the percentage of Fe and Mn in the specific fractions with respect to the total content are Mn>Fe and addition of organic matter caused Zn and Fe to move from less soluble forms to more plant available fraction which was always favoured by organic amendments.

The AFeOX-Mn reported significantly higher concentration in the treatments where organic manure and chemical fertilizers were applied in combination to the rice crop. Among the treatments, organically treated plots showed higher release of AFeOX-Mn in solution. The AFeOX-Mn ranged from 22.40 to 23.67 mg kg^{-1} in the treatments where organic manure @ 6 t ha^{-1} was incorporated along with N @ 80 kg ha^{-1} and P_2O_5 @ 30 kg ha^{-1} were applied. Similar increase was observed in the treatments where organic manure @ 6 t ha^{-1} was incorporated along with N @ 80 kg ha^{-1} was applied without incorporation of phosphatic fertilizer where it ranged from 20.03 to 21.87 mg kg^{-1}. However, it varied from 15.77 to 16.53 mg kg^{-1} in the treatments where no organic manure was incorporated and only N @ 120 kg ha^{-1} was applied without P_2O_5 application to the rice crop and 17.30 to 21.87 mg kg^{-1} in the other treatments where no manure was incorporated and only chemical fertilizers like N @ 120 kg ha^{-1} and P_2O_5 @ 30 kg ha^{-1} were applied. However, the significant difference was also observed in AFeOX-Mn fraction in the wheat crop where different levels of P_2O_5 (0, 30 and 60 kg ha^{-1}) were applied. The interaction between rice and wheat crops was found as non significant. Sekhon *et al.,* (2006) reported that addition of organic manure to rice increased AFeOX form of Mn under rice-wheat rotation. Agbenin (2003) reported a similar increase in AFeOX-Mn fractions fertilized with NPK, FYM and FYM+NPK. Singh *et al.,* (1988) in a study on 11 soils reported that 9 and 5 per cent of total Fe and Mn was associated with AFeOX fraction. The significantly higher concentration of CFeOX-Mn fraction was reported in the treatments where organic manure was incorporated along with chemical fertilizers (Table 5).

Table.1 Treatment details of long term experiment on rice-wheat system

Treatments	Rice			Wheat
	Manure	N	P_2O_5	P_2O_5
	(t ha^{-1})	(kg ha^{-1})		(kg ha^{-1})
T_1	0	120	0	0
T_2	0	120	0	30
T_3	0	120	0	60
T_4	6	80	30	0
T_5	6	80	30	30
T_6	6	80	30	60
T_7	0	120	30	0
T_8	0	120	30	30
T_9	0	120	30	60
T_{10}	6	80	0	0
T_{11}	6	80	0	30
T_{12}	6	80	0	60

Table.2 Chemical fractions of Fe (WSEX, SpAd, MnOX and AFeOX) in surface soil (0-15cm) under rice-wheat system

Treatments of rice	**Rates of P applied to wheat (kg P_2O_5 ha^{-1})**			**Mean**
	P_0	P_{30}	P_{60}	
WSEX-Fe (mg kg^{-1})				
M_0 N_{120} P_0	0.11	0.12	0.12	0.11
M_6 N_{80} P_{30}	0.15	0.15	0.16	0.15
M_0 N_{120} P_{30}	0.11	0.11	0.12	0.11
M_6 N_{80} P_0	0.11	0.13	0.15	0.13
Mean	0.12	0.13	0.14	-
LSD *(p<0.05)*	R=0.01, W=NS, RxW=NS			
SpAd-Fe (mg kg^{-1})				
M_0 N_{120} P_0	0.22	0.20	0.24	0.22
M_6 N_{80} P_{30}	0.25	0.25	0.27	0.26
M_0 N_{120} P_{30}	0.24	0.22	0.23	0.23
M_6 N_{80} P_0	0.26	0.25	0.27	0.26
Mean	0.24	0.23	0.25	-
LSD *(p<0.05)*	R=0.02, W=0.009, RxW=0.02			
MnOX-Fe (mg kg^{-1})				
M_0 N_{120} P_0	36.72	37.84	38.35	37.64
M_6 N_{80} P_{30}	44.77	45.60	48.73	46.37
M_0 N_{120} P_{30}	39.12	41.03	40.78	40.31
M_6 N_{80} P_0	42.86	41.58	43.25	42.56
Mean	40.87	41.51	42.78	-
LSD *(p<0.05)*	R=1.62, W=1.15, RxW=NS			
AFeOX-Fe (mg kg^{-1})				
M_0 N_{120} P_0	366.5	368.5	376.8	370.6
M_6 N_{80} P_{30}	386.8	389.9	390.1	388.9
M_0 N_{120} P_{30}	377.1	378.5	382.0	379.2
M_6 N_{80} P_0	385.9	383.4	385.1	384.8
Mean	379.1	380.1	383.5	-
LSD *(p<0.05)*	R=1.55, W=0.85, RxW=1.70			

Table.3 Chemical fractions of Fe (CFeOX, OM, RES and Total) in surface soil (0-15cm) under rice-wheat system

Treatments of rice	Rates of P applied to wheat (kg P_2O_5 ha^{-1})			Mean
	P_0	P_{30}	P_{60}	
CFeOX-Fe (mg kg^{-1})				
M_0 N_{120} P_0	564.8	570.2	569.5	568.2
M_6 N_{80} P_{30}	616.5	626.7	631.3	624.8
M_0 N_{120} P_{30}	579.8	582.3	585.0	582.4
M_6 N_{80} P_0	587.8	598.4	607.4	597.9
Mean	587.2	594.4	598.3	-
LSD *(p<0.05)*	R=1.80, W=1.32, RxW=NS			
OM-bound Fe (mg kg^{-1})				
M_0 N_{120} P_0	22.62	22.84	24.68	23.38
M_6 N_{80} P_{30}	24.60	26.28	26.02	25.63
M_0 N_{120} P_{30}	23.78	24.58	24.71	24.36
M_6 N_{80} P_0	24.99	25.09	24.65	24.91
Mean	23.99	24.70	25.02	-
LSD *(p<0.05)*	R=1.15, W=0.64, RxW=NS			
RES-Fe (%)				
M_0 N_{120} P_0	1.33	1.30	1.28	1.30
M_6 N_{80} P_{30}	1.37	1.36	1.34	1.36
M_0 N_{120} P_{30}	1.29	1.26	1.25	1.26
M_6 N_{80} P_0	1.41	1.39	1.38	1.39
Mean	1.35	1.33	1.31	-
LSD *(p<0.05)*	R=0.01, W=0.01, RxW=NS			
Total-Fe (%)				
M_0 N_{120} P_0	1.43	1.40	1.38	1.40
M_6 N_{80} P_{30}	1.47	1.47	1.45	1.46
M_0 N_{120} P_{30}	1.39	1.36	1.35	1.37
M_6 N_{80} P_0	1.51	1.49	1.48	1.49
Mean	1.45	1.43	1.42	-
LSD *(p<0.05)*	R=0.01, W=0.01, RxW=NS			

Table.4 Chemical fractions of Mn (WSEX, SpAd, MnOX and AFeOX) in surface soil (0-15cm) under rice-wheat system

Treatments of rice	Rates of P applied to wheat (kg P_2O_5 ha^{-1})			Mean
	P_0	P_{30}	P_{60}	
WSEX-Mn (mg kg^{-1})				
M_0 N_{120} P_0	3.99	3.77	3.81	3.86
M_6 N_{80} P_{30}	4.41	4.79	4.64	4.61
M_0 N_{120} P_{30}	4.01	3.98	4.27	4.08
M_6 N_{80} P_0	4.17	4.25	4.39	4.27
Mean	4.14	4.20	4.28	-
LSD *(p<0.05)*	R=0.41, W=NS, RxW=NS			
SpAd-Mn (mg kg^{-1})				
M_0 N_{120} P_0	2.25	2.36	2.47	2.36
M_6 N_{80} P_{30}	2.77	2.80	2.89	2.82
M_0 N_{120} P_{30}	2.55	2.67	2.76	2.66
M_6 N_{80} P_0	2.85	2.91	2.69	2.82
Mean	2.60	2.69	2.70	-
LSD *(p<0.05)*	R=0.20, W=NS, RxW=NS			
MnOX-Mn (mg kg^{-1})				
M_0 N_{120} P_0	52.87	54.80	56.63	54.77
M_6 N_{80} P_{30}	61.43	62.43	66.20	63.36
M_0 N_{120} P_{30}	50.97	55.40	54.00	53.46
M_6 N_{80} P_0	68.20	68.80	73.90	70.30
Mean	58.37	60.36	62.68	-
LSD *(p<0.05)*	R=1.23, W=0.81, RxW=1.62			
AFeOX-Mn (mg kg^{-1})				
M_0 N_{120} P_0	15.77	16.13	16.53	16.14
M_6 N_{80} P_{30}	22.40	22.53	23.67	22.87
M_0 N_{120} P_{30}	17.30	19.23	19.63	18.72
M_6 N_{80} P_0	20.30	20.03	21.87	20.73
Mean	18.94	19.48	20.43	-
LSD *(p<0.05)*	R=1.08, W=1.16, RxW=NS			

Table.5 Chemical fractions of Mn (CFeOX, OM, RES and Total) in surface soil (0-15cm) under rice-wheat system

Treatments of rice	**Rates of P applied to wheat (kg P_2O_5 ha^{-1})**			**Mean**
	P_0	P_{30}	P_{60}	
CFeOX-Mn (mg kg^{-1})				
M_0 N_{120} P_0	19.47	19.33	20.14	19.65
M_6 N_{80} P_{30}	20.20	20.27	20.70	20.39
M_0 N_{120} P_{30}	16.70	17.13	18.40	17.41
M_6 N_{80} P_0	20.50	20.90	21.67	21.02
Mean	19.22	19.41	20.23	-
LSD *($p<0.05$)*	R=1.11, W=0.70, RxW=NS			
OM-bound-Mn (mg kg^{-1})				
M_0 N_{120} P_0	1.26	1.28	1.30	1.28
M_6 N_{80} P_{30}	1.44	1.48	1.55	1.49
M_0 N_{120} P_{30}	1.30	1.33	1.35	1.33
M_6 N_{80} P_0	1.37	1.37	1.43	1.39
Mean	1.35	1.37	1.41	-
LSD *($p<0.05$)*	R=0.07, W=0.05, RxW=NS			
RES-Mn (mg kg^{-1})				
M_0 N_{120} P_0	93.20	92.45	86.22	90.62
M_6 N_{80} P_{30}	84.27	81.24	73.55	79.69
M_0 N_{120} P_{30}	95.28	87.09	84.63	89.00
M_6 N_{80} P_0	84.11	77.37	71.05	77.51
Mean	89.22	84.54	78.86	-
LSD *($p<0.05$)*	R=2.07, W=2.80, RxW=NS			
Total-Mn (mg kg^{-1})				
M_0 N_{120} P_0	192.1	190.1	187.1	189.8
M_6 N_{80} P_{30}	196.9	195.5	193.2	195.2
M_0 N_{120} P_{30}	188.1	186.8	185.0	186.7
M_6 N_{80} P_0	201.5	198.9	197.0	199.2
Mean	194.7	192.9	190.6	-
LSD *($p<0.05$)*	R=0.30, W=0.48, RxW=NS			

The significant higher concentration that ranged from 20.20 to 20.70 mg kg^{-1} of this fraction was noticed in the treatments where organic manure @ 6 t ha^{-1} was incorporated along with N @ 80 kg ha^{-1} and P_2O_5 @ 30 kg ha^{-1} were applied. Similar increase was observed in the treatments where organic manure @ 6 t ha^{-1} was incorporated along with N @ 80 kg ha^{-1} was applied without incorporation of phosphatic fertilizer where it ranged from 20.50 to 21.67 mg kg^{-1}. The higher concentrations of CFeOX-Mn reported that Mn requirement can be maintained from this fraction as Mn associated with CFeOX released more concentration of Mn in the soil solution. Singh *et al.*, (1988) in a study on 11 soils reported that 11 and 17 per cent of total Mn were associated with CFeOX fraction.

The significant increase was noticed in organically bound fraction (OM-Mn) in the treatments where organic manure was incorporated along with chemical fertilizers were applied where it ranged from 1.44 to 1.55 mg kg^{-1} and 1.37 and 1.43 mg kg^{-1} as compared to the treatment which were treated inorganically and it ranged from 1.26 to 1.30 mg kg^{-1} and 1.30 to 1.35 mg kg^{-1}, also no organic manure was incorporated in these treatments. However, the significant difference was also observed in OM-Mn fraction in case of wheat crop where different levels of P_2O_5 (0, 30 and 60 kg ha^{-1}) were applied. The interaction between rice and wheat crops was found as non-significant. The higher concentration of OM-Mn in the soil solution indicated that the micronutrients associated with the OM bound fraction may play a beneficial role in the uptake of these nutrients by the plants.

Sekhon *et al.*, (2006) reported that OM bound fraction of Mn increased with application of organic manure in rice-wheat system.

The RES-Mn varied from 71.05 to 93.20 mg kg^{-1} in all the treatments. The concentration for RES-Mn was observed higher as compared to all other fractions except total Mn fraction. The higher level of Mn in these fractions under rice-wheat cropping system may be due to effect of submergence.

In conclusion, the inter conversion of Fe and Mn from one fraction to the other was accelerated with the addition of the manure. So, the concentration of Fe and Mn were found higher in the treatments where biogas slurry was incorporated along with inorganic fertilizers in different fractions viz. WSEX, SpAd, MnOX, AFeOX, CFeOX, OM-bound, RES and Total. The residual (RES) fraction is the dominating fraction among all the different fractions. Water soluble and exchangeable (WSEX) fraction contributes little as compared to the other fractions viz. Crystalline Fe-oxide (CFeOX) and Amorphous Fe-oxide (AFeOX) fractions. The organic compounds released during decomposition of manures enhanced the availability of Fe and Mn by preventing fixation, oxidation, precipitation and leaching.

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