



Risk assessment through rice (*Oryza sativa*) in sludge treated soil

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ABSTRACT

An experiment was carried out on basmati rice (*Oryza sativa*, var. Pusa Basmati 1121) using IARI soil to assess the effect of applied sludge on different soil properties, metals content in rice grain and computed hazard quotient for suitability of rice grain for human consumption. The different doses of sludge were used as treatments 0, 10, 20, 30, 40 and 50 g/kg. The results indicated that applied sludge @ (50 g/kg) reduced the pH from 7.98 (control) to 7.21 of sludge amended soil. Whereas, the EC increased from 0.22 (control) to 1.33 dS/m under applied @ 50 g sludge/kg of soil. The organic carbon content and cation exchange capacity were found highest in 50 g sludge/kg soil treatment as compared to control. DTPA Extractable metals in post-harvest soil were significantly higher in sludge application @ ≥ 30 g/kg treatments. The value of HQ ranged from 0.05 to 0.49 and 0.02 to 0.09 for Pb and Cd, respectively across the various treatments of sludge addition. It can be concluded that risk assessment for intake of metals through consumption of rice by human indicated that application of sludge should be restricted up to 20 g/kg unfit for rice cultivation.

Key words: DTPA extractable metals, Grain metal content, Hazard quotient, Risk assessment, Sludge

The application of sewage sludge in agriculture as a source of plant nutrients and as soil conditioner is increasingly being favoured by the farmers not only in our country but across the globe. As per the records of Central Pollution Control Board (2016) the total sewage generated from urban areas was estimated 62000 million litres per day (MLD) while the treatment capacity across India is only 23277 MLD, or 37% of sewage generated. Sewage sludge having a relatively variable composition, it is rich in organic matter, primary nutrients, secondary nutrients and may also contain micronutrients (Golui *et al.* 2014). These waste materials add substantial amount of organic matter, besides improving physico-chemical properties of soil like bulk density, water holding capacity, and cation exchange capacity (Meena and Patel 2018). Significant enhancement in microbial biomass carbon and dehydrogenase activity have been reported particularly when higher amount of sludge was applied (Mondal *et al.* 2015). Long term addition of sludge in agricultural land creates adverse changes such as decreases in pH, increase in salinity and heavy metals contents (Singh and Agrawal 2008). Usually, prominent concentrations of heavy metals were observed in soils amended with sewage sludge (Golui

et al. 2014). The addition of sludge to soils could also affect potential availability of heavy metals (Moreira *et al.* 2013). Total content of toxic trace elements in soil gives an insight into their inventory. Besides this, extractable metal pools in sludge amended soils have been considered as a good hazard index (Ray *et al.* 2013). Currently, 43 million ha of land is under the cultivation of rice crop in our country with the total production of 110.15 million tonnes (Annual Report 2017-18, Ministry of Agriculture and Farmer Welfare). Limited information is available on suitability of use of sludge in rice crop in relation to soil quality and food chain contamination by metals. The present investigation was undertaken to study the effect of sewage sludge used in rice on metal uptake by rice, transference of metal from soil to human food chain and soil properties.

MATERIALS AND METHODS

An experiment was conducted on basmati rice var. Pusa Basmati 1121 in 2016 to study the effect of sludge application on metal uptake by rice. Sludge was collected from sewage treatment plants of Delhi, i.e. Okhla, in the month of March and analysed for chemical properties using standard procedure and sludge consisted pH (6.73 ± 0.02) and EC (3.92 ± 0.03 dS/m). Total C, N, P, K and S contain in sludge was 13.9 ± 0.01 , 1.67 ± 0.01 , 1.60 ± 0.14 , 0.18 ± 0.01 and $1.92 \pm 0.01\%$, respectively. Total trace elements in sludge were $0.95 \pm 0.03\%$ Fe, 1186 ± 87.1 mg/kg Zn, 94.1 ± 2.29 mg/kg Cu, 128 ± 2.11 mg/kg Mn, 32.7 ± 0.58 mg/kg Pb and 4.75 ± 0.66 mg/kg Cd.

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Experiment was conducted on rice using IARI soil (28°30' N, 77°10' E; 250 m amsl), New Delhi. The pH and electrical conductivity (EC) of IARI soil was 8.0 and 0.26 dS/m, respectively. The soils of IARI belong to sandy loam textural class. Organic carbon and cation exchange capacity were 0.32% and 16.9 [cmol (+)/kg], respectively. Available N, P, K, DTPA Extractable Zn, Fe, Cu, Mn, Pb and Cd content in IARI soil were 94.6, 8.51, 139, 2.06, 4.11, 1.24, 7.26, 0.94 mg/kg and 38.3 µg/kg, respectively. For this purpose plastic pots of 8 kg capacity were filled with 5 kg of soil. The sludge was added @ 0, 10, 20, 30, 40 and 50 g/kg of soil. All six treatments were arranged in completely randomised design (CRD) replicated thrice. A dose of N: P₂O₅: K₂O @ 75: 40: 30 mg/kg soils were added in solution form to the soil of each pot through urea, DAP and muriate of potash, respectively. Half of N and full dose of P and K were applied at the time of transplanting of rice and remaining N fertilizer was applied in two equal splits at tillering and flower initiation stage. Plants were harvested at maturity and recorded yield data. After harvest of the crop, soil samples were collected and analysed for pH (1:2:: soil: water), electrical conductivity (EC), cation exchange capacity (Jackson 1973), organic carbon (OC)(Walkley and Black 1934) and dehydrogenase activity (Klein *et al.* 1971). Available nitrogen by Subbiah and Asija (1956), available phosphorus (Olsen *et al.* 1954), available potassium (Jackson 1973) and DTPA Extractable Zn, Fe, Cu, Mn, Cd and Pb (Lindsay and Norvell 1978) in soil samples were analysed. Metal contents in the samples were determined using ICP-MS. Rice grain samples were digested with HNO₃ in a microwave digester and content of metals in the digested materials were determined using ICP-MS.

Hazard quotient is the ratio of the average daily dose (ADD; mg/kg/d) of metals to their reference dose (RfD; mg/kg/d (Datta and Young 2005, Meena *et al.* 2016, Mishra *et al.* 2019). The values of RfD used for Pb and Cd were 0.0035 and 0.001 mg/kg/d, respectively (WHO 1982). Daily intake of rice grain was assumed to be 0.2 kg/d which is the recommended amount from a nutritional point of view. Average body weight for an adult was assumed to be 70 kg (USEPA 1991). Statistical analysis of data was done adopting through completely randomized design (Snedecor and Cochran 1967).

RESULTS AND DISCUSSION

Soil pH, electrical conductivity (EC), organic carbon (OC), cation exchange capacity (CEC) and dehydrogenase activities (DHA) in post-harvest soil

Application of sludge @ 50 g/kg soil reduced the pH from 7.97 (control) to 7.21 in post-harvest soil samples. Significant decrease in pH was observed in all the treatments over control but no significant difference in pH was observed within treatments, *i.e.* sludge applied @ 30, 40 and 50 g/kg. The range of EC value was recorded from 0.24 (control) to 1.33 dS/m (50 g sludge/kg soil). The highest value of

EC was observed in 50 g sludge/kg of soil application followed by 40, 30, 20 and 10 g/kg soil (Table 1). Soil OC of 0, 10, 20, 30, 40 and 50 g/kg of sludge treated soil was 0.29, 0.35, 0.44, 0.55, 0.76 and 0.94%, respectively. The significant increase in OC content was found when sludge was applied more than 10 g/kg soil over control. The highest value of CEC was recorded with 50 g/kg (24.3 [cmol(+)/kg]) followed by treatments 40 g/kg (21.8 [cmol(+)/kg]) and 30 g/kg (20.2 [cmol(+)/kg]). Moreover, sludge applied 30 g/kg treatment was statistically at par with all treatments except control in term of CEC. Lowest value of CEC was observed in control (16.8[cmol(+)/kg]). The dehydrogenase activity (DHA) in soil ranged from 4.25 to 6.17 µg TPF/g/h. Significant highest DHA in post-harvest soil was recorded under sludge applied @ 50g/kg as compared to other treatments, whereas sludge applied @ 10, 20, 30 and 40 g/kg treatments were statistically at par with each other. The significant increase in dehydrogenase activity was observed under 40 and 50 g sludge/kg soil over control (Table 1). The consistent decrease in soil pH with increase in the rate of sludge over control was recorded. The reason of the decline in pH of sludge treated soil may be lower pH (6.73) of sludge as compared to that of soil and various organic acids might have released from sludge during the period of experimentation (Singh and Agrawal 2010, Golui *et al.* 2014). Soil properties like EC, OC, CEC and dehydrogenase activity increased in post-harvest soil of different treatments due to application of sludge contain higher soluble salts and TOC content (13.9%)(Latere *et al.* 2014, Mondal *et al.* 2015).

Available nitrogen, phosphorus and potassium in post-harvest soil

Available N content in post-harvest soil was found to be significantly higher under sludge applied @ 50 g/kg (158 mg/kg) as compared to control. No significant changes in available N was noticed up to sludge applied @ 30 g/kg but treatment sludge applied @ 40g/kg was statistically at par with the sludge applied @ 30 and 50 g/kg treatments. Similar trends were observed in available P and K. Increasing doses of applied sludge also increased the available P and available K content in soil from 8.22(control) to 18.7mg/kg (50 g/kg) and from 141(control) to 186 mg/kg (50 g/kg), respectively in post-harvest soil (Table 1). Several earlier studies reported that, addition of sludge undoubtedly improved the fertility status of soil in respect of major nutrients N, P and K (Nikzad *et al.* 2015).

DTPA Extractable metals in post-harvest soil

DTPA Extractable Zn content in soil was recorded higher as 3.73, 6.00, 8.44, 8.61 and 13.5 mg/kg when sludge applied @ 10, 20, 30, 40, and 50 g/kg, respectively compared to control (2.45 mg/kg) (Table 2). Significant increase in DTPA Extractable Zn was obtained beyond 10 g/kg of sludge application over control. Similar results were obtained in case of DTPA Extractable Fe. While DTPA extractable Cu content in soil ranged from 1.65 to 4.01 mg/kg. Significant

Table 1 Effect of sludge application on chemical and biological properties of soil

Rates of sludge addition (g/kg)	Properties					Available nutrients (mg/kg)		
	pH _{1:2}	EC _{1:2} (dS/m)	Organic carbon (%)	CEC [cmol (+) /kg]	DHA (µg TPF/g/ h)	Available N	Available P	Available K
Control	7.97 ^a	0.24 ^d	0.29 ^e	16.8 ^c	4.25 ^c	93.0 ^c	8.22 ^d	141 ^b
10	7.62 ^b	0.72 ^c	0.35 ^{de}	17.7 ^{bc}	4.38 ^{bc}	98.0 ^c	10.2 ^{cd}	160 ^{ab}
20	7.48 ^c	0.98 ^{bc}	0.44 ^{cd}	19.1 ^{bc}	4.74 ^{bc}	106 ^c	10.5 ^{cd}	171 ^b
30	7.32 ^d	1.03 ^b	0.55 ^c	20.2 ^{abc}	5.09 ^{bc}	117 ^{bc}	12.3 ^{bc}	177 ^a
40	7.25 ^d	1.13 ^{ab}	0.76 ^b	21.8 ^{ab}	5.23 ^b	135 ^{ab}	15.2 ^b	181 ^a
50	7.21 ^d	1.33 ^a	0.94 ^a	24.3 ^a	6.17 ^a	158 ^a	18.7 ^a	186 ^a
LSD (P=0.05)	0.11	0.29	0.14	4.48	0.92	24.5	2.99	32.9

highest DTPA-Cu was noticed in sludge applied @ 50 g/kg as compared to 20 g/kg treatment. There was no significant difference within the treatment of 30, 40 and 50 g/kg of sludge treatments with respect to DTPA Extractable Cu. DTPA Extractable Mn content in soil ranged from 8.02(control) to 24.1 mg/kg (50 g/kg) (Table 2). Significantly higher DTPA extractable Mn was recorded among all the sludge applied treatments over control. Beyond application of 20 g sludge/kg, significant increment was observed in Mn content with increase dose of sludge up to 50 g/kg of soil. In case of DTPA Extractable Pb and Cd in post-harvest soil varied from 0.87 (control) to 1.65 mg/kg (50 g/kg) and from 27.7 (control) to 55.9 µg/kg(50 g/kg), respectively (Table 2). Significant highest DTPA Extractable Pb and Cd were recorded with sludge applied @50 g/kg (1.65 mg/kg and 55.9 µg/kg, respectively) over sludge applied @30 g/kg treatment. There were no significant changes found in DTPA extractable Pb in soil below 10 g/kg sludge. DTPA extractable Zn, Cu, Fe, and Mn increased with increase rates of sludge application. Such increase in extractable micronutrient pool as a result of sludge application can be considered as a positive effect. However, DTPA extractable Zn exceeded the phyto toxicity limit, i.e. 10 mg/kg (Rattan *et al.* 2005). There was significant increase in Cd and Pb content in sludge amended soil over control. Several researchers also reported that sludge application increased the extractable metals content in post-harvest soil (Ray *et al.* 2013, Golui *et al.* 2014, Latore *et al.* 2014).

Biomass yield and metal content in rice grain

The highest dry biomass yield of rice was recorded as 57.7 g/pot (50 g/kg) and lowest 25 g/pot (control with no amendments application). Increased in dry biomass was statistically at par with successive increasing dose of sludge (data not presented in table). Total Zn, Fe, Cu and Mn content in rice grain ranged from 21.5, 6.61, 2.82 and 9.58 mg/kg (control) to 38.4, 19.5, 6.53 and 19.8 mg/kg (50 g/kg), respectively. Significant highest Zn, Fe, Cu and Mn in rice grain were recorded under 50 g/kg soil treatment while treatment @ 40 g/kg soil was statistically similar to 30 and 50 g/kg soil treatments in Zn, Mn and Fe content in rice grain. However, sludge @ 30 g/kg recorded similar Cu

content in rice grain with 20 and 40 g/kg sludge application treatment. Lead content in grain was 66, 134, 221, 338, 539 and 595 µg/kg with addition of sludge at the rate of 0, 10, 20, 30, 40 and 50 g/kg, respectively. Significant increase in Pb content in grain of rice was recorded when sludge was applied @ ≥ 10 g/kg. Cadmium content in rice grain was increased by 162, 225, 262, 287 and 536% over control in sludge added @ 10, 20, 30, 40 and 50 g/kg, respectively (Table 2). Treatments with application of sludge @ 40 and 50 g/kg soil recorded significant increment in Cd content in rice grains. By and large, a consistent increase in metal content in rice grains was observed as a result of concomitant increased rate of sludge application. This is attributed to the fact that considerable amount of Zn, Cu, Fe, Mn, Cd and Pb was added to soil through sludge, which in turn enhances the available pool of metals in soil. Apart from extractable metal in soil, pH and organic carbon have been considered as the most important factors govern the solubility of metals in soil (Datta and Young 2005; Golui *et al.* 2014).

Hazard quotient (HQ) for intake of metals by human through consumption of rice grain grown on sludge treated soil

Risk to human health for intake of metals by human through consumption of rice grain grown on sludge treated soils was assessed in term of hazard quotient (HQ) is presented (Fig 1). Results indicate that HQ ranged from 0.05 to 0.49 and 0.02 to 0.09 for Pb and Cd, respectively across the various treatments of sludge addition. The value of HQ for Pb particularly at higher rates of sludge addition, i.e. 30, 40 and 50 g/kg should be cause concern. Hence, there is an indication that application of sludge should be limited to 20 g/kg of soil. These results need verification under field condition, but we found that when sludge applied more than 30 g/kg of soil, rice grain becomes unfit for human consumption.

Conclusions

Based on findings, it can be concluded that sludge is a good source of plant nutrients to enhanced crop productivity and soil fertility. A substantial improvement in soil fertility and important biological properties was observed with

Table 2 Effect of sludge application on DTPA Extractable micronutrients and heavy metals in soil and rice grain

Rates of sludge addition (g/kg)	Micronutrients and heavy metals in soil (mg/kg)						Micronutrients and heavy metals in rice grain (mg/kg)					
	Zn	Fe	Cu	Mn	Pb	Cd*	Zn	Fe	Cu	Mn	Pb*	Cd*
Control	2.45 ^d	4.75 ^e	1.65 ^c	8.02 ^e	0.87 ^b	27.7 ^c	21.5 ^d	6.61 ^d	2.82 ^e	9.58 ^d	66.0 ^e	5.33 ^d
10	3.73 ^d	6.48 ^e	1.93 ^c	10.6 ^d	0.94 ^b	28.7 ^c	25.3 ^d	9.28 ^c	3.26 ^{de}	11.5 ^d	134 ^d	8.67 ^c
20	6.00 ^c	9.11 ^d	2.79 ^b	12.8 ^d	1.37 ^a	34.0 ^c	26.8 ^{cd}	10.6 ^c	4.07 ^{cd}	14.5 ^c	221 ^c	12.0 ^{bc}
30	8.44 ^b	14.2 ^c	3.43 ^{ab}	15.6 ^c	1.38 ^a	41.4 ^b	31.1 ^{bc}	15.1 ^b	4.84 ^{bc}	16.7 ^{bc}	338 ^b	14.0 ^b
40	8.61 ^b	17.8 ^b	3.89 ^a	19.7 ^b	1.50 ^a	43.3 ^b	35.2 ^{ab}	17.2 ^{ab}	5.46 ^b	18.4 ^{ab}	539 ^a	15.3 ^b
50	13.5 ^a	23.6 ^a	4.01 ^a	24.1 ^a	1.65 ^a	55.9 ^a	38.4 ^a	19.5 ^a	6.53 ^a	19.8 ^a	595 ^a	28.6 ^a
LSD (P=0.05)	1.88	2.07	0.78	2.36	0.37	7.39	6.54	2.38	0.85	2.43	62.5	3.81

* (µg/kg)

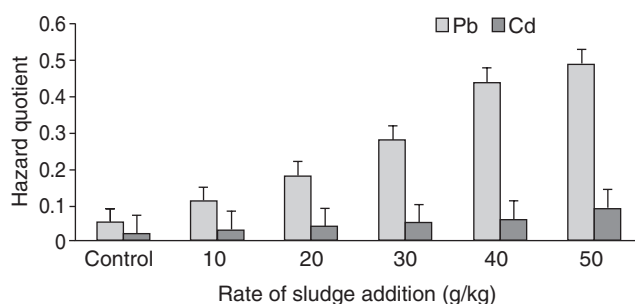


Fig. 1 Human health risk in terms of hazard quotient (HQ) for dietary intake of rice grown on sludge amended soils. Error bar indicate LSD value.

sludge application. Rice crop responded positively to sludge application. However, elevated level of toxic traces elements namely, Pb and Cd particularly at higher rates of sludge application may be a cause of concern. Risk assessment for intake of metals through consumption of rice by human indicated that application of sludge should be restricted up to 20 g/kg of soil.

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