Research article

Phosphorous and lime-induced zinc fractions transformations in Zn deficit tropical paddy soils

Shahram Mahmoud Soltani1*, Mohamed Musa Hanafi2, 3, Abdul Wahid Samsuri3, Sharifah Kharidah4

Article Info

ABSTRACT

The soil geochemical Zn controlling factors in soil solid-solution equilibrium influence the phyto-available forms of Zn in soil Zinc fraction pools. A laboratory incubation experiment was conducted on two highly weathered tropical soil orders (Ultisols and Entisols) to investigate the effects of some of the most important of these factors (various levels of Phosphorus (P), lime, and Zn) on Zn fractions in six tropical paddy soils. The sequential fractionation procedure was used to determine soils Zn fractions after 30 days of submerged incubation threat by the aforementioned factors. The non-residual and available fractions (water-soluble plus exchangeable (WE) and organic complexes (Org)) increased by Zn treatments about 1.29 to 2.65 times, and 1.7 to 4 times, respectively and decreased with increasing lime application levels about 9 to 30%, and 3 to 37%, respectively, whereas the non-residual and non-available (amorphous (Amor) and manganese oxides (MN)) and the residual fractions increased with the increasing level of applied P and lime. The effects of P, lime, and Zn applications on Zn fractions were similar for all soils. The application of P plus lime was more effective in reducing available Zn fractions and increasing non-available fractions than the separate application of P or lime between 30 to 70%. It can be concluded that at the studied soil conditions and to avoid Zn deficiency in rice paddy fields due to P and lime application, Zn fertilizer should be applied more than rice nutrition requirements to overcome converted Zn to the unavailable fractions.

Keywords: Highly weathered soils, Organic zinc, Residual fraction, Sequential analysis

INTRODUCTION

The capacity of Zinc (Zn) fractions to supply available Zn for optimum growth of crops varies widely among soil types. Zinc deficit soils are developing seriously across tropical paddy soils which are heavily weathered and leached (Alloway, 2008). Soil zinc presents in a number of chemical fractions with varying solubility (Almendros et al. 2008). The phyto-available Zn in soils depends greatly on its specific fractions rather than its total concentrations (Fan et al. 2009). The availability of Zn in soils depends on the equilibrium among different Zn fractions (Cao et al. 2013; Lee et al. 2012).

The main controlling factors that affect the equilibrium between plant-available and other Zn fractions of soils are the total amount of soil Zn content, soil pH, reduction and oxidation conditions, calcium carbonate (CaCO3) content (liming), organic matter (OM) percentage, and the concentrations of micro- and macro-elements especially phosphorus (P) (Alloway 2009). In tropical paddy soils, liming and P content are the most crucial factors controlling plant available Zn. (Alloway 2009).

Excess amount of P application have been reported to induce Zn deficiency in rice (Ranjha et al. 2001; Nawaz et al. 2012) due to the interactions between P and Zn (Gao et al. 2012). High available P in paddy soils causes a change in soil surface charges and a change in soil pH due to P adsorption by the soil colloids (Mandal and Mandal 1986; Shuman 1985, 1998). Rupa et al. (2003)

---

1 Assistant Professor of Rice Research Institute of Iran, Agricultural Research, Education and Extension Organization, Rasht, Iran.
2 Laboratory of Plantation Crops, Institute of Tropical Agriculture, Universiti Putra Malaysia, 43400, Serdang, Selangor, Malaysia.
3 Department of Land Management, Faculty of Agriculture, Universiti Putra Malaysia, 43400, Serdang, Selangor, Malaysia.
4 Department of Food Science, Faculty of Food science and technology, Universiti Putra Malaysia, 43400, Serdang, Selangor, Malaysia.
* E-mail: shmsoltani@gmail.com
reported that the different soil Zn fractions indicated various response to the added P levels. Application of more than 40 mg kg\(^{-1}\) P decreased the concentration of plant-available Zn fractions (water soluble plus exchangeable (WE) and organo-complexes (Org)), whereas the concentration of non-soluble forms (crystalline (Cry), manganese oxide (MN)) and residual fractions increased in soils. Also, in spite of high reduction in the residual fraction due to P application, the only lower transformation of added Zn to WE and Org fractions negatively affected Zn uptake by crops (Rieuwerts 2007).

Application of lime increases the pH and the amount of basic cations (Ca\(^{2+}\) and Mg\(^{2+}\)) in tropical paddy soils (Chen et al. 2000) and reduced the Al\(^{3+}\) and Fe\(^{3+}\) toxicity (Alloway 2008). However, the bioavailability of soil Zn within a short-term use of lime sharply decreased due to the increase in soil pH which causes Zn precipitation (Adhikari and Rattan 2007; Wang et al. 2001; Zhang et al. 2010b). Wang (2001) showed that the concentrations of extractable Zn were significantly reduced by 96% after 30 days of CaCO\(_3\) application. Lime often can stabilize the Zn exchangeable fractions due to precipitating of this fraction to insoluble compounds (Cakmak et al. 2010) and increasing the cation exchange capacity of soil (Hong et al. 2002). In the rare situations, high soil pH can promote the dissolution of soil organic matter (Laor et al. 1998; Zhang et al. 2010a), and increase in the availability of soil Zn (Wang et al. 2001). Keeping the above facts in view, management of Zn tropical paddy soil is very important in order to avoid Zn deficiency. Therefore, this study was conducted to determine the interactive effects of Zn, lime and P applications on Zn fractions in tropical paddy soils.

MATERIALS AND METHODS

The incubation experiment was conducted on two highly weathered tropical soil orders (Ultisols and Entisols) that divided into six soil series. Available zinc concentrations in the all selected soils were below critical zinc level (double acid method) and soils are Zn deficit.

The soils from the surface layer (0-30 cm) of the rice paddy fields of Kelantan and Kedah states-two largest rice production areas in Malaysia-were sampled. The soil samples were air-dried, ground to pass through a 2-mm metal sieve and stored for the current experiment. The physical and chemical properties of the selected soil samples, such as: texture, organic carbon, pH (1:2.5), cation exchange capacity (CEC) (C mole kg\(^{-1}\)), available Zn, Phosphorus, Fe\(^{2+}\) and Mn\(^{2+}\) were determined. A pipette method was used to analyse soil particle-size (Gee et al., 1986). Soil pH and electrical conductivity were measured in a 1:2 ratio (w: v) soil to water suspension. Cation-exchange capacity was measured by leaching and replacing procedure (Bower et al., 1952). Phosphorus was measured by Bary and Krutz (1945) method. Oxidizable organic matter was measured using the Walkley–Black (Jackson 1964) and available zinc by double acid method (Amacher 1996).

10 grams portions of the stored soil samples were placed in 294 of 100 mL plastic vials and treated with: (i) three levels of P (0, 50, and 100 mg kg\(^{-1}\)) as KH\(_2\)PO\(_4\), (ii) three levels of Zn (0, 2.5 and 5 mg kg\(^{-1}\)) as zinc sulphate, and two levels of liming requirement (LR) (0 and LR to reach pH to 5.5), all in the form of solutions, which are mixed uniformly with the soils. The flooded moisture regimes were maintained for all of the treatments. A randomized complete design with four factors factorial triplicate experiment and 3 levels of P, 6 types of soil series, 3 levels of Zn and 2 levels of LR were carried out. The containers were incubated at room temperature (25°C ± 0.5) in the laboratory of agricultural faculty in University of Putra Malaysia. The soil samples were sequentially extracted after 30 days (reproductive stages initiation). Zinc fractions (fertility fractions), water soluble plus exchangeable, organically complexes, amorphous sesquioxides bound and crystalline sesquioxides bound fractions were determine by modified Mandal and Mandal (1986) procedure and residual (Res) and total Zn with aqua regia method. Data analyses were done using the IBM SPSS 21 and SAS 9.2 software.

RESULTS AND DISCUSSION

Classification and general properties of soils

According to USDA soil taxonomy, selected soils, Telemong, Lubok Itek and Kundur soil series belong to Entisols orders and Chengai, Tok Yang, and Tepus soil series classified in Ultisols orders, whereas parent material of all soils almost were recent alluvial. Available Zn ranged from 0.56 mg Z/kg soil in Tepus soil series of acid sulphate soils, to 1.185 mg Zn/kg in Telemong soil series as a neutral soils, which all Zn contents of soils series were less than the critical limit in paddy soils (2 mg kg\(^{-1}\)) (Dobermann 2000).

Concentration and distribution of Zinc fractions in soil

According to the soil native Zn sequential fractionation total Zn were low to medium level (10.136 to 52.018 mg kg\(^{-1}\)), with highest content of residual fractions ranged from 5.167-38.700 mg kg\(^{-1}\) (54 to 82% of total Zn), whereas WE form varied from 0.273 to 0.517 mg kg\(^{-1}\) (0.5-3.4%, an average of 2.13% of total Zn) which indicated low bioavailability of Zn for rice uptake in all soils. These data emphasized that studied paddy soils were Zn deficit. Between these two extreme
Table 1. Concentrations of water soluble and exchangeable Zn in six tropical soil series in incubation experiment receiving various Zn, P and lime levels

| Treatment | Tropical Soil Orders and Series | Lubok Itek | Kundur | Tok Yong | Chengai | Tupus | Telemong |
|-----------|---------------------------------|------------|--------|---------|---------|-------|----------|
|           | mg Zn kg⁻¹ soil                 |------------|--------|---------|---------|-------|----------|
| Zn0 L0    |                                 | 0.52       | 0.52   | 0.27    | 0.42    | 0.34  | 0.35     |
| Zn0 L1    |                                 | 0.42       | 0.33   | 0.18    | 0.34    | 0.31  | 0.35     |
| Zn5 L0    |                                 | 1.17       | 0.94   | 0.50    | 0.63    | 0.45  | 0.35     |
| Zn5 L1    |                                 | 0.68       | 0.61   | 0.43    | 0.41    | 0.19  | 0.45     |
| Zn10 L0   |                                 | 1.31       | 1.67   | 0.64    | 0.60    | 0.82  | 0.45     |
| Zn10 L1   |                                 | 1.01       | 1.49   | 0.47    | 0.60    | 0.14  | 0.19     |

ranges, Mn (0.243-1.193 mg kg⁻¹), Org (0.584-3.04 mg kg⁻¹), Amor (0.875-6.280 mg kg⁻¹) and crystalline bound zinc fractions (0.259-3 mg kg⁻1) were located. Therefore, the fractionation distributions were in the following order: WE< MN< Org< Amor< Cry< RES. Thus, Zn in these soils seems to have predominantly been fixed with mineral crystalline lattices.

Zn fractions transformations

Water soluble plus exchangeable zinc fraction

Analysis of variance showed that the concentration of WE fraction were significantly affected by soil type (P<0.01), Zn levels (P<0.01), lime application (P<0.01), P (P<0.01) and also by treatments combination interactions (p<0.01) (Table 7). By increasing the Zn level from 0 to 2.5 mgkg⁻¹, the concentrations of WE fractions in all soils were elevated from 1.29 times in Tepus and Telemong (low total Zn soil series) to 2.65 times in Tok Yong with the highest amount of total Zn; nevertheless, compared to the 2.5 mgkg⁻¹, more Zn concentration was observed in 5 mg kg⁻¹ treatments but the slope of increasing did not follow any regular pattern (Table 1). Also, the results of Table 1 showed a distinct decrease of native NH₄OAc-extractable fraction (WE) due to P and lime application under submerged soil condition either separately or in combination, the reduction in WE concentration being more accelerated in combination conditions. Although, applied Zn treatments would increase WE availability, particularly by increasing Zn values from 0 to 5 mg kg⁻¹ in comparison with non-treated soils, but the most decreasing percentage in concentration of WE fraction vs P and lime application were observed in the highest level of Zn. It meant that the excess amount of Zn would accelerate the immobilization of Zn in the soils. The effect of treatments on reduction of WE fractions are in the following order: P + lime > P > lime in all incubated soils.

In addition, applied P and lime also caused a drop in the transformation of applied Zn into WE fraction. As mentioned before about native soil Zn (untreated soils), the depressing effect was more pronounced under P plus lime treatments than these two alone. The change in applied Zn in WE vs P and lime followed the same trend as native Zn fractionation. Soil solution thermodynamic findings for Zn behaviour in flooded soils showed soluble P was the most effective in Zn immobilization through lower soluble Zn products formation (Mandal and Mandal 1990; Rupa et al. 2003). Integrated amendment of soluble P materials followed by the excessive P immobilized with carbonaceous materials, for example, CaCO₃ or (Ca Mg) CO₃, could promote excessive Zn reduction. Furthermore, the native soil P solubility increased by the submerged condition due to the reduction and/or hydrolysis of Fe and Al phosphate. The final product diffusion combination with Zn caused the depression of WE Zn fraction(Cao et al. 2013). Liming of previous acid soils by relatively increase pH values and a high content of strongly adsorptive CaCO₃, can retain Zn through chemisorption (Alloway 2009; Kiekens 1995; Singh et al. 2005; Vondráčková et al. 2013). In line with the original calcareous soils,
respectively. Surprisingly, two times increasing in comparison with control at 2.5 and 5 mg kg⁻¹ the Zn levels, around 1.7 to 4, and 2 to 5 times in 

had the lowest value. Organic complexes Zn fraction 

bound zinc was considerably increased by 

sulphate soil which it 

non- 

treated 

the Zn that already present in the exchange 

sites and thus restricts the entry of applied Zn into 

flooded condition, compete with Zn 

formation of Fe³⁺ and Mn²⁺ in soil under the 

flooded condition, compete with Zn²⁺ for exchange 

sites and thus restricts the entry of applied Zn into 

the exchange complexes sites. The increase in 

soluble Fe³⁺ and Mn²⁺ may also displace some of 

the Zn that already present in the exchange complex 

leading to its reaction with P in the 

solution phase with formation of compounds with lower solubility (Mandal and Mandal 1990; Shuman 1985; Shuman 1998).

Organic complexes Zn fraction 

The mean value of Org fraction in selected 

paddy soils and its variations across applied 

treatments was shown in Table 2. The organically 

bound zinc was considerably increased by 

increasing organic carbon content in non-treated 

soils except in Tepus acid sulphate soil which it 

had the lowest value. Org elevated by increasing 

the Zn levels, around 1.7 to 4, and 2 to 5 times in 

comparison with control at 2.5 and 5 mg kg⁻¹, 

respectively. Surprisingly, two times increasing in 

applied Zn levels have not caused two times 

increases in Org concentration, and also, the 

highest increase percentage (4 and 5 times) 

observed at the lowest organic matter (0.68% OC) 

background (Tepus acid sulphate soil series). The 

first might be as a consequence of several soil 

factors involve in Zn chemistry, and the second 

might be due to strongly acidic nature of acid 

sulphate soil (pH=4) compared to others.

Lime application permanently decreased the 

Org concentration in all soils and all levels of 

added Zn treatments, nevertheless, compared to 0 

Zn level, the Org concentration reduction in 2.5 and 

5 mg kg⁻¹ were not significant. Similar to lime, P 

application decreased the Org Zn concentration for 

both 50 and 100 mgkg⁻¹ levels, but two times 

increasing in applied P levels have not caused two 

times reduction in Org concentration. As expected 

from lime and P separate effects, combination of P 

and lime vs Zn levels reduced Org fractions in all 

treated soils, too. Among all treatments, single 

lime had the greatest and significant effect on 

decreases of Org concentration than P, while P plus 

lime effect was not significant. As reported in the 

previous studies, the phosphates could also reduce 

the Zn mobility, likely through surface 

precipitation or co-precipitation of apatite or 
tertiary metal phosphates (Ownby et al. 2005). 

These newly formed precipitates compound have a 

tertia metal phosphates 

precipitation or co 

ry metal phosphates 

involve in Zn chemistry, and the second 

might be due to strongly acidic nature of acid 

sulphate soil (pH=4) compared to others.

Table 2. Concentrations of organic complexes Zn in six tropical soil series in incubation experiment receiving various Zn, P and lime levels

| Treatment | Lubok Itek | Kundur | Tok Yong | Chengai | Tupus | Telemong |
|-----------|-----------|-------|---------|--------|------|---------|
| P(0)      |           |       |         |        |      |         |
| Zn0 L0    | 3.04      | 1.48  | 0.66    | 1.15   | 0.58 | 1.38    |
| Zn0 L1    | 1.89      | 1.24  | 0.64    | 0.86   | 0.41 |         |
| Zn5 L0    | 5.30      | 3.05  | 1.98    | 2.61   | 2.23 | 3.69    |
| Zn5 L1    | 3.98      | 2.83  | 1.84    | 2.41   | 2.04 |         |
| Zn10 L0   | 6.44      | 6.11  | 3.30    | 3.75   | 2.77 | 5.77    |
| Zn10 L1   | 5.61      | 5.38  | 2.91    | 3.50   | 2.55 |         |
| P(50)     |           |       |         |        |      |         |
| Zn0 L0    | 2.46      | 1.52  | 0.76    | 1.10   | 0.52 | 1.27    |
| Zn0 L1    | 2.18      | 1.38  | 0.73    | 0.83   | 0.40 |         |
| Zn5 L0    | 5.50      | 3.77  | 1.94    | 2.56   | 1.69 | 3.38    |
| Zn5 L1    | 4.44      | 3.47  | 1.62    | 2.37   | 1.05 |         |
| Zn10 L0   | 6.50      | 5.78  | 2.88    | 3.60   | 2.68 | 5.66    |
| Zn10 L1   | 5.81      | 5.20  | 2.48    | 2.88   | 2.57 |         |
| P(100)    |           |       |         |        |      |         |
| Zn0 L0    | 2.54      | 1.42  | 0.67    | 1.04   | 0.28 | 1.14    |
| Zn0 L1    | 2.18      | 1.40  | 0.63    | 0.97   | 0.25 |         |
| Zn5 L0    | 4.365     | 3.63  | 1.71    | 2.55   | 1.52 | 3.32    |
| Zn5 L1    | 3.92      | 3.88  | 1.60    | 2.33   | 1.30 |         |
| Zn10 L0   | 5.57      | 6.26  | 2.80    | 3.46   | 2.48 | 4.73    |
| Zn10 L1   | 5.04      | 4.32  | 2.30    | 3.25   | 2.29 |         |
transforming the Org fractions into those bound to oxy-hydroxides, therefore, this Zn fractions are clearly decreased (Zhang et al. 2010). Also, lime is often conductive to stabilizing the Zn phyto-available fractions as well as those bound to ferric oxides, hydroxides and inner sphere surface complex on the soil minerals, and by facilitating of the dissolution of more soil organic matter (Laor et al. 1998; Wang et al. 2001; Zhang et al. 2010). Therefore, the high pH condition facilitates the dissolution of soil organic matter, and under this condition, more Zn to be adsorbed or co-precipitated with Fe oxy-hydroxides (Miretzky and Fernandez-Cirelli 2008).

Amorphous sesquioxide bound Zn fractions

Concentrations of Amorphous sesquioxide Zn in six tropical soil series in incubation experiment receiving various Zn, P and lime levels and the treatment interactions listed in Tables 3 and 7, respectively. Similar to the WE and Org fractions, Amor Zn concentration, increased more in the Zn treated environments in much larger absolute value. As shown in Table 2, in contrast with the two more previous soluble fractions, the concentration values increased significantly by increasing the added Zn from 2.5 to 5 mg kg⁻¹ and it is more linear than the former ones. The soil pH seemed to be more effective in the elevation of this fraction, because the highest increase was observed in Tepus soil series with the lowest pH (six times) compared to the other neutral or near to neutral soils (two times).

As illustrated in Table 3, although added P increased the Amor Zn fractions concentration and the increment also positively related to the P levels, but was not significant (Tables 9 and 10). It can be explained by Zn adsorption with hydrous oxides due to low or moderate enrichment of the oxides with phosphate (Benjamin and Bloom 1981; Mandal and Mandal 1990). Phosphorus sorption can increase the surface negative charge or potential (or reduced surface positive charge), or precipitation a new adsorbing plane of Fe phosphate on the surface of the oxides. Sorption process was more active under flooded compare to non-flooded moisture regimes through changing of the crystalline forms of sesquioxides to the amorphous sesquioxides (Hazara et al. 1987), and by increasing of crystalline oxide surface area could facilitated greater P enhanced Zn adsorption (Logeragan et al. 1982).

Similar to P, the added lime also increases the Amor fractions in all soils and the greater increase belongs to Tepus acid sulphate soil. Zhang et al. (2010) indicate that by using lime as an amendment material in soils Zn bound to Fe/ Mn oxides of low crystallinity fraction concentration increased around 2 times. The results of Tables 9 and 10 illustrated the combined effect of phosphate and lime on Amor Zn fraction was not significant. The findings were in line with Nascimento et al. (2007) that showed addition of lime significantly increased this fraction in Ultisols.

Manganese oxide Zn faction

As listed in Table 4 and similar with pervious fractions, the Mn oxide fraction increased by increasing applied Zn levels. In contrary to 2.5 mg

Table 3. Concentrations of amorphous sesquioxide Zn in six tropical soil series in incubation experiment receiving various Zn, P and lime levels

| Treatments | Tropical Soil Orders and Series | Entisols | Ultisols | Entisols |
|------------|---------------------------------|----------|----------|----------|
|            |                                 | Lubok Itek | Kundur | Tok Yong | Chengai | Tupus | Telemong |
| P(0)       |                                 | mg Zn kg⁻¹ | soil   |         |         |       |         |
| Zn0 L0     |                                 | 2.53 | 1.67 | 6.28 | 2.20 | 0.88 | 1.9 |         |
| Zn0 L1     |                                 | 2.63 | 1.62 | 2.10 | 2.17 | 2.70 |         |         |
| Zn5 L0     |                                 | 4.55 | 3.90 | 2.23 | 4.30 | 2.39 |         |         |
| Zn5 L1     |                                 | 4.24 | 3.53 | 4.63 | 4.05 | 3.85 |         |         |
| Zn10 L0    |                                 | 6.11 | 5.92 | 4.48 | 6.880 | 3.74 |         |         |
| Zn10 L1    |                                 | 5.62 | 5.64 | 7.13 | 6.32 | 6.20 |         |         |
| Zn0 L0     |                                 | 2.64 | 1.75 | 6.55 | 2.08 | 1.04 | 2.05 |         |
| Zn0 L1     |                                 | 2.58 | 1.61 | 1.90 | 2.03 | 1.34 |         |         |
| Zn5 L0     |                                 | 4.72 | 4.06 | 2.13 | 4.59 | 2.22 |         |         |
| Zn5 L1     |                                 | 4.29 | 3.12 | 4.56 | 3.50 | 3.99 |         |         |
| Zn10 L0    |                                 | 6.04 | 6.31 | 4.77 | 6.27 | 4.01 |         |         |
| Zn10 L1    |                                 | 5.66 | 5.67 | 6.74 | 6.13 | 6.70 |         |         |
| P(50)      |                                 | mg Zn kg⁻¹ | soil   |         |         |       |         |
| Zn0 L0     |                                 | 2.70 | 1.84 | 7.17 | 2.36 | 1.07 | 2.55 |         |
| Zn0 L1     |                                 | 2.57 | 1.68 | 2.34 | 2.30 | 1.03 |         |         |
| Zn5 L0     |                                 | 4.90 | 4.25 | 4.63 | 3.65 | 2.44 |         |         |
| Zn5 L1     |                                 | 4.31 | 3.88 | 4.64 | 4.61 | 5.11 |         |         |
| Zn10 L0    |                                 | 6.62 | 6.63 | 5.39 | 6.13 | 3.99 |         | 6.77    |
| Zn10 L1    |                                 | 6.00 | 5.64 | 7.18 | 5.60 | 6.64 |         |         |
| P(100)     |                                 | mg Zn kg⁻¹ | soil   |         |         |       |         |
| Zn0 L0     |                                 | 2.70 | 1.84 | 7.17 | 2.36 | 1.07 | 2.55 |         |
| Zn0 L1     |                                 | 2.57 | 1.68 | 2.34 | 2.30 | 1.03 |         |         |
| Zn5 L0     |                                 | 4.90 | 4.25 | 4.63 | 3.65 | 2.44 |         |         |
| Zn5 L1     |                                 | 4.31 | 3.88 | 4.64 | 4.61 | 5.11 |         |         |
| Zn10 L0    |                                 | 6.62 | 6.63 | 5.39 | 6.13 | 3.99 |         | 6.77    |
| Zn10 L1    |                                 | 6.00 | 5.64 | 7.18 | 5.60 | 6.64 |         |         |
Zn kg⁻¹, the 5 mg kg⁻¹ Zn treatment showed higher increases in the overall concentration of MN, although, two times increasing in applied Zn levels have not caused two times increases in its concentration. The slightly acidic soils (pH 5-5.5)-Lubok Itek, Kundur and Tok Yong soil series - showed the lesser increase in MN fraction concentration under Zn levels in comparison with the Tepus acid sulphate soil series (pH 3.8) due to lower acidic condition of former soil. A sequential extraction procedure findings in major soils (Entisols order) of Indian paddy fields indicated that all applied Zn treatments increased the Mn oxide Zn fraction in all soils studied (Adhikari and Rattan 2007). Almost all of treatments, separately or in combination, significantly increased Mn concentration. The slightly acidic soils (pH 5-5.5) Lubok Itek, Kunbur and Tok Yong soil series and neutral Telemong soil series showed the highest increase in Cry fraction (around 2 times) under either Zn level application or P and lime treatments. As noted before, the increase in Cry-Zn content was related to the greater adsorption of Zn by the higher negative charges developed on the surfaces of Fe and Al oxides as a result of P adsorption (Mandal and Mandal 1990).

**Residual Zinc fraction**

As illustrated in Zn fractions and distribution of the original soils (Non-treated soils) section, around 54 to 82% portion of Zn belonged to residual forms. Furthermore, as presented in WE and Org fractions sections, their concentrations decreased due to P and lime application. Thus, the redistribution of decreased fractions to Res pool and elevation of Res was acceptable. In comparison to the control treatment, lime application as well as P application (singly and in combination) increases the concentration of Res, but the rising rate was not significant at any treatments (Tables 6, and 7). Also, applied Zn treatments had significant effect on Res content at 5% confidence level. Thus, the more added Zn was retained very less in residual fraction (Tables 1 to 8). These findings were in line with the pervious works that suggested the short term Zn application to soils increased more proportions of Zn in the most available forms (We and Org) in comparison with residual and non-residual non- mobile Zn fraction (Ma and Uren 1997; Nascimento et al. 2007; Nyamangara and Mzezewa 1999). It can be concluded that, incubation time or duration after application are an important factors on the Zn availability in the soil and, longer reaction times

| Treatments | Tropical Soil Orders and Series | Entisols | Ultisols | Entisols |
|------------|---------------------------------|----------|----------|----------|
|            | Tropical Soil Orders and Series | Lubok Itek | Kundur | Tok Yong | Chengai | Tepus | Telemong |
| P(0)       | Zn0 L0  | 0.94 | 0.42 | 1.19 | 0.95 | 0.24 | 0.38 |
| Zn0 L1  | 1.42 | 0.71 | 0.41 | 1.02 | 0.80 |
| Zn5 L0  | 1.40 | 1.14 | 1.57 | 1.35 | 0.72 | 1.09 |
| Zn5 L1  | 1.42 | 1.17 | 0.75 | 1.49 | 1.15 |
| Zn10 L0 | 2.78 | 1.04 | 1.35 | 2.04 | 1.49 | 1.03 |
| Zn10 L1 | 2.63 | 1.73 | 1.42 | 1.69 | 1.62 |
| P(50)     | Zn0 L0 | 1.39 | 0.67 | 0.53 | 1.53 | 1.03 | 0.48 |
| Zn0 L1 | 1.17 | 0.37 | 0.62 | 0.48 | 0.80 |
| Zn5 L0 | 1.82 | 1.24 | 1.03 | 1.71 | 0.43 | 1.18 |
| Zn5 L1 | 1.56 | 1.56 | 1.41 | 1.90 | 1.45 |
| Zn10 L0 | 1.96 | 1.65 | 1.92 | 2.07 | 1.14 | 1.35 |
| Zn10 L1 | 2.50 | 1.51 | 1.28 | 2.23 | 1.54 |
| P(100)    | Zn0 L0 | 1.06 | 0.71 | 0.59 | 0.85 | 0.57 | 0.44 |
| Zn0 L1 | 1.50 | 1.07 | 0.75 | 1.75 | 0.55 |
| Zn5 L0 | 1.70 | 0.99 | 1.63 | 1.71 | 0.81 | 1.34 |
| Zn5 L1 | 1.84 | 0.99 | 0.90 | 1.50 | 0.93 |
| Zn10 L0 | 2.33 | 1.20 | 1.63 | 1.65 | 0.48 | 1.09 |
| Zn10 L1 | 2.15 | 1.18 | 1.11 | 1.56 | 1.79 |
Table 5. Concentrations of crystalline sesquioxide Zn in six tropical soil series in incubation experiment receiving various Zn, P and lime levels

| Treatments | **Entisols** | **Ultisols** | **Entisols** |
|------------|--------------|--------------|--------------|
| Zn0 L0     | 3.08         | 2.58         | 4.21         | 4.66         | 1.10         | 0.97         |
| Zn0 L1     | 3.53         | 3.86         | 4.65         | 5.38         | 2.52         |
| Zn5 L0     | 3.61         | 3.90         | 4.73         | 5.83         | 2.517        | 2.02         |
| Zn5 L1     | 3.89         | 4.49         | 5.38         | 6.49         | 3.227        |
| Zn10 L0    | 4.90         | 5.09         | 5.67         | 6.18         | 3.443        |
| Zn10 L1    | 5.22         | 5.51         | 6.00         | 6.50         | 4.387        | 3.57         |
| Zn0 L0     | 3.28         | 3.08         | 5.05         | 5.250        | 3.21         | 1.65         |
| Zn0 L1     | 4.06         | 3.49         | 5.15         | 5.75         | 3.50         |
| Zn5 L0     | 4.08         | 4.29         | 5.25         | 6.58         | 3.81         | 3.68         |
| Zn5 L1     | 4.49         | 5.51         | 5.55         | 7.13         | 3.88         |
| Zn10 L0    | 5.38         | 5.63         | 6.14         | 6.58         | 4.34         | 4.97         |
| Zn10 L1    | 5.69         | 6.00         | 6.46         | 7.83         | 6.16         |
| Zn0 L0     | 4.08         | 3.70         | 6.14         | 6.51         | 3.48         | 2.38         |
| Zn0 L1     | 4.53         | 4.28         | 6.46         | 7.68         | 4.27         |
| Zn5 L0     | 4.67         | 5.07         | 6.64         | 8.47         | 5.74         | 4.89         |
| Zn5 L1     | 5.37         | 6.36         | 6.93         | 8.82         | 6.40         |
| Zn10 L0    | 8.10         | 5.92         | 7.00         | 7.43         | 5.66         | 5.43         |
| Zn10 L1    | 10.62        | 6.48         | 8.03         | 9.67         | 6.99         |

Table 6. Concentrations of residual Zn in six tropical soil series in incubation experiment receiving various Zn, P and lime levels

| Treatments | **Entisols** | **Ultisols** | **Entisols** |
|------------|--------------|--------------|--------------|
| **Zn**     | **Lubok Itek** | **Kundur** | **Tok Yong** | **Chengai** | **Tupus** | **Telemong** |
| P(0)       | 31.30        | 10.70        | 38.70        | 9.03        | 13.77     | 5.17         |
| Zn0 L0     | 31.63        | 11.70        | 40.43        | 14.23       | 12.33     |
| Zn5 L0     | 31.43        | 11.90        | 39.50        | 17.77       | 6.83      | 5.47         |
| Zn5 L1     | 32.63        | 12.40        | 36.60        | 17.97       | 10.40     |
| Zn10 L0    | 31.90        | 12.00        | 39.70        | 19.60       | 7.73      | 6.34         |
| Zn10 L1    | 31.00        | 13.90        | 41.73        | 14.27       | 7.57      |
| P(50)      |              |              |              |             |           |              |
| Zn0 L0     | 28.43        | 11.20        | 39.83        | 16.43       | 13.03     | 6.40         |
| Zn0 L1     | 32.47        | 12.50        | 37.93        | 15.13       | 10.77     |
| Zn5 L0     | 30.27        | 13.20        | 42.93        | 16.67       | 23.73     | 6.10         |
| Zn5 L1     | 31.97        | 13.80        | 45.97        | 14.13       | 8.73      |
| Zn10 L0    | 29.80        | 14.30        | 40.80        | 16.80       | 11.87     | 6.60         |
| Zn10 L1    | 34.20        | 15.00        | 45.27        | 17.03       | 7.57      |
| P(100)     |              |              |              |             |           |              |
| Zn0 L0     | 26.47        | 12.60        | 41.17        | 18.53       | 8.97      | 5.50         |
| Zn0 L1     | 31.97        | 13.30        | 45.20        | 16.80       | 13.10     |
| Zn5 L0     | 28.87        | 13.50        | 42.17        | 16.60       | 11.80     | 5.13         |
| Zn5 L1     | 31.37        | 13.90        | 45.07        | 19.67       | 12.30     |
| Zn10 L0    | 30.37        | 14.40        | 42.43        | 20.67       | 12.77     | 7.67         |
| Zn10 L1    | 31.20        | 15.34        | 43.73        | 22.03       | 12.53     |

transformed more Zn into stable fractions (Barrow 1993; Nascimento et al. 2007).

Total and Summation (SUMF) of Zinc fractions

As listed in Tables 7, these fractions showed the same reactions aservous fractions for various amounts of added Zn. They increased 4-6% for 2.5 mg kg$^{-1}$ and 8-14% for 5 mg kg$^{-1}$ added Zn treatment, averagely. The total Zn concentration increased through Zn application in submerged soils and elevation amount is close to but is not equal to the summation of untreated and applied Zn concentration at 30 days data collection, while by increasing the incubation time makes it more close to summation (Wijebandara 2007). Phosphorus and lime treatments showed different effects on total and SUMF; when the total Zn significantly (p≤0.01) (Table 7) affected by P treatments, the SUMF did not affect significantly. Also, the combination of treatments showed no significant effects in almost of the condition.
Table 7. Summary of analysis of variances of zinc fractions in 6 soils series

| Sources          | df | WE       | ORG       | AMOR      | MN        | CRY       | RES       | SUMF      | Total      |
|------------------|----|----------|-----------|-----------|-----------|-----------|-----------|-----------|------------|
| Combination (Com)| 10 | 1.19**   | 32.27***  | 11.08***  | 3.39**    | 4.80**    | 5134.92   | 6016.83** | 6393.93**  |
| P                | 2  | 1.37**   | 2.209**   | 3.60**    | 0.32**    | 5.05**    | 46.86*    | 252.91**  | 133.44**   |
| Zn               | 2  | 5.02**   | 253.67**  | 334.98**  | 17.72**   | 22.10**   | 56.12**   | 3470.19** | 4011.60**  |
| Com*P            | 20 | 0.09**   | 0.28**NS  | 0.87**    | 0.10**    | 0.08**    | 33.27**   | 38.58**   | 36.09**    |
| Com*Zn           | 20 | 0.30**   | 2.30**    | 10.04**   | 0.25**    | 0.92**    | 16.23**NS | 41.87**   | 42.66**    |
| P*Zn             | 4  | 0.02**   | 0.57**NS  | 0.39**NS  | 0.41**    | 0.75**    | 3.03**NS  | 4.72**NS  | 5.88**NS   |
| Com*P*Zn         | 40 | 0.02**   | 0.22**NS  | 0.45**    | 0.31**    | 0.06**    | 15.61**   | 17.02**   | 33.17**    |

Note: Com=comination of soil and lime levels as an one level in ANOVA analysis

CONCLUSIONS

Overall, not only the phyto- available fractions (WE and Org) but also the concentration of the non-residual and non-available fractions such as, Amor, MN and the residual and residual fraction greatly increased by applying Zn sulphate in all soils studied. Moreover, the phyto- available fractions decreased with increasing the P levels and lime application, whereas, the non-residual and non-available fractions increased with the increase of applied P and lime treatments and this trend accelerated by increase of Zn levels. The changes in Zn concentrations vs P and lime levels was similar in all soils, further supporting that the effect of P and lime was responsible for Zn immobilization in all soils. In addition, sequential fractionation of Zn vs Zn, P and lime application showed that a substantial portion of the applied Zn had reverted to plant-unavailable fractions. Thus, our study helped to confirm the knowledge concerning the controlling mechanisms of Zn availability under high rates of P and lime application in agricultural fields. It also showed that Org fractions play the most important role in adding Zn to the plant-available pool. However, the attention should be paid to the relation of the results of this study with plant morphology, growth stages and Zn uptake in future works.

ACKNOWLEDGMENTS

This research was financially supported by Long-term Research Grant Scheme (LRGS 5525001), Food Security Project, under the Ministry of Higher Education Malaysia (MOHE) and to Universiti Putra Malaysia for providing laboratory and other research facilities.

CONFLICT OF INTEREST

The authors declare that there are no conflicts of interest regarding the publication of this manuscript.

ORCID

Shahram Mahmoud Soltani: https://orcid.org/0000-0002-1300-9536

Mohamed Musa Hanafi: https://orcid.org/ 0000-0003-1898-6819

Abdul Wahid Samsuri: https://orcid.org/ 0000-0002-5450-0604

Sharifah Kharidah: https://orcid.org/ 0000-0002-9908-1503

REFERENCES

Adhikari T, Rattan R.K. (2007) Distribution of Zn fractions in some major soils of India and impact on nutrition of rice. Communication in soil science and plant analysis, 38:2779–2798.

Alloway B.J. (2009) Soil factors associated with zinc deficiency in crops and humans. Environmental geochemistry and health, 31(5):537-48.

Barrow N.J. (1993) Mechanisms of reaction of zinc with soil and soil components. In Zinc in Soils and Plants; Robson, A.D. (ed.) Kluwer Academic Publishers, Dordrecht, Netherlands.

Cao X, Liang Y, Zhao L, et al. (2009) Immobilization of Zinc Fertilizer in Flooded Soils Monitored by Adapted DTPA Soil Test. J. (2009) Soil factors associated with zinc deficiency in crops and humans. Environmental geochemistry and health, 31(5):537-48.

Dobbermann A, Fairhurst T. H. (2000) Nutrient disorders and nutrient management Potash and Phosphate Institute of Canada and International Rice Research Institute, Singapore.

Fageria N.K., Stone L.F. (2008) Micronutrient deficiency problems in South America. In Micronutrient deficiencies in global crop production, ed. B. J. Alloway, 247-268 Springer, New York.

Hazara G.C., Mandal B., Mandal L.N. (1987) Distribution of zinc fractions and their transformation in submerged rice soils. Plant and Soil 104:175-181.

Johnson-Beebout S.E, Sarah E, Lauren J, Duxbury J. (2009) Immobilization of Zinc Fertilizer in Flooded Soils Monitored by Adapted DTPA Soil Test. Communication in soil science and plant analysis, 40:1842-1861.

Kiekens L. (1995) Zink. In: Alloway, B.J. (Ed.), Heavy Metals in Soils, 2nd ed. Blackie Academic & Professional, Glasgow, UK.

Laor Y, Farmer W.J, Aochi Y, Strom P.F. (1998) Phenanthrene binding and adsorption to
dissolved and to mineral-associated humic acid. Water Research, 32(6):1923-1931.

Logeragan J.K., Grunes D.L., Welch R.M. et al. (1982) Phosphorus accumulation and toxicity in leaves in relation to zinc supply. Soil Science Society of America Journal, 46:345-352.

Ma Y.B., Uren N.C. (1997) The effects of temperature, time, and cycles of drying and rewetting on the extractability of zinc added to a calcareous soil. Geoderma, 75:89-97.

Mandal B., Mandal L.N. (1990) Effect of phosphorus application on transformation of zinc fractions in soil and on the zinc nutrition of low land rice. Plant Soil, 121:115-123.

Miretzky P., Fernandez-Cirelli A. (2008) Phosphates for Pb immobilization in soils: a review. Environmental Chemistry Letters, 6:121-133.

Masciamento C.W.A.D., de Melo E.E.C., Nascimento R.S.D.M.P., Leite P.V.V. (2007) Effect of Liming on the Plant Availability and Distribution of Zinc and Copper among Soil Fractions. Communications in Soil Science and Plant Analysis, 38:545-560.

Nyangarara J., Mzezewa J. (1999) The effect of long-term sewage sludge application on Zn, Cu, Ni and Pb levels in a clay loam soil under pasture grass in Zimbabwe. Agriculture, Ecosystems & Environment, 73:199-204.

Ownby D.R., Galvan K.A., Lydy M.J. (2005) Lead and zinc bioavailability to Eisenia fetida after phosphorus amendment to repository soils. Environmental Pollution, 136:315-321.

Rupa T.R., Srinivasa Rao C., Subba Rao A., Singh M. (2003) Effects of farmyard manure and phosphorus on zinc transformations and phyto-availability in two alfisols of India. Bioresource Technology, 87:279-288.

Shuman L.M. (1985) Fractionation method for soil micronutrients. Soil Sci, 140:11-22.

Shuman L.M. (1998) Micronutrient fertilizers. Journal of Crop Production, 2:165-195.

Singh B., Natesan S.K.A., Singh B.K., Usha K. (2005) Improving zinc efficiency of cereals under zinc deficiency. Curr Science, 88(36-44).

Vondračková S., Hejcman M., Tlustos P., Szakova J. (2013) Effect of quick lime and dolomite application on mobility of elements (Cd, Pb, As, Fe, and Mn) in contaminated soils. Polish Journal of Environmental Studies, 22(2):577-589.

Wang Y., Chen T.C., Yeh K.J., Shue M.F. (2001) Stabilization of an elevated heavy metal contaminated site. Journal of Hazardous Materials, 88(1):63-74.

Wijebandara D.M.D.I. (2007) Studies on distribution and transformation of soil zinc and response of rice to nutrients in traditional and system of rice intensification (Sri) methods of cultivation. DHARWAD University of agricultural sciences.

Zhang W.H., Huang H., Tan F.F., Wang H., Qiu R.L. (2010) Influence of EDTA-washing on the species and mobility of heavy metals residual in soils. Journal of Hazardous Materials, 173(1-3):369-376.