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Evaluation of lead toxicity in maize (*Zea mays* L.) as influenced by organic and inorganic sources of nitrogen

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Abstract

Number of human health risks and environmental degradation is associated with lead toxicity. It is imperative to examine and underpin sound solutions. There fore, present study was undertaken at Agronomic Research Area, University of Agriculture, Faisalabad with aim to investigate the effect of lead toxicity in maize (*Zea mays* L.) as influenced by organic and inorganic sources of nitrogen. Experiment was laid out in randomized complete block design (RCBD) with factorial arrangement having three replications and two factors i.e. nitrogen sources (control, 100 % N from urea, 100 % N from poultry manure, 75 % N from urea and 25 % N from poultry manure and 50 % N from urea and 50 % N from poultry manure) and lead management (0 kg Pb ha^{-1} and 100 kg Pb ha^{-1}) with a plot size of 3 m × 6 m. Data on yield and quality parameters were recorded by following the standard procedures. Results showed that maximum cob length (19.78 cm), cob diameter (3.87 cm), no. of grains per cob (486.67), biological yield (18.17 tons ha^{-1}), grain yield (6.51 tons ha^{-1}) and seed protein content (8.81 %) were recorded in N_3L_0 (75 % N from urea and 25% N from poultry manure with 0 kg Pb ha^{-1}), while minimum results were recorded in N_0L_1 (0 kg N with 100 kg Pb ha^{-1}). Highest seed oil contents (4.3 %) were found in N_0 (0 kg N ha^{-1}). It is concluded that nitrogen application through combined use of organic and inorganic sources significantly increased the yield and also in the presence of organic manure toxicity of lead greatly reduced.

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Introduction

Maize (*Zea mays* L.) is a staple food in many countries and it is important cereal crop belongs to Poaceae family. It is major source of income for poor farmers in developing countries (Tagne *et al.,* 2008). It is a versatile crop that is used as food for human being and feed for animals (poultry and livestock). Nutritional value of its grain is very high that's why it is used as raw material for industries (Afzal *et al.,* 2009). It is short duration crop, utilizes inputs more efficiently and capable of producing more yield per unit area. It is cultivated as spring and autumn crop twice in a year. It is more nutritious and contains about 10% proteins, 4.8% oil, 72% starch, 8.5% fiber, 1.7% ash and 3% sugar (Chaudhary, 1993).

Environmental pollution has been increasing due to abundant use of heavy metals mainly in industries and agriculture (Sinhal *et al.,* 2010). Waste water releasing from different industries and its use for irrigation is a major source of heavy metal in Pakistani soils. This water is very harmful because it contains various heavy metals, microorganisms, different organic compounds and has high electric conductivity. The permanent use of this water results in building up of heavy metals in soil along with other soil problems (Hussain, 2000). Lead is comparatively static and not available to plants. It is unnecessary element and noxious to living organisms even in small quantity (Walker *et al.,* 2006).

Pb toxicity causes decrease in germination percentage, length of root and shoot and also dry weight (Munzuroglu and Geckil, 2002). Disturbance in mineral nutrition of plants (Paivoke, 2002) and decreased cell division is mainly due to Pb toxicity (Eun *et al.,* 2000). Heavy metal treatment at higher concentration produces toxic impact on germination and seedling growth and brought up changes in most of the growth parameters of Maize (Valasang *et al.,* 2012). High levels of Pb in soil resulted in decreased biomass of maize (Singh *et al.,* 2010). Severe Pb toxicity resulted in impaired growth with small number of leaves which are more brittle having dark purple coloration (Gupta *et al.,* 2010).

Nitrogen (N) is a macronutrient which is required by plants for better growth and it has vital role in crop yield (Arshad, 2003). It plays a vital role in increasing crop yields (Massignam *et al.,* 2009) and also improving the quality of crops (Ullah *et al.,* 2010). The use of inorganic fertilizer is the most efficient and suitable method to recover the soil fertility and to maintain the soil health (Akande, 2005). The combined use of organic manures and chemical fertilizers resulted in better growth, yield and quality parameter of maize (Boateng *et al.,* 2006). Addition of nitrogen in the soil by combining organic manures and chemical fertilizers can be helpful to remove lead from the soil. The ammonium ion causes the desorption of heavy metal from soil through replacing ions (Lorenz *et al.,*1994) and manure application improves the soil fertility and increases plant resistance to heavy metal toxicity (Adejumo *et al.,* 2010).Manures were helpful in reducing lead accessibility to plants and thus lower its uptake (Ye *et al.,* 1999). Growth and yield of maize was significantly higher and accumulation of lead lower under compost application than control and inorganic fertilizer (Adejumo *et al.,* 2010) because compost supply required nutrient to soil (Sadovnikova, 2002).

From the above study it is clear that combined use of organic and inorganic manures not only improves the soil health and yield of maize crop but also help to minimize the lead toxicity in maize by reducing the uptake of metal. Keeping in view the above mentioned facts the proposed study is therefore planned with the objective is to evaluate different integrated nitrogen management approaches on lead toxic sites for enhancing maize productivity.

Materials and methods

Experimental Site

The proposed study was conducted at Agronomic Research Area; University of Agriculture Faisalabad during the year 2015.

Soil Analysis

A composite soil sample to a depth of 30 cm was obtained from the experimental area with soil Auger before sowing and after harvesting of crop. The sample was analyzed for its physio-chemical properties as presented in Table 1.

Chemical analysis of poultry manure

Poultry manure was analyzed for its various chemical properties (NPK %) in laboratory of Animal Nutrition, University of Agriculture Faisalabad (Table 2). After grinding the sample, poultry manure was dried to a constant weight in an oven and then stored in labeled plastic bottles and then analyzed for its NPK %.

Experimental design and Treatments

The experiment was laid out using RBCD with factorial arrangement with three replications. The plot size was 6 m x 3m.

Treatments

This experiment comprised of two factors i.e. nitrogen sources (control, 100% N from urea, 100% N from poultry manure, 75% N from urea and 25% N from poultry manure and 50% N from urea and 50% N from poultry manure) and lead management (0 kg Pb ha^{-1} and 100 kg Pb ha^{-1}).

Crop husbandry

Seed bed preparation

The field was given two fallow cultivations, thereafter; pre-soaking (rauni) irrigation of 10 cm depth was applied. At "wattar" conditions (workable moisture level), the field was tilled twice with a cultivator each followed by planking.

Sowing of crop

This crop was sown on 3rd March, 2015. Sowing was done with the help of hand drill at 75 cm spaced rows using seed rate of 25 kg ha^{-1}.

Fertilization

Fertilizers applied at the rate of 250, 125 and 125 kg ha^{-1} of nitrogen, potassium and phosphorus, respectively. Lead was applied at the rate of 100 kg ha^{-1}. Lead sulphate was used as a source of lead and applied at the time of sowing.

Thinning

Thinning was done at 3-4 leaf stage to maintain plant to plant distance of 25 cm and plant population density.

Plant protection measures

Crop was kept weed free by hoeing twice to avoid weed-crop competition. Insect pest were kept under the threshold level through chemical control. Two doses of Carbofuran granules at the rate of 18 kg ha^{-1} were applied to control the stem borers.

Irrigation

In addition to rain fall received during the growing period of crop, a total of 6 irrigations were applied as and when needed at different plant growth stages till the physiological maturity of the crop. First irrigation was applied 20 days after sowing and subsequent irrigations were applied with fifteen days interval till flowering and from flowering to seed formation irrigation were applied with seven days interval due to high water requirement of spring maize crop.

Crop harvest

The crop was harvested manually after its maturity in last week of June. The crop plants were tied into piles and kept for sun drying.

Shelling

The cobs were removed and were kept for sun drying to minimize moisture content of grain and easy shelling. After seven days of sun drying, the cobs were shelled with the help of maize sheller to separate grains from pith.

Procedure for recording observations

A. *Agronomic parameters*

Cob length (cm)

The length of ten randomly selected cobs from each plot was determined with the help of "a measuring tape" and then average of cob length was recorded.

Cob diameter (cm)

The diameter of ten randomly selected cobs from each plot was determined with the help of vernier caliper and then average diameter was recorded.

Number of grains per cob

Ten cobs were selected at random from each plot, number of grains per cob was counted individually and then average was calculated.

Biological yield (tons ha^{-1})

When the crop was fully mature, it was harvested and sun dried plot wise.

The stalks along with cobs were weighed with the help of spring balance to calculate biological yield per plot. The biological yield per plot was recorded and then converted into t ha^{-1}.

Grain yield (tons ha^{-1})

All the cobs in each sub plot were sun dried, shelled and weighed. The grain yield was weighed in kg from a plot size of 18 m^2 and then converted into t ha^{-1}.

B. Quality parameters

Seed oil content (%)

Oil contents were determined by Soxhlet Fat Extraction method (A.O.A.C., 1990).Percent oil content was calculated using the following equation;

$$Oil\ Content\ (\%) = \frac{Weight\ of\ flask + oil - Weight\ of\ flask}{Weight\ of\ flask + seed - Weight\ of\ flask} X100$$

Seed protein content (%)

Protein in seed was determined according to Kjeldhal method (Bremner, 1964). Percent crude protein was calculated using the formula;

$$(Crude\ Protein\%) = \frac{(V1 - V2)N}{100\,W} X\ 14 X\ 6.25 X\ 100$$

Statistical analysis

The data collected was analyzed by using standard procedure of Fisher's analysis of variance technique. Individual comparison of treatment means were made by using least significant difference test (LSD) at 5% probability level (Steel *et al.*, 1997).

Results and discussion

Cob length (cm)

Both the factors (nitrogen sources and lead) and their interaction (N × Pb) significantly affected the cob length as shown in table 3.

Combination of Organic and inorganic sources of nitrogen with 0 kg Pb ha^{-1} resulted in maximum cob length mainly due to least N loses and more availability of nutrients during the crop growth period which were essential for proper cob development inmaize. Under the optimum supply of nutrients maize plant has the ability to express its potential effectively and produce more cob length.

Table 1. Soil physio-chemical analysis of soil.

Chemical Analysis		
	Before sowing	After Harvest
pH	8.2	7.8
EC (dSm^{-1})	1.94	3.49
Nitrogen (%)	0.032	0.048
Available Phosphorus (ppm)	19.4	8.5
Potassium (ppm)	190	160
Organic Matter (%)	0.98	1.26
Lead (mg kg^{-1})	4.4	
Mechanical analysis		
Textural class	Loam	Loam

Soil analysis for lead after harvesting of crop.

Treatments	Pb mg kg^{-1}
N_0 (Control) + 100 kg Pb ha^{-1}	6
N_1 (100 % N from urea) + 100 kg Pb ha^{-1}	8
N_2 (100 % N from PM) + 100 kg Pb ha^{-1}	10.14
N_3 (75 % N from urea &25 %Nfrom PM) + 100 kg Pb ha^{-1}	14.23
N_4 (50 % N from urea &50% N from PM) + 100 kg Pb ha^{-1}	13.47

The minimum results obtained due to the unavailability of nutrients under the application of heavy metals which are required for the growth and development of plants.

These results are in accordance with Sims *et al.* (1995) and Ayoola and Adeniyan (2006) who reported that cob length increased by combining organic and inorganic sources.

Table 2. Chemical analysis of poultry manure.

Nutrients	Unit	Value
N	%	2.63
P	%	1.75
K	%	1.25

Cob diameter (cm)

It can be affirmed that both the individual factors (nitrogen sources and lead) and their interaction significantly affected the parameter under study (Cob diameter) as shown in table 3.

The maximum cob diameter might be due to high and continued nutrient supply potential of organic manure in addition to supply of nitrogen from inorganic fertilizer. Increased availability of nutrient and uptake at combination of organic and inorganic N might have increased photosynthetic rate and net assimilation rate, which resulted in more cob yield. Minimum results obtained due to reduced photosynthetic activity under the application of lead which lowers the cob yield. Boateng *et al.* (2006) reported the same results that organic and inorganic sources have significant effect on cob diameter. Eun *et al.* (2000) also found similar results that lead application significantly affected the cob diameter.

Number of grains per cob

Nitrogen sources, lead and their interaction (N × Pb) significantly affected the number of grains per cob as shown in table 3. Maximum number of grains per cob might be due to availability of N at proper time, which is required for better growth and development of plants and increase improvement in moisture retention and soil structure by poultry manure. The minimum no. of grains that was found in N_0L_1 might be due to unavailability of nutrients required for optimum growth and yield and due to the inhibition of silking in maize under lead toxicity which results in less number of grains per cob.

These results are in agreement with Kumar *et al.* (2002) who concluded that combined use of organic and inorganic sources had significant effect on number of grains per cob. Liu *et al.* (2008) also reported that number of grains per cob significantly affected by lead.

Biological yield (tons ha^{-1})

The result regarding the effect of lead management under organic and inorganic sources of nitrogen on biological yield showed that nitrogen sources, lead and their interaction had significant effect on biological yield as shown in table 3. The Maximum biological yield through combined use of urea and poultry manure might be due to better supply of nutrients and their uptake which increase the plant height and over all vegetative growth. While the decrease in biological yield might be attributed to deficiency of nutrients and lead toxicity that cause reduction in stomatal frequency, water uptake, gas exchange and photosynthetic rate that results in decreased plant biomass. These results are in agreement with Dilshad *et al.* (2010) who reported that application of integrated plant nutrient significantly increased the various yield parameters that produced higher biological yield. Kasim, (2001) also reported that lead caused the reduction in biological yield of plants.

Grain yield (tons ha^{-1})

Effect of lead management under organic and inorganic sources of nitrogen on grain yield showed that both the factors (nitrogen sources and lead) and their interaction had significant effect on grain yield as shown in table 3.

These results might be due to proper supply of nutrients from both sources during the plant development and also due to greater cob length, cob diameter and better grain development with the same treatment (N_3L_0). Reason for lower yield in plot where no nitrogen and 100 kg Pb ha^{-1} was applied is that lead affected the various physiological functioning of plant. It reduced the photosynthesis due to which fewer grains were produced and ultimately reduction in yield was occurred. Papadakis *et al.,* 2004 reported that heavy metals reduced the yield of crop. Rasool *et al.* (2008) also reported that grain yield and uptake of nutrients significantly increased under integrated use of poultry manure and inorganic fertilizer.

Table 3. Effect of lead management on yield and yield components under organic and inorganic sources of nitrogen.

Treatments	Cob length (cm)	Cob diameter (cm)	No. of grains per cob	Biological yield (tons ha^{-1})	Grains yield (tons ha^{-1})	Seed protein content (%)	Seed oil content (%)
Factor A: Nitrogen sources							
N_0 (Control)	13.547 C	2.90 C	264.50 C	9.915 D	2.40 E	6.98 C	4.3 A
N_1 (100 % N from urea)	17.947 A	3.41 AB	391.50 B	15.72 B	4.41 C	7.94 B	3.80 BC
N_2 (100 % N from PM)	16.930 B	3.31 B	341.83 C	13.57 C	3.57 D	7.27 C	3.93 B
N_3 (75 % N from urea & 25 % N from PM)	18.715 A	3.59 A	451.00 A	17.40 A	5.60 A	8.36 A	3.66 C
N_4 (50 % N from urea & 50 % N from PM)	18.105 A	3.43 AB	428.67AB	16.435 B	5.08 B	8.08 AB	3.79 BC
LSD Value	0.77	0.22	39.22	0.847	0.388	0.305	0.229
Factor B: Lead management							
L_0 (0 kg Pb ha^{-1})	17.837 A	3.45 A	406.33 A	15.65 A	4.87 A	8.09 A	3.92
L_1 (100 kg Pb ha^{-1})	16.261 B	3.20 B	344.67 B	13.56 B	3.55 B	7.37 B	3.88
LSD Value	0.487	0.1405	24.80	0.536	0.245	0.193	NS
Interaction							
N_0L_0	13.72 e	3.10 d	276.67 e	10.34 g	2.57 f	7.09 de	4.34
N_1L_0	18.96 ab	3.64 ab	450 ab	17.34 ab	5.58 b	8.55 a	3.83
N_2L_0	17.98 bc	3.4 bc	344.66 d	15.06 de	3.61 de	7.38 cd	3.91
N_3L_0	19.78 a	3.87 a	486.67 a	18.17 a	6.51 a	8.81 a	3.68
N_4L_0	19.08 a	3.66 ab	473.66 a	17.37 ab	6.11 ab	8.60 a	3.84
N_0L_1	13.37 e	2.7 e	252.33 e	9.48 g	2.24 f	6.88 e	4.3
N_1L_1	16.93 cd	3.19 cd	333 d	14.11 e	3.25 e	7.33 cd	3.78
N_2L_1	15.88 d	3.22 cd	339 d	12.08 f	3.54 de	7.16 cde	3.96
N_3L_1	17.64 c	3.32 cd	383.66 cd	16.63 bc	4.70 c	7.91 b	3.65
N_4L_1	17.13 c	3.20 cd	415.33 bc	15.50 cd	4.06 d	7.57 bc	3.74
LSD: Value	1.089	0.314	55.47	1.198	0.549	0.432	NS

Seed protein content (%)

The results regarding the impact of different sources of nitrogen and lead on seed protein content showed that not only nitrogen sources and lead management but also their interaction significantly affected the seed protein content % as shown in table 3. Highest value of seed protein content (8.81%) was obtained in N_3L_0 (75 % N from urea and 25 % N from poultry manure with 0 kg Pb ha^{-1}) which was statistically at par with N_4L_0 (50 % N from urea & 50 % N from poultry manure with 0 kg Pb ha^{-1}) and N_1L_0 (100 % N from poultry manure with 0 kg Pb ha^{-1}) by keeping protein content of 8.60 % and 8.55 % respectively and minimum seed protein content (6.88 %) was observed in N_0L_1 (0 kg N ha^{-1} with 100 kg Pb ha^{-1}).

It has also been reported that by increasing N supply through integration of organic and inorganic sources protein content in seed increased.

While the lowest protein content in N_0L_1 might be due to unavailability of nitrogen and also the strong affinities of heavy metal ions for side chain ligands of protein indicated enzyme. Lead combine with protein forming enzymes and reduced its production. Ghani (2010) reported that under heavy metal toxicity protein content of seed greatly reduced. Our results that nitrogen supplies in proper amount increased the protein content of seed are similar to the findings of Uribelarra *et al.* (2004) and Khan *et al.* (2008).

Seed oil content (%)

The data related to seed oil content showed that nitrogen sources had significant effect on seed oil content. While interactions of nitrogen sources and lead management had non-significant effect on seed oil contents as shown in table 3.

Maximum seed oil content (4.3 %) was found in N_0 (0 kg N ha^{-1}). Minimum seed oil content (3.66 %) was observed in N_3 where 75 % N from urea and 25% N from poultry manure was applied. These results might be due to better supply of nitrogen from the higher rate of organic and inorganic sources. Low seed oil contents may be due to availability of nitrogen to plant at proper time and in proper proportion because if protein content is more, then oil content decreases. These results narrate the findings of Witt and Pasuquin (2007).

Conclusion

In view of above stated findings it has been concluded that under agro-ecological conditions of Faisalabad, maize showed maximum growth and yield on those sites where minimum lead toxicity was reported and 75 % N from urea and 25 % N from poultry manure was applied.

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