

# INFLUENCE OF ZINC AND BORON SUPPLEMENT UPON SOME TRACE NUTRIENTS DEPOSITION IN RICE GRAIN

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خلاصه

چاول کا ٹار پاکستان کی اہم غذائی اجناس کے طور پر ہوتا ہے جو کے باتی دنیا کی تھی بنیادی خوراک ہے۔ غذائیت سے بھر پور ہونے کی وجہ سے بیر دن ممالک میں چاولوں کی برآ مدگی زیادہ رہی ہے۔ اگر اسکی یوائی کے وقت فر ٹیلا ئیزر کی مقرر دہ مقدار اور کا شت کاری کے ایکھے طریقے استعمل کئے جامی تو اسکی غذائیت ندید برٹھ سکتی ہے۔ درمن ذیل تجربہ میں پارٹی تھنم کے چاولوں رہی ہے۔ اگر اسکی یوائی کے وقت فر ٹیلا ئیزر کی مقرر دہ مقدار اور کا شت کاری کے ایکھے طریقے استعمل کئے جامی تو اسکی غذائیت ندید برٹھ سکتی ہے۔ درمن ذیل تجربہ میں پارٹی تھنم کے چاولوں (تی ہے۔ اگر اسکی یوائی کے وقت فر ٹیلا ئیزر کی مقرر دہ مقدار استعمل کی گئی۔ (تی ہو اس کی برآ مدگی کی تعلیم کی تک ہو اوں پر N اور P کی خاص مقرر دہ مقدار استعمل کی گئی۔ تجربہ کی حاص مقرر مقدار استعمل کی گئی۔ (تی استعمال کی جائیں اول کی N میں یوائی کی حاص مقرر مقدار استعمل کی گئی۔ (PH:8.12) تعرب کے اپنی استعمال کی جائیں اول کی جائیں (PH:8.12) تعرب کی خاص مقرر استعمال کی گئی۔ (PH:8.12) تعرب کی جائیں اول میں نامیاتی رادوں کی کی جب ای وال کی جائیں اول میں مالک میں پر N ملکر معدار استعمال کی گئی۔ (PH:8.12) تعرب کے لئے استعمال کی جائیں (PH:8.12) تی جہ ہے والی مٹی سا خت کے اعتبار سے (PH:8.12) تی میں نامیاتی رادوں کی کی جائی (PH:8.12) تعرب کی حوال میں نامیاتی رادوں کی کی جائی (PH:8.12) تعرب کی حاص کی گئی۔ (PH:8.12) تعرب کی اصافی سور سے کی طور پر جبکہ (PH:8.12) تعرب کی اضافی سور سے طور پر جبکہ (PH:8.12) تعرب کی اضافی سور سے کی طور پر جبکہ (PH:8.12) تعرب کی اضافی سور سے کی طور پر جبکہ (PH:8.12) تعرب کی اضافی سور سے کی طور پر استعمال کیا گیا۔ نتیجہ سے معلوم ہوا کہ جائی ہے اسکن کی اسمافی سور سے کی طور پر جبکہ ہوں کی اضافی سور سے کی طور پر جبکہ دور پر جبکہ پر طور پر جبکہ ہوں ہوں کی گئی۔ (PH:8.12) کی گئی۔ (PH:8.12) کی تک کی اضافی سور سے کی طور پر جبکہ میں میں خول ہوں کی اضافی سور سے کی طور پر جبکہ دور پر جبکہ ہوں کی تعرب کی اضافی سور سے کی طور پر اسکن کی گیا۔ تی جہ ہو اول کی اقسام اور ٹر ٹینٹ کا Pow KgHa-1 ہوں کی ہوں ہوں ہوں کی اضافی سور سے کی ہوں کی کی ہوں کی کی اضافی سور سے کی ہوں کی کی اضافی سی کی ہوں کی کی جب ہو یوں کی ایک ہوں ہوں کی اول کی تی کی کی ہوں کی ہوں ہوں کی کی کی طور پر جبکہ ہوں کی کی گئی ہو

#### Abstract

Rice is major cereal crop of Pakistan and staple food for rest of the world. The nutritional enriched rice grain is demanded by exporting countries and the consumers. The objective of nutritional rice may be achieved by ensuring judicious supply of fertilizers followed by application of good field practices. In order to investigate the influence of Zinc (Zn) and Boron (B) supplement applied along with recommended doses of Nitrogen (N) and Phosporous (P) upon health required trace nutrient accumulation into rice grain, the experiment was conducted. Five varieties (IR6, IR8, DR92, DR83 and Shahkar) were selected for the study. Soil of the experimental field were clayey in texture (clay > 52.75 %), non-saline (E.C < 0.40 dSm<sup>-1</sup>), moderately alkaline (pH:8.12) and deficient in organic matter contents and trace nutrients viz. B: 0.7  $\mu$ gg<sup>-1</sup>; Zn:0.46 $\mu$ gg<sup>-1</sup>:Cu: 0.56 $\mu$ gg<sup>-1</sup> ; Fe: 0.59  $\mu$ gg<sup>-1</sup>and Mn: 0.91 at both the depths (0-15 and 15-30cm). In addition to this zinc sulphate (ZnSO<sub>4</sub>) as Zn source and Boric acid (H<sub>3</sub>PO<sub>3</sub>) as B source were applied. Result of the study indicated that influence of the treatment and variety upon Zn, B, Copper (Cu) and Iron (Fe) contents in grain with difference of magnitude was found to be highly significant (*P*<0.001) whereas upon Mn it was found to be significant at (*P*<0.01). It was concluded that not only the supplement of Zn at 10 Kgha<sup>-1</sup> but B at 2 Kgha<sup>-1</sup> along with N at 135 KgHa<sup>-1</sup> and P at 90 KgHa<sup>-1</sup> are necessarily required in flooded rice regimes to obtain nutritional grain. **Key words:** Rice, Nutrition, Trace elements, Zinc, Boron, supplement

### Introduction

Rice (*Oryza sativa*, *L*) among all cereals is one of the major crops fulfilling domestic food demand and remaining huge quantities are also exported to the rest of world. It is a main source of energy which provides more than seventy five percent calories to nearly two billion population of Asia and about three percent of calories to about a billion people of Africa and Latin America. Food security has become major concern due to increasing world's population. It is estimated that the population scores may be increased upto two billion in the twenty years of which half of this total increase may be found in Asia where rice is a staple food to avoid hunger (Gangaiah, B. *et al.*, 1999).

Pakistan is blessed with seasonal variation resulting in cultivation of important crops include; cotton, sugarcane, banana, rice, wheat, different type of fruits and vegetables. Rice provides huge export earnings and has become second most important food followed by wheat. It is cultivated at an average of about 10 % of countries' total area that stands seventeen percent among other cereals crops. Rural economy and livelihood there is considerably depends upon cultivation of this crop around two and half million hectares with climate range. In Sindh with main regions such as upper (Larkana, Jacobabad, Shikarpur and some part of Dadu district) and the lower (Badin and Thatta) rice contribute about twenty five percent of total cultivation. According to an estimate its production is about one and half million tons (Mollah, M. Z. *et al.*2009; Joyo, M.*et al.* 2018).

Food growth rate of country is only one percent over increasing population growth which is about 3%. According to recent estimates, by 2025 around seven hundred sixty million tons of rice shall be the regular food stuff for hungry world. Contrary to the future needs, rice production is about 35% less than the actual requirement. It has been emerging as a pressing need of time to increase the total area of rice cultivation for a better yield in future. The nutritional enriched rice may be achieved by executing good agricultural practices and balanced supply of nutrients elements to the plant (GOP, 2018). The study therefore was designed to assess the influence of Zn and B supplement upon trace nutrient uptake and accumulation into polished rice grain.

#### **Materials and Methods**

The experiment was performed to assess the effect of Zinc (Zn) and Boron (B) application allowing with recommended doses of Nitrogen (N) and Phosphorus (P) on micronutrient uptake in rice varieties. The experiment was conducted in the research field of RRI (Rice Research Institute, Dokri, Larkana). Before sowing, fifteen composite soil samples from experimental fields at both depths 0-15 and 15-30 cm were drawn to assess textural class, electrical conductivity, pH, organic matter and some micronutrients (Zn, Cu, Fe, Mn and B) before cropping. Soil of the experimental area wasclayey in texture (clay > 52.75 %), non-saline (E.C < 0.40dSm<sup>-1</sup>), moderately alkaline (pH: 8.12) and deficient in organic matter contents and trace nutrients viz. B: 0.7  $\mu gg^{-1}$ ; Zn: 0.46 $\mu gg^{-1}$ : Cu: 0.56 $\mu gg^{-1}$ ; Fe: 0.59  $\mu gg^{-1}$  and Mn: 0.91  $\mu gg^{-1}$  at both the depths (0-15 and 15-30cm). Urea (46% N) as N and DAP (48% P and 18 N) as P fertilizer at 135 Kgha<sup>-1</sup> and 90 Kgha<sup>-1</sup> respectively were applied. Nursery bed was prepared for seedlings with application of DAP (168.75g) and Urea (116.5g), when tillers reached to length of 3cm (116.5g) urea was again added. After 30 days, rice seedlings of varieties were transplanted to their respective plots. Sowing was done following the recommended procedures which include sowing in living with 0.9cm plant to plant distance. For each experimental plot of 15m<sup>2</sup> all DAP 281g was applied as basal dose before transplanting of rice seedlings whereas Urea 390.2 g was applied in two equal split doses; 195.1g after 20 days of transplanting and remaining 195.1g was applied after 65 days of transplanting or at panicle initiation. Zinc Sulphate  $(ZnSO_4)$  150g as Zn source and Boric acid  $(H_3PO_3)$  17g as B source were mixed with 1.0Kg of dry soil and broadcasted after 20 days of transplanting and along with first doze of Urea. Five rice varieties (IR6, IR8, DR92, DR83 and Shahkar) were selected for the study and each treatment was designed in triplicate. At harvest, drawn paddy samples from each plot were de-husked by hand and white polished rice grain were preserved into polyethylene bags for trace nutrient analysis (Zn Cu, Fe, Mn, B) contents.

### **Results and Discussion**

Nutrients accumulation begins soon after germination of rice seedlings in nursery bed and in main fields after transplantation. The content of accumulation and resultant effects are dependable of root growth. Uptake of these nutrient elements is in low amounts at early stage but as the plants continue to grow, their need for specific nutrient raises. Phosphorus and nitrogen helps improving germination and vegetation. Some trace elements are also as important as macro.

**Boron** (ug/g):Data in supplementary Table 1 shows the analysis of variance to determine the effects of treatments upon (B) uptake. Result indicated that the influence of the treatment on B within grain was found to be highly significant (P<0.001). Data presented in fig. 1 revealed that in all varieties B contents of rice grain found to be increased significantly (p<0.05) at T2. Upon T2 highest increase in the contents was found in a variety IR8 followed by Shahkaar, DR92, IR6 and DR83 whereas upon T1, highest increase was found in varieties in DR83 followed by IR8, DR92 and IR6 respectively.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.				
Corrected Model	13568.706	3568.706 44		73.197	.000				
Intercept	223527.790	1	223527.790	53056.596	.000				
Treatment (T)	7252.962	2	3626.481	860.782	.000				
Error	1516.682	360	4.213						
Total	238613.178	405							
Corrected Total	15085.388	404							
a R Squared = .899 (Adjusted R Squared = .887)									

Supplementary Table-1: Analysis of variance for Boron (B)

Boron uptake is apparently passive due to reduced temperature regimes and the addition of respiratory inhibitors (Hussain, S. *et al.*, 2010). Another factor that inhibits the uptake of nutrients is influenced by the contents available in soil solution and the transpiration flux potential and ratio by plant (Shafiq, M., Maqsood, T. 2010). Application of nitrogen and phosphorus may have no or non-significant impact upon boron contents within rice. (Yuan, J. C. *et al.* 2005) investigated that among trace nutrient; Zn, Cu, Fe and Mn, boron (B) could not attain much importance rather remained ignored. Result of this study showed that varieties upon application of recommended doses of nitrogen and phosphorus had varied response in boron accumulation into grains.

The significant variation of contents into grains upon control among some varieties such as IR6, IR8 and DR83, did not reflect only the impacts of some soil and environmental factors but also showed improper supply of nutrients from soils to crop plants. The differential uptake may have been occurred due to varietal potential and their genetic behaviors. Addition of zinc fertilizer along with recommended doses of nitrogen and phosphorus have shown positive growth effect on plant due to mineralization nutrients in plant tissues. Sufficient amount of zinc applied with nitrogen and phosphorus increased boron (B) translocation as well by reducing deficiency within plant but the increase was non-significant (Aref, F. 2010). In this work it is evident that overall impact of zinc application irrespective of variety was found to be insignificant. Negatively strong correlation response of DR83 was found that may reflect genetic behavior of the variety. Addition of zinc and boron fertilizers along with application of recommended doses of nitrogen and phosphorus showed significant impact on grain yield and quality. Nutrient concentration into grain also increased whenthese fertilizers are applied in combination. (Malakouti, M. A. 2008). Uptake of boron (B) within rice plant is enhanced with application of boron fertilizer but only less than 40 % of B is stored into grain, remaining is rendered into leaves and stems (Katyal, J.C., Singh, B. 1992; Bashir, K. et al. 2013). (Shafiq, M., Maqsood, T. 2010) found that B content of paddy rice significantly increased with increasing B doses, highest uptake  $(1.03\mu gg^{-1})$  was found at the dozes of B 3.5 Kgha<sup>-1</sup> and lowest 0.9 at no B application. Similar trend of increase in B content in grain of lentil, chickpea and wheat over B application was also investigated by (Flurani, M. C. et al. 2003; Johnson, S. E. et al. 2005). Application of B at 0.75 Kgha<sup>-1</sup> showed significant impacts on quality of rice grain (Rashid, A., Ryan, J. 2009). In present study, addition of boron was found to be significant impacts upon soil physicochemical factors and on availability and translocation of nutrient elements within plant system. Results of the study are in conformity also with those obtained by (Arif, M. et. al. 2012) who reported that application of Zn and B showed increase in chlorophyll content, grain Zn and B accumulation. All the varieties had shown no response over addition of Zn only with nitrogen and phosphorus but when B was added, accumulation of B increased significantly as well. Upon addition of boron fertilizer irrespective of varieties, boron (B) content of rice grain increased significantly may be due to improvement in soil boron reserves and resultant translocation and uptake within plant system that increases root growth. Higher and lower responses in percent over each treatment may have been the influence of specific genetic characteristic of the variety.



Fig. 1. Influence of treatments upon B uptake within grain. Markers labeled with same letters are nonsignificant (p>0.05)

**Zinc** (*ug/g*): Zn is necessarily required for functioning of numerous physiological functions and serves as metal activator for number of enzymes within plant system. Its uptake within plant system and accumulation into grains, is regulated by availability in rhizo sphere (Welch, R. M., Graham, R.D. 2004; Hussain, S., *et al.* 2010). Data in supplementary Table 2 showed that influence of treatment on zinc content within grain was highly significant (P<0. 001) upon control (C). Fig. 2 reveals that at T1 (Zn addition alone) and T2 (Zn and B) increased the contents of Zn into rice grain upon C (control). The difference between T1 and T2 was found to be statistically non-significant (P<0.05) whereas the differences between these treatments upon control was significant (P<0.05). The increase was higher upon T2 with exception IR and Shahkar where slight decrease was recorded.

Source	Type III Sum of Squares	Df	Mean Square	F	Sig.		
Corrected Model	5118.163	44	116.322	67.115	.000		
Intercept	52341.910	1	52341.910	30200.168	.000		
Treatment (T)	4177.424	2	2088.712	1205.142	.000		
Error	623.940	360	1.733				
Total	58084.013	405					
<b>Corrected Total</b>	5742.103	404					
a R Squared = .891 (Adjusted R Squared = .878)							

Supplementary	Table-2:	Analysis	of variance	e for Zin	c (Zn)
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Besides other soil factors, both of these nutrients if applied in heavier doses, may reduce Zn availability in soil and the translocation within plant system may be reduced due to increased amount of ammonium ion in soil solution which cause yield losses (Cakmak, I. *et al.* 2010). As a result of metabolic disorder within plant cell, imbalance between P and Zn may be occurred causing different problems in plant growth. Approximately 50% of the cereal-grown areas have low Zn within soil required for nutritional quality crop production (Barman, H. *et al.* 2018). Uptake of Zn within rice grain may vary significantly among varieties upon application of nitrogen and phosphorus. Varied responses on Zn concentration into all grain varieties was recorded. It could be due to prevalent soils attributes that plays conducive or depressive role in availability of nutrients to crop during its growth cycle.



Fig.2. Influence of treatments upon Zn uptake within grain. Markers labeled with same letters are nonsignificant (*p*>0.05)

Difference in Zn content in rice grain may also reflect genetic potential of rice of multiple variety with respect to nutrient uptake. In this work the trend was also recorded over application of nitrogen and phosphorus. Significant difference of Zn (lower and higher) uptake into grain among varieties IR6, DR83 and Shahkaar was found upon N and P only. Increase in Zn uptake by a variety Shahkaar and reduction in IR8 and DR83 on same soil and environmental condition doesn't reflect only the impact of applied fertilizers but it may also be due to differential genetic potential of varieties. Further from variance study, it was revealed that overall influence of treatments was found to be highly significant. All the varieties has shown increase in yield over addition of Zn but when B was added, varieties showed non-significant increase rather decrease of Zn content in Shahkaar variety was recorded. Increased Zn uptake within plant system and accumulation into grains is regulated by the increased availability of Zn in the rhizo sphere (Welch, R. M., Graham, R.D. 2004). Within monocotyledon plants such as wheat and rice fair transport of Zn from leaves, stem and grains is investigated (Pearson, J. N.*et al.* 1996). In a research experiment conducted by Jiang, W. *et al.* 2008 and Wissuwa, M.*et al.* 2008, it was

determined that most of Zn contents in grain was a result of increased availability in soil and uptake through roots system after flowering. This study also showed significant increase of Zn content into rice grain and increased availability and translocation within plant. Higher and lower responses in percent over each treatment could be effect of specific genetic characteristic of the variety. Findings are also in agreement with the results revealed by Pervaiz, K.*et al.* 2012 and Mollah, M. Z. *et al.* 2009.

*Copper (ug/g)*:Copper play vital role in several biochemical functions within plants. These include chlorophyll formation and protein and carbohydrate metabolism (Ortiz, M. L., Camara, M. F.2018). Supplementary Table 3 indicated that influence of the treatment on Cu accumulation within rice grain was highly significant (P<0.001). Fig. 3 showed that, overall influence of T1 (Zn addition alone) upon (C) depressed Cu in all varieties except IR8 whereas impact of T2 (Zn and B) on Cu contents in varieties IR6, DR92 and DR83 was found to be significant (P<0.05) whereas, T1 (Zn addition alone) in all varieties was found to be non-significant (P>0.05) upon (C).

Investigations have revealed that higher doses of N: P (100:90 Kgha<sup>-1</sup>) enhanced bioavailability of Zn that further may be the cause of depressed Cu transportation within plants (Fageria, N. K. 2002; Couto, W. 2017). Nitrogen and phosphorus are major nutrient elements required in developing vegetative growth. N- Fertilizer application may reduce the uptake of other trace nutrient elements such as Mg and Cu due to loss of vigor by plants, and perhaps lower demand for other nutrients with increased N shortage. On the other hand high soil P levels can depress Cu uptake, especially when other Cu limiting conditions are present, such as high soil pH and high soil organic matter. Cu has a strong affinity for N atom of amino groups and it appears quite likely that soluble N compounds like amino acids act as Cu carriers in xylem and phloem (White, P. J., Brown, P.H.2010). Uptake of Cu by the rice root, straw and grains was found to be more in the rice plants grown under waterlogged condition than that in saturated soil condition. The higher Cu uptake by the different plant parts of rice (root, straw and grain) under waterlogged condition may be explained due to favorable chemical environment of the root medium leading to higher root proliferation and nutrient absorption by the rice due to healthy reducing conditions (Das, D.K., Mandal, L.N. 1986; Couto, W. 2017). In present study it was found that application of N and P (Control), turned Cu content significantly depressive in IR6. Similar behavior among IR8 and DR92 when compared with DR83 and Shahkaar was found. Differential Cu uptake may be reflecting potential of individual varieties under submerged soilconditions. Variance studies of the data revealed that influence of treatments was found to be highly significant. Impact of Zn application with recommended doses of N, P depressed Cu storage into grain of varieties. This negative impact may have been occurred due to higher doses of N and P and enhanced bioavailability of Zn that ultimately depressing Cu transportation within plants. All the varieties except Shahkaar had shown no increase in Cu contents over addition of Zn applied N and P. Similar trend was shown by varieties IR8 and Shahkaar when B fertilizer was added but IR6, DR92 and DR83 shown to have significant increase. Higher and lower responses in percent over each treatment may have been occurred due to effect of specific genetic characteristic of the variety (Fageria, N. K. 2002; Ortiz, M. L., Camara, M. F.2018).

Source	Type III Sum of Squares	df	Mean Square	F	Sig.		
Corrected Model	84.408	44	1.918	6.115	.000		
Intercept	5303.880	1	5303.880	16905.804	.000		
Treatment (T)	7.228	2	3.614	11.520	.000		
Error	112.943	360	.314				
Total	5501.231	405					
Corrected Total	197.352	404					
a R Squared = $.428$ (Adjusted R Squared = $.358$ )							

Supplementary Table-3: Analysis of variance for Copper (Cu)



Fig.3. Influence of treatments upon Cu uptake within grain. Markers labeled with same letters are nonsignificant (*p*>0.05)

**Iron** (ug/g): Heavy doses of N and P and higher availability of manganese (Mn) and copper (Cu) in soils and within plant reduced iron (Fe) mobility in soils and transport within plants (Rout, G.R., Sahoo, S.2015). Supplementary Table 4 shows the analysis of variance to determine the effects of treatment. Results indicated that influence of the treatment on Fe content into rice grain was highly significant (p<0.001). Fig. 4 showed that impact of T2 on Fe accumulation within grain of each variety was found to be significant (p<0.05) whereas at T1 the response was non-significant (P>0.05) upon C in all varieties with exception of IR8 where contents reduced.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.			
Corrected Model	13568.706	44	308.380	73.197	.000			
Intercept	223527.790	1	223527.790	53056.596	.000			
Treatment (T)	7252.962	2	3626.481	860.782	.000			
Error	1516.682	360	4.213					
Total	238613.178	405						
Corrected Total	15085.388	404						
R Squared = .899 (Adjusted R Squared = .887)								

Su	pp	lementarv	Table-4:	Analysis	of	variance	for	Iron (	Έe	9)
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Fig.4. Influence of treatments upon Fe uptake within grain. Markers labeled with same letters are nonsignificant (*p*>0.05)

Application of N and P in soil may promote uptake and translocation of Fe within rice plant under submerged conditions. Increasing rate of N increased the Fe uptake by rice (Rout, G.R., Sahoo, S. 2015). The reason of higher uptake may have been occurred due to most apparent change resulting in reduced availability of Fe. This type of reduction exhibit under anaerobic soil regime (Vigani, G.*et al.* 2013). Soil of the experimental area was calcareous and moderately alkaline in nature and trial was conducted under submerged soils regime. Urea application as N in these condition may have exerted an acid effect resulting in decrease in soil pH, Fe (III) oxide hydrates are reduced to Fe (II) compounds. On the other hand, it has been revealed that heavy N and P application, higher availability of Mn and Cu in soils and plant system reduced Fe mobility both in soils and plants (Conn, S., Gilliham, M.2010). It is evident from the results of present study that application of N and P significantly increased Fe accumulation into grain of some varieties. Differential response of uptake over control (C) may indicate the potential of that variety in accumulating micronutrients into grain that is being regulated by homeostatic mechanism in plant system. This mechanism within plant system perform functions such as metal uptake, transportation, phloem sap loading unloading extent. Results of present study are in agreement with (Mollah, M. Z. *et al.*2009; Malakouti, M. A. 2008; Ortiz, M. L., Camara, M. F.2018). Variance studies of the data revealed that influence of treatments was found to be highly significant. It could be derived

that impact of Zn application with N and P showed no impact on Fe contents into rice grains of varieties. The interaction among Zn and Fe is as complex as in case of Zn and P resulting in little or no effect on Fe accumulation and translocation within plants. As a result of submergence soil regime and increase in sulphate ions in soil may have decreased soils pH resulting in speedy dilution and reduced uptake ability Fe by rice plant roots. The findings are in line with (Norvell, W.A., Welch, R. M. 1993; Safaya, N. M. 1976; White, P. J., Brown, P.H. 2010). Furthermore the present study also revealed that Zn and B combination increased Fe-content significantly into grain of all varieties. Highest significant storage of Fe percent into grain over addition of Zn and B was observed in a variety Shahkaar. Fageria, N. K. 2002 also reported that higher and lower responses over each treatment may have been occurred due to effect of specific genetic characteristic of the variety.

*Manganese (ug/g):* Differential behavior of rice varieties in Mn uptake in cereal grain over Zn and B in addition to N and P was also found by (Bashir, K.*et. al* 2013) who also investigated that this trend existed due to genotype impact factors for uptake of micronutrients. Supplementary Table 5 analysis of variance indicated that influence of the treatments only on Mn content into grain of rice was found to be significant (p<0.01). Fig. 5 showed that treatment 2 (T2) decreased in Mn contents of varieties IR8 and DR83, significantly (p<0.05) whereas IR6 and Shahkar varieties with exception of DR92 upon T2, Mn contents were found to be statistically indifferent (p>0.05). Among all the varieties lowest increase upon T2 was found in Shahkar whereas upon T1 highest increase in the contents was found in a variety IR6 followed by DR92, IR8 and Shahkar.

Source	Type III Sum of Squares	Df	Mean Square	F	Sig.		
Corrected Model	784.325	44	17.826	1.547	.018		
Intercept	399215.273	1	399215.273	34644.149	.000		
Treatment (T)	115.061	2	57.530	4.993	.007		
Error	4148.392	360	11.523				
Total	404147.990	405					
Corrected Total	4932.717	404					
a R Squared = .159 (Adjusted R Squared = .056)							

Supplementary Table-5: Analysis of variance for Manganese (Mn)

Mn plays a key role in splitting of water molecules, development of oxygen in photosynthesis, assimilation of carbon dioxide and in nitrogen metabolism (Luo, B.*et al.* 2018). Influence of N and P upon mineralization of Mn is less understood rather indirect effect of application these fertilizers may be found resulting an increase in the contents in soils and grains (Devika, O. S. *et al.* 2018). This change in soil reaction may occur due to release of ammonium ions from the fertilizer.

Results of present study revealed that behavior of rice varieties was different at application of N and P. On same soil and environment, average higher uptake of Mn at control (C) by IR6 and the lower in IR8 shown may have been occurred due to genetic potential of that variety. Differential behavior of rice varieties in Mn uptake in cereal grain over Zn and B in addition to N and P was also found by (Couto, W. 2017) who reported that varied responses due to genotype impact for uptake of micronutrients. Variance studies revealed that influence of treatment was found to be significant upon Mn accumulation into grain. Balanced supply of mineral nutrients to shoot of plants like iron to zinc, magnesium to zinc, phosphorus to zinc and phosphorus and manganese to zinc and copper to zinc , may have been more vitality than Zn concentration within plant tissue (Cayton, M. T.*et al.* 1985; Qadar, A. 2002; Tripathi, D. K. *et al.* 2014). In this study, prevalence of Zn content in plants was due to greater specific ion effect resulting in reduced Mn uptake and accumulation into rice grain. Results are in agreement with (Kumar, D. *et al.* 2016; Neue, H. U., Lantin, R.S. 1994; Cakmak, I. *et al.* 1997; Ghori, N. H. *et al.* 2019) who reported that increased uptake and translocation of Zn may restrict translocation of Fe, Mg, P, Mn, and Cu to plant due to disturbance of enzyme functions. Differential behavior of rice varieties in Mn uptake was found. Highest significant storage of Mn over addition of Zn was observed in a variety IR8 and lowest negative in DR83 was recorded. (Rengel, Z. *et al.* 1999) also reported similar trend.



Fig.5. Influence of treatments upon Mn uptake within grain. Markers labeled with same letters are nonsignificant (*p*>0.05)

# Conclusion

From results of present study it is concluded that soil with physicochemical characteristics of heavy texture, non-saline nature, alkaline and having low organic matter. If supplement of Zn at 10 Kgha<sup>-1</sup> and Boron at 2.0 Kgha<sup>-1</sup> along with Nitrogen at 135 Kgha<sup>-1</sup> and Phosphorous at 90 Kgha<sup>-1</sup> applied, can maximize the yield and nutritional value of rice grain.

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