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Effect of different methods of Zn application on rice growth, yield and nutrients dynamics in plant and soil

ABSTRACT

Zn deficiency is widely spread in Egyptian paddy soils and has negative impact on national rice production. Field experiments were conducted at the research farm of the Rice Research and Training Center, Sakha, Egypt, to evaluate the effects of different methods of Zn application on rice growth, yield of Sakha 104 (Oryza sativa L.) and nutrients dynamics in soil and plant. The experiment included four treatments no Zn, root soaking, foliar and soil application. The results indicated that Zn application by different methods, significantly increased number of tillers, panicles, plant height, 1000-grain weight, filled grains% and grains yield of Sakha 104. Among the different of Zn application, soil application of 15 kg ha^{-1} $ZnSO_4.H_2O$ caused greater total N, total K and available Zn content in both grains and straw, however the P% content decreased significantly. Zinc content in soil after harvesting time was significantly affected by different of Zn application. Application of different methods of Zn tended to increase the total N and total K contents soil but decreased P concentration significantly.

Key words: Zn fertilizers, soils properties, rice varieties, yield, nutrients.

1. INTRODUCTION

Rice (*Oryza sativa* L.) is one of the most important crops in the world, providing over 21% of the dietary calories for more than half of the global world population [1]. Zinc (Zn) deficiency in rice has been widely reported in many rice-growing regions of the world [2, 3]. Zinc deficiency in crop plants results in not only yield reduction but also Zn malnutrition in humans, where a high proportion of rice is consumed as a staple food [4, 5]. Low bioavailability of Zn in soil generally results in Zn deficiency in rice plants, and thus becomes one of the common constraints for Zn biofortification in rice grain. Zn deficiency in rice can be alleviated through Zn fertilization, which is considered to be a cost-effective method to alleviate Zn malnutrition [6].

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Recently, water crisis in Egypt, has caused moves towards water-saving cultivation, from flooded to alternate wetting and drying to aerobic rice systems [7, 8]. Importantly, adoption of these water saving systems may decrease Zn availability [9]. High-yielding varieties and greater fertilizer inputs in Egypt were the strategies to raise the crop yield potential and feed increasing population. The increase in the application of fertilizers were not sufficient to compensate over-use of cultivated land. High-yielding varieties rapidly depleted soil micronutrients, therefore recent literature witnessed Zn deficiency, along with Fe deficiency. Zinc deficiency is considered as a major threat to the global and regional food security [10]. Amongst several factors of low plant Zn availability in paddy fields are high soil pH and high $CaCO_3$ content and low redox potential [11]. Widespread occurrence of Zn deficiency in traditional lowland [12] and newly developed alternate wetting and drying or aerobic rice systems [13] necessitates harnessing of breeding efforts to increase Zn uptake [14, 15].

After NPK, widespread zinc (Zn) deficiency has been found responsible for yield reduction in rice [16]. Globally, more than 30 % of soils are low in plant-available Zn [17]. Nonetheless, frequency of Zn deficiency is greater in rice than other crops, with more than 50 % of the crop worldwide prone to this nutritional disorder [18]. Zinc application methods and sources are aimed at improving Zn availability for plant uptake. Zn can be applied to soil, seed and leaves [18] and by dipping seedlings into a fertilizer solution. Zn use efficiency is often determined based on the ratio of shoot dry matter or grain yield produced under Zn deficiency to that produced with Zn fertilization. Zinc fertilization to cereal crops improves productivity and grain Zn concentration [19, 20] and thus contributes to grain nutritional value for human beings. However, the vast majority of Zn fertilizer trials and resulting fertilizer recommendations in rice have been in the context of managing the Zn deficiency, with very few studies related to Zn bio fortification [21]. Selection of appropriate Zn sources for soil application is considered to be an alternative strategy to improve plant availability of Zn under lowland conditions Generally, ZnSO4 is the most widely applied Zn source for its high solubility and low cost. In addition, Zn-EDTA (Ethylene Diamine Tetra acetic Acid) is also being recommended due to its efficiency of Zn availability for the plants [22].

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Because of the water crisis, rice production in Egypt is now undergoing changes, on a gradient from continuous flooding to continuously aerobic [23, 24]. Among several water-saving strategies, alternate wetting and drying (AWD) management is one of the most commonly practiced water-saving technologies, which is being widely adopted in Egypt. It has been reported that AWD potentially results in decreased water inputs by 5%-35% when compared with contentious flooding, with the yield of rice grain either being maintained [25] or even increasing [26]. This study was aimed to investigate the effects of different methods of Zn applied through soil, root soaking and foliar on rice growth, yield and nutrients dynamics status in grains and straw of Sakha 104 rice variety. The obtained results of this study will help to mitigate Zn deficiency in rice and improve zinc use efficiency.

2. Materials and Methods

2.1 Area description and soil sampling

Field experiment was carried out at the research area of Rice research and Training Center, Sakha, Kafr El-Sheikh, Egypt (31° 6′ 42″ N, 30° 56′ 45″ E). Represented soil samples were taken in bulk from 0-25 cm depth before the growing season. The samples were air-dried, ground and passed through 2-m sieve. Then composite samples were taken and analyzed for physical and chemical characteristics namely EC, pH, texture, cations and anions following standard methods [27]. The characterization of the soil used is presented in Table 1.

Table 1. Selected soil characteristics of the experimental site

pH	EC (dS m^{-1})	Cations (meq L^{-1})				Anions (meq L^{-1})				Particles size %			Zn (mg kg^{-1})*
		Ca^{2+}	Mg^{2+}	Na^{+}	K^{+}	Cl^{-}	HCO_3^{-}	CO_3^{2-}	SO_4^{2-}	Sand	Silt	Clay	
7.89	1.60	2.70	1.65	3.75	2.69	3.80	2.25	3.69	2.67	12.0	28.9	59.1	0.29

* Extracted by using Diethylene Triamine Pentaacetic Acid (DTPA).

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2.2 Field experiment

Field experiment was laid out in RCBD with four replications with plot size of 2x5m^2. The experiment included four treatments as given in Table 2.

Table 2. Treatments

Symbolic names	Recommended NPK (kg ha^{-1})	Method of Zn application	Zn application rate (kg ha^{-1})	Zn sources
T1 (no Zn applied)	Only 150-81-72	-	-	-
T2 (Root soaking)	150-81-72	Root soaking in 1.50% solution	7.50	$ZnSO_4.H_2O$
T3 (Foliar spray)	150-81-72	Spray at 20 DAT	2.50	$ZnSO_4.H_2O$
T4 (Soil application)	150-81-72	Soil application just after transplanting	15.0	$ZnSO_4.H_2O$

DAT, Days after transplanting

Nursery of Sakha 104 was transplanted in pots. All plots expect T1 received 150 kg ha^{-1} N as urea, 81 kg ha^{-1} of P as P and 72 kg ha^{-1} K as K. Full dose of P and K was applied just before transplanting. Nitrogen fertilizers as applied in two split application: the first split was applied at the time of finial land preparation and second split was added and panicle imitation stage. All other cultural practices were kept same and uniform for all treatments. The following observations were recorded during growing season: plant height and number of tillers. At harvesting stage, rice plants were manually harvested from 1 m^2 in the center of each plot, and these plants were used to determine grain yield and its components [28].

2.3 Zn analysis

Samples (0.10 g) of each treatment were digested with 5.0 ml HNO_3-H_2O_2 (4:1, v/v) using a hot block heater. After cooling, the digest was transferred to a 20-ml volumetric flask composed of distilled water and then filtered. Zn concentrations of these samples were determined using inductively coupled plasma mass spectrometry (ICP-MS; Agilent Modell 7500a, Agilent Technologies, USA).

All plants were harvested at maturity, and separated into straw and grains. The dry weight of the straw was determined after oven-drying at 70℃ to constant weight. Yield

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component included panicle number, percentage of filled grains, 1000-grain weight, and grain yield of each treatment were recorded.

2.4 Soil Samples

Samples of rhizosphere soil were collected by gently shaking off from the root system and were extracted available Zn by DTPA according to method of [29] and determined using ICP-MS (Agilent 7500a, USA).

2.5 Statistical analysis

All the data including soil and plant samples were analyzed for critical differences using the ANOVA statistical package [30].

3. Results and Discussion

3.1 Growth characters

The height of Sakha 104 rice plant as affected by different methods of Zn application are shown in Figure 1. It is observed from the figure that methods of Zn application significantly affected the plant height. Maximum plant height of 100 cm was obtained from the treatment T4 which receiving 5 kg Zn ha^{-1} as soil application which differs significantly from the other treatments. Minimum plant height was recorded with T1 as control. The tillers numbers/m^2 of Sakha 104 rice plant as affected by different of methods of Zn application are shown in Figure 2. The data indicated that application of different methods of Zn application, significantly affected on number of tillers. Application of different of methods of Zn application increased the tillers numbers/m^2 of Sakha 104

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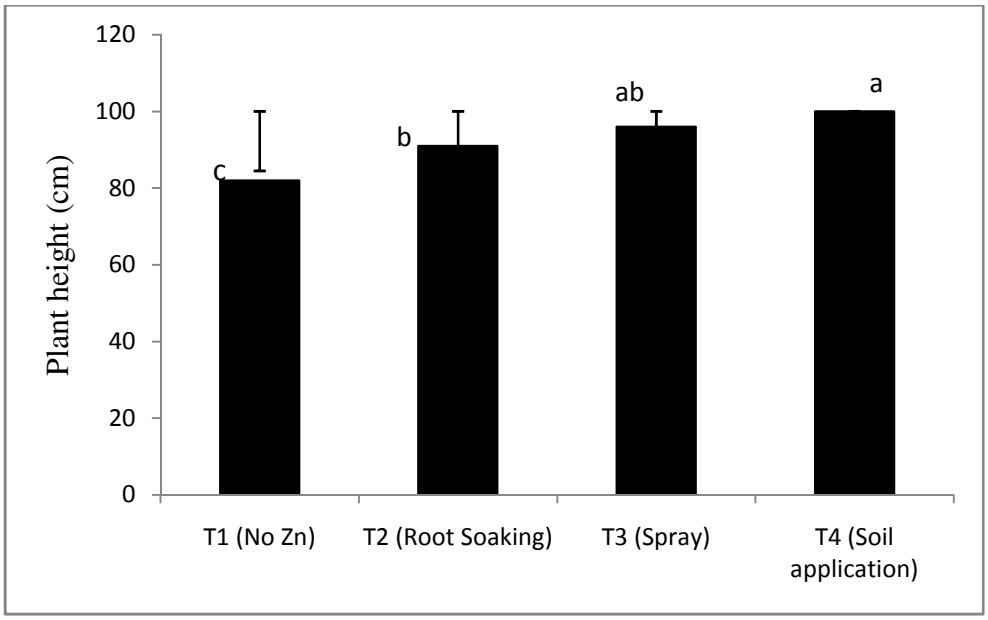

Fig. 1. Plant height of Sakha 104 as affected by different methods of Zn application. Means at followed by the same letters are not significantly different at 5% level of probability. Vertical bars represent ±standard error (SE, n=4)

rice plant compared with control. It is known that, tillering capacity is one of the most important rice components which responsible for yield production. Maximum tillers numbers/m^2 of 430 were recorded by T4 (soil application) followed by T3 (Zn was applied as foliar); while the minimum tillers numbers/m^2 was recorded in T1 (as control). The increase of tillers numbers/m^2 by soil application of Zn may be attributed due to increases of nutrients availability in soil compared with other treatments. Similar results were reported by [31, 32].

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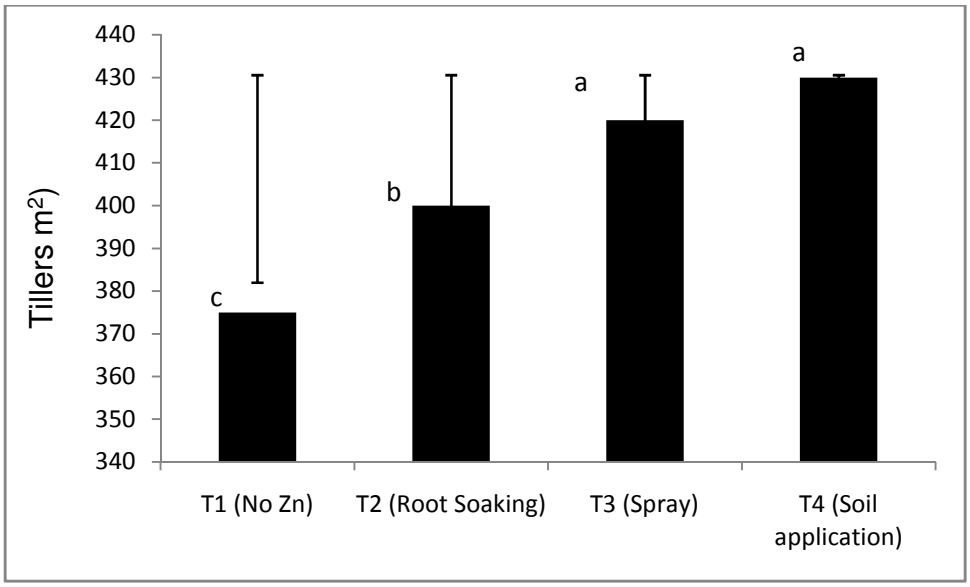

Fig. 2. Number of tillers of Sakha 104 as affected by different methods of Zn application. Means followed by the same letters are not significantly different at 5% level of probability. Vertical bars represent ±standard error (SE, n=4)

3.2 Yield components

Table 3 shows number of panicles/m^2, number of spikelet/panicle, filled grains% and 1000-grain weight as affected by different methods of Zn application. The results indicated that number of panicles/m^2 is significantly affected by methods of Zn application. All the different c methods of Zn application (T2, T3 and T4) increased the number of panicles/m^2 compared with T1 (no Zn applied). The maximum number of panicles/m^2 was recorded in T4 (soil application) followed by T2 (root soaking) and T3 in which Zn applied as foliar, while the minimum number of panicles/m^2 of Sakha 104 rice plant was recorded in T1 (control). The results of number of spikelet/panicle as affected by different of Zn application (Table 3) indicated that different of methods of Zn application increased the number of spikelet/panicle compared to control, however no significant differences were found among the methods of Zn application on number of spikelet/panicle. Both filled grains% and 1000-grain weight of Sakha 104 as affected by different methods of Zn application follows the same trend in which the different of Zn application increased the filled grains% and 1000-grain weight compared with T1 (control), however no significant differences among the methods of Zn application on filled grains% and 1000-grain weight. Similar results were reported by [33, 34, 35].

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Table 3. Number of panicles/m^2, Number of spikelet/panicle, filled grains% and 1000-grain weight as affected by different methods of Zn application

Treatments	Number of panicles/m^2	Number of spikelet/panicle	Filled grains%	1000-grain weight (g)
T1 (No Zn)	325 d	110 b	80 b	20.1 b
T2 (Root soaking)	348 b	130 a	83 a	25.9 a
T3 (Foliar spray)	339 c	128 a	81 a	26.5 a
T4 (Soil application)	350 a	129 a	82 a	27.1 a

Means values by each column followed by the same letters are not significantly different at 5% level of probability.

3.3 Grain and straw yield of Sakha 104

The impact of different methods of Zn application on grains and straw yield of Sakha 104 are shown in (Figure 3). The results indicated that the different methods of Zn application significantly increased the grain yield of Sakha 104 compared to no Zn application (T1). Maximum grain yield of 9.60 tones ha^{-1} recorded by applied 15 kg Zn ha^{-1} as soil application. No significant differences were observed in grains yield between plots receiving Zn either root soaking (T2) or foliar (T3). The increases in Sakha 104 yield by Zn applied as soil application could be attributed to its effectives and has more residual effect compared with root soaking or foliar. The data of straw yield as affected by different methods of Zn application are shown in (Figure 3). The results indicated that the different methods of Zn application (T2, T3 and T4) significantly increased the straw yield compared with T1, however no significant differences were observed among o[illegible] application. Similar findings were reported by [36, 37, 38].

3.4 Effect of different methods of Zn application on plant Zn, N, P and K content

Zinc content in grains and straw of Sakha 104 rice variety as affected by different methods of Zn application are presented in Table 4. The results indicated that, the highest Zn content of 31.1 mg kg^{-1} was recorded in T3 (Zn applied as foliar), followed by T4 and T2. The variation of Zn content was significant difference at 5% level among the treatments. The lowest Zn content in grain and straw was recorded in T1 (no Zn applied). Although foliar application is effective in increasing grains Zn content [39, 40,

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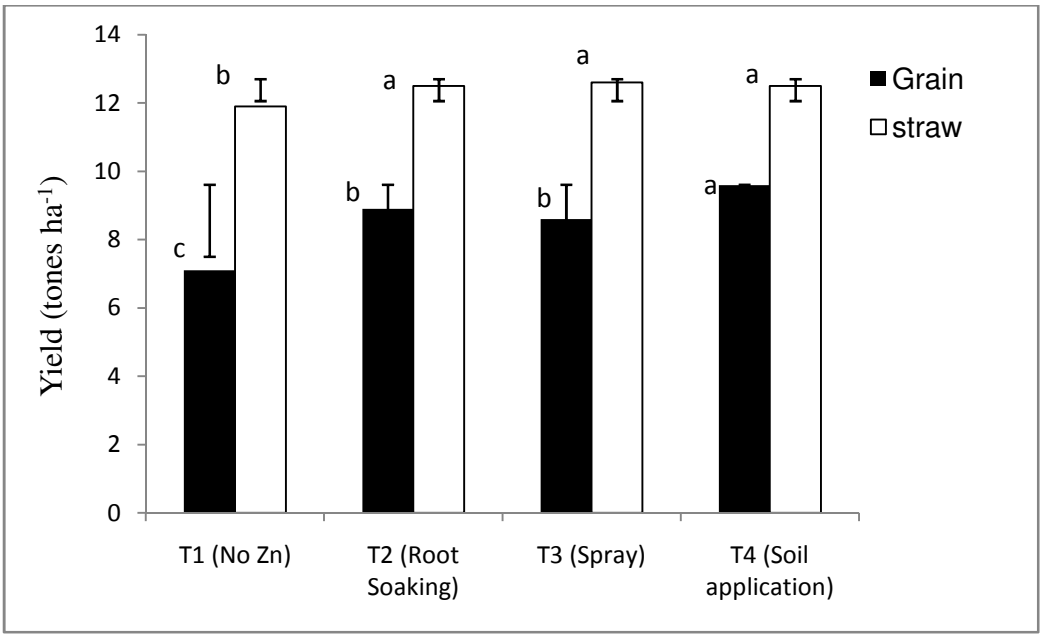

Fig. 3. Grain and straw yield of Sakha 104 as affected by different methods of Zn application. Means followed by the same letters are not significantly different at 5% level of probability. Vertical bars represent ±standard error (SE, n=4)

41], time of foliar Zn application is an important factor in this regard [42]. The increases in Zn content by grains occur when it is applied as foliar at later stages of plant growth. [43] evaluated Zn translocation towards rice grains in a nutrient solution using aerobic rice genotypes. They reported that, when Zn was applied to roots or as foliar spray; under sufficient Zn supply, Zn partitioning from grains was greater from root-supplied than Zn applied as foliar form.

Table 4 showed that, N% in rice grains and straw of Sakha 104 affected significantly by different methods of Zn application compared with no Zn (T1). The highest N% content in grains (0.97%) was recorded in T2, followed by T4 and T2, respectively. The increases in N% in grains by adding the different methods of Zn application may be attributed to the enhancing the micrograms in N recycling and organic N mineralization. The results are in agreement with findings of [44].

Phosphorus (P) content of plant as affected by different methods of Zn application is shown in Table 4. The results indicated that, application of different methods of Zn significantly decreased the P% content in grains and straw of Sakha 104 compared with T1 (no Zn applied). The highest P% of 0.26% and 0.041% were observed in T1,

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Table 4. Effect of different methods of Zn application on total N, P, K, and Zn content of rice plant

Treatments	Zn (mg kg^{-1})		N		P		K	
			%					
	Grains	Straw	Grains	Straw	Grains	Straw	Grains	Straw
T1 (No Zn)	22.1 c	23.2 b	0.73 d	0.33 b	0.26 a	0.041a	0.160 c	1.40 d
T2 (Root soaking)	29.1 b	27.3 a	0.97 a	0.50 a	0.23 b	0.036 b	0.210 a	0.165 c
T3 (Foliar spray)	32.1 a	28.3 a	0.94 c	0.48 a	0.24 b	0.031 b	0.207 a	0.175 b
T4 (Soil application)	30.1 ab	29.1 a	0.96 b	0.47 a	0.24 b	0.026 b	0.180 b	1.80 a

Means values by each column followed by the same letters are not significantly different at 5% level of probability.

however no significant difference was observed between the methods of Zn application. All the Zn applied by different methods decreased the P% content. This may be attributed the antagonistic effect of Zn on P uptake. [45] reported that application of P, not only decreased water-soluble and exchangeable-Zn, but also increased bound forms of soil-Zn. These effects were more pronounced under flooded than non-flooded water regime. Other studies also showed that P application influenced Zn uptake by rice and translocation into shoots

The data of K% content in grains and straw of Sakha 104 are presented in Table 4. The results showed that with application of different methods of Zn sources, K increased significantly compared with T1 as absolute control. These results are confirmation findings of [46], they reported that K concentration of both rice grain and straw showed increasing trend with Zn fertilizer application.

3.5 Nutrients dynamics in soil

Zinc content of soil after harvesting time as influenced my different methods of Zn application are presented in Table 5. The results indicated that the application of different methods of Zn application significantly increased Zn of soil compared with control (T1). Maximum Zn content were recorded in the order T4>T3>T2. However, the Zn content of soil after harvesting time was increased compared with the initial value

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Table 5. Effect of different of methods of Zn application on available Zn, total N, P and K concentrations of soil after harvesting time

Treatments	Zn content (mg kg^{-1})	N	P	K
		%		
T1 (No Zn)	0.35 c	0.73 d	0.26 a	0.15 c
T2 (Root soaking)	0.69 b	0.96 b	0.21 b	0.16 b
T3 (Foliar spray)	0.75 ab	0.91 c	0.22 b	0.18 a
T4 (Soil application)	0.81 a	0.99 a	0.23 b	0.19 a

Means values by each column followed by the same letters are not significantly different at 5% level of probability.

before the starting the experiment as presented in Table 1. Zinc content in soil is present in a number of chemical forms with varying solubility. These forms include soluble Zn present in soil solution (water soluble), adsorbed on exchange sites (exchangeable), associated with OM, co-precipitated as secondary minerals or associated with sesquioxides [47]. These different forms mainly control solubility and availability of Zn to plants [48]. In addition, Soil chemical properties such as pH, Rh, soil OM, have strong influence on Zn adsorption-desorption reactions and play a critical role in regulating Zn solubility and its fractionation in soils. Varied responses of rice to the Zn fertilizers have been reported, depending on the source, application time, methods of application and soil chemical properties [49].

Nitrogen (N) content of soil at harvesting time as affected by applying different methods of Zn are presented in Table 5. The data showed that there is significant different in N% content among the methods of Zn application. The maximum N% of 0.99 was recorded by T4 (Zn applied as soil application), followed by T2 and T3, while the T1 has the lowest N% value. Maximum N% of soil content was observed with combined application of N and Zn. Among the Zn application methods, soil application was considered superior [50].

Phosphorus (P) content of soil at harvesting time as affected by applying different methods of Zn are presented in Table 5. It clear that with application of different methods of Zn application, P content in soil decreased significantly compared with no Zn (T1). The highest P% content observed in T1, however no significant differences were recorded among the methods of Zn application. Phosphorus interacts with Zn in soil

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which reduces Zn translocation from roots to the shoots [51] and also imbalanced P: Zn ratio has a negative effect on rice yield. Other studies also indicated that P application influenced Zn uptake by rice and translocation into shoots [52].

The content of K% of soil at harvesting time as affected by different of methods of Zn application are presented in Table 5. The results indicated that the application of different of methods of Zn application has a little effect of increased the K% content compare with no Zn added (T1). The maximum K% content of 0.19% was recorded with T4, followed by T3 and T2. Similar results are reported by [53].

REFERENCES

1. Fitzgerald MA, Mccouch SR, Hall RD. Not just a grain of rice: the quest for quality. Trends Plant Sci., 2009; 14(3): 133–139.

2. Mandal B, Hazra GC, Mandal LN. Soil management influences on zinc desorption for rice and maize nutrition. Soil Sci. Soc. Am. J., 2000; 64: 1699–1705.

3. Tiong J, McDonald GK, Genc Y, et al. HvZIP7mediates zinc accumulation in barley (*Hordeum vulgare*) at moderately high zinc supply. New Phytol., 2014; 201(1): 131–143.

4. Yao F, Huang J, Cui K, et al. Agronomic performance of high-yielding rice variety grown under alternate wetting and drying irrigation. Field Crops Res., 2012; 126: 16–22.

5. Zhang H, Xue Y, Wang Z, et al. An alternate wetting and moderate soil drying regime improves root and shoot growth in rice. Crop Sci., 2009; 49(6): 2246–2260.

6. Zhao FJ, Mcgrath SP. Biofortification and phytoremediation. Curr. Opin. Plant Biol., 2009; 12(3): 373–380.

7. Farooq M, Kobayashi NK, Wahid A, Ito O, Basra SMA. Strategies for producing more rice with less water. Adv Agron., 2009; 101: 351–388.

UNDER PEER REVIEW

8. Farooq M, Rehman A, Aziz T, Habib M. Boron nutripriming improves the germination and early seedling growth of rice (Oryza sativa L.). J Plant Nutr., 2011a; 34: 1507–1515.

9. Gao XP, Zou CQ, Fan XY, Zhang FS, Hoffland E. From flooded to aerobic conditions in rice cultivation: consequences for zinc uptake. Plant Soil, 2006; 280: 41–47.

10. Rana WH, Kashif SR. Effect of different Zinc sources and methods of application on rice yield and nutrients concentration in rice grain and straw. Journal of Environmental and Agricultural Sciences, 2014; 1–9.

11. Alloway BJ. Soils factors associated with zinc deficiency in crops and humans. Environ Geochem Health, 2009; 31(5): 537–548.

12. Singh B, Natesan SKA, Singh BK, Usha K. Improving zinc efficiency of cereals under zinc deficiency. Curr Sci., 2003; 88: 36–44.

13. Dobermann A, Fairhurst TH. Nutrient disorders and nutrient management. Potash and Phosphate Institute, PPI of Canada and IRRT, Singapore, 192 pp, 2000.

14. Sharma PK, Lav B, Ladha JK, Naresh KK, Gupta RK, Balsubrananian BV, Bouman, BAM. Crop water relation in rice-wheat cropping under different tillage systems and water management practices in a marginally sodic, medium textured soil. In: Bouman, B.A.M., Tuong, T.P., Ladha, J.K. (Eds) Water-wise production. IRRI, Los Baños, Philippines, 2002.

15. Rehman HU, Aziz T, Farooq M, et al. Zinc nutrition in rice production systems: a review. Plant Soil, 2012; 361(1-2):203–226.

16. Fageria NK, Baligar VC, Clark RB. Micronutrients in crop production. Adv Agron., 2002; 77: 185–268.

17. Hajiboland R, Yang ZE, Romheld V. Effects of bicarbonate and high pH on growth of Zn efficient and Zn inefficient genotypes of rice, wheat and rye. Plant Soil, 2003; 250: 349–357.

18. Johnson SE, Lauren JG, Welch RM, Duxbury JM. A comparison of the effects of micronutrient seed priming and soil fertilization on the mineral nutrition of

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chickpea (Cicer arietinum), lentil (Lens culinaris), rice (Oryza sativa) and wheat (Triticum aestivum) in Nepal. Exp. Agric., 2005; 41: 427–448.

19. Cakmak I. Enrichment of cereal grains with zinc: agronomic or genetic biofortification. Plant Soil, 2008; 302, 1–17.

20. Phattarakul N, Rerkasem B, Li LJ, et al., Biofortification of rice grain with zinc through zinc fertilization in different countries. Plant Soil, 2012; 361(1-2): 131-141.

21. Impa SM, Schulin R, Ismail A, Beebout JS. Unravelling the mechanisms influencing grain-zn content in rice genotypes. In: Abstracts, International Rice Congress, Hanoi Veitnam, Nov 8-12, 2010.

22. Quijano-Guerta C, Kirk GJD, Portugal AM, Bartolome VI, McLaren GC. Tolerance of rice germplasm to zinc deficiency. Field Crops Res., 2002; 76: 123–130.

23. Bouman BAM, Hengsdijk H, Hardy B, et al. Water-Wise Rice Production. International Rice Research Institute, Los Baños, Philippines, p.89–102, 2002.

24. Rehmani MIA, Wei G, Hussain N, Ding C, Li G, Liu Z, Wang S, Ding Y. Yield and quality responses of two Indica rice hybrids to post-anthesis asymmetric day and night open-field warming in lower reaches of Yangtze River delta. Field Crops Res., 2014; 156: 231–241.

25. Chapagain T, Yamaji E. The effects of irrigation method, age of seedling and spacing on crop performance, productivity and water-wise rice production in Japan. Paddy Water Envion., 2010; 8(1): 81–90.

26. Yang XE, Chen WR, Feng Y. Improving human micronutrient nutrition through biofortification in the soil plant system: China as a case study. Environ Geochem Health, 2007; 29 (5): 413–428.

27. Page AI, Miller RH, Keeney DR. Methods of soil analysis. Madison, Wisconsin, USA, 1982.

28. Ramesh SA, Shin R, Eide D, Schachtman DP. Differential metal selectivity and gene expression of two zinc transporters from rice. Plant Physiol., 2003; 133: 126–134.

UNDER PEER REVIEW

29. Lindsay WL, Norvell WA. Development of a DTPA soil test for Zn, Fe, Mn, and Cu. Soil Sci. Soc. Am. J. 1978; 42: 421–428.

30. Gomez KA, Gomez AA. Statistical Procedure for Agricultural Research, 2nd Edition. A Wiley Interscience Pub., John Wiley and Sons, New York. USA. p. 628, 1984.

31. Ghoneim AM, Ueno H, Asagi N, Watanabe T. Indirect ^{15}N Isotope Techniques for Estimating N Dynamics and N Uptake by Rice from Poultry Manure and Sewage Sludge. Asian Journal of Earth Sciences, 2012; 5 (2): 63–69.

32. Ghoneim AM. Nitrogen and carbon uptake by some Rice cultivars from $^{15}NH_4Cl$ and ^{13}C-U-glucose labeling fertilizer. International Journal of Agronomy and Agricultural Research, 2014; 4 (4): 20–27.

33. Cakmak I. Enrichment of fertilizers with zinc: an excellent investment for humanity and crop production in India. J Trace Elem Med Biol., 2009; 23(4): 281–289.

34. Beebout SJ, Tuyogon D, Rubianes F, Castillo O, Larazo W, Bunquin M, Laureles, E. Improved zinc management strategies for rice scientists and farmers. In: Proceedings of 2010 International Annual Meetings of ASA-CSSA-SSSA, October 31 to November 04, 2010, Long Beach, California, USA, 2010.

35. Farooq M, Wahid A, Siddique KHM. Micronutrients application through seed treatments - a review. J Soil Sci. Plant Nutr., 2012; 12: 125–142.

36. Khan MU, Qasim M, Subhan M, Jamil M, Ahmad RD. Response of rice to different methods of Zn application in calcaerous soils. Pak J Appl. Sci., 2003; 3:524–529.

37. Jiang W, Struik PC, Zhao M, van Keulen H, Fan TQ, Stomph TJ. Indices to screen for grain yield and grain zinc mass concentration in aerobic rice at different soil Zn levels. NJAS Wageningen J Life Sci., 2008b; 55: 181–197.

38. Bhaduri D, Purakayastha TJ. Soil available Zn: a potent soil quality indicator in a rice–wheat system. 3rd International Zinc Symposium, 10–14 October 2011, Hyderabad, India.

39. Welch RM. Breeding strategies for biofortified staple plant foods to reduce micronutrient malnutrition globally. J Nutr., 2002; 132:495–499.

UNDER PEER REVIEW

40. Hajiboland R, Salehi SY. Characterization of Zn efficiency in Iranian rice Genotypes I. Uptake efficiency. Gen Appl Plant Physiol. 2006; 32: 191–206.

41. Chasapis CT, Loutsidou AC, Spiliopoulou CA, et al. Zinc and human health: an update. Arch. Toxicol., 2012; 86(4): 521–534.

42. Stomph TJ, Hoebe N, Spaans E, van der, Putten PEL. The relative contribution of post-flowering uptake of zinc to rice grain zinc density. 3rd International Zinc Symposium 10-14 October 2011, Hyderabad, India.

44. Gao X, Hoffland E, Stomph TJ, Grant CA, Zou C, Zhang F. Improving zinc bioavailability in transition from flooded to aerobic rice. A review. Agron Sustain Dev., 2012; 32: 465–478.

45. Lal B, Majumdar B, Venkatesh MS. Individual and interactive effects of phosphorus and zinc in lowland rice. Indian J Hill Farming, 2002; 13: 44–46.

46. Iqbal M, Aslam M, Ranja AM, Akhtar J. Salinity tolerance of rice as affected by Zn application. Pakistan J. Biol. Sci., 2000; 3: 2055–2057.

47. Arnold T, Kirk, GJD, Wissuwa M, Frei M, Zhao FJ, Mason TFD, Weiss DJ. Evidence for the mechanisms of zinc uptake by rice using isotope fractionation. Plant Cell Environ., 2010; 33: 380–381.

48. Almendros P, Gonzalez D, Obrador A, Alvarez JM. Residual zinc forms in weakly acidic and calcareous soils after an oilseed flax crop. Geophysical Research Abstracts. EGU General Assembly, 2008; 10, EGU2008-A-12479.

49. Takrattanasaran N, Chanchareonsook J, Johnson PG, et al. Amelioration of zinc deficiency of corn in calcareous soils of Thailand: zinc sources and application methods. J. Plant Nutr., 2013; 36(8): 1275–1286.

50. Depar N, Rajpar I, Memon MY, Imtiaz M, Zia-ul-hassan. (2011). Mineral nutrient densities in some domestic and exotic rice genotypes. Pak J Agric. Eng Vet Sci., 2011; 27: 134–142.

51. Haldar M, Mandal LN. Influence of soil moisture regimes and organic matter application on the extractable Zn and Cu content in rice soils. Plant Soil, 1979; 53: 203–213.

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52. Lonergan PF, Pallotta MA, Lorimer M, et al. Multiple genetic loci for zinc uptake and distribution in barley (*Hordeum vulgare*). New Phytol., 2009; 184(1):168–179.

53. Wu CY, Feng Y, Shohag MJ, et al. Characterization of 68Zn uptake, translocation, and accumulation into developing grains and young leaves of high Zn-density rice genotype. J. Zhejiang Univ.-Sci. B (Biomed. & Biotechnol.), 2011; 12(5): 408–418.