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SCREENING OF SHORT DURATION RICE GENOTYPES FOR ZINC EFFICIENCY

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ABSTRACT

Zn (Zn) deficiency in tropical soils affects nutritional quality of food grains and to secure nutritional quality, one potential option is the 'agronomic bio-fortification' which depends mainly on genotypic efficiency to absorb and accumulate more of Zn into grains. To identify the rice genotypes having better ability to absorb and translocate more Zn into grains, a field experiment was conducted with 15 short duration rice genotypes on a Zn deficient sandy clay loam soil with and without Zn addition. A split plot design was adopted using Zn treatment in main plots, as M₁: control (only recommended dose of N,P, and K without Zn) and M₂: recommended dose of N, P, and K with Zn (100 kg ZnSO₄ ha⁻¹ as soil application + 0.5% as foliar application thrice at 50% flowering, milk and dough stages) and the selected genotypes as sub plots. Results of present study revealed that, Zn application significantly increased the average grain yield (13.5%) and grain Zn content (37 to 55 %) over control. The rice genotypes, CO 47 performed better with higher grain yield of 5980 and 6750 kg ha⁻¹ respectively under both with and without Zn fertilization. Higher grain yield index was noted with CO 47, and CO 51 (92.7) followed by ADT 45 (88.0) while the highest grain Zn uptake index was noted with CO 47 (40.4). Based on the yield and Zn uptake efficiency, the genotypes CO 47, CO 51, ADT 36, ADT 37, MDU 5, MDU 6, TKM 12, IR 50 were found efficient and responsive to Zn fertilization thus can be utilized for Zn bio-fortification. The rice genotypes TPS 5, Anna 4, CB 14508 are highly inefficient and susceptible to Zn deficiency which needs Zn fertilization without it the yield loss is unavoidable.

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1 Introduction

Micronutrients such as Zinc (Zn), copper (Cu), manganese (Mn), iron (Fe), boron (B), and molybdenum (Mo) are essential but required in small quantities for the growth and development of many plants (Alloway, 2008). These micronutrients are very crucial for plant, animal and human health and play a key role in various biochemical processes. It is estimated that Zn deficiency in Indian soils is likely to increase from 49 to 63% by 2025. A substantial genetic variability in tolerance to Zn deficiency exists in many crops and this variability has been used significantly to improve the Zn acquisition by plants (Sudha & Stalin, 2015a). The inherent soil micronutrients status mainly depends on the geological substrate and weathering processes (White & Zasoski, 1999). Though the total concentration of micronutrients generally vary in soils due to diversity in parent materials, their availability to plants depends on pH, organic matter, texture, soil moisture and clay mineralogy (Martens & Lindsay 1990; Fageria, 2001). Generally, micronutrient content is high in the surfaces soils and decreases with depth (Gupta, 2005). The soil pH and oxidation-reduction conditions decide micronutrient form in which these are present in soil and determine their bioavailability by influencing fixation reactions (Fageria et al., 2011). Moreover, their content in the soils is also influenced by the type of cultivated crops which differ in their uptake of micronutrients. In the tropical soils of India, micronutrient deficiencies in soils is attributed to the larger removal from soil by fertilizer responsive high yielding crops, use of micronutrient free high analysis fertilizers, mono cropping, increased cropping intensity and reduction in the use of organic manures etc.

Zinc is considered as the fourth most important yield-limiting nutrient after, nitrogen, phosphorus and potassium (Shukla et al., 2018) and plays an important role in starch formation, synthesis of auxin and essential for the production of growth hormones like IAA. It also influences RNA levels and ribosome contents in cells, component of protein and chloroplast and also necessary for various enzymes that are responsible for driving many metabolic reactions in plants. In addition, Zn is a co-factor for many enzymes and proteins involved in cell division, nucleic acid metabolism and protein biosynthesis (Figueiredo, et al., 2012).

Continuous intensive cropping along with high yielding crop varieties has further aggravated the depletion of soil Zn leading to low Zn concentration in edible grains. The reported use efficiency of micronutrients in Indian soils seldom exceeds 5 per cent (Shukla et al., 2014). Reliance on cereal-based diets with low Zn may induce Zn deficiency in human beings resulted in serious health problems such as growth retardation, susceptibility to infectious diseases, hypogonadism, iron deficiency anemia, and poor birth outcome in pregnant women (Graham et al., 2000; Graham et al., 2012). The potential options to correct

micronutrient deficiency in human beings may be food supplementation, food fortification or bio-fortification (White & Broadley, 2009). “Agronomic bio-fortification” cannot be successfully achieved through the application of fertilizers nutrients which is proved effective in enriching micronutrients content in rice grain by controlling the availability of soil micronutrients (White & Broadley, 2009). Studies showed that certain plant species, as well as their genotypes exhibit a significant genetic variation in their tolerance to Zn deficiency and this ability of a genotype to grow and yield well in a Zn deficient soil is termed as “Zn efficiency” (Hacisalihoglu & Kochian, 2003). Genotypic differences for Zn use efficiency have been reported in several crops species (Rengel, 2005; Cakmak et al., 2010) and related to various mechanisms operating in the rhizosphere and within the plant system.

Rice is an important staple food and energy source for more than half of the world population, however, it is a poor source of essential micronutrients such as Fe and Zn (Welch, 2008; Tripathy et al., 2017). It is highly sensitive crop to Zn deficiency with drastic yield reduction and lesser Zn concentration in the grains yield when Zn is limited in soils. Application of Zn fertilizers is essential for keeping sufficient available Zn in soil solution and maintaining adequate transport during critical growth stages of crop which leads increased grain Zn concentration to a considerable amount. Thus the present study was proposed to study the effect of Zn fertilization on grain yield and Zn content in rice and to identify the Zn uptake efficient short duration rice genotypes for bio-fortification.

2 Materials and Methods

2.1 Experimental site

Field experiment was conducted in naturally Zn deficient soil at wetland farms of Tamil Nadu Agricultural University, Coimbatore. The farm is situated at an altitude of 426.72 m above MSL in the Western agro climatic Zone of Tamil Nadu at 11° North latitude and 77° East longitude.

2.2 Climatic condition

Annual rainfall of 641 mm in 37 rainy days and cropping period rainfall of 302 mm in 18 rainy days were recorded during 2016-17 at the principal observatory, Agro Climate Research Centre, Tamil Nadu Agricultural University, Coimbatore. The North East Monsoon (NEM) contributes 375 mm of rainfall. The relative humidity ranged from 26 to 94 per cent with mean sunshine hours of 7.4 hours. The mean maximum and minimum temperatures prevailed during the cropping period was 30.3°C and 21.3 °C which were favourable for growth and development of rice crop (AMFU - Coimbatore, 2017).

2.3 Soil characteristics

Surface soil samples were drawn from 0-15 cm air dried, crushed and passed through two mm sieve. The initial soil properties were analyzed as per the standard methods which were furnished in table 1. The soil of the experimental field was slightly alkaline in pH (8.32) with low salt (0.30 dS m^{-1}) and available N status (222 kg ha^{-1}), medium organic carbon (5.60 g ha^{-1}), high in available P (24.0 kg ha^{-1}) and K (410 kg ha^{-1}). The soil of the study area was deficient to Zn (0.98 mg ha^{-1}) but sufficient for other micronutrients.

2.4 Experiment details

The experiment was conducted during the *kharif* season of 2017 at wetland farms of Tamil Nadu Agricultural University, Coimbatore to screen short duration rice genotypes (15 nos.) for higher grain Zn content and Zn efficiency using soil and foliar Zn fertilization practices. Rice genotypes included in the study were ADT 36, ADT 37, ADT 43, ADT (R) 45, MDU 5, MDU 6, CO 47, CO 51, TPS 5, ASD 16, TKM 12, Anna (R) 4, IR 50, CB 14508 and Shabdhagithan. Two Zn treatments *viz.* M_1 : NPK Control (150: 50: 50 kg NPK ha^{-1}) and M_2 : M_1 + Zn at $100 \text{ kg ZnSO}_4 \text{ ha}^{-1}$ as soil application + 0.5% as thrice foliar application at 50% flowering, milk and dough stages. The fertilizers were applied basally and the crop was grown to maturity and harvested. The grain and straw yield of crop, Zn content and uptake was recorded besides measuring the yield and growth attributes. The post harvest soil and plant samples were collected and analysed for Zn status using atomic absorption spectrophotometer (Lindsay & Norvell, 1978). The Zn efficient and inefficient rice genotypes were identified using uptake and yield efficiency indices marked on X and Y axis, by drawing a scattered diagram (Graham et al., 1992; Fageria & Baligar, 1993). All the data were analysed by using OPSTAT model (Sheoran et al., 1998)

A perpendicular and parallel line to X axis were drawn with an average yield and uptake efficiency to divide the scattered diagram so as to classify the genotypes into four groups *viz.* efficient and responsive (ER), efficient and non responsive (ENR), inefficient and responsive (IER) and inefficient and non responsive (IENR) as described by Graham et al. (1992) and Fageria et al (2008). The ER genotypes have high yield as well as high uptake efficiency index, ENR genotypes have high yield and low uptake efficiency index, IER genotypes have low yield and high uptake efficiency index and IENR have low yield as well as low uptake efficiency index. The efficient and responsive genotypes would be most suitable for enriching Zn in grains through biofortification.

3 Results and Discussion

3.1 Effect of Zn fertilization on Yield

Spectacular differences were witnessed on the grain and straw yield of rice owing to the interaction between the Zn fertilization and genotypes (Table 2). The grain and straw yield differed from 4492 to 7200 kg and 6980 to 9297 kg ha^{-1} respectively. Application of 150:50:50 kg +100 kg $\text{ZnSO}_4 \text{ ha}^{-1}$ as soil application + 0.5% as foliar application thrice at 50% flowering, milk and dough stages recorded the highest grain and straw yield of 7200 kg and 9297 kg ha^{-1} . This increase in yield might be attributed to the greater influence of Zn on basic plant life processes, such as nitrogen metabolism (uptake of nitrogen and protein quality) and photosynthesis (chlorophyll synthesis and carbonic anhydrase activity) (Hazra et al., 2015; Kumar et al., 2018). The genotype CO 47 registered the highest grain yield in both control (6000 kg ha^{-1}) and Zn applied plots (7200 kg ha^{-1}), while the lowest grain yield was recorded in IR 50 in control (4500 kg ha^{-1}) and Zn applied treatments (5142 kg ha^{-1}). However with regard to straw yield, highest straw yield was recorded with

Table 1 Details of analytical procedures employed in soil analysis

Estimations	Procedure	Reference
<i>Physico-chemical properties</i>		
Soil reaction (pH)	1:2.5 soil water suspension	Jackson (1973)
Electrical conductivity (EC)	1:2.5 soil water suspension	Jackson (1973)
<i>Chemical properties</i>		
Organic carbon	Chromic acid wet digestion	Walkley & Black (1934)
Available nitrogen	Alkaline permanganate method	Subbiah & Asija (1956)
Available phosphorus	0.5 M NaHCO_3 (pH-8.5)	Olsen et al. (1954)
Available potassium	Neutral N NH_4OAc	Stanford & English (1949)
DTPA micronutrients	DTPA extraction and AAS method	Lindsay & Norvell (1978)

Table 2 Effect of Zn fertilization on grain and straw yield of rice genotypes

Genotypes	Yield (kg ha ⁻¹)					
	Grain		Straw			
	(-Zn)	(+Zn)	(-Zn)	(+Zn)		
Anna (R) 4	5417	6292	8063	8605		
ADT 37	5083	5583	8147	8830		
MDU 5	4500	5142	7147	7788		
CO 47	6000	7200	7480	9297		
CO 51	5850	6733	8230	9205		
TPS 5	5000	6583	8047	8788		
CB 14508	5317	6092	7855	8897		
ASD 16	5167	6200	8147	8997		
ADT 43	4850	5833	7880	8880		
ADT 36	4667	5250	6980	7747		
TKM 12	5142	5483	8038	9097		
Shabhagithan	4892	5750	7680	8705		
IR 50	4492	5183	7597	8030		
ADT (R) 45	5025	5708	7705	8063		
MDU 6	5275	5575	7630	8655		
Mean	5112	5907	8063	8605		
	SE(d)	CD (P=0.05)	SE(m)	SE(d)	CD (P=0.05)	SE(m)
M	56.4	113.2	39.9	55.5	111.4	39.3
V	154.5	310.1	109.2	152.2	305.2	107.6
M X V	218.5	438.6	154.5	215.1	431.7	152.1

M = Zn application V = Rice genotypes

CO 47 and the lowest yield was evident in MDU 5. The overall data revealed 13.5 % increase in grain yield and 11 per cent increase in straw yield due to Zn application over NPK alone and the yield increase in genotypes might be due to increased activity of enzymes and auxin metabolism in the plant which was reported by Sudha & Stalin (2015a).

3.2 Effect of Zn fertilization on Zn content

Crop species have differential micronutrient density in grains when grown under similar conditions and the Zn concentration ranged from 11.5 to 37.2 mg kg⁻¹ and 22.8 to 41.4 mg kg⁻¹ in whole rice grains and straw respectively in Zn applied treatment and the increase in Zn content was 37 to 55 per cent over control. In the present study also genotypes differed significantly in Zn absorption and CO 47 recorded the maximum Zn concentration in both grain and straw (37.2 and 41.4 mg kg⁻¹) while the lowest Zn

content was recorded in CB 14508 (11.5 mg kg⁻¹) and TPS 5 (22.8 mg kg⁻¹) in grain and straw respectively (Table 3). It was statistically at par with CO 51(35.6 and 40.7 mg kg⁻¹) and ASD 16 (35.4 and 40.1 mg kg⁻¹) in grain and straw Zn contents respectively. The difference in micronutrient concentration in rice genotypes may be attributed to micronutrient loading in grains and according to Impa et al. (2013), some genotypes showed continued root uptake which is the predominant source of Zn loading in grain, whereas in some genotypes, net remobilization of Zn from shoot and root to grain was predominant. Further, the difference was also might be due to genotype and environment interaction, water management and soil factors like pH, EC, organic carbon, phosphorus and micronutrient availability (Sudha & Stalin, 2015a). Increased root proliferation further increased the Zn uptake, enhanced synthesis of carbohydrates and its transport to grain (Sudha & Stalin, 2015b; Tripathy et al., 2017).

Table 3 Effect of Zn fertilization on Zn content in rice genotypes

Genotypes	Zn content (mg kg ⁻¹)					
	Grain			Straw		
	(-Zn)	(+Zn)		(-Zn)	(+Zn)	
Anna (R) 4	14.1	34.5		25.4	36.4	
ADT 37	14.8	34.3		27.9	38.9	
MDU 5	14.9	33.7		29.2	38.7	
CO 47	16.2	37.2		30.4	41.4	
CO 51	14.9	35.6		24.8	40.7	
TPS 5	11.6	33.5		22.8	36.8	
CB 14508	11.5	18.3		24.8	37.6	
ASD 16	15.6	35.4		29.2	40.1	
ADT 43	15.1	32.5		25.8	36.8	
ADT 36	12.6	30.0		26.2	37.2	
TKM 12	14.4	34.2		30.2	38.4	
Shabhagithan	12.7	31.1		23.4	36.4	
IR 50	14.2	31.7		35.0	34.2	
ADT (R) 45	11.8	34.5		23.4	35.8	
MDU 6	14.8	31.3		27.4	33.5	
Mean	13.9	32.5		27.1	37.5	
	SE(d)	CD (P=0.05)	SE(m)	SE(d)	CD (P=0.05)	SE(m)
M	0.36	0.72	0.25	0.41	0.82	0.29
V	0.98	1.98	0.69	1.13	2.26	0.80
M X V	1.39	2.80	0.98	1.59	3.20	1.13

M = Zn application V = Rice genotypes

3.3 Effect of Zn fertilization on Zn uptake

Zn application had positive influence on the Zn uptake by rice grains varied from 58 to 96.6 g ha⁻¹ in control and 153 to 269 g ha⁻¹ in Zn applied plot in grain yield while in straw it ranged from 179 to 242 in control and from 274 to 385 g ha⁻¹ in Zn applied treatment (Table 4). Genotypes exerted their differential response on Zn removal, in which CO 47 recorded higher Zn uptake (269 and 385 g ha⁻¹) and the lowest Zn uptake was witnessed in IR50 in both grain and straw (153 and 274 g ha⁻¹). Total Zn uptake by rice genotypes ranged from 241 to 324 g ha⁻¹ in control treatment and 428 to 654 g ha⁻¹ in Zn applied treatment. The increased in Zn uptake could be attributed to better Zn absorption and root to shoot transport by Zn efficient genotypes and also probably due to more efficient transport system such as ion channel or ion pump, compared with the Zn-inefficient genotypes (Malewar et al., 1993; Kumar et al., 2018).

3.4 Zn Efficiency

To assess the Zn efficiency of rice genotypes, yield and Zn uptake index was worked out by taking the ratio between yield and uptake in control and Zn applied treatments (Figure 1). The grain yield index varied from 76 to 92.7, grain Zn index ranges from 34.6 to 46.5 and grain Zn uptake index varied from 26.3 to 40.4. The results showed that, CO 47 and CO 51 had higher grain yield index (92.7) followed by ADT 45 (88.0) while the highest grain Zn uptake index was noted with CO 47 (40.4). The lowest grain yield index of 76.0 and grain Zn uptake index of 26.3 were recorded in TPS 5, whereas the lowest grain Zn index of 34.2 was noticed in ADT 45. This was perhaps due to the abundant supply of Zn nutrition and balanced NPK, which increased the protoplasmic constituents, accelerates the process of cell division and elongation, photosynthesis processes, respiration, nitrogen metabolism-protein synthesis, other biochemical and physiological activates (Sudha & Stalin, 2015a).

Table 4 Effect of Zn fertilization on the Zn uptake by rice genotypes

Genotypes	Zn uptake (g kg ⁻¹)								
	Grain			Straw			Total		
	(-Zn)	(+Zn)		(-Zn)	(+Zn)		(-Zn)	(+Zn)	
Anna (R) 4	77.0	216		204	319		282	537	
ADT 37	74.0	191		227	343		302	535	
MDU 5	67.3	170		208	301		276	472	
CO 47	96.6	269		227	385		324	655	
CO 51	87.9	239		204	374		292	614	
TPS 5	58.0	219		183	316		241	536	
CB 14508	60.7	187		195	334		256	523	
ASD 16	81.1	221		236	361		318	583	
ADT 43	72.1	188		203	327		275	515	
ADT 36	58.8	160		182	288		242	449	
TKM 12	74.6	185		242	349		317	534	
Shabhagithan	72.3	174		179	315		252	490	
IR 50	61.8	153		271	274		333	429	
ADT (R) 45	58.6	186		180	289		239	475	
MDU 6	78.0	170		209	289		287	460	
Mean	71.9	195		210	324		282	520	
	SE(d)	CD (P=0.05)	SE(m)	SE(d)	CD (P=0.05)	SE(m)	SE(d)	CD (P=0.05)	SE(m)
M	2.28	4.50	1.61	4.14	8.32	2.93	4.97	9.98	3.51
V	6.24	12.5	4.41	11.3	22.7	8.02	13.6	27.3	9.63
M X V	8.83	17.7	6.24	16.1	32.2	11.3	19.2	38.6	13.6

M = Zn application V = Rice genotypes

Figure 1 Grain yield and Zn uptake indices in rice grain

Table 5 Genotypes categorization based on yield and uptake efficiency

Genotypes	Responsive	Non responsive
Efficient	ADT 36, ADT 37, MDU 5, MDU 6, Co 47, CO 51, TKM 12, IR 50	ADT 45, Shabhagithan
Inefficient	ADT 43, ASD 16	TPS 5, Anna 4, CB 14508

Figure 2 Grain Yield and uptake efficiency index of rice genotypes

The Zn uptake and efficiency varied widely among the genotypes which might be due to the ability of rice genotypes to increased Zn availability in the rhizosphere for subsequent uptake by releasing active Zn mobilizing substance such as phytosiderophores. Similar results with the resistant rice varieties under Zn stress condition in extracting Zn from complex and organically bound Zn forms was reported by Jin et al. (2008). However the susceptible varieties absorbed Zn only from readily available sources.

Based on the yield and Zn uptake efficiency, grouping of genotypes was made into efficient responsive, efficient non responsive, inefficient responsive and inefficient non responsive (Table 5 & Figure 2) as per the classification of Fageria & Baligar (1993) for nutrient use efficiency and average yield at low Zn supply. The first group comprised of efficient and responsive genotypes that produced more than the average yield of all the genotypes under Zn deficiency condition and their Zn efficiency was also higher than the average efficiency. Among the genotypes it was found that

genotypes ADT 36, ADT 37, MDU 5, MDU 6, Co 47, CO 5, TKM 12 and IR 50 were grouped under efficient responsive.

The second group of efficient and non-responsive genotypes produced more than average yield at low Zn supply but response to Zn application was lower than the average. The genotypes ADT 45 and Shabhagithan are found to be efficient non responsive. Third category, inefficient and responsive genotypes which produced less than average yield, but their response to Zn application was above the average. The genotypes falls this category are ADT 43 and ASD 16 and the fourth group of genotypes produced less than average yield at low Zn level and also less than average response to added Zn. These genotypes are inefficient and non-responsive. The genotypes, TPS 5, Anna 4 and CB 14508 were grouped under inefficient non responsive genotypes. This indicates that genotypes with high efficiency are desired as they will be efficient scavengers of Zn under low level of Zn supply. Similar findings were reported by Kumar et al., (2018) in rice.

Conclusions

The present investigation is concluded that, basal soil application of 150: 50: 50 kg NPK + 100 kg ZnSO₄ ha⁻¹ along with 0.5% as foliar spray thrice during 50% flowering, milk and dough stages of the rice crop significantly increased the rice grain and straw yield, Zn content and its uptake. Based on the efficiency indices, the genotypes, CO 47, CO 51, ADT 36, ADT 37, MDU 5, MDU 6, TKM 12, IR 50 were found efficient and responsive to Zn fertilization thus can be utilized for bio-fortification of Zn. The genotypes ADT 45 and Shabhagithan are found to be Zn efficient but non-responsive to Zn application thus indicates that they are genetically efficient to utilize the native soil Zn thus suitable for Zn low condition. The rice genotypes TPS 5, Anna 4, CB 14508 are highly inefficient and susceptible to Zn deficiency.

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Conflict of interest

The corresponding author declares that there is no conflict of interest.

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