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Genotypic Variation for Micronutrient Content in Traditional and Improved Rice Lines and its Role in Biofortification Programme

Vishnu Varthini Nachimuthu[1], S. Robin[1], D. Sudhakar[2], S. Rajeswari[1], M. Raveendran[2], K. S. Subramanian[3], Shalini Tannidi[1], Balaji Aravindhan Pandian[1]

[1] Centre for Plant Breeding and Genetics, Tamil Nadu Agricultural University, Coimbatore, India; popvarun@gmail.com, robin.tnau@gmail.com, rajisundar93@gmail.com, tannidi.shalini@gmail.com, aaron.agri@gmail.com

[2]Centre for Plant Molecular Biology and Biotechnology, Tamil Nadu Agricultural University, Coimbatore, India; dsudhakar@hotmail.com, raveendrantnau@gmail.com

[3] Department of Nano science and technology, Tamil Nadu Agricultural University, Coimbatore, India; kssubra2001@rediffmail.com

Abstract

Biofortification is an emerging cost-effective strategy to address global malnutrition especially in developing countries. This strategy involves supplying of micronutrients such as iron and zinc in the staple foods by using conventional plant breeding and biotechnology methods. Initial step in conventional plant breeding is to screen the natural gene reservoir for existing variation. The objective of this study is to estimate iron and zinc in the brown rice of 192 germplasm lines and to define its role in biofortification programme. Substantial variations among 192 lines existed for both iron and zinc content. Iron concentration ranged from 6.6 µg/g to 16.7 µg/g and zinc concentration from 7.1 µg/g to 32.4 µg/g in brown rice. Iron and zinc concentration were positively correlated implying the chance for concurrent selection for both the micronutrients. Micronutrient-rich genotypes identified in this study opens up the possibilities for the identification of genomic regions or QTLs responsible for mineral uptake and translocation that can be used as donor for developing nutrient enriched varieties.

Keywords: Biofortification, Germplasm, Iron, Micronutrient, Variability, Zinc

1. Introduction

Zinc is an important micronutrient which has critical role in tissue growth, wound healing, connective tissue growth & maintenance, immune system function, prostaglandin production, bone mineralization, proper thyroid function, blood clotting, cognitive functions, fetal growth & sperm production. It is required for metabolic activity of enzymes (as a cofactor) involved in repair brain function & replacement of body cells. It's essential for cell division & synthesis of DNA & proteins. Zinc is a mineral that promotes immunity, resistance to infection, and proper growth and development of the nervous system, and is integral to healthy pregnancy outcomes. 17.3% of the global population is at risk for zinc deficiency due to dietary inadequacy, though up to 30% of people are at risk in some regions of the world[5].

Hence, micronutrient rich lines can be selected from the existing variation in germplasm of rice. The current study was conducted in a collection of 192 genotypes of diverse origin to assess the variability in iron and zinc

**Author for correspondence*

content in dehusked rice grains for its utilization in micronutrient biofortification program.

Iron and zinc concentrations in rice samples were estimated by colorimetric method[13] or by Atomic Absorption Spectrometry (AAS), X-ray Fluorescence (XRF) and Inductively Coupled Plasma Optical Emission Spectrometry (ICP-OES). Colorimetric method is the qualitative method where as in quantitative methods, AAS and ICP-OES are destructive methods.

The only non destructive method for the estimation of Iron and zinc concentrations is X-Ray florescence. In XRF, the preselected wavelength of incident X-rays expel an electron from the innermost orbit followed by the transfer of one of the electrons from the outermost orbit to innermost orbit leading to release of specific wavelength of X-rays. The energy of the emitted radiation is specific for a particular atom. Therefore, it is simultaneously identified and quantified by the detector.

Different types of X-ray spectrometry are used for analysis of mineral elements. Laboratory bench top Energy Dispersive X-ray Florescence (ED-XRF) is the most commonly used technique because of its precision and rapid and cost effective screening for the estimation of large number of samples[14,15]. Hence this study was proposed to estimate iron and zinc content in a population panel of 192 lines using ED-XRF method.

2. Materials and Methods

2.1 Experimental Site

A set of 192 genotypes were grown in Paddy Breeding Station, Department of Rice, Tamil Nadu Agricultural University, India during Rabi 2013 as few accessions are photosensitive. This area is situated at latitude of 11° N and longitude of 77° E with clayey soil of pH 7.8. The experiment was laid out in randomized complete block design with a spacing of 20 × 20 cm. Normal cultural practices were followed as per standard recommendation.

2.2 Plant Materials

192 accessions of land races and varieties collected from nine different states of India as well as from Argentina, Bangladesh, Brazil, Bulgaria, China, Colombia, Indonesia, Philippines, Taiwan, Uruguay, Venezuela and United States were used in this study (Table 1). The seeds harvested from these lines were dehusked using lab dehusker.

2.3 Iron and Zinc Content Estimation

EDXRF (OXFORD Instruments X-Supreme 8000) was performed in Directorate Rice Research, Hyderabad using an Oxford Instruments X-supreme 8000 which has 10 place auto sampler. Dehusked rice was cleaned for broken and debris and 5g of each sample was weighed and transferred to sample cups. The sample cups were gently shaken for uniform distribution of samples and kept for analysis.

For each set of sample, it has taken 3.1 minutes which included 60s acquisition time for the separate Zn and Fe conditions as well as 66s 'dead time' during which the XRF will establish each measurement condition. Scans were conducted in sample cups assembled from 21 mm diameter and the cup combined with polypropylene inner cups was sealed at one end with 4 μm Poly-4 XRF sample film. Concentration was expressed in microgram per gram (μg/g). The statistical analysis was done using software Past 3.0[16].

3. Results

192 accessions consisting indigenous and exotic lines were analyzed for iron and zinc concentration in brown rice. Iron concentration ranged from 6.6 μg/g to 16.7 μg/g and zinc concentration from 7.1 μg/g to 32.4 μg/g (Figure 1) (Table 2). The mean value of iron in the germplasm lines is 10.1 μg/g and Zinc, 15.4 μg/g. The lowest concentration of iron was recorded in RG1 (Mapillai samba) which is a Tamil Nadu landrace and the lowest zinc in variety RG22 (IR36).

Among the landraces, Nootripathu (RG192) had highest iron content of 13.3 μg/g and the line CHIR8 (RG8) from west Bengal has the highest iron content of 16.7 μg/g. The line RG149 (RH2-SM-2-23) which is a cross between Swarna and Moroberakan has higher zinc content of 16.4 μg/g. Among harvest plus trial lines, RG69 (Bindli, a landrace from Uttarkhand) has higher iron content of 16.3 μg/g. In IRRI germplasm lines, RG75 (Hongjeong X IRGC 73052-1) has higher iron content of 16.1 μg/g (Figure 2).

In zinc estimation, the IRRI germplasm line RG130 (Honduras) has the highest zinc content of 32.4 μg/g. In landraces, vadakathi samba (RG187) has higher zinc content (27.6 μg/g). RPHP 90 (RG131, 182(M) from Andhra Pradhesh) and RPHP 163 (RG183, Seeta sail from West Bengal) have higher zinc content of 28.6 μg/g and 28 μg/g respectively in harvest plus trial lines (Figure 3).

Table 1. Germplasm Accessions used in the study

G. NO.	Genotypes	Parentage	Origin
RG1	Mapillai samba	Landrace	Tamil Nadu, India
RG2	CK 275	CO50 X KAVUNI	Tamil Nadu, India
RG3	Senkar	Landrace	Tamil Nadu, India
RG4	Murugankar	Landrace	Tamil Nadu, India
RG5	CHIR 6	Improved chinsurah	West Bengal
RG6	CHIR 5	Improved chinsurah	West Bengal
RG7	Kudai vazhai	Landrace	Tamil Nadu, India
RG8	CHIR 8	Improved chinsurah	West Bengal
RG9	Kuruvai kalanjiyam	Landrace	Tamil Nadu, India
RG10	Nava konmani	Landrace	Tamil Nadu, India
RG11	CHIR 10	Improved chinsurah	West Bengal
RG12	Vellai chithiraikar	Landrace	Tamil Nadu, India
RG13	CHIR 2	Improved chinsurah	West Bengal
RG14	Jothi	variety	Kerala, India
RG15	Palkachaka	Landrace	Tamil Nadu, India
RG16	Thooyala	Landrace	Tamil Nadu, India
RG17	Chivapu chithiraikar	Landrace	Tamil Nadu, India
RG18	CHIR 11	Improved chinsurah	West Bengal
RG19	Koolavalai	Landrace	Tamil Nadu, India
RG20	Kalvalai	Landrace	Tamil Nadu, India
RG21	Mohini samba	Landrace	Tamil Nadu, India
RG22	IR 36	IR 1561 X IR 24 X Oryza nivara x CR 94	IRRI, Philippines
RG23	Koombalai	Landrace	Tamil Nadu, India
RG24	Tadukan	Landrace	Tamil Nadu, India
RG25	Sorna kuruvai	Landrace	Tamil Nadu, India
RG26	Rascadam	Landrace	Tamil Nadu, India
RG27	Muzhi karuppan	Landrace	Tamil Nadu, India
RG28	Kaatukuthalam	Landrace	Tamil Nadu, India
RG29	Vellaikattai	Landrace	Tamil Nadu, India
RG30	Poongar	Landrace	Tamil Nadu, India
RG31	Chinthamani	Landrace	Tamil Nadu, India
RG32	Thogai samba	Landrace	Tamil Nadu, India
RG33	Malayalathan samba	Landrace	Tamil Nadu, India
RG34	RPHP 125	NDR 2026 (RICHA)	UTTAR PRADHESH
RG35	CK 143	CO50 X KAVUNI	Tamil Nadu, India
RG36	Kattikar	Landrace	Tamil Nadu, India
RG37	Shenmolagai	Landrace	Tamil Nadu, India
RG38	Velli samba	Landrace	Tamil Nadu, India
RG39	Kaatu ponni	Landrace	Tamil Nadu, India
RG40	kakarathan	Landrace	Tamil Nadu, India
RG41	Godavari samba	Landrace	Tamil Nadu, India
RG42	Earapalli samba	Landrace	Tamil Nadu, India
RG43	RPHP 129	Kamad	JAMMU & KASHMIR
RG44	Mangam samba	Landrace	Tamil Nadu, India
RG45	RPHP 105	Moirang phou	MANIPUR
RG46	IG 4 (EC 729639- 121695)	TD2: :IRGC 9148-1	IRRI, Philippines
RG47	Machakantha	Landrace	Orissa, India

Continued

Table 1. (*continued*)

G. NO.	Genotypes	Parentage	Origin
RG48	Kalarkar	Landrace	Tamil Nadu, India
RG49	Valanchennai	Landrace	Tamil Nadu, India
RG50	Sornavari	Landrace	Tamil Nadu, India
RG51	RPHP 134	NJAVARA	Kerala
RG52	ARB 58	Variety	Karnataka
RG53	IR 68144-2B-2-2-3-1-127	IR 72 X ZAWA BONDAY	IRRI, Philippines
RG54	PTB 19	Variety	Kerala, India
RG55	IG 67(EC 729050- 120988)	IR 77384-12-35-3-12-l-B::IRGC 117299-1	IRRI, Philippines
RG56	RPHP 59	Taroari Basmati/karnal local	HARYANA
RG57	RPHP 103	Pant sugandh dhan -17	UTTARKHAND
RG58	Kodaikuluthan	Landrace	Tamil Nadu, India
RG59	RPHP 68	Subhdra	Orissa, India
RG60	Rama kuruvaikar	Landrace	Tamil Nadu, India
RG61	Kallundai	Landrace	Tamil Nadu, India
RG62	Purple puttu	Landrace	Tamil Nadu, India
RG63	IG 71(EC 728651- 117588)	TEPI BORO::IRGC 27519-1	IRRI, Philippines
RG64	Ottadaiyan	Landrace	Tamil Nadu, India
RG65	IG 56 (EC 728700- 117658	BICO BRANCO	Brazil
RG66	Jeevan samba	Landrace	Tamil Nadu, India
RG67	RPHP 106	akut phou	MANIPUR
RG68	IG 63(EC 728711- 117674)	CAAWA/FORTUNA	IRRI, Philippines
RG69	RPHP 48	Bindli	UTTARKHAND
RG70	Karthi samba	Landrace	Tamil Nadu, India
RG71	IG 27(IC 0590934- 121255)	ARC 11345::IRGC 21336-1	IRRI, Philippines
RG72	Aarkadu kichili	Landrace	Tamil Nadu, India
RG73	Kunthali	Landrace	Tamil Nadu, India
RG74	ARB 65	Variety	Karnataka
RG75	IG 21(EC 729334- 121355)	HONGJEONG::IRGC 73052-1	IRRI, Philippines
RG76	Matta kuruvai	Landrace	Tamil Nadu, India
RG77	Karuthakar	Landrace	Tamil Nadu, India
RG78	RPHP 165	Tilak kachari	West Bengal
RG79	Manavari	Landrace	Tamil Nadu, India
RG80	IG 66 (EC 729047- 120985)	IR 71137-243-2-2-3-3::IRGC 99696-1	IRRI, Philippines
RG81	CB-07-701-252	Improved line	Tamil Nadu, India
RG82	Thooyamalli	Landrace	Tamil Nadu, India
RG83	RPHP 93	Type-3 (Dehradooni Basmati)	UTTARKHAND
RG84	Velsamba	Landrace	Tamil Nadu, India
RG85	RPHP 104	Kasturi (IET 8580)	UTTARKHAND
RG86	RPHP 102	Kanchana	Kerala, India
RG87	IG 40 (EC 728740- 117705)	DEE GEO WOO GEN	TAIWAN
RG88	Saranga	Landrace	Tamil Nadu, India
RG89	IR 83294-66-2-2-3-2	DAESANBYEO X IR65564-44-5-1	IRRI, Philippines
RG90	IG 61(EC 728731- 117696)	CRIOLLO LA FRIA	Venezuela
RG91	IG 23(EC 729391- 121419)	MAHA PANNITHI::IRGC 51021-1	IRRI, Philippines
RG92	IG 49(EC 729102- 121052)	MENAKELY ::IRGC 69963-1	IRRI, Philippines
RG93	uppumolagai	Landrace	Tamil Nadu, India
RG94	Karthigai samba	Landrace	Tamil Nadu, India
RG95	Jeeraga samba	Landrace	Tamil Nadu, India

RG96	RP-BIO-226	IMPROVED SAMBHA MAHSURI	ANDHRA PRADESH
RG97	Varigarudan samba	Landrace	Tamil Nadu, India
RG98	IG 5(EC 729642- 121698)	IR 65907-116-1-B::C1	IRRI, Philippines
RG99	IG 31(EC 728844- 117829)	ORYZICA LLANOS 5	Colombia
RG100	IG 7(EC 729598- 121648)	VARY MAINTY::1RGC 69910-1	IRRI, Philippines
RG101	RPHP 52	SEBATI	Orissa, India
RG102	Varakkal	Landrace	Tamil Nadu, India
RG103	Mattaikar	Landrace	Tamil Nadu, India
RG104	IG 53(EC 728752- 117719)	CAROLINA RINALDO BARSANI	URUGUAY
RG105	IG 6(EC 729592- 121642)	SOM CAU 70 A::1RGC 8227-1	IRRI, Philippines
RG106	Katta samba	Landrace	Tamil Nadu, India
RG107	RH2-SM-1-2-1	SWARNA X MOROBERAKAN	Tamil Nadu, India
RG108	Red sirumani	Landrace	Tamil Nadu, India
RG109	Vadivel	Landrace	Tamil Nadu, India
RG110	Norungan	Landrace	Tamil Nadu, India
RG111	IG 20(EC 729293- 121310)	CHIGYUNGDO::IRGC 55466-1	IRRI, Philippines
RG112	IG 35(EC 728858- 117843)	PATE BLANC MN 1	Cote D'Ivoire
RG113	IG 45(EC 728768- 117736)	FORTUNA	Puerto Rico
RG114	RPHP 159	Radhuni Pagal	BANGLADESH
RG115	IG 43(EC 728788- 117759)	IR-44595	IRRI, Philippines
RG116	RPHP 27	Azucena	HARYANA
RG117	IG 65(EC 729024- 120958)	GODA HEENAT1::IRGC 31393-1	IRRI, Philippines
RG118	Ponmani samba	Landrace	Tamil Nadu, India
RG119	Ganthasala	Landrace	Tamil Nadu, India
RG120	Thattan samba	Landrace	Tamil Nadu, India
RG121	IG 74(EC 728622- 117517)	KINANDANG PATONG::IRGC 23364-1	IRRI, Philippines
RG122	Kaliyan samba	Landrace	Tamil Nadu, India
RG123	IG 2(EC 729808-121874)	BLUEBONNET 50::IRGC 1811-1	IRRI, Philippines
RG124	IG 29(EC 728925- 117920)	TOX 782-20-1	NIGERIA
RG125	RPHP 55	Kalinga -3	Orissa
RG126	Kallimadayan	Landrace	Tamil Nadu, India
RG127	IG 10(EC 729686- 121743)	HASAN SERALIRGC 79564-C1	IRRI, Philippines
RG128	IG 75(EC 728587- 117420)	AEDAL::IRGC 55441-1	IRRI, Philippines
RG129	IG 38(EC 728742 - 117707)	DELREX	UNITED STATES
RG130	IG 39(EC 728779- 117750)	HONDURAS	HONDURAS
RG131	RPHP 90	182(M)	Andhra Pradesh
RG132	IG 33(EC 728938- 117935)	WC 3397	JAMAICA
RG133	IG 42(EC 728798- 117774)	KALUBALA VEE	SRILANKA
RG134	IG 9(EC 729682- 121739)	GEMJYA JYANAM::IRGC 32411-C1	IRRI, Philippines
RG135	RPHP 161	Champa Khushi	
RG136	IG 8(EC 729601- 121651)	XI YOU ZHAN::1RGC 78574-1	IRRI, Philippines
RG137	IG 37(EC 728715- 117678)	CENIT	ARGENTINA
RG138	Sigappu kuruvikar	Landrace	Tamil Nadu, India
RG139	RPHP 138	EDAVANKUDI POKKALI	Kerala, India
RG140	Raja mannar	Landrace	Tamil Nadu, India
RG141	IG 44(EC 728762- 117729)	EDITH	UNITED STATES
RG142	Sasyasree	TKM 6 x IR 8	West Bengal
RG143	IG 46(IC 471826- 117647)	BABER	INDIA
RG144	Chetty samba	Landrace	Tamil Nadu, India

Continued

Table 1. (*continued*)

G. NO.	Genotypes	Parentage	Origin
RG145	IG 60(EC 728730- 117695)	CREOLE	Belize
RG146	IR 75862-206	IR 75083 X IR 65600 -81-5-3-2	IRRI, Philippines
RG147	IG 58(EC 728725- 117689)	CI 11011	UNITED STATES
RG148	Chinna aduku nel	Landrace	Tamil Nadu, India
RG149	RH2-SM-2-23	SWARNA X MOROBERAKAN	Tamil Nadu, India
RG150	IG 14(IC 517381- 121422)	MALACHAN::IRGC 54748-1	IRRI, Philippines
RG151	IG 32(EC 728838- 117823)	NOVA	United States
RG152	RPHP 47	Pathara (CO-18 x Hema)	India
RG153	Sembilipiriyan	Landrace	Tamil Nadu, India
RG154	IG 48(EC 729203- 121195)	DINOLORES::IRGC 67431-1	IRRI, Philippines
RG155	Sona mahsuri	Landrace	Tamil Nadu, India
RG156	IG 12(EC 729626- 121681)	SHESTAK::IRGC 32351-1	IRRI, Philippines
RG157	Karungan	Landrace	Tamil Nadu, India
RG158	IG 13(EC 729640- 121696)	CURINCA::C1	IRRI, Philippines
RG159	Sembala	Landrace	Tamil Nadu, India
RG160	IG 72(EC 728650- 117587)	TD 25::IRGC 9146-1	IRRI, Philippines
RG161	Panamarasamba	Landrace	Tamil Nadu, India
RG162	IR 64	IR-5857-33-2-1 x IR-2061-465-1-5-5	IRRI, Philippines
RG163	Mikuruvai	Landrace	Tamil Nadu, India
RG164	Thillainayagam	Landrace	Tamil Nadu, India
RG165	ARB 64	Variety	Karnataka
RG166	RPHP 140	VYTILLA ANAKONDAN	Kerala
RG167	IG 70(EC 729045- 120983)	IR43::1RGC 117005-1	IRRI, Philippines
RG168	Haladichudi	Landrace	Orissa, India
RG169	IG 24(EC 728751- 117718)	DNJ 140	BANGLADESH
RG170	RPHP 42	Shalimar Rice -1	JAMMU & KASHMIR
RG171	RPHP 44	BR- 2655	KARNATAKA
RG172	IG 25(EC 729728- 121785)	LOHAMBITRO 224::GERVEX 5144-C1	IRRI, Philippines
RG173	IG 73(EC 728627- 117527)	MAKALIOKA 34::1RGC 6087-1	IRRI, Philippines
RG174	IG 51(EC 728772- 117742)	GOGO LEMPUK	Indonesia
RG175	Vellai kudaivazhai	Landrace	Tamil Nadu, India
RG176	Kodai	Landrace	Tamil Nadu, India
RG177	Kallundaikar	Landrace	Tamil Nadu, India
RG178	IG 17(EC 728900- 117889)	SIGADIS	INDONESIA
RG179	Avasara samba	Landrace	Tamil Nadu, India
RG180	IG 59(EC 728729- 117694)	COPPOCINA	BULGARIA
RG181	IG 52(EC 728756- 117723)	DOURADO AGULHA	BRAZIL
RG182	ARB 59	Variety	Karnataka
RG183	RPHP 163	Seeta sail	West Bengal
RG184	IG 18(EC 728892- 117880)	SERATOES HARI	INDONESIA
RG185	RPHP 36	TKM-9	Tamil Nadu, India
RG186	IG 28(EC 728920- 117914)	TIA BURA	INDONESIA
RG187	Vadakathi samba	Landrace	Tamil Nadu, India
RG188	RPHP 80	24(K)	Andhra Pradesh
RG189	IG 41(EC 728800- 117776)	KANIRANGA	Indonesia
RG190	IG 26(IC 0590943- 121899)	BASMATI 370::IRGC 3750-1	IRRI, Philippines
RG191	IG 15(EC 728910- 117901)	SZE GUEN ZIM	CHINA
RG192	Nootri pathu	Landrace	Tamil Nadu, India

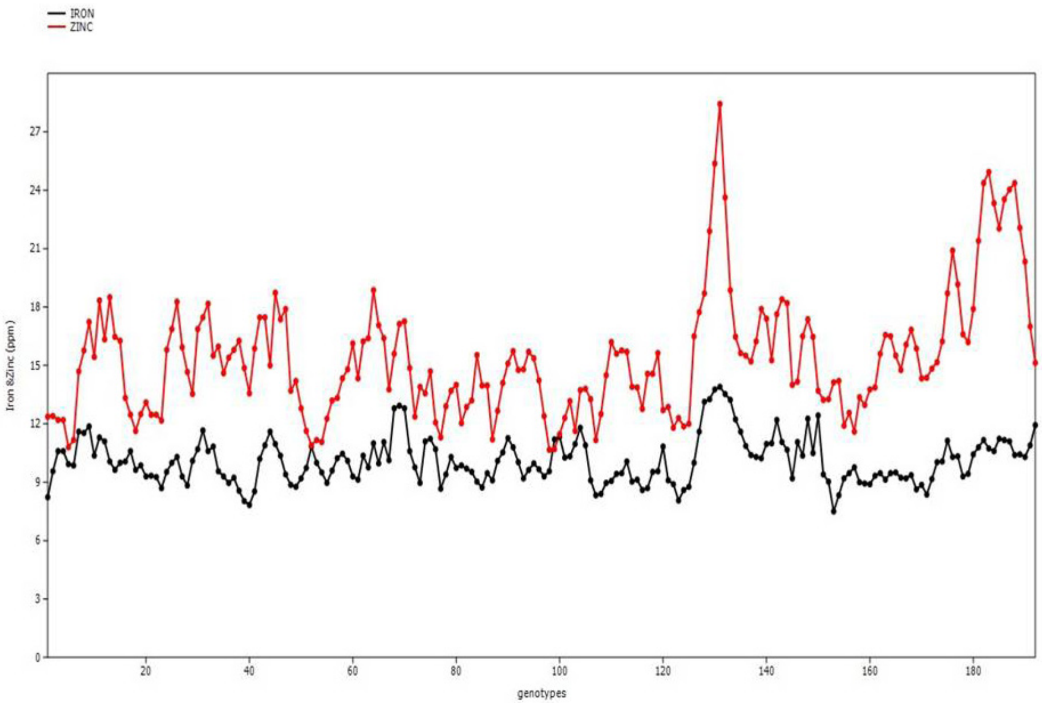

Figure 1. Iron and Zinc content in 192 genotypes.

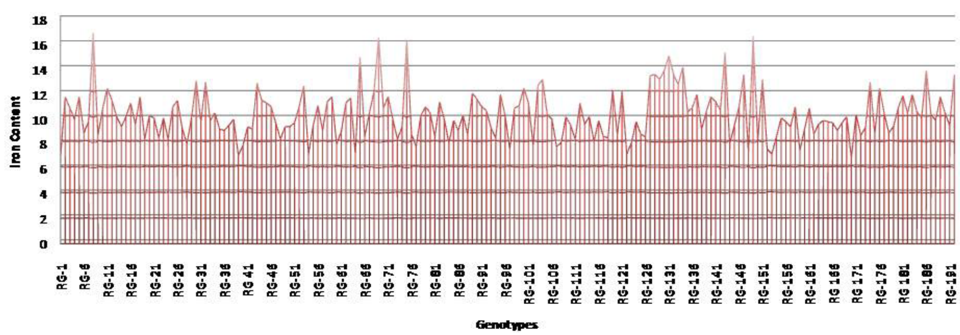

Figure 2. Variation in Iron content.

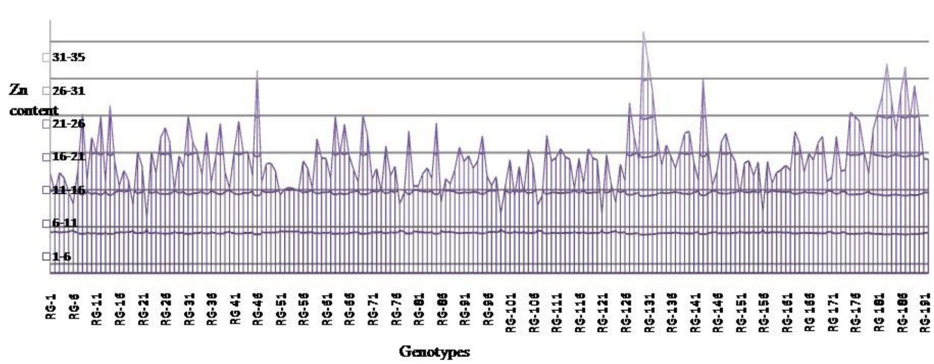

Figure 3. Pattern of Zinc Content in germplasm.

Table 2. Iron and Zinc content in Germplasm lines

S. No	Variety name	Iron (μg/g)	Zinc (μg/g)
1	RG-1	6.6	13.2
2	RG-2	11.5	10.7
3	RG-3	10.6	13.3
4	RG-4	9.7	12.6
5	RG-5	11.5	10.7
6	RG-6	8.6	9.1
7	RG-7	9.5	13.7
8	RG-8	16.7	21.3
9	RG-9	8.4	12.3
10	RG-10	10.5	18.1
11	RG-11	12.2	15.9
12	RG-12	11.2	21
13	RG-13	9.9	12.1
14	RG-14	9.1	22.4
15	RG-15	9.9	14.9
16	RG-16	11	11.5
17	RG-17	9.3	13.6
18	RG-18	11.5	12.3
19	RG-19	8.1	9
20	RG-20	10	16.2
21	RG-21	9.8	14.1
22	RG-22	8.2	7.1
23	RG-23	9.8	16.2
24	RG-24	8.1	13.2
25	RG-25	10.7	18
26	RG-26	11.2	19.4
27	RG-27	9	17.4
28	RG-28	7.7	11
29	RG-29	9.8	15.6
30	RG-30	12.8	14
31	RG-31	9.5	21
32	RG-32	12.7	17.4
33	RG-33	9.6	16.1
34	RG-34	10.2	13
35	RG-35	8.9	18.8
36	RG-36	8.8	12
37	RG-37	9.2	15.4
38	RG-38	9.7	20
39	RG 39	6.8	13.4
40	RG 40	7.6	11.2
41	RG 41	9.1	16.1
42	RG 42	8.9	20.3
43	RG 43	12.6	16
44	RG 44	11.2	16.1
45	RG 45	11	12.9
46	RG-46	10.7	27.2
47	RG 47	9.4	12
48	RG 48	8.1	14.5
49	RG-49	9.1	14.6
50	RG-50	9.1	13.5
51	RG-51	9.4	10.3
52	RG-52	10.7	11.1
53	RG-53	12.4	11.3
54	RG-54	6.9	11.1
55	RG-55	9.2	10.8
56	RG-56	10.8	14.9
57	RG-57	8.8	13.9
58	RG-58	11.1	11.2
59	RG-59	11.5	17.9
60	RG-60	7.7	15.3
61	RG-61	8.7	15.2
62	RG-62	11	12.5
63	RG-63	11.4	21
64	RG-64	6.9	15.7
65	RG-65	14.7	19.9
66	RG-66	8.3	15.6
67	RG-67	10.2	13.7
68	RG-68	11.9	12
69	RG-69	16.3	21.1
70	RG-70	10.6	18.3
71	RG-71	11.5	12.4
72	RG-72	9.7	13.9
73	RG-73	8.1	10.8
74	RG-74	9.1	17
75	RG-75	16.1	12.9
76	RG-76	8.5	14.2
77	RG-77	7.5	9.1
78	RG-78	10	10.6
79	RG-79	10.7	19
80	RG-80	10.2	11.5
81	RG-81	8.3	11.5
82	RG-82	11.1	13.1
83	RG-83	9.7	14
84	RG-84	7.8	12.5

85	RG-85	9.6	20.1
86	RG-86	8.8	9.3
87	RG-87	10	12.5
88	RG-88	8.5	11.8
89	RG-89	11.8	13.7
90	RG-90	11.3	16.8
91	RG-91	10.7	14.8
92	RG-92	10.4	15.6
93	RG-93	9	13.9
94	RG-94	8.2	14.9
95	RG-95	11.7	18.3
96	RG-96	10	12.9
97	RG-97	7.3	11.5
98	RG-98	10.6	12.8
99	RG-99	10.8	7.7
100	RG-100	12.2	11.6
101	RG-101	11	15.1
102	RG-102	7.6	10.2
103	RG-103	12.4	14.2
104	RG-104	12.9	10.5
105	RG-105	10.1	16.5
106	RG-106	9.7	14.4
107	RG-107	7.5	8.9
108	RG-108	7.8	10.2
109	RG-109	9.9	18.4
110	RG-110	9.2	14.9
111	RG-111	8.1	15.3
112	RG-112	11	16.6
113	RG-113	9.3	15.4
114	RG-114	9.9	15.1
115	RG-115	7.9	11.2
116	RG-116	9.6	15.3
117	RG-117	8.3	11.8
118	RG-118	8.2	16.6
119	RG-119	12.1	15.3
120	RG-120	8.4	15
121	RG-121	12	7.8
122	RG-122	6.9	15.8
123	RG-123	7.8	11.8
124	RG-124	9.5	9.3
125	RG-125	8.5	14.5
126	RG-126	8.3	12.2
127	RG-127	13.2	22.8
128	RG-128	13.3	18.2
129	RG-129	12.9	15.1
130	RG-130	13.6	32.4
131	RG-131	14.8	28.6
132	RG-132	13.3	24.3
133	RG-133	12.5	18
134	RG-134	13.9	14.3
135	RG-135	10.3	17.1
136	RG-136	10.6	15.5
137	RG-137	11.7	13.9
138	RG-138	8.9	16.2
139	RG-139	10.3	18.6
140	RG-140	11.5	18.9
141	RG-141	11.1	14.7
142	RG-142	10.4	12.2
143	RG-143	15.1	26
144	RG-144	7.7	17
145	RG-145	9.2	11.6
146	RG-146	10.7	13.4
147	RG-147	13.3	17.5
148	RG-148	7.1	18.6
149	RG-149	16.4	16
150	RG-150	8	14.8
151	RG-151	12.9	10.3
152	RG-152	7.3	14.6
153	RG-153	6.9	14.9
154	RG-154	8.3	12.9
155	RG-155	9.8	14.8
156	RG-156	9.5	8
157	RG-157	9.1	14.9
158	RG-158	10.7	11.9
159	RG-159	7.2	13.3
160	RG-160	8.9	13.7
161	RG-161	10.6	14.3
162	RG-162	8.5	13.6
163	RG-163	9.3	18.9
164	RG-164	9.6	17.2
165	RG-165	9.5	13.4
166	RG 166	9.4	15.9
167	RG 167	8.8	15
168	RG 168	9.4	17.3
169	RG 169	9.9	18.2

Continued

Table 2. (*continued*)

S. No	Variety name	Iron (μg/g)	Zinc (μg/g)
170	RG 170	6.6	12.1
171	RG 171	10.1	12.7
172	RG 172	8.4	18.3
173	RG 173	9	13.5
174	RG 174	12.7	13.7
175	RG 175	8.5	21.5
176	RG-176	12.2	20.9
177	RG 177	10.2	20.3
178	RG 178	8.6	16.3
179	RG 179	9.1	13.2
180	RG 180	10.6	19.1
181	RG 181	11.6	21.4
182	RG 182	10.2	23.7
183	RG 183	11.7	28
184	RG 184	10.3	23.1
185	RG 185	9.8	18.9
186	RG-186	13.6	24.1
187	RG-187	10.1	27.6
188	RG-188	9.6	20.4
189	RG-189	11.5	25.1
190	RG-190	10.2	20.7
191	RG-191	9.2	15.2
192	RG-192	13.3	15.1

The genotype RG131 has higher iron and zinc content (14.8 μg/g & 28.6 μg/g). Few other genotypes RG127, RG130, RG132, RG143 and RG186 have higher iron as well as zinc content.

The ellipse shows the area where 95 % of the total genotypes falls based on its iron and zinc content. Convex hull indicates the genotypes which have extreme variation for iron and zinc content (Figure 4). A positive correlation (☐ - +0.364) was observed between iron and zinc contents of 192 genotypes indicated the possibility of simultaneous effective selection for both the micronutrients. Few of the commercially cultivated rice cultivars included in this study are deficient in iron and zinc compared to the other staple food crops such as wheat and maize.

Based on the iron and zinc content, these 192 genotypes can be classified into three categories, low, moderate and high. For iron content, the genotypes(58 genotypes) with the iron content of 0–9 μg/g was considered in low category, iron content from 9.1 to 12 μg/g were grouped in moderate (105 genotypes) and more than 12 μg/g (29 genotypes) were placed in high category. (Figure 5a). The genotypes with the zinc content of 0–12 μg/g (40 genotypes) was categorized under low zinc content, 126 genotypes with the zinc content from 12.1 to 20 μg/g was grouped in moderate category and the genotypes (26 genotypes) with more than 20 μg/g to 32.4 μg/g was placed in high category (Figure 5b).

4. Discussion

Iron and Zinc deficiency is probably the most widespread micronutrient deficiencies in cereals. Since rice is the staple food for more than 50% of the population especially in developing countries, a lot of efforts are being made to enrich the nutritional status of rice to prevent malnutrition. Many researchers have studied the feasibility of breeding for enhancing bio-available micro nutrients in grains by increasing the concentrations of metal-binding proteins[17].

The first pre-requisite for initiating a breeding program to develop micronutrient-rich genotypes is to screen the available germplasm and identify the source of genetic variation for the target trait, which can be used in crosses, genetic studies, molecular marker development and to understand the basis of micronutrient uptake process. Iron and Zinc contents in grains also depend on the micronutrient uptake and translocation efficiency from root to grains.

A large genetic variation for grain iron and zinc has been observed in different germplasm of rice and maize and it was exploited in breeding programs[18]. Gregorio[19] has screened rice lines in IRRI germplasm for high iron and zinc content. Among a subset of 1,138 samples analyzed, iron concentrations ranged from 6.3 to 24.4 μg/g, and for zinc, the range was 15.3–58.4 μg/g[19]. Traditional varieties Jalmagna and Zuchen contained almost twice as much iron and 50% more zinc compared to widely grown varieties, IR36 and IR64. Forty six cultivated and wild accessions of rice was screened for grain Fe and Zn and found that wild rice accessions have higher grain Fe and Zn content than cultivars[12]. In the current study, iron and zinc contents in dehusked grains varied from 6.6 μg/g to 16.7 μg/g and 7.1 μg/g to 32.4 μg/g respectively.

Brar[9] has surveyed in 220 rice genotypes for Fe and Zn content and reported that *indica* and aromatic rice varieties have high Fe and Zn content[9]. Anandan[20] have shown that traditional rice cultivars have high Fe and Zn content than improved cultivars[20]. A similar result was obtained in

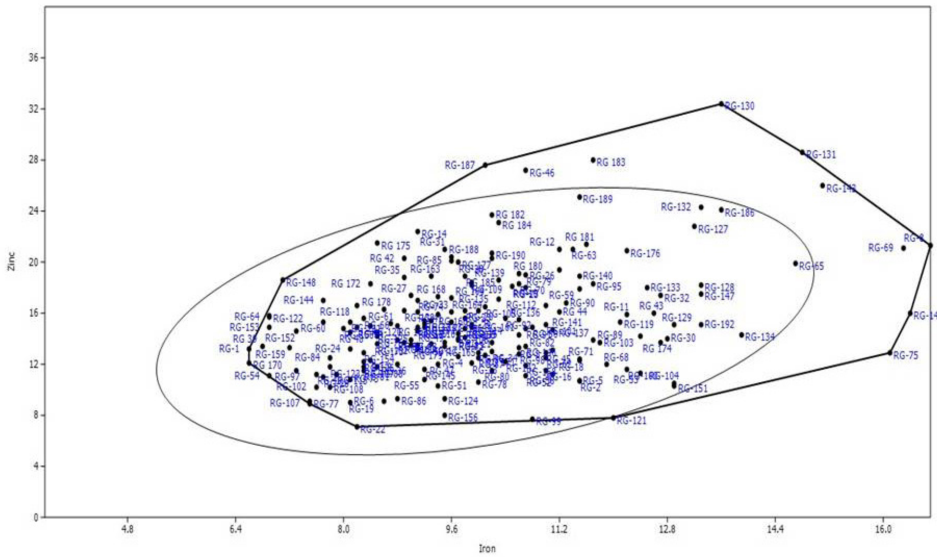

Figure 4. Scatter diagram showing variation for iron and zinc in 192 genotypes.

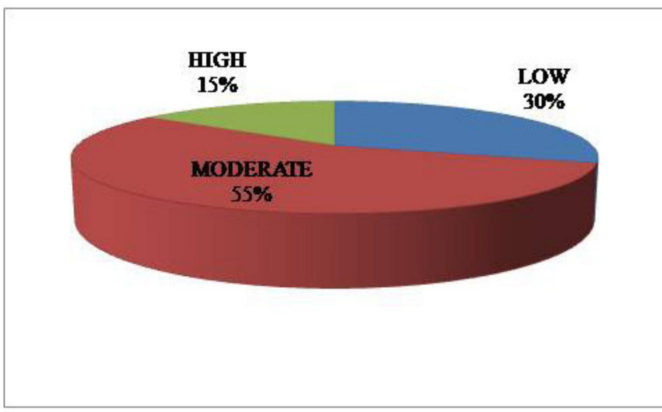

Figure 5a. Genotypes classification based on Iron content.

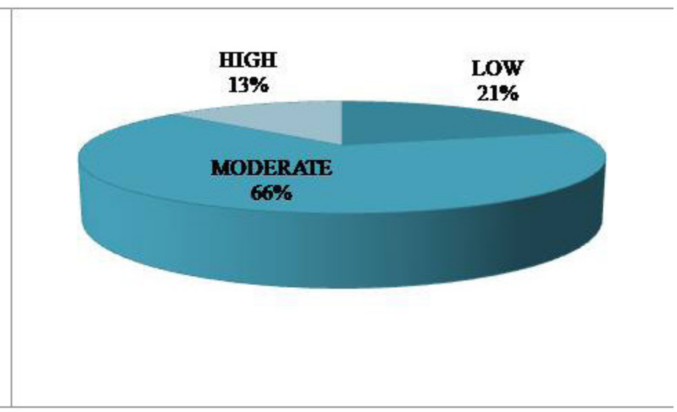

Figure 5b. Genotypes classification based on Zinc content.

the current study in which traditional cultivars has higher iron and zinc content compared to commercial cultivars.

Roy and Sharma[11] has screened 84 landraces for iron and zinc content. Iron content varied between 0.25 µg/g to 34.8 µg/g and Zinc content from 0.85 µg/g to 195.3 µg/g. Local cultivar Swetonunia had highest iron content of 34.8 µg/g followed by the other cultivars Gobindobhog 3.1 µg/g, and Attey 2.05 µg/g. Nepali Kalam had the highest Zinc content 195.3 µg/g followed by Govindobhog 138.6 µg/g, Begunbeej 20.4 µg/g and Ghiosh16.15 µg/g. In our study, CHIR8 (RG8) has the highest iron content of 16.7 µg/g and RG130 (Honduras) has the highest zinc content of 32.4 µg/g. The landrace Nootripathu (RG192) had higher iron content of 13.3 µg/g and Vadakathi Samba (RG187) has higher zinc content (27.6 µg/g).

Previous studies and present study has indicated that there is no consistent value of iron and zinc for a genotype. The variation depends on different factors such as micronutrient homeostasis, sampling method, grain nature, soil properties, analytical methods, environment, genotype and genotype X environment interaction[8].

Though the variation in micronutrient content depends on several factors, the germplasm stock in rice has sufficient variation to exploit for developing stable lines[18,21]. As micronutrient malnutrition poses a significant global challenge, the development of micronutrient enriched genotypes serves as the need of the hour. The extreme genotypes identified in this study will be useful for selecting and breeding lines with enriched micronutrient status.

5. Conclusion

Existing genetic variation in rice germplasm offers scope for developing nutrient rich rice varieties. Notably, there was about multiple fold difference in Fe and Zn content suggesting the existence of genetic potential to increase the concentration of these micronutrients in rice grain. Micronutrient-rich and poor rice genotypes identified in this study may be used in breeding program for the identification of genomic regions or QTLs responsible for mineral uptake and translocation and it can be used as donor for developing nutrient enriched varieties.

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