

Characterization of Lowland Rice Genotypes (F₆₋₇) for Variability in Agronomic and Morphological Traits in Iron-Toxic Environment

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Abstract – One of the most devastating abiotic stresses in the Inland Valley Swamps (IVS) for the production of rice is iron toxicity. Indigenous rice farmers find it very difficult to cope and manage these constraints in the IVS like; prolong flooding, pest and diseases. Reports indicated that in W/Africa, rice yield reduced as much as 12-100% as a result of iron toxicity.

This investigation was conducted at the Rokupr Agricultural Research Center, in one of the rain-fed IVS swamps proven to be iron toxic, during the 2016 wet season. The planting materials consisted of 58 entries of F₆₋₇ population and with three lowland check varieties which possess diverse traits of (susceptibility and tolerance) to iron toxicity, RYMV and Rice Blast. Due to iron excess effect, six of the genotypes tested failed to survive in the permanent field after the seedlings had been transplanted.

Results showed that 59.3% of the total 61 genotypes investigated had grain yield lower than the grand mean, which indicated drastic grain yield reduction. Impressively, G26 produced significantly the highest grain yield component than Rok-24 (tolerance check) and most importantly, 40.7% of the genotypes produced higher grain yield and yield component above the grand mean and more than the (susceptible checks) and the other genotypes. Stress parameters score rating classified G5, G7, G9, G12, G14, G18, G26, G37, G454, and G55 as significantly tolerant to iron toxicity.

The evaluated rice genotypes exhibited a considerable morphological variability. However, the dendrogram produced six cluster groups where cluster (I), (II) and (III) consist 15, 9 and 3 genotypes respectively which produced the best trait performance of all the clusters. Meanwhile, genotypes identified as high iron toxicity tolerant with better yield and agronomic performance should be re-conducted in multi-location field trails in iron toxic environments to confirm the conclusions before making concrete recommendations.

Keywords – Genotype, Inland Valley Swamp, Rice Mottle Virus Disease, Iron, Rokupr Rice Research Center.

I. INTRODUCTION

The quickest growing basic food in most parts of Africa is rice where Sierra Leone is no exemption. A little more than half of the world's population depends on rice for its prime daily source of food energy and protein and therefore the value of rice in relation to food security and socio-economic stability is obvious [1].

Sierra Leone naturally comprised of five agro-ecological zones, (Upland, River-rain, Boli, Inland Valley Swamp and Mangrove Swamp). The Inland Valley Swamp is distributed in the three provinces and the western area of the country. The cultivated area for lowland rice is estimated to

be around 128 million hectare of irrigated and rain-fed lowland in West Africa [2]. Moreover, rice as a cereal crop is regarded extensively as the staple food for the inhabitants of Sierra Leone and most other parts of Africa. It has to be intensively cultivated to meet with the growing demand and terrific growth rate of the world's population. Rice is widely grown almost by all farmers in Sierra Leone and agriculture largely account for the employment of the country's low earning population. Farmers in the interior lowland and some part of the coastline, depend on the IVS ecology for their rice farming activities.

The yield produced from the IVS ecology is relatively higher (37%) than the other agro-ecologies under improve agronomic or even under traditional agronomic management practices [3], [4], [5], although much of the available Inland Valley Swamp is not cultivated. The ecology is utilized for different crop cultivation both in the rainy season and in the dry season. It has been used for rice cultivation in the rainy season and for vegetable production in the dry season in most parts of the country, unlike the other agro-ecologies.

The Inland Valley Swamp as a high yielding agro-ecology is faced with many agronomic and socio-economic constraints. Indigenous rice growing farmers find it very difficult to cope and manage these constraints in the IVS such as; iron toxicity, prolong flooding, pest and diseases, high weed infestation and little or no fertilizer application. Further exploration of the lowland ecology is constrained chiefly by iron toxicity [6]. Iron toxicity, RYMV and Rice Blast are major problems in rice cultivation in Sierra Leone. However, iron (Fe) is an essential element in plants that is involved in many physiological processes, but that can also be toxic when occurred in surpluses. Iron excesses is found mainly in waterlogged or flooded soils where anaerobic conditions occur, may be in some irrigated rice fields in the country. Fe³⁺ is readily reduced to more soluble Fe²⁺ under these conditions. Fe is present as ferric hydroxides with low plant availability [7]. Report by [8], said that iron toxicity caused reduced plant height and tillering, while loss of chlorophyll, leaf discoloration and reduced root growth was reported by [9]. One possible illumination for this is the opposed effect of iron uptake on the availability of other essential nutrients like potassium, zinc and manganese, which could cause nutritional disorder. The other effect is eliciting reactive oxygen radical and the accumulation of oxidized polyphenol, with their cytotoxic effect on macro molecules, such as peroxidation of lipids, denaturation of proteins and DNA [10], leading to a disruption in the

cellular structural organization. Iron toxicity alone has made many hectares of Inland Valley Swamps remain useless for rice cultivation.

Furthermore, diseases are among the most considerable limiting factors that affect rice production, causing annual yield losses conservatively estimated at 5% [11]. RYMV is the common way of abbreviating Rice Yellow Mottle Virus and is a plant virus disease. It is endemic in Africa, which was first detected in Kenya in 1966. RYMV was first picked up in West Africa particularly, Sierra Leone in 1975. By 1990, it had been recorded in all West and Central African countries, excluding Mauritania. It has turned into a principal problem in irrigated rice systems, for the last 20 years or so, chiefly in Burkina Faso, Côte d'Ivoire, Mali and Niger, and in lowlands in Burkina Faso, Côte d'Ivoire, Senegal and Sierra Leone. It will, however, attack rice in any lowland situation [12]. The severe nature of rice yellow mottle disease poses a serious threat to the cultivation of susceptible rice in the wetlands of Africa. The disease mainly affects irrigated rice production of lowland rice in sub-Saharan countries [13], [14]. More than Seventy diseases caused by fungi, bacteria, viruses or nematodes have been documented on rice [15], among which rice blast (*Magnaporthe grisea*) is the most serious constraint on rice productivity [11].

Resistant cultivars and the application of pesticides have been used for blast control. The first objective of this study is to identify high yielding genotypes with implausible agronomic performances and secondly, to characterize genotypes for agronomic and variability of traits in environment with iron toxicity.

II. MATERIALS AND METHODS

Description of Experimental Area.

Experimental Site

The investigation was conducted at the Rokupr Agricultural Research Center, formally Rice Research Station Rokupr, in one of the IVS swamps which was proven as a hot spot for iron toxicity. Rokupr where the experimental site is found, is located on the earth/ globe at latitude 9°01' North and longitude of 12°57' with an elevation of 7.92 meters above sea level. The town lies at the river bank of the Great Scarcies River, in Magbema Chiefdom, Kambia District, north-west Serra Leone. This locality has an average annual rainfall of 3000mm, and its distribution is 85 percent from June – October. This trial experiment was conducted during the 2016 wet season. Soil sample from the experimental site was collected for nutrient or chemical analysis. The soil physical and chemical properties are indicated in table 1 below.

Table 1. Mango Stan IVS swamp soil characteristics.

Soil Characteristics	Analyzed Result (%)
pH (Per Hydrogen)	4.03
Electric Conductivity (mS/cm)	1.2
Texture	Clay Loam
Total Nitrogen (mg/kg soil)	1.2
Available Phosphorus (P ⁺) (mg/kg soil)	5.2
Available Potassium (K ⁺) (ppm)	13.2
Available Carbon	1.12%

Soil Characteristics	Analyzed Result (%)
Available iron (mg/kg soil)	650

In Kambia District, the rainfall pattern is not evenly distributed with the year clearly divided into the rainy and dry seasons. The rainy season begins in April/May and ends in November/ December that is, lasting for six-seven months in a year. Rainfall amount ranges from 5000 mm along the coast to 2000 mm in the north away from the coast in the district. The dry season begins in November/ December and ends in March/ April. The average temperature ranges from 19.7 °C in the dry harmattan season in December/ January to 32.3°C at the peak temperatures in March. This area experiences a remarkable relative humidity ranging from 50% in March to 90% in August [16].

Planting Materials, Trial Design, Fertilizer Application, Treatment and Planting.

Source of Plant Materials

The experimental planting materials consist of 58 entries of F₆₋₇ populations and three lowland check varieties with diverse traits, (susceptible and tolerance) to iron toxicity, RYMV and Blast. Table two shows the parentage of planting materials used in the experiment. These planting materials were sourced from the prominent Rokupr Rice Research Center (RARC). Consequently, the conventional breeding method of spikelet emasculation was used to develop these lines. Germination test was conducted to ascertain the germination percentage of F₆ planting materials which proved to be as high as 95 percent germination. All the planting materials (lines) were primed in separate appropriate containers in order to hasten their germination rate, via soaking them in water for twelve hours. Rok-24 used as a parent of the lines evaluated in this study as planting materials have been proven to be tolerant to iron toxicity after it test at different iron toxic locations in farmers' fields in the Kambia, Port Loko, Moyamba, Bo and Kenema Districts by Rice Research Station, Rokupr [17]. Rok-24 is a progeny from one of the parents (*O. glaberrima*). Consequently, *O. glaberrima* constitutes a rich reservoir of adaptive traits essential for new rice varieties for Africa. It has sources of resistance to African rice gall midge [18], for weed competitiveness [19], for tolerance to iron toxicity [20], and for drought tolerance [21] - [23]. *O. glaberrima* is also tolerant to RYMV 2 and 1[24].

Table 2. Parentage, objectives of crosses made in 2010, and number of F₅₋₆ plants selections per cross.

Parents [(♀) / (♂)]	Objective	No. of Hills selected
ROK24/ROK32	To obtain Fe-Tox plant types with good grain.	28 (28plots-2360 hills)
ROK24/Cisadane	To obtain Fe-Tox plant types with good grain that are Resistant to AFRGM.	13 (13plots-1560 hills)
ROK24/ROK36	To obtain Fe-Tox plant types with good grain and aroma.	19 (19plots-2280 hills)
ROK30/WAB 450-I-B-P-32-HB	To obtain early, blast resistant plant types with good grain.	10 (10plots-1200 hills)
Total		70

Field Experiment

The F₆ seeds were nursed for 21 days in the upland and transplanted to the lowland field (IVS), in Alpha Lattice Design in three replications. Plant spacing of 20 by 20 cm between - row and within - row respectively at a density of a single plant per hill in four blocks was implemented. A plot size was measured 60cm by 200cm, 40cm between plots and 60cm between blocks. The recommended fertilizer rate of 80-60-60- Kg/ha- N-P₂O₅-K₂O was applied: 60-60-60 kg/ha applied two weeks after seedlings transplanted and was top-dressed with 30kg/ha N at panicle initiation. Weeds were controlled by manual method (hand weeding).

Data Collection

Iron-toxicity responses were scored by subjective visual assessment of Fe-toxicity symptoms on fully expanded leaves (bronzing symptoms) for the entire plant and expressed as percentage leaf area affected. The scoring system used was, the Standard Evaluation System for scoring leaf blast (*Pyricularia oryzae*) lesions and rice yellow mottle virus provided by the International Rice Research Institute [25] was adapted for Fe toxicity as follows:

Rice yellow mottle virus; 1 = No symptom observed, 3 = Leaves green but with sparse dots or streaks and less than 5% of height reduction, 5 = Leaves green or pale green with mottling and 6% to 25% of height reduction, flowering slightly delayed, 7 = Leaves pale yellow or yellow and 26-75% of height reduction, flowering delayed and 9 = Leaves turn yellow or orange, more than 75% of height reduction, no flowering or some plants dead.

Blast; 0 = No lesions observed, 1 = Small brown specks of pin-point size or larger brown specks without sporulating center. 2 = Small roundish to slightly elongated, necrotic gray spots, about 1-2 mm in diameter, with a distinct brown margin, 3 = Lesion type is the same as in scale 2, but a significant number of lesions are on the upper leaves, 4 = Typical susceptible blast lesions 3 mm or longer, infecting less than 4% of the leaf area, 5 = Typical blast lesions infecting 4-10% of the leaf area, 6 = Typical blast lesions infection 11-25% of the leaf area, 7 = Typical blast lesions infection 26-50% of the leaf area, 8 = Typical blast lesions infection 51-75% of the leaf area and many leaves are dead and 9 = More than 75% leaf area affected.

Iron score; 0 = Growth and tillering nearly normal, 1 = Growth and tillering nearly normal; reddish-brown spots or orange discoloration on tips of older leaves, 3 = Growth and tillering nearly normal; older leaves reddish-brown, purple, or orange yellow, 5 = Growth and tillering retarded; many leaves discolored, 7 = Growth and tillering ceases; most leaves discolored or dead and 9 = Almost all plants dead or dying [25].

Morphological traits were measured at the physiological maturity stage taking on individual plants. Plant heights from ten tagged hills, number of tillers from five randomly selected hills, number of panicles per hill from five randomly selected hills per plot were recorded. Number of grains per panicle from five randomly sampled panicles per plot was determined; number of fertile grains per panicle, and number of unfilled grains per plant was counted and determined separately after harvesting. Grain yield per plot

(kilogram/hectare) and 1000 grain weight was determined in grams after harvesting. Days to 50 percent flowering, and days to physiological maturity of each line was recorded. However, the grain yield was determined for each plot in the experiment. Soil sample from the experimental site was collected for nutrient or chemical analysis.

Data Analysis

Statistical data Analysis was performed with all experimental data using the package from International Rice Research Institute (IRRI) - Statistical Tool for Agricultural Research (STAR), whereby Mean performance of genotypes, dendrogram, euclidean clustering of genotypes using Ward's method, Cluster Means and Correlation of Traits analyses were carried out.

III. DISCUSSION

Crop improvement via selection can be successful based on having an in-depth knowledge in the relationship between the rice crop traits and its yield which have direct effect on the overall yield outcome and therefore it is significant to be exploited in the identification of such characters for breeding activities.

Iron is an essential element critical for plant life [26]. It is a critical component of proteins and enzymes, also in basic biological processes such as photosynthesis, chlorophyll synthesis, respiration, nitrogen fixation and uptake [27], and the synthesis of the DNA through the action of the ribonucleotide reductase [28], but when it is available in excess then growth and good yield productivity face bottlenecks.

However, growing rice plant in natural soil conditions exhibited diversity in all rice parameters. The difference in genetic potential in response to growth environment due to high dissolved iron caused the diversity [29] - [31]. Cultivation in the lowland (IVS) ecology is constrained chiefly by iron toxicity [6]. Available literature indicated that in West Africa, rice yield is reduced by as much as 12-100 percent as a result of iron toxicity [32] - [34].

In agreement with such observation, the mean grain yield in the present investigation recorded was 915.1kg/ha, with 59.3% of the total 61 genotypes investigated in this study had grain yield lower the grand mean, this indicated drastic grain yield reduction. Iron toxicity is one of the chief yield limiting factors in low lands by inhibiting plant growth and root development, which influences the uptake and retention of nutrients. This yield reduction displays depressing affects and also expresses how iron toxicity is devastating on rice production in lowlands. Reference [35], [36], also emphasized that, the severity of iron toxicity in lowland ecology is dependent on the variety, stage of crop growth and the soil nutrient status.

The impact of iron toxicity in rice crop may be minimized significantly by the improvement of soil, water and nutrient management practices [37] (a); [38]. Moreover, it remains very expensive for peasant farmers in countries of the developing world (such as Sierra Leone), who stand to be reluctant to adopt other iron toxicity correction practices. The promising approach to improve the yield under iron toxicity and disease conditions is the selection of more

resistant and/or tolerant rice cultivars [39] - [41]. Thus the development of iron toxic tolerant rice genotypes may be the only alternative affordable to many small scale and poor farmers in West Africa and therefore, improved tolerance to iron toxicity is a significant breeding objective to aid small scale and poor farmers in West Africa.

There is considerable evidence that yield potentials contributes to yield under iron toxic circumstances. The present study revealed that G26 produced significantly the highest grain yield component than Rok-24 (improved tolerance check) and most importantly, the mean grain yield recorded was 915.1 kg/ha, with 40.7% of the genotypes had higher grain yield and yield component above the grand mean than the (susceptible checks) Rok-5, L-20 and the other genotypes. Comparably, most of the genotypes from results gave yield as high as the iron tolerance check Rok 24. The Rok-24 rice crop variety possess the characteristics of being very adaptable to the IVS ecology, very good tillering ability, tolerant to iron toxicity, resistant to Lodging and Fairly resistant to stem borer/reaction Lambert Delimini, 2012. Stress parameters were rated using visual observation recommended by IRRI, SES revealed that G5, G7, G9, G12, G14, G18, G26, G37, G454, and G55 were observed with a score of 3 implying - growth and tillering nearly normal; older leaves were found to be reddish-brown, purple or orange yellow and therefore exhibited significantly high tolerance to iron toxicity. Whereas G1, G3, G13, G15, G16, G17, G19, G20, G22, G24, G25, G27, G28, G30, G31, G32, G33, G34, G35, G36, G38, G40, G41, G42, G43, G46, G47, G48, G49, G51, G52, G53, G56, G57 and NERICA-L20 were observed with a score of 5 indicating - growth and tillering retarded; many leaves were observed discolored consequently, produced moderate tolerance to iron toxicity similar to the tolerant check Rok-24 to iron toxicity. The present outcome is consistent with Africa Rice Center [42], which reported that amongst the 269 lines tested in the five observational yield trails, 25 lines significantly ($P < 0.05$) out yielded WITA 4 in the range of 200% to 41%. The line with the largest percent increase over WITA 4, namely, WAT 1410-27-2-2-1, had the lowest iron toxicity score of 3 with the remaining lines scoring from 5 to 7. The same organization also revealed that across the three sets of trials 45 lines significantly ($P < 0.05$) out yielded WITA 4. Of these only three lines had a maximum resistant iron toxicity score of 3 (7%) whilst 17 (38%) recorded a maximum score of 5 (25%) and (56%) a maximum score of 7.

Resistant lowland rice varieties had been implicated in the reduction of the negative impact of iron toxicity [29], [43], [10]. Various mechanisms had been reported for this resistance mechanism; avoidance/ inclusion (compartmentalization and exclusion of iron from the symplast) [29], avoidance /exclusion (rhizospheric oxidation and root ion selectivity) [44], [45], tolerance (inclusion) (detoxification by enzymes and the activity of phytoferritin) [46], [47] in the maintenance of Fe^{2+} homeostasis. Similarly, twenty lowland rice cultivars for tolerance to iron toxicity were subjected at an iron-toxic site in Korhogo, Côte d'Ivoire, under irrigated conditions and the evaluated cultivars differed in tolerance of iron toxicity. Grain yields varied

from 0.10 to 5.04 t ha⁻¹ and iron toxicity scores, [33]. The high yielding G26 result of the present study is similar to [33] results who evaluated rice cultivars during 1992-97 and reported that among three promising iron-tolerant cultivars, CK 4 was the top yielder (mean grain yield 5.33 t ha⁻¹), followed by WITA 1 (4.96 t ha⁻¹) and WITA 3 (4.46 t ha⁻¹), and tolerant check Suakoko 8 (3.80 t ha⁻¹). In conjunction with the present study outcome with others, implies that high yields and iron toxicity tolerance are physiologically compatible [29].

In the present study iron toxicity stress drastically reduce yield and yield components of the surviving progenies with the number of tillers harvested. The highest number of tillers was recorded for G48 than the tolerant improved check Rok-24 whereas G4, G6, G13, G16, G22, G24 and G27 showed the least number of tillers below the tolerant improved check Rok-24. The mean number of tillers harvested was 6.63, with 66.7% of the genotypes had significantly higher number of tillers above the grand mean. This study also observed that G14, G36, G43 and G47 recorded the maximum number of panicles while G2, G4, G6, G10, G11, G13, G16, G20, G22, G24, G27, G30, G32, G44 and G45 exhibited minimum number of panicles. The mean number of panicles produced was 4.24, with 44.4% of the genotypes had higher number of panicles above the grand mean. The total number of tillers and panicles are parameters that determine the yield per unit area of the rice plant. Iron toxicity affects rice plant by severe stunted growth, extremely limited tillering and panicles. Observed results substantiated the fact that iron toxicity reduced number of tillers and effective tillers as this was also indicated in the findings of [48] who said iron toxicity reduces growth, tillering and spikelet fertility, while [8], showed that iron toxicity resulted in reduced plant height and tillering. Similarly, [49], [50] noted that in West Africa, rice yield losses associated with iron toxicity are estimated at 12 to 100% depending on the rice cultivars used and the prevailing iron toxicity levels. The present study clearly revealed such observation and is consistent with [31], [41], [51] that, the tiller number were formed extremely modest (Table 4), which were averagely 6.63, this was probably because of high levels of iron in the soil that caused toxicity and the use of lines that were sensitive to metal stress and were unable to adapt well to the extreme conditions of site. The high dissolved ferrous iron in the soil might result in mineral nutrient imbalances, which will affect plant growth, especially to lines (genotypes) susceptible to iron stress. Tiller formation was adversely affected by iron toxicity as observed in the susceptible checks.

Results in Table 4 also uncovered that G40 showed the minimum 1000 grain weight whereas the maximum 1000 grain weight was recorded for G42. The mean of the 1000 grain weight harvested was 20.47g, with 48.1% of the genotypes showed grain weight above the grand mean weight. The lowest number of filled grains was recorded for G30 whereas G26 recorded the highest number of filled grains; the mean number of the filled grains was 186.1, with 38.9% of the genotypes accounted for higher number of filled grains above the grand mean. Results also revealed that G38 exhibited the least number of unfilled grains

whereas the highest number of unfilled grains was recorded for G40. The mean number of unfilled grains was 81.9, with 50% of the genotypes accounted for higher number of unfilled grains above the grand mean. The above rice traits made up the yield component and which have a direct effect on the yield of the rice crop.

Verily, iron toxicity affects rice growth and yield approximately 30% of the lowland swamp soils in rain-fed and irrigated lowland areas in West and Central Africa [34]. Iron toxicity symptoms are manifested as tiny brown spots starting from the upper tips and spreading toward the bases of the leaves with advancement in iron toxicity. By means of increasing iron toxicity stress, the whole affected leaves appear purplish brown, followed by drying of the leaves, which gives the rice plant a scorched manifestation [33], [34]. The disorder may be expressed as reduced plant height, reduced tillering, leaf discoloration and loss of chlorophyll, and reduced root growth [8]. As a result of the leaf discoloration and loss of chlorophyll, and reduced root growth, which had impacted the photosynthesis and photosynthetic rate translocation processes adversely. These may have led to higher percentage (50%) of the genotypes accounted for number of unfilled grains above the grand mean, a low percentage (38.9%) of the genotypes accounted for number of filled grains above the grand mean and 1000 thousand grain weight, consequently had direct impact on the grain yield. Consistent with these results, [29] found out that at any given concentration of iron in the leaves, net photosynthetic rates were lower in the iron-toxicity susceptible genotypes than in the iron-toxicity tolerant rice cultivar.

Reference [29] carried out field experiments on iron-toxic soils in an effort to establish the mechanisms involved in cultivar differences in iron-toxicity tolerance. Their outcome was that iron-tolerant rice cultivar absorbed less iron or translocated less iron from the roots to leaves. In agreement with [29], G26 recorded the highest number of filled grains and 38.9% of the genotypes accounted for higher number of filled grains, 48.1% of the genotypes showed grain weight above the grand mean (186.1) and (20.47g) respectively as compared to the improved check Rok-24. These traits obviously showed the potentials of these genotypes tolerance to iron stress. Reference [29] declared that iron-tolerant rice cultivar owed its superior performance under iron-toxic conditions partly to avoidance (less iron accumulation in the photosynthesizing leaves) and iron tolerance, by maintaining

superior photosynthetic potential in the presence of absorbed iron in the leaves.

Iron toxicity can affect rice plant at different growth stages and this may affect rice at the seedling stage, during the vegetative growth, and at the early and late reproductive stages. This investigation revealed that Rok-24 the improve check produced the tallest plant height whereas the shortest plant height was recorded for G19. The plant height mean was 63.98 cm; however, 50% of the genotypes investigated showed taller plant heights above the grand mean. Observed outcome registered reduce heights under iron excess field which correspond with several authors. Reference [52] reported iron toxicity happening during seedling stage, the rice plants remain stunted with extremely limited tillering. Similarly, iron toxicity during the vegetative stages is associated with reduced plant height and dry-matter accumulation [53], with the shoot being more affected than the root biomass [54].

Under irrigated or rain-fed conditions, rice growth is suppressed when a large amount of iron is mobilized in soil solution or when interflow brings iron ions from upper slopes. Moreover, observed results in the present study on days to 50% flowering, exhibited that G9, G41 and the susceptible check NERICA L-20, flowered very early while G44 registered very late flowering. The mean days to 50% flowering was 121.93 days, with 59.3% of the genotypes had mean days to 50% flowering above the grand mean. The observed physiological maturity in this study showed, G9, G41 and NERICA L-20 expressed very early days to physiological maturity while G44 recorded very long period of days to physiological maturity. The mean days to physiological maturity recorded was 148.2 days, with 63% of the genotypes expressed longer period of days to physiological maturity. Both days to 50% flowering and days to physiological maturity of the genotypes registered significantly delay or longer periods in completing these respective stages. These findings were in agreement with [55], who related delayed flowering and maturity by up to 20-25 days and an increase in spikelet sterility and [56] also found out that in highly susceptible cultivars, no flowering at all will occur; this observation was evident in most of the genotypes in this investigation. Lack of flowering resulted after booting, root growth stops, and the aerenchyma starts to senesce and decay. This occurs because the oxidation power of the root breaks down, and the root surface is coated with dark brown to black precipitates of $Fe(OH)_3$, and many roots die [57].

Table 4. Shows means of traits of 54 lowland rice genotypes evaluated under iron toxic environment.

No.	Progenies/ Genotypes (G)	Number of tillers	Number of Panicles	Plant height	Days to 50% flowering (days)	Days to maturity (days)	1000 grain weight (g)	Yield grain (kg/ha)	Number of filled grains	Number of unfilled grains
1	G1	8	5	67	120	145	20.5	1201.8	213	74
2	G2	6	2	49	124	149	15.7	93.8	41	28
3	G3	6	3	67	118	143	14.5	556.1	83	102
4	G4	3	2	61	118	144	15.3	92.1	32	40
5	G5	7	6	73	129	155	22	1285.7	235	88
6	G6	3	2	64	124	149	30.4	176.8	31	45
7	G7	8	5	69	121	158	14.7	962	96	108
8	G9	7	5	81	96	121	15.2	893.1	463	142



No.	Progenies/ Genotypes (G)	Number of tillers	Number of Panicles	Plant height	Days to 50% flowering (days)	Days to maturity (days)	1000 grain weight (g)	Yield grain (kg/ha)	Number of filled grains	Number of unfilled grains
9	G10	6	2	39	123	150	18.8	88.8	19	60
10	G11	4	2	49	126	153	17.8	172.6	24	114
11	G12	9	7	94	127	154	20.4	1341.3	512	126
12	G13	3	2	40	129	155	26	802.6	117	79
13	G14	8	8	89	127	154	17	1163.7	537	128
14	G15	5	3	69	124	149	21	187.6	235	125
15	G16	3	2	53	117	142	21	633.3	90	70
16	G17	7	6	37	124	151	26	918.8	123	44
17	G18	9	7	82	124	151	18.2	2416.1	318	105
18	G19	9	3	35	120	145	20	730.4	80	63
19	G20	10	2	58	127	154	17	545.3	80	110
20	G22	3	2	50	124	151	23	915.5	150	25
21	G24	3	2	56	119	144	20	213.31	64	124
22	G25	7	4	74	121	158	18.8	2116.5	475	109
23	G26	7	6	86	124	149	18	2494.2	672	152
24	G27	3	2	42	129	155	28	293.8	38	105
25	G28	9	7	77	117	142	16	1784.5	270	130
26	G30	10	2	57	121	146	22.2	203.4	18	135
27	G31	6	4	46	121	147	19	932.1	228	105
28	G32	7	2	42	123	150	17	296.3	68	141
29	G33	7	6	54	121	147	21.5	2005.3	290	90
30	G34	8	5	46	120	145	20	1018.9	279	154
31	G35	6	3	71	124	149	18	703.8	119	39
32	G36	9	8	95	120	145	19.4	2114.8	351	111
33	G37	9	5	73	129	155	22	1971.3	255	88
34	G38	9	3	41	128	154	21	224.9	40	16
35	G40	6	4	67	116	141	14	927.1	144	218
36	G41	6	5	69	96	121	22	752	94	39
37	G42	6	5	78	124	149	25	1194.4	132	40
38	G43	10	8	59	129	155	21	957	274	74
39	G44	5	2	47	134	161	19.7	173.5	30	45
40	G45	4	2	45	123	153	21.2	178.5	35	27
41	G46	4	3	47	123	150	17	307.1	40	35
42	G47	10	8	46	116	141	26.5	947.9	239	90
43	G48	11	7	47	128	154	34	959.5	243	36
44	G49	4	3	72	129	155	23	703	94	32
45	G51	7	5	65	128	154	24	811.7	140	37
46	G52	8	6	54	129	155	21.5	1220.1	368	65
47	G53	7	4	54	126	153	18	638.3	83	135
48	G54	8	4	86	124	150	20.8	906.4	350	25
49	G55	10	7	88	116	141	20	598.4	95	25
50	G56	4	3	67	124	150	24	792.7	101	30
51	G57	7	7	60	123	150	16.8	1985.4	446	108
52	Rok-5	5	3	105	120	145	16	664	49	97
53	Rok-24	7	5	118	121	146	20.5	2431.9	393	54
54	L-20	5	3	95	96	121	25	713.8	53	35
Mean				63.9						
		6.63	4.241	8	121.93	148.22	20.47	915.1	186.1	81.89
Standard error of mean		0.306	0.273	2.56	1.02	1.12	0.548	89.6	21.6	6.08
CV (%)				29.4						
		33.96	47.37	3	6.17	5.54	19.67	71.94	85.32	54.56
Range				35-				88.8-		
		3 – 11	2 – 8	188	96 -134	121 -161	14 – 34	2494.2	18 - 672	16 – 218

Standard Evaluation System (SES) from International Rice Research Institute (IRRI) was employed to score selected stress parameters of 54 out of the 61 genotypes, two susceptible checks and one tolerance check of lowland rice evaluated under iron toxic environment Table 5. It was clearly observed that G8, G21, G23, G29, G39 and G50 where significantly susceptible and did not survive iron toxicity stress to the late stages of the rice plant. However, visual observation using SES showed that all other

progenies evaluated were observed to be resistant/ tolerant to both rice yellow mottle virus (RYMV) and rice blast with established green leaves but with sparse dots or streaks and less than five percent height reduction for RYMV score rating. Rice blast rating genotypes expressions were observed to establish small roundish to slightly elongated, necrotic gray spots about 1 – 2 mm in diameter, which as well exhibited with a distinct brown margin.

Table 5. Shows scores of selected stress parameters of 54 lowland rice genotypes evaluated under iron toxic environment.

VAR.	Fe score	RYMV	BLAST	VAR.	Fe score	RYMV	BLAST
G1	5	3	2	G32	5	3	2
G2	7	3	2	G33	5	3	2
G3	5	3	2	G34	5	3	2
G4	7	3	2	G35	5	3	2
G5	3	3	2	G36	5	3	2
G6	7	3	2	G37	3	3	2
G7	3	3	2	G38	5	3	2
G9	3	3	2	G40	5	3	2
G10	7	3	2	G41	5	3	2
G11	7	3	2	G42	5	3	2
G12	3	3	2	G43	5	3	2
G13	5	3	2	G44	7	3	2
G14	3	3	2	G45	7	3	2
G15	5	3	2	G46	5	3	2
G16	5	3	2	G47	5	3	2
G17	5	3	2	G48	5	3	2
G18	3	3	2	G49	5	3	2
G19	5	3	2	G51	5	3	2
G20	5	3	2	G52	5	3	2
G22	5	3	2	G53	5	3	2
G24	5	3	2	G54	3	3	2
G25	5	3	2	G55	3	3	2
G26	3	3	2	G56	5	3	2
G27	5	3	2	G57	5	3	2
G28	5	3	2	Rok-5	7	3	2
G30	5	3	2	Rok-24	3	3	2
G31	5	3	2	L-20	5	3	2
Mean	5	3	2		5	3	2
Range	3-7	3-3	2-2		3-7	3-3	2-2

Principle Component Analysis of Genotypes

Morphological characterization of rice genotypes is a fundamental breeding procedure and it provides basic information of the germplasm for its future use [58]. The rice genotypes used in the present study displayed variability for all the studied traits. Reference [59] also observed reasonable amount of genetic variation for different traits.

Principal Component analysis (PCA) undoubtedly indicates the genetic variation of the progenies or genotypes in (Table 6). It measures the important characters which have a bigger impact to the total variables and each coefficient of proper vectors indicated the degree of contribution of every original variable with which each principal component. The first 5 components in PCA analysis contribute 91% of the total variation among total genotypes for nine quantitative traits of 61 genotypes are represented in Table 6. Moreover, PC1, PC2 and PC3 had Eigen value greater than one and PC4 and PC5 had Eigen value below one. However, PC1 clearly demonstrated with Eigen value of 3.29 contributed 37% of the total variability,

PC2 with Eigen value of 2.10 contributed 23% of the total variability, PC3 with Eigen value of 1.30 contributed 14% of the total variability and PC4, and PC5 with Eigen value of 0.87 and 0.61 contributed 10% and 7% of the total variability respectively.

Table 6 explicitly showed that in the first Principal Component, 1000 grain weight had positive and the largest contribution followed by days to 50% flowering, days to maturity and number of unfilled grains, number of tillers, plant height, number of panicles, grain yield and number of filled grains contributed negatively to the component; as a result, both vegetative and reproductive characters gave contribution to the component. In the second Principal Component most of the traits had positive contribution which include days to 50% flowering, days to maturity, 1000 grain weight, number of unfilled grains, number of tillers, grain yield and number of filled grains contributed positively but plant height and number of unfilled grains negatively contributed in this component. The major characters that contributed highly to the variation include days to 50% flowering, days to maturity and 1000 grain

weight while number of tillers, number of panicles, grain yield and number of filled grains contributed least to the variation. In the third Principal Component, 1000 grain weight, panicle numbers, number of tillers, and grain yield had positive effect while days to 50% flowering, days to maturity number of unfilled grains, and number of filled grains contributes negatively to the component and these are fully reproductive traits. 1000 grain weight and number of panicles contributed mainly in this component.

In the fourth Principal Component, traits which had positive contribution in the component include plant height, grain yield, Number of filled grains, days to maturity and days to 50% flowering. The highly contributing characters include, Plant height and grain yield. The remaining component traits exhibited negative and reproductive characters. In the fifth Principal Component

number of tillers exhibit the highest positive role to the component which is a reproductive character followed by Plant height, with least positive contribution offered by days to 50% flowering, days to maturity and number of panicles while negative contribution was offered by 1000 grain weight, Grain yield, Number of filled grains and Number of unfilled grains. According the above discussion in most of the cases the traits contributes both vegetative and reproductively (PC1, PC2, PC4 and PC5). Thus through Principal Component Analysis it could be used to identify the number of plant characters which are responsible for the genotypic variation within a group. Principal Component Analysis as well helps to identify the characters which have greater impact of phenotypic difference in rice progenies, and this is very much important for the selection procedure in breeding programmes.

Table 6. Shows Principal Component Analysis of 54 lowland rice genotypes evaluated under iron toxic environment.

Statistics	PC1	PC2	PC3	PC4	PC5
Standard deviation	1.82	1.45	1.14	0.93	0.78
Proportion of Variance	0.37	0.23	0.14	0.10	0.07
Cumulative Proportion	0.37	0.60	0.74	0.84	0.91
EigenValues	3.29	2.10	1.30	0.87	0.61
EIGENVECTORS					
Variables					
Number of tillers	-0.35	0.17	0.11	-0.62	0.46
Number of Panicles	-0.47	0.13	0.24	-0.23	0.04
Plant height	-0.35	-0.20	0.08	0.60	0.30
Days to 50% flowering	0.09	0.65	-0.15	0.15	0.07
Days to maturity	0.08	0.65	-0.20	0.16	0.09
1000 grain weight	0.12	0.21	0.69	-0.07	-0.54
Grain yield	-0.47	0.11	0.07	0.25	-0.18
Number of filled grains	-0.48	0.11	-0.02	0.16	-0.26
Number of unfilled grains	-0.24	-0.08	-0.62	-0.26	-0.53

Cluster Means Analysis and Association of Traits

Quantitative agronomic and morphological traits were examined as recommended by [60], [61]. Table 7 presents cluster means of 54 out of the 61 lowland rice genotypes evaluated under iron toxic environment. The present study revealed that the evaluated rice genotypes exhibited a considerable morphological variation. Also the dendrogram obtained from this study agreed with the findings of [62], in terms of similarity which exist among rice genotypes, they noted that cluster analysis has the remarkable efficacy and ability to identify crop with highest level of similarity. Dendrogram showing Euclidean clustering of 54 out of the 61 lowland rice genotypes evaluated in (Fig. 1). The dendrogram exhibited six different main cluster groups, of which the tool brought together evaluated traits which are similar. With regards to the above established fact, cluster (I) and cluster (IV) had 15 genotypes each, clusters (II) nine, cluster III three, cluster V four and cluster (VI) eight genotypes. Each main cluster is divided into sub-cluster, which consist of close similarity. Therefore, cluster (I) had five sub-clusters, clusters (II) and (IV) three sub-clusters each and clusters (III), (V) and (VI) had two sub-clusters each.

Genotypes in cluster (I) and (IV) each accounted for 27.8 percent of total genotypes. The unique characteristics which are peculiar to genotypes in cluster (I) were higher number of tillers, number of panicles, number of filled grains, grain weight and grain yield, and longer days to 50 percent flowering and physiological maturity. This cluster presents better yield components that may create the opportunity and potential to breeders for selection. Also the genotypes might have avoided the iron stress or take it but create a tolerance mechanism within it system. Characteristics such as low number of tillers, number of panicles, and number of filled grains, plant height and lower grain yield were the peculiar features that characterized genotypes in cluster (IV). The genotypes may have absorbed greater quantity of the environmental stress (excess iron) and manifest itself with bizarre negative effect to genotypes in this cluster. Genotypes in cluster (II) accounted for 16.7 percent of total genotypes. The present study agreed with [63] who studied the genetic relationship of some rice varieties and observed that origin, habitat and breeding background contributed to variation in the rice population. The outstanding characteristics which are specific to genotypes in this cluster were highest number of panicles, number of filled

grains, plant height, grain yield and number of unfilled grains, higher number of tillers and longer period to physiological maturity. Cluster (II) gave similar desirable traits as cluster I but the iron stress might have hindered the genotypes to produced highest values of infertile grains.

Furthermore, genotypes in cluster (III) claimed 5.6 percent of the total genotypes and the unique features which characterized genotypes in this cluster were moderate number of tillers, number of panicles, 1000 grain weight, number of filled grains, grain yield, and the earliest period to days to 50% flowering and physiological maturity. The life cycle of genotypes in cluster (III) uncovered to be the shortest as a plant escape strategy, which might have reduced the results of the other yield component in the group. Results also revealed that genotypes in cluster (V) accounted for 7.4 percent of the total genotypes. The clear characteristics which were specific to genotypes in this cluster are similar to cluster (IV) and they had the lowest number of panicles, number of filled grains, plant height,

grain yield and longer period to days to 50% flowering and physiological maturity and moderate 1000 grain weight and number of unfilled grains. Genotypes in cluster (VI) accounted for 14.8 percent of the total genotypes in the dendrogram. The traits in this cluster (VI) were varied but produce poor trait outcome. The clear characteristics which are specific to genotypes in cluster (VI) were longest period to days to 50% flowering and physiological maturity, highest 1000 grain weight, and also produced the lowest number of tillers, number of panicles and low number of filled grains and grain yield. According to [64], genetic variability in crop species should be exploited so as to develop new rice varieties with high stability to resist or tolerate adverse environments and biotic conditions. As a result, the disclosed variations among the genotypes and the identified traits that contributed to variability significantly in the population of the present study could be utilized in breeding actions.

Table 7. Shows cluster means of 54 lowland rice genotypes evaluated under iron toxic environment.

Cluster	Number of tillers	Number of Panicles	Plant height	Days to 50% flowering	Days to maturity	1000 grain weight	Gain yield	Number of filled grains	Number of unfilled grains
I	8.13	5.8	61.27	123.87	149.6	22.92	135.91	230.93	69
II	8	6.56	86.11	122.67	149.89	18.34	238.93	441.56	113.67
III	6	4.33	81.67	96	121	20.73	94.73	203.33	72
IV	5.27	2.8	59.87	121.47	148	17.25	52.5	79.13	95.73
V	9.5	2.5	47.75	124	149.75	20.05	51.33	54.5	81
VI	3.62	2.25	53.38	127	153.62	24.41	60.79	74.5	48.5

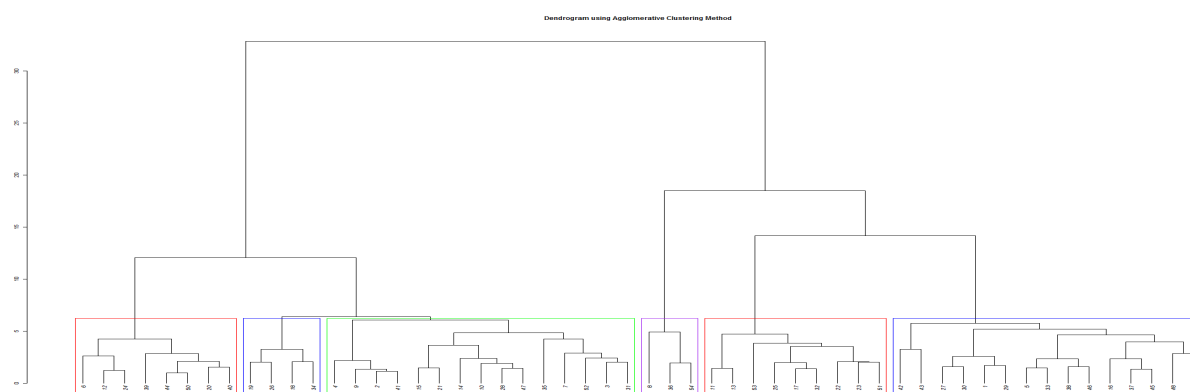


Fig. 1. Dendrogram showing Euclidean clustering of 54 lowland rice genotypes using Ward's method.

Correlation of Traits

Person's correlation coefficient of 54 out of the 61 lowland rice genotypes evaluated under iron toxic environment is presented in Table 8. Observation showed that number of tillers, number of panicles (effective tillers), plant height and number of filled grains exhibited very strong ($p \leq 0.01$) and positive relationship with grain yield. Several authors have noted similar findings, [65] reported comparable answers were with plant height, number of effective tillers per meter, panicle length, grains per panicle and 1000 grains weight had significant and positive correlation with grain yield. In addition, positive and

significant correlation of grain yield per plant with number of productive tillers, panicle length, number of filled grains per panicle, straw yield and 1000 seed weight was reported by [66]. [67] Reported significant and positive correlation of grain yield per plant with plant height, flag leaf area, number of grains per panicle and panicle length. [68] Also revealed significantly positive association of grain yield per plant with number of productive tillers per plant. [69] With panicle length and number of effective tillers, and [70] with filled grains per plant, grains per panicle and 1000 grain weight. [71] With plant height, number of grains per panicle and flag leaf area.

Number of tillers had positive and very strong correlation with number of panicles, grain yield and number of filled grains. However, it had recorded non-significant association with plant height, days to 50% flowering and days to maturity. Furthermore, number of panicles revealed positive and very strong association with plant height, number of filled grains and grain yield. Related results were revealed by [72]. However, in improving grain yield, genotypes which produce significant and positive relationships are useful for selection

Plant height correlated positively and strongly with the number of filled grains and grain yield. In a similar outcome, [68] also reported plant height exhibited positive and significant correlation with panicle length, number of filled grains per panicle. Conversely, days to physiological maturity and 50 percent flowering expressed negative and strong $P (< 0.05)$ correlation with plant height. Principally, negative relationships come up from competition for a general option, for instance nutrient availability. In

supportive of the above, [73] related that if one component gets advantage over the other, a negative relationship may come up.

Results of this investigation showed days to physiological maturity and 50 percent flowering displayed positive and strong association with each other. This result coincided with [74] who indicated days to 50 per cent flowering exhibited a positive and significant association with days to maturity furthermore, [68] reported parallel findings. Finally, 1000 grain weight showed negative and significant relationship with number of unfilled grains, whereas number of filled grains associated positively and significantly with the trait number of unfilled grains.

Observed traits which show direct and mutual relationship to yield may be considered for selection in breeding activities. The investigation has projected a reason that an increase in grain yield is observed when yield component characters exhibited positive and significant correlation with grain yield.

Table 8. Shows Pearson's correlation coefficient of 54 lowland rice genotypes evaluated under iron toxic environment.

Traits	Number of tillers	Number of Panicles	Plant height	Days to 50% flowering	Days to maturity	1000 grain weight	Gain yield	Number of filled grains
Number of tillers								
Number of Panicles	0.68**							
Plant height	0.17	0.38**						
Days to 50% flowering	0.04	-0.04	-0.27*					
Days to maturity	0.05	-0.04	-0.27*	0.96*				
1000 grain weight	-0.04	0.05	-0.21	0.17	0.10			
Grain yield	0.41**	0.68**	0.52**	0.00	0.03	-0.06		
Number of filled grains	0.43**	0.70**	0.49**	0.01	0.03	-0.11	0.75**	
Number of unfilled grains	0.19	0.19	0.11	-0.09	-0.06	-0.43**	0.26*	0.36**

*, **= significant at 5% and 1% probability levels, respectively. Values without asterisk are not significant at both probability levels.

IV. CONCLUSIONS

Significant variations were evident on iron toxic conditions and its implications on the traits at the different growth stages are diverse. Out of the 61 rice genotypes evaluated under iron toxic environment, 54 genotypes survived to maturity stage, and the other seven genotypes seedlings died after transplanting. This indicates that iron toxicity tolerant genotypes (lines) will provide a new and unique opportunity for sustainable agricultural development in lowland environment.

Under iron toxicity condition, G26 produce the highest grain yield a little over the tolerant check Rok-24 but G10 produce the least and 40.7% of the genotypes produced higher grain yield and yield components above the grand mean which indicates iron toxicity tolerance of the genotypes. The lines with high tolerance to iron toxicity stress also had high yield potentials, confirming the evidence that yield potential contributes to yield under iron toxicity.

The dendrogram produced six different main Euclidean cluster groups. These mentioned clusters produced the best trait performance of all the clusters. Thus, grain yield could be improved considerably through selecting its components characters like plant height, number of panicles, number of

filled grains and 1000 grain weight. Therefore, genotypes in these clusters could be identified as iron tolerance genotypes and also could be selected for further crop improvement.

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