Screening of F₂ Population under Higher Iron Toxic Levels of Hydroponics in Rice

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A B S T R A C T

F₂ population obtained from F₁ cross between Tulasi (Most tolerant genotype) and CUL-8709 (Most susceptible genotype). 300 F₂ plants and their parents were screened at 800 ppm of Fe. Phenotyping screening of F₂ plants under iron toxic levels indicated presence of wide variability for shoot length, root length, total number of roots, number of fresh roots, shoot weight, root weight and visual scoring for iron-toxicity symptoms. The measures of skewness and kurtosis for various traits revealed a large quantitative variability. All the above traits except iron content in root of F₂ lines exhibited a positive platykurtic distribution pointing to presence of gene interaction in trait expression. Measures of skewness and kurtosis also indicated occurrence of transgressive segregation in the F₂ population. Leaf bronzing the typical symptom of Fe toxicity, showed a strong negative correlation with shoot length, root length, total number of roots, number of fresh roots, shoot weight and root weight. The results indicated that leaf bronzing is associated with growth reduction due to Fe²⁺ toxicity in this F₂ population.

Keywords
Range, Variability, Skewness, Curtosis, Transgressive segregation

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Introduction

Globally, rice is the most important food crop, serving as staple food for more than half of the world’s population (Khush, 2005). It occupies almost one-fifth of the total land area cropped with cereals. During 2015, the total global rice production reached 740.2 million tonnes from an area of 161.1 Mha (FAO, 2016). Rice and wheat are the major food crops grown in India. In 2015, the total rice production in the country reached 104.8 million tonnes with a production of 44.16 Mha and productivity of 2373 kg/ha (Indiastat, 2015).

In acidic soils of Kerala, iron content of the root to the order of 50,000 ppm under submerged conditions was found to inhibit morphological and physiological development leading to low yield (Bridgit, 1999). During recent years, the problem of iron toxicity has become even more severe due to the introduction of modern high-input rice varieties susceptible to excess iron. Several management and cultural practices have been proposed for the control of iron toxicity in the field. Great inter-varietal differences in iron toxicity tolerance in rice have been reported.
Mohanty and Panda, 1991). Therefore, exploiting the varietal tolerance to iron toxicity is accepted as the most cost-effective and practical means for increasing rice production under iron toxic soils (Shimizu, 2009).

Rice varieties are different in their tolerance for iron toxicity and this selection of rice variety with better iron tolerance is important to avoid yield reduction. Genetic differences in adaptation and tolerance for iron toxic soil conditions have been exploited for rice variety with tolerance for iron toxicity (Gunawardena et al., 1982; Fageria et al., 1990). The existence of genetic variability for various desirable maturity and yield related traits in segregating generations is of utmost importance in crop breeding programs to develop desirable recombinant inbred lines and cultivars. Breeders have developed a wide array of cultivars with various degrees of adaptation, using both traditional breeding methods (Akbar et al., 1987; Gunawardena et al., 1982; Luo et al., 1997; Mahadevappa et al., 1991) and quantitative trait loci (QTL) analysis combined with marker-assisted breeding (Bennett, 2001; Wan et al., 2003a and 2003b; Wissuwa, 2005).

Materials and Methods

The experimental material for the study comprised of thirty rice genotypes selected from the KAU rice germplasm maintained at Regional Agricultural Research Station (RARS), KAU, Pattambi. The 30 rice genotypes were subjected to further screening to confirm their tolerance or susceptibility to iron toxicity. One most tolerant genotype (Tulasi) and most susceptible genotype (CUL-8709) selected and used for development of F2 population. 300F2 plants and their parents were screened at 800 ppm of Fe through hydroponics. In the present study, an attempt has been made to understand the influence of iron at toxic level (800ppm) on growth parameters viz., shoot length, root length, total number of roots, number of fresh roots, shoot weight, root weight and visual scoring for iron-toxicity symptoms of F2 plants. The amount of iron reversibly adsorbed on root surface, iron content in root and leaf were also assessed.

Results and Discussion

Results (Table 1, Fig. 1 and 2) indicated presence of wide variability for these traits among the F2 plant population studied. Wu et al., (1997) had also observed wide variability among double haploid (DH) populations for leaf bronzing index and shoot weight in confirmation with the results of the present study.

Mean visual scoring for iron-toxicity symptoms of 300 F2 plants after 4 weeks of 800ppm of Fe treatment was 5. Mean visual scoring for iron-toxicity symptoms of 300 F2 plants after 6 weeks of 800ppm of Fe treatment was 8. Visual scoring for iron-toxicity symptoms ranged from 1 to 9 after both after 4 weeks and 6 weeks of 800ppm of Fe treatment.

Skewness and kurtosis of visual scoring for iron-toxicity symptoms after 4 weeks of 800ppm of Fe treatment is -0.14 and -1.41 respectively. Frequency distribution (Fig. 1) was used to determine the number of individuals in the segregating F2 population that had visual scoring for iron-toxicity symptoms close to parent PGC 14 (Tulasi) (1) and PGC 31 (Cul-8709) (9) as well as intermediate between the two. F2 individuals with visual scoring for iron-toxicity symptoms ≥ 9 were designated as having higher visual scoring for iron-toxicity symptoms; those with values between 3 and 7 as intermediate and individuals with visual scoring for iron-toxicity symptoms ≤ 1 as low.
Results indicated that out of the 300 F2 plants, 87 F2 plants possessed high visual scoring for iron-toxicity symptoms (29%), 163 F2 plants had intermediate values (54.33%) while visual scoring for iron-toxicity symptoms was low in 50 F2 individuals (16.67%).

Skewness and kurtosis of visual scoring for iron-toxicity symptoms after 6 weeks of 800ppm of Fe treatment is -1.67 and 1.55 respectively. Frequency distribution (Fig. 1) was used to determine the number of individuals in the segregating F2 population that had visual scoring for iron-toxicity symptoms close to parent PGC 14 (Tulasi) (1) and PGC 31 (Cul-8709) (9) as well as intermediate between the two. F2 individuals with visual scoring for iron-toxicity symptoms ≥ 9 were designated as having higher visual scoring for iron-toxicity symptoms; those with values between 3 and 7 as intermediate and individuals with visual scoring for iron-toxicity symptoms ≤ 1 as low. Results indicated that out of the 300 F2 plants, 206 F2 plants possessed high visual scoring for iron-toxicity symptoms (68.67%), 79 F2 plants had intermediate values (54.33%) while visual scoring for iron-toxicity symptoms was low in 15 F2 individuals (5%).

Frequency distribution (Fig. 1 and 2) of for the parameters studied indicated existence of clear difference b/w Tulasi and Cul 8709 with respect to the traits studied. Most F2 individuals recorded phenotypic values between the susceptible and resistant parent under iron stress.

The measures of skewness and kurtosis for various traits revealed existence of a large quantitative variability. However, none of the traits showed a perfect symmetrical data or skewness of zero. According to Fisher et al., (1932), the study of distribution using skewness provides information about nature of gene action while Robson (1956) opined that kurtosis is indicative of the number of genes controlling the traits. Distribution of root length, iron content in leaf and visual scoring for iron-toxicity symptoms of F2 plants after 4 weeks was approximately symmetrical as skewness of these characters ranged from -0.5 to 0.5 indicating a fairly normal frequency distribution under iron toxic conditions.

All these traits exhibited a negative platykurtic distribution. A near zero skewness and negative value of kurtosis points to the absence of gene interaction (Ashwini et al., 2011).

However, after 6 weeks of exposure to iron stress, the distribution of LBS was highly skewed with too many iron sensitive individuals. A negative skewness is indicative of duplicate (additive x additive) gene interactions while positive skewness is associated with complementary gene interactions (Ashwini et al., 2011).

The distribution was also platykurtic and positive. The traits with platykurtic distribution are considered to be controlled by a large number of genes (Kotch et al., 1992). The results thus pointed out that the LBS after 6 weeks was controlled by multigenes that exhibit duplicate gene action. The efficiency of selection in a breeding programme depends on the amount of gene interaction. According to Choo and Reinberos (1982), improvement in population performance may be greater under complementary interaction rather than under duplicate gene interaction.

In case of total number of roots, shoot length, and iron content in root of F2 plants, the distribution was moderately skewed (0.5 to 1.0) while a highly skewed (< -1 or > +1) distribution was observed for number of fresh roots, shoot weight, root weight, iron reversibly adsorbed on root surface and visual scoring for iron-toxicity symptoms of F2 plants after 6 weeks.
Table 1: Variability in F2 population screened for response at 800 ppm of iron

| Sl. No. | Trait                                | Mean  | Range  | Standard deviation | Coefficient of variation | Skewness | Kurtosis |
|---------|--------------------------------------|-------|--------|--------------------|--------------------------|----------|----------|
| 1       | Leaf bronzing after 4 weeks          | 5.43  | 8.00   | 2.97               | 54.69                    | -0.14    | -1.41    |
| 2       | Leaf bronzing after 6 weeks          | 7.65  | 8.00   | 2.37               | 30.90                    | -1.67    | 1.55     |
| 3       | Root length (cm)                     | 19.52 | 10.30  | 2.25               | 11.51                    | 0.39     | -0.48    |
| 4       | Shoot length (cm)                    | 55.29 | 13.80  | 2.69               | 4.87                     | 0.73     | 0.45     |
| 5       | Root weight (g)                      | 4.24  | 5.35   | 1.25               | 29.43                    | 1.42     | 1.20     |
| 6       | Shoot weight (g)                     | 6.02  | 9.95   | 2.13               | 35.44                    | 1.44     | 1.28     |
| 7       | Total number of roots                | 25.32 | 14.00  | 2.68               | 10.58                    | 0.99     | 1.03     |
| 8       | Number of fresh roots                | 5.24  | 32.00  | 8.20               | 156.53                   | 1.56     | 1.41     |
| 9       | Iron adsorbed on root surface (g)    | 5.02  | 12.48  | 2.75               | 54.81                    | 1.67     | 2.10     |
| 10      | Iron content in root (g)             | 8746.52 | 7772.31 | 1813.20          | 20.73                    | 0.70     | -0.37    |
| 11      | Leaf iron content (g)                | 1633.15 | 2667.97 | 614.40           | 37.62                    | 0.19     | -0.84    |
Table 2: Skewness and kurtosis of leaf bronzing score and growth traits in F₂ population

| Sl. No. | Trait                              | skewness | kurtosis |
|--------|------------------------------------|----------|----------|
| 1      | Leaf bronzing after 4 weeks        | -0.14    | -1.44    |
| 2      | Leaf bronzing after 6 weeks        | -1.67    | 1.55     |
| 3      | Root length (cm)                   | 0.39     | -0.48    |
| 4      | Shoot length (cm)                  | 0.73     | 0.45     |
| 5      | Root weight (g)                    | 1.42     | 1.20     |
| 6      | Shoot weight (g)                   | 1.44     | 1.28     |
| 7      | Total number of roots              | 0.99     | 1.03     |
| 8      | Number of fresh roots              | 1.56     | 1.41     |
| 9      | Iron adsorbed on root surface (g)  | 1.67     | 2.10     |
| 10     | Iron content in root (g)           | 0.70     | -0.37    |
| 11     | Leaf iron content (g)              | 0.19     | -0.84    |

Fig.1: Frequency distribution of F₂ plants for screening observations (I)

A) Leaf bronzing score after 4 weeks    B) Leaf bronzing score after 6 weeks

C) Total number of roots    D) Number of Fresh roots
**E) Root length**

**F) Shoot length**

*Fig. 2* Frequency distribution of F₂ plants for screening observations (II)

**G) Root weight**

**H) Shoot weight**

**I) Iron reversibly adsorbed on root surface**

**J) Iron content in root**
K) Iron content in leaf

The genes controlling the trait with skewed distribution tend to be predominantly dominant irrespective of whether they have increasing or decreasing effect on the trait (Ashwini et al., 2011). Maximizing the genetic gain in respect of traits with positively skewed distribution requires intense selection from the existing variability while genetic gain in respect of all the traits exhibiting negative skewed distribution will be rapid under mild selection from the existing variability (Roy, 2000).

All the above traits except iron content in root of F2 lines exhibited positive platykurtic distribution. The platykurtic distribution for this trait was near zero (-0.37). Kurtosis is negative or close to zero in the absence of gene interaction and is positive in the presence of gene interactions (Choo and Reinbergs, 1982; Kotch et al., 1992).

Measures of skewness and kurtosis also indicated that the performance of a few F2 individuals were better than the resistant parent (Tulasi) while some were lower than that of susceptible parent (Cul-8709) for all observations except leaf bronzing score. This indicated occurrence of transgressive segregation in the F2 population as observed in the variation in normal distribution of traits confirming the polygenic control of traits. Skewness and kurtosis values of screening observations presented in the table 2. In consonance with the study, Shimizu et al., (2005) and Dufey et al., (2015) had observed transgressive variation in segregating populations for leaf bronzing index (LBI) and all correlated parameters. According to Miles and Wayne (2008), the parental lines need not be phenotypically different for traits controlled by several genes; rather, they must simply contain different alleles at various loci, which are then reassorted by recombination in the derived population to produce a range of phenotypic values. Transgressive segregation indicated that the subset of F2 population comprising of 300 individuals in the present study contained sufficient genetic variation for mapping QTLs for resistance to Fe toxicity.

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