Morphogenetic Response of Assorted Rice Genotypes to Salinity in Tanzania

Barnabas Sitta, Sophia Kashenge, Sang-Bok Lee, Kang Kyung-Ho, Victoria Bulegeya, Mbaraka Batare, and Rebecca Mwakapala

ABSTRACT

Salinity, where salts concentrate on the soil surface causing severe decline of crop yields, is a worldwide problem. In Tanzania salinity is one of the major soil degradation challenges affecting over 3.5 million hectares limiting agriculture productivity of various crops including rice. Most of the varieties grown are sensitive to salts and inadequate tolerant cultivars available in the country. A hydroponics mass screening technique using Yoshinda Solution was used to test the 102 genotypes in NaCl- saline treated and non-treated solution at Tanzania Agricultural Research Institute (TARI-Dakawa Centre). Different salt concentrations (4 dSm⁻¹, 6 dSm⁻¹, 8 dSm⁻¹ and 10 dSm⁻¹) were used and the experiment was done in three replications. The genotypic variability for salinity tolerance was observed as less salt injury symptoms, low Na⁺ accumulation and Na⁺/K⁺ ratio in plant tissues and high biomass accumulation (fresh weight and dry weight). Results revealed further those 28 genotypes (28.45%) out of 102 showed tolerance to salinity, at high salinity level of 10dSm⁻¹. Lines namely SR35266-2-18-2-1, SR35250-1-19-1-1, SR23364-128-1762-1-HV-1-1, SR35230-1-12-1-1, SR23364-128-1986-1-HV-1-1, SR34590-HB3433-4-1-1, SR35266-2-7-1-1, PBR1000922-1 and SR34053 (#5-52)-1-4-2-10-3-3 showed high performance under high salt conditions. Others includes SR35266-3-1-5-1, SR34574-2-10-3-1-2-1, SR35278-2-10-1-1, SR35250-2-3-1-1, SR35266-3-2-3-1, SR35266-3-2-4-1, SR23364-133-184-1-HV-1-1, SR34592-HB-1-HV-1, and SR34042F3-22-1-1-5-3 indicated tolerance to salt and had high dry matter as well. All the genotypes had increased levels of Na⁺ and differential performance was observed in some genotypes under saline and non-saline conditions. Among these three lines namely SR35266-2-7-1-1, SR33364-128-1762-1-HV-1-1, and SR34590-HB3433-4-1-1 expressed high dilution ability as the K⁺ and Na⁺ concentrations were lower compared to other genotypes. The study, therefore, suggests that the lines can be used in breeding programs to develop varieties with potential to salt tolerance and other traits.

Keywords: Genotypes, Hydroponics, K⁺ and Na⁺ Concentrations, Salinity.

I. INTRODUCTION

Rice (*Oryza sativa* L.) is one of the most important crops which is consumed by more than half of the world’s population [1]. In Tanzania, the crop accounts as the second most consumed after maize; thus, many farmers are venturing in rice production due to the influence of the premium price offered by the market [6]. Both quantitative and qualitative rice production is needed to meet the fast-growing population to assure national and global food security [10]. The current rapid population increase year by year suggests a need to increase production by more than 80% of the current production especially food crops such as rice [8]. However, the climate is rapidly changing globally resulting in the emergence of several biotic and abiotic stresses such as drought and flooding which reduces yield potential of the current rice varieties grown by farmers [5]. Among the abiotic stresses, salt is the second most environmental devastating stress in rice production after drought [7]. The world is facing a challenge of increased Salt which results in to decline in yields under irrigated agriculture and quality as well [1].

Scientists define saline stress as the concentration of the ions such as sodium, chloride, and sulfate in the rhizosphere in a way that disrupts the natural growth of the plants [11]. Various reports indicated that plants that are under stress usually suffer from cellular homeostasis imbalance, due to the
production of high-energy state successive electrons. Alteration of these successive electrons transfers electrons to oxygen molecules which leads to the formation of reactive oxidative species that normally act as signaling molecules and may induce oxidative damage at high concentrations and reduce the rate of photosynthesis [13]. Saline soils are estimated to cover about 5-10 % of the world’s arable land [9]. However, 20% of the world’s irrigated land is adversely increased because of poor drainage and climate changes [22], [9].

In Tanzania, the challenge is increasing rapidly as reported by FAO in 2003 about 3.5 million hectares are prone to salinity covering both the semi-arid, low land and irrigated and non-irrigated areas [14]. Salinity will be a constraint to sustainable rice production, therefore combating salinity is urgently needed to reduce the risk. Developing saline adaptive plants and a better understanding of its mechanism, especially rice, is urgently needed since mere soil remediation management will be difficult in Tanzania [9], [19]. Unfortunately, the tolerant mechanism to salt stresses not fully understood. Problems in breeding techniques also show that it has lower reliability and time consuming, and it is costly; some criteria used in screening usually do not correlate to salinity [8], [19]. However, breeding for salt tolerance seems to be an economically viable and long-term solution for the farmers, the reason being many farmers cannot afford to pay for the costs of soil amendment to make it suitable for plant growth. The current study suggests identified lines to be introgressed into the breeding program that will enhance the development of new varieties tolerant to salt.

II. MATERIALS AND METHODS

A. Screening Environment

The study was conducted in a screenhouse at Tanzania Agricultural Research Institute (TARI) Dakawa Center in Morogoro. The study was conducted in a screen house that had a well-covered top, and insect-proof net around all sides. There was no problem with sunlight. Seeds of selected genotypes were germinated in a sterile mixture of soil, sand, and manure at a 1:1:1 ratio, and seedlings were grown for 14 days. A hydroponics experiment was prepared using a Plastic container of 40 × 25 × 20 cm. A styrofoam sheet was cut to fit the top of each container. Five rows with five holes each were made on each styrofoam sheet, and a nylon net was placed at the bottom of each styrofoam sheet to prevent the seedling from falling into the solution as described by Kashenge-Killenga et al. [8], [20]. Each styrofoam sheet was floated in a container filled with 4 L of distilled water as indicated in Fig. 1. After two weeks, the seedlings (at two to three leaf stages) were uprooted, rinsed with sterilized deionized water to remove the soil, and were transferred to the prepared containers. Each container had five rows consisting of five genotypes (one genotype per row), and each hole had three seedlings (Fig. 1).

B. Preparation of a Working Solution

The working solution was prepared using the following stocks: NH₄NO₃ (91.4 g/L), Na₂HPO₄ (35.6 g/L), CaCl₂ (117.4 g/L), MgSO₄ (324 g/L) and K₂SO₄ (70.65 mg/L) for macronutrient stocks and a combination of MnCl₂ (1.5 g/L), H₃BO₃ (0.934 g/L) ZnSO₄ (0.035 g/L), FeSO₄ (7.7 g/L), CuSO₄ (0.031 g/L) [(NH₄)6MoO₄O₃] (0.13 g/L) and H₂C₁₂H₁₇O₇ (11.9 g/L) was used to make stock solution for required micronutrients [8], [20], [21]. Seedlings were grown in the nutrient solution for seven days prior to salinization to allow proper establishment. The nutrient solution was renewed every 8 days and the pH of 5.0 was maintained daily by adding either NaOH or HCl.

![Fig. 1. Seedling's growth in styrofoam sheets floated on modified Yoshinda solution [8].](image-url)
C. Genotypes Screening

Hydroponic containers were used to screen a total of 102 lines from Africa Rice Centre in Sahel Station Senegal and nine (9) local germplasms against salinity tolerance at the seedling stage. Improved salt tolerant variety (SATO 1), medium tolerant variety (SATO 9), and a susceptible check (Table I) were used as checks. Fifty-one (51) containers prepared with hydroponic solution were used. In each hole of the Styrofoam two seedling were planted and floated in containers consist of Yoshinda Solution and left to grow for 10 days on eleventh day the seedling was salinized by adding table salt (NaCl) and the pH of culture solution maintained at 5.0 through adding both NaOH and HCl. The trial was set in three replications in Randomized Complete Block Design (RCBD). The genotypes were screened at the different salt concentrations (4 dSm-1, 6 dSm-1, 8 dSm-1 and 10 dSm-1). Salt Injury scoring was done as per the modified Evaluation System used in rating visual symptoms of salt toxicity injury [24]. The injury score was recorded when the sensitive checks indicated injury or when almost dead.

D. Data Collection

Data collection was done 10 days after the maximum desired stress level was achieved. Plant growth (vigour), injury symptoms, shoot fresh and dry weight, shoot Na+, K+, and Na+/K+ were determined in both control and salt stressed plants. Scoring for salt tolerance on the basis of seedling vigour and salt injury was done on a scale at a respective growth stage of the plant (IRRI, 1988); where 1 - Germination, 2 - Seedling, 3 - Tillering, 4 - Stem elongation, 5 - Booting, 6 - Heading, 7 - Milk stage, 8 - Dough stage, and 9 - Mature grain. Salt injury rating was done between growth stage 3-4 and was rated as following modified standard evaluation scores (SES) in Table II.

Plant shoots were harvested for the determination of Na+ and K+ concentrations at 20d after the start of salt stress treatments. Few seedlings were left in the treated solution to understand the maximum survival days for each of the tested genotypes. Harvested plant samples were dried and ground to understand the maximum survival days for each of the tested treatments. Few seedlings were left in the treated solution and K+ concentrations at 20d after the start of salt stress growth stage of the plant (IRRI, 1988); where 1 - Germination, 2 - Seedling, 3 - Tillering, 4 - Stem elongation, 5 - Booting, 6 - Heading, 7 - Milk stage, 8 - Dough stage, and 9 - Mature grain. Salt injury rating was done between growth stage 3-4 and was rated as following modified standard evaluation scores (SES) in Table II.

Plant shoots were harvested for the determination of Na+ and K+ concentrations at 20d after the start of salt stress treatments. Few seedlings were left in the treated solution to understand the maximum survival days for each of the tested genotypes. Harvested plant samples were dried and ground to a fine powder and about 0.1 g was transferred to a test tube containing 10 mL of 0.1 N acetic acid and heated in a water bath at 80 °C for 2 h as described by Ansari and Flowers [25].

TABLE II: MODIFIED STANDARD EVALUATION SCORE (SES) OF VISUAL SALT INJURY

| Source | Observation | Tolerance level |
|--------|-------------|----------------|
| 1      | Normal growth | High tolerant |
| 3      | Nearly normal growth leaf tips or few leaves whithed and rolled | Tolerant |
| 5      | Growth severely retarded, most leaves rolled; only a few are elongating | Moderately tolerant |
| 7      | Completely cessation of Growth most leaves dry; some plants drying | Susceptible |
| 9      | Almost all plants dead or dying | High susceptible |

(Source: [20])

E. Plant Materials

29 samples of dry leaves packed in paper envelopes were received on 23rd November 2019 with a request of analyzing salt tolerance. Received samples were stored at 40°C.

F. Collection of leaf samples for molecular analysis

Fresh leaf samples were collected from 21-day-old seedlings to extract genomic DNA. Initially, a healthy portion of the youngest leaves of the tiller was cut apart with sterilized scissors and washed in distilled water and ethanol (70%) and dried on fresh tissue paper to remove spore of microorganisms and any other source of foreign DNA. The collected leaf samples were then kept in white polythene bags containing silica gel. The leaf samples were then taken to the Laboratory at SUA and stored for a week and then DNA extraction was performed.

G. DNA Extraction

Genomic DNA was isolated from leaves of 21-day-old plant based on the DNA isolation protocol of [23]. Samples were ground to powder using the geno-grinder. A 1000 μl ice cold extraction buffer was added, and then incubated in a boiling water bath for 7 minutes and then incubated on ice for 5 minutes. Six microliters (6 μl) Ranse —Al was added and incubated at 37 °C for 40 minutes then spun at maximum speed for 10 minutes and supernatants transferred to new tubes. One tenth (1/10) volume of 7.5 M AOAC and equal volume of ice-cold isopropanol was added and then incubated at -20 °C for 30 minutes. Samples were then centrifuged at regular speed and supernatant discarded and pellets dissolved in 600 μl of 2 MAOAC for 30 minutes then centrifuged, and supernatant was transferred into new tubes. Equal volume of isopropanol was added, mixed by inversion, and incubated at -20°C for 45 minutes; and afterward centrifuged then supernatant discarded, and pellets washed in 70% ethanol, centrifuged and ethanol discarded, and DNA pellet air-dry. The DNA pellets were resuspended in 60μl of 1x T.E buffer for use later.

H. Polymerase Chain Reaction (PCR)

PCR was performed with SSR marker in 29 total reactions. The expected sizes were 190bp amplifying UPR1 95-17 gene and 150 bp amplifying UPR93-287R gene. Once double bands are obtained, we expected the PSD3 gene. The PCR total volume reaction was 20 with the following components shown in Table III.

TABLE III: PCR COMPONENTS AND REACTIONS

| Item             | 1 reaction | 29 Reactions |
|------------------|------------|--------------|
| PCR water        | 17         | 493          |
| Forward primer   | 1          | 29           |
| Reverse primer   | 1          | 29           |
| DNA              | 1          | -            |
| TOTAL            | 20μl       | -            |

I. Agarose Gel Electrophoresis

Primer RM336 was used for the study. Amplified microsatellite loci were analyzed for polymorphism using 2% Agarose Gel Electrophoresis and the result revealed that the primer detected clear polymorphism among the rice genotypes analyzed. The primer was polymorphic and showed clear bands for most rice genotypes.

III. RESULTS

Variability was observed in plant turgidity immediately after seedlings imposition in treated solution. Less turgidity loss was observed in tolerant checks (SATO 1) and some of
treated lines. And strong turgidity loss was observed in a susceptible check (TXD 306). Previously, Munns (2002) reported that saline solutions impose both ionic and osmotic stresses on plants. Kashenge-Killenga et al. (8) reported the importance of vigorous growth at early stages of plant development. They reported that vigorous growth provides a dilution effect of the salts concentration in plant tissues therefore increases survival of the plant under saline condition.

The tested lines showed wide phenotypic variations in reaction to salt stress. At 10 d Sm-1 29 lines out of 102 (28.43%) indicated phenotypic tolerance to salt injury (Table I and Table II).

The lines namely SR35266-2-18-2-1, SR35250-1-19-1-1, SR23364-128-1762-1-HV-1-1, SR35230-1-12-1-1, SR35236-128-1986-1-HV-1-1, SR34590-HB3433-4-1-1, SR35266-2-7-1-1, PBR1009922-1, SR34053(#5-52)-1-4-2-10-3-3, others includes SR35266-3-1-5-1, SR34574-2-10-3-1-2-1, SR35278-2-10-1-1, SR35250-2-3-1-1, SR35266-3-2-3-1, SR35266-3-2-4-1, SR23364-133-184-1-HV-1-1, SR34592-HB-1-HV-1 and SR34042F3-22-1-1-5-3 indicated tolerance to salt. These lines also indicated having high dry matter.

The genotypic variability for salinity tolerance was assessed and expressed as less salt injury symptoms, low Na+ accumulation and Na+/K+ ratio in plant tissues and high biomass accumulation (fresh weight and dry weight). The Na+ / K+ ratio indicated significant variation among the genotypes tested. The lines namely SR35266-2-7-1-1, SR23364-128-1762-1-HV-1-1 and SR34590-HB3433-4-1-1 indicated low Na+ concentration expressing similar trend shown during the morphological characterization as damage of leaves associate with the concentration level of sodium ions in the plant tissues.

29 lines that expressed phenotypic tolerance to salt stress responded differently under molecular screening using primer RM 336 where 11 genotypes were heterozygous to salt tolerance allele indicating the potential tolerance to salinity stress.

| Source of variation | d.f. | 4dSm⁻¹ | 6dSm⁻¹ | 8dSm⁻¹ | 10dSm⁻¹ | FRESH WEIGHT | DRY WEIGHT |
|---------------------|------|--------|--------|--------|----------|--------------|------------|
| REP                 |      | 2.00   | 1.00   | 0.67   | 0.50     | 1.64         | 0.38       |
| Entry               | 100  | 0.80   | 1.40   | 1.35   | 2.71*    | 0.19         | 0.05       |
| Residual            | 202  | 0.68   | 1.79   | 1.59   | 2.62     | 0.17         | 0.06       |
| Total               | 305  |        |        |        |          |              |            |

| GENOTYPE            | 4dSm⁻¹ | 6 dSm⁻¹ | 8 dSm⁻¹ | 10dSm⁻¹ | FRESH WEIGHT | DRY WEIGHT |
|---------------------|--------|---------|---------|----------|--------------|------------|
| SR35266-2-18-2-1    | 2.33   | 3.00    | 5.00    | 7.00     | 0.82         | 0.58       |
| SR35250-1-19-1-1    | 2.33   | 3.00    | 2.67    | 2.00     | 0.92         | 0.61       |
| SR35250-2-19-1-1    | 1.67   | 4.33    | 5.67    | 7.67     | 0.61         | 0.63       |
| SR35230-2-9-2-2     | 2.33   | 3.00    | 5.67    | 8.33     | 0.62         | 0.48       |
| SR35266-2-12-2-1    | 1.67   | 3.00    | 4.33    | 6.33     | 0.93         | 0.66       |
| SR34598-HB-8-HV-1   | 3.00   | 5.00    | 7.00    | 9.00     | 0.51         | 0.36       |
| SR35266-3-3-1-1     | 2.33   | 3.67    | 5.67    | 7.00     | 0.91         | 0.58       |
| SR23364-128-1762-1-HV-1-1 | 2.33 | 2.33 | 4.33 | 5.00 | 1.64 | 0.85 |
| SR35230-1-12-1-1    | 1.67   | 3.67    | 5.00    | 6.33     | 0.84         | 0.60       |
| SR23364-128-1982-1-HV-1-1 | 2.33 | 3.67 | 5.67 | 7.00 | 0.73 | 0.53 |
| SR34590-HB3433-4-1-1 | 1.67 | 3.67 | 5.00 | 6.33 | 0.97 | 0.56 |
| SR35266-2-7-1-1     | 1.67   | 3.00    | 5.67    | 7.00     | 1.06         | 0.66       |
| SR34054-1-21-4-3-1-3 | 2.33 | 3.67 | 5.67 | 7.00 | 0.91 | 0.61 |
| SR34590-HB3433-1-3-1 | 2.33 | 4.33 | 5.67 | 7.67 | 0.76 | 0.50 |
| SR35263-HB3415-26-1 | 3.00 | 5.00 | 5.00 | 7.67 | 0.73 | 0.54 |
| SR34590-HB3433-8-1-1 | 2.33 | 5.00 | 5.67 | 7.67 | 0.67 | 0.51 |
| SR33705F2-60-1-2-HV-1-2 | 3.00 | 5.00 | 5.67 | 7.67 | 0.67 | 0.48 |
| SR34053(#5-52)-1-4-2-10-3-1 | 3.00 | 4.33 | 6.33 | 9.00 | 0.46 | 0.30 |
| PBR1009922-1        | 1.00   | 3.00    | 4.33    | 5.67     | 1.18         | 0.81       |
| SR34053(#5-52)-1-4-2-10-3-3 | 1.67 | 3.00 | 5.00 | 6.33 | 1.08 | 0.87 |
| SR35266-3-1-5-1     | 1.67   | 3.00    | 5.00    | 6.33     | 0.90         | 0.61       |
| SR34053(#5-52)-1-4-2-10-3-2 | 2.33 | 4.33 | 6.33 | 8.33 | 0.54 | 0.44 |
| SR35250-2-19-3-1    | 2.33   | 4.33    | 6.33    | 8.33     | 0.56         | 0.39       |
| SR34574-2-10-3-1-2-1 | 2.33 | 4.33 | 6.33 | 8.33 | 0.56 | 0.38 |
| SR35278-2-10-1-1    | 2.33   | 3.00    | 5.67    | 7.00     | 0.83         | 0.60       |
| SR35250-2-3-1-1     | 1.67   | 3.67    | 5.00    | 6.33     | 1.10         | 0.67       |
### Cont. of Table V

| GENOTYPE           | 4dSm$^1$ | 6 dSm$^1$ | 8 dSm$^1$ | 10dSm$^1$ | FRESH WEIGHT | DRY WEIGHT |
|--------------------|----------|-----------|-----------|-----------|--------------|------------|
| SR35266-2-7-2-1    | 2.33     | 3.67      | 5.67      | 7.00      | 1.10         | 0.77       |
| SR35266-3-2-3-1    | 2.33     | 3.67      | 5.67      | 7.00      | 0.88         | 0.54       |
| SR35266-2-7-3-1    | 2.33     | 3.67      | 5.67      | 7.67      | 0.85         | 0.59       |
| SR35266-3-2-4-1    | 1.67     | 3.67      | 5.00      | 6.33      | 1.02         | 0.66       |
| ARICA3             | 3.00     | 4.33      | 6.33      | 9.00      | 0.46         | 0.41       |
| SR35278-2-10-1-2   | 3.00     | 3.67      | 6.33      | 9.00      | 0.48         | 0.42       |
| SR34042F3-22-1-1-5-2| 3.00   | 5.00      | 6.33      | 9.00      | 0.46         | 0.38       |
| SR34042F3-22-1-1-5-3| 3.00 | 5.00      | 6.33      | 9.00      | 0.53         | 0.44       |
| SR34590-HB3433-1-2-1| 3.00 | 5.00      | 6.33      | 9.00      | 0.55         | 0.41       |
| SR23364-133-261-1-HV-1-2| 3.00 | 4.33      | 6.33      | 9.00      | 0.38         | 0.30       |
| SR23364-133-184-1-HV-1-1| 2.33 | 3.67      | 5.67      | 7.00      | 0.90         | 0.59       |
| SR34054-1-21-4-1-2-3| 2.33    | 3.67      | 5.67      | 7.00      | 1.01         | 0.69       |
| SR34592-HB-1-HV-1  | 1.67     | 3.67      | 5.00      | 6.33      | 0.88         | 0.55       |
| PBR1000922-2       | 2.33     | 3.67      | 5.67      | 7.00      | 1.01         | 0.62       |
| SR23364-128-1938-1-HV-1| 2.33 | 3.67      | 5.67      | 7.00      | 0.76         | 0.51       |
| SR34590-HB3433-3-1-1| 1.67    | 2.33      | 4.33      | 5.00      | 1.38         | 0.80       |
| SR35230-1-13-3-1   | 1.67     | 3.67      | 5.67      | 7.67      | 0.62         | 0.48       |
| SR33705F2-59-2-2-HV-1| 3.00   | 5.00      | 7.00      | 9.00      | 0.49         | 0.41       |
| SR35266-2-18-1-1   | 2.33     | 5.00      | 6.33      | 8.33      | 0.56         | 0.44       |
| SR34590-HB3433-5-1-1| 1.67    | 3.00      | 4.33      | 6.33      | 1.15         | 0.68       |
| SR34042F3-22-1-1-1-3| 2.33    | 4.33      | 5.67      | 8.33      | 0.52         | 0.41       |
| SR34590-HB3433-4-2-1| 2.33    | 4.33      | 6.33      | 8.33      | 0.39         | 0.31       |
| PBR1000653-2       | 2.33     | 3.67      | 6.33      | 8.33      | 0.70         | 0.47       |
| SR33705F2-60-2-2-HV-1-1| 2.33    | 2.33      | 5.67      | 5.00      | 0.88         | 0.55       |
| SATO 1             | 2.33     | 3.67      | 5.00      | 6.33      | 1.45         | 0.79       |
| SATO 9             | 2.33     | 3.67      | 5.00      | 7.00      | 1.19         | 0.74       |
| TXD 85             | 1.67     | 2.33      | 3.67      | 5.00      | 1.54         | 0.87       |
| TXD 88             | 2.33     | 4.33      | 6.33      | 8.33      | 0.64         | 0.45       |
| KOMBOKA            | 2.33     | 4.33      | 6.33      | 8.33      | 0.61         | 0.43       |
| TXD 306            | 3.00     | 5.00      | 7.00      | 9.00      | 0.52         | 0.38       |
| 2805               | 2.33     | 4.33      | 6.33      | 8.33      | 0.53         | 0.34       |
| 2810               | 2.33     | 4.33      | 6.33      | 8.33      | 0.54         | 0.40       |
| 2823               | 2.33     | 3.67      | 6.33      | 8.33      | 0.52         | 0.38       |
| 2826               | 2.33     | 4.33      | 6.33      | 8.33      | 0.46         | 0.33       |
| 2851               | 1.67     | 4.33      | 5.67      | 7.67      | 0.62         | 0.43       |
| 2851               | 1.00     | 3.67      | 5.00      | 7.00      | 0.69         | 0.53       |
| 2898               | 1.67     | 3.67      | 5.67      | 7.67      | 0.72         | 0.49       |
| 2289               | 1.67     | 3.67      | 5.67      | 7.67      | 0.71         | 0.51       |
| 2855               | 1.00     | 3.67      | 5.00      | 7.00      | 0.90         | 0.66       |
| 2853               | 1.67     | 3.67      | 5.67      | 7.67      | 0.59         | 0.45       |
| 2889               | 1.67     | 3.67      | 5.67      | 7.67      | 0.76         | 0.54       |
| 2700               | 1.67     | 3.00      | 5.67      | 7.00      | 0.74         | 0.56       |
| 2784               | 2.33     | 3.00      | 6.33      | 7.67      | 0.79         | 0.60       |
| 2714               | 2.33     | 4.33      | 6.33      | 8.33      | 0.66         | 0.49       |
| 2748               | 3.00     | 4.33      | 7.00      | 9.00      | 0.45         | 0.40       |
| 2754               | 2.33     | 4.33      | 6.33      | 8.33      | 0.50         | 0.43       |
| 2729               | 2.33     | 4.33      | 5.67      | 7.67      | 0.75         | 0.49       |
| 2782               | 1.67     | 3.00      | 5.67      | 7.67      | 0.75         | 0.50       |

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| GENOTYPE      | 4dSm³ | 6 dSm³ | 8 dSm³ | 10dSm³ | FRESH WEIGHT | DRY WEIGHT |
|--------------|-------|--------|--------|--------|--------------|------------|
| 2796         | 1.67  | 3.00   | 5.67   | 7.67   | 0.75         | 0.47       |
| 2733         | 2.33  | 4.33   | 6.33   | 8.33   | 0.56         | 0.44       |
| 2711         | 1.67  | 3.67   | 5.00   | 6.33   | 1.25         | 0.84       |
| 5788         | 2.33  | 3.67   | 5.67   | 7.67   | 0.74         | 0.58       |
| 6079         | 2.33  | 3.67   | 5.67   | 7.67   | 0.64         | 0.51       |
| 6150         | 1.67  | 4.33   | 6.33   | 8.33   | 0.68         | 0.50       |
| 5668         | 2.33  | 3.67   | 6.33   | 8.33   | 0.62         | 0.45       |
| 6729         | 2.33  | 4.33   | 6.33   | 8.33   | 0.57         | 0.38       |
| 5646         | 2.33  | 4.33   | 6.33   | 8.33   | 0.52         | 0.37       |
| 5281         | 1.00  | 3.67   | 5.00   | 7.00   | 0.90         | 0.56       |
| 6155         | 1.67  | 3.67   | 5.67   | 7.67   | 0.60         | 0.42       |
| 6149         | 1.67  | 4.33   | 5.67   | 7.67   | 0.72         | 0.49       |
| 6137         | 1.67  | 3.00   | 5.67   | 7.67   | 0.95         | 0.57       |
| 6198         | 2.33  | 4.33   | 6.33   | 8.33   | 0.57         | 0.38       |
| 5662         | 1.67  | 5.00   | 5.67   | 7.67   | 0.60         | 0.43       |
| 5859         | 2.33  | 4.33   | 6.33   | 8.33   | 0.65         | 0.49       |
| 6183         | 3.00  | 4.33   | 7.00   | 9.00   | 0.44         | 0.36       |
| 5289         | 3.00  | 5.00   | 7.00   | 9.00   | 0.34         | 0.28       |
| 4424         | 3.00  | 5.00   | 7.00   | 9.00   | 0.48         | 0.36       |
| 3740         | 2.33  | 4.33   | 6.33   | 8.33   | 0.61         | 0.49       |
| 6151         | 2.33  | 5.00   | 6.33   | 8.33   | 0.61         | 0.43       |
| 6135         | 2.33  | 3.67   | 5.67   | 8.33   | 0.63         | 0.43       |
| 6050         | 2.33  | 4.33   | 6.33   | 8.33   | 0.52         | 0.39       |
| 4444         | 1.00  | 4.33   | 5.00   | 7.00   | 0.66         | 0.46       |
| 5117         | 1.67  | 4.33   | 5.67   | 7.67   | 0.59         | 0.43       |
| 59806        | 2.33  | 3.67   | 6.33   | 8.33   | 0.59         | 0.43       |

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| ZAMBIA       | 2.33  | 4.33   | 5.67   | 7.67   | 0.68         | 0.50       |

| %CV          | 37.80 | 34.10  | 21.80  | 21.20  | 56.10        | 46.00      |
| LSD          | 1.33  | 2.15   | 2.03   | 2.61   | 0.67         | 0.38       |

| TABLE VI: K⁺ AND Na⁺ CONCENTRATIONS IN REACTION TO SALINITY TOLERANCE |
|---------------------------------------------------------------|
| Genotypes | K (%) | Conc K (mg/l) | Na (%) | Conc Na (mg/l) | K: Na |
| SR35266-2-18-2-1  | 0.77  | 77.00         | 14.96  | 1496.00        | 0.59  |
| SR35250-1-19-1-1  | 0.77  | 77.00         | 10.38  | 1038.00        | 0.07  |
| SR23364-128-1762-1-HV-1-1 | 0.69 | 69.00         | 7.33   | 733.00         | 0.09  |
| SR35230-1-12-1-1  | 1.15  | 115.00        | 10.76  | 1076.00        | 0.11  |
| SR34590-HB3433-4-1-1 | 0.85 | 85.00         | 9.74   | 974.00         | 0.09  |
| SR35266-2-7-1-1   | 0.69  | 69.00         | 7.07   | 707.00         | 0.10  |
| PBR1000922-1      | 1.00  | 100.00        | 12.54  | 1254.00        | 0.08  |
| SR34053(-5.52)-1-4-2-10-3-3 | 1.38 | 138.00        | 13.30  | 1330.00        | 0.10  |
| SR35266-3-1-5-1   | 1.23  | 123.00        | 13.30  | 1330.00        | 0.09  |
| SR34053(-5.52)-1-4-2-10-3-2 | 1.15 | 115.00        | 19.28  | 1928.00        | 0.06  |
| SR35250-2-19-3-1  | 1.38  | 138.00        | 12.92  | 1292.00        | 0.11  |
| SR34574-2-10-3-1-2-1 | 1.23 | 123.00        | 16.61  | 1661.00        | 0.07  |
| SR35278-2-10-1-1  | 1.38  | 138.00        | 16.74  | 1674.00        | 0.08  |
| SR35250-2-3-1-1   | 0.85  | 85.00         | 19.53  | 1953.00        | 0.04  |
| SR35266-2-7-2-1   | 1.61  | 161.00        | 19.02  | 1902.00        | 0.08  |
| SR35266-3-2-3-1   | 1.91  | 191.00        | 17.37  | 1737.00        | 0.11  |
| SR35266-2-7-3-1   | 1.15  | 115.00        | 12.79  | 1279.00        | 0.09  |
| SR35266-3-2-4-1   | 1.38  | 138.00        | 16.61  | 1661.00        | 0.08  |
| SR23364-133-184-1-HV-1-1 | 1.53 | 153.00        | 15.59  | 1559.00        | 0.10  |
### Cont. of Table VI

| Genotype | K (mol L⁻¹) | Na (mol L⁻¹) | K+ (mg L⁻¹) | Na+ (mg L⁻¹) | K: Na |
|----------|-------------|--------------|-------------|--------------|-------|
| SR34054-1-21-4-1-2-3 | 1.15 | 115.00 | 18.90 | 1890.00 | 0.06 |
| SR34592-HB-1-HV-1 | 1.61 | 161.00 | 11.78 | 1178.00 | 0.14 |
| PBR1000922-2 | 1.68 | 168.00 | 15.46 | 1546.00 | 0.11 |
| SR23364-128-1938-1-HV-1 | 1.64 | 164.00 | 23.16 | 2316.00 | 0.07 |
| SR34590-HB3433-3-1-1 | 1.84 | 184.00 | 12.54 | 1254.00 | 0.15 |
| PBR1000653-2 | 1.15 | 115.00 | 10.25 | 1025.00 | 0.11 |
| SR3402F3-22-1-1-5-3 | 1.23 | 123.00 | 11.01 | 1101.00 | 0.11 |

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**Fig. 2.** K+ and Na+ concentrations of 28 genotypes with morphological tolerance to salinity.

**K: Na**

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**Fig. 3.** K+ and Na+ concentrations of 11 genotypes with genetic tolerance to salinity.
The molecular study indicated the genotypes SR35266-2-18-2-1, SR35250-1-19-1-1, SR35230-1-12-1-1, SR35264-128-1982-1-HV-1-1, SR34590-HB3433-4-1-1, SR35266-2-7-1-1, SR34053(#5-52)-4-10-3-1, SR34053(#5-52)-1-4-2-10-3-3, SR35266-3-1-5-1, SR34053(#5-52)-1-4-2-10-3-2, SR35250-2-19-3-1, SR34574-2-10-3-1-2-1, SR35278-2-10-1-1, SR35250-2-3-1-1 and SR35266-2-7-2-1 high level of tolerance to salinity (Fig. 2 and Fig. 3).

IV. DISCUSSION

Rice is sensitive to salt stress particularly at the seedling stage and during reproduction [16]. The stress constrains sustainable rice production in various rice agro-ecologies worldwide [19]. Rice among the important food crop in Tanzania is facing similar circumstances, in many irrigation schemes conditions are becoming worse as the salinity level increased as EC was raised to 8 dSm⁻¹ and SR34590 and SR35266 used in molecular characterization to identify potential lines with high level of tolerance to salinity stress. The 11 identified genotypes can be potential sources of salt tolerance and can be used in hybridization program for generating new salt tolerant genotype. The genotypes can also be used in construction of mapping population to study the genetic architecture of salt tolerance followed by cloning of candidate gene and maker assisted introgression of such gene.

V. CONCLUSION

The present study has employed both morphological and molecular characterization to identify potential lines with high level of tolerance to salinity stress. The 11 identified genotypes can be potential sources of salt tolerance and can be used in hybridization program for generating new salt tolerant genotype. The genotypes can also be used in construction of mapping population to study the genetic architecture of salt tolerance followed by cloning of candidate gene and maker assisted introgression of such gene.

VI. RECOMMENDATION

The identified tolerant lines should be tested in salt affected soils in different rice agro-ecologies of Tanzania. The aim is to study their performance in under field environment.

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CONFLICT OF INTEREST

Authors declare that they do not have any conflict of interest.

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Fig. 4. Gel profile of 48 rice genotypes with marker RM 336.
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