Climate Smart Agriculture for a Resilient Coastal BangladeshRECSA-OKP-BGD-103561

Project for a collaborative research project (WP3)

Screening of landraces rice cultivars for salinity tolerance and utilization of organic soil amendments for mitigating soil salinity in rice

Abstract

Bangladesh is one of the most densely populated, agro-based developing countries in the world. Agriculture is an important sector of Bangladesh for job opportunities, gross domestic growth, and nutrient supply, which are contributing to the food security of Bangladesh. High soil salinity causes a significant reduction in growth and yield of rice in Bangladesh. Salinity causes osmotic stress and ionic imbalances, and thus limits crop productivity. The accumulation of organic compounds is one of the adaptive mechanisms to salinity in plants. The present investigation will be carried out to screen landrace rice genotypes for salt tolerance at seedling stage and to evaluate the effects of soil organic amendments to minimize negative effects of soil salinity on rice. In plate and hydroponics experiments, growth parameters like root length, shoot length and plant biomass will be measured after 14 days of exposure to four different levels of saline solution (EC's of 0, 4, 8 and 12 dS m^{-1}). Soil amendments like farmyard manure (FYM) with the rate of 5 and 10 t ha^{-1}) will be applied to evaluate the effects on growth, grain and straw yield of target rice genotypes under saline conditions. Nutrient uptake status and the K^+/Na^+ ratio in rice will also be measured and any effect of the application of organic manures will be assessed. After salt tolerance screening of seedlings through plate and hydroponic experiments, the most salt tolerant rice genotypes will be grown in pot culture containing sandy loam soil for measuring growth parameters and yield.

1. **Project Title**: **Screening of landraces rice cultivars for salinity tolerance and utilization of organic soil amendments for mitigating soil salinity in rice**
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3. Introduction, identification of problem and justification of the project

Rice (*Oryza spp*.) is an important cereal crop and is mainly used for human consumption. 100 g of rice provides 345.0 kcal, 78.2 g of carbohydrates and 6.8 g of protein (Gopalan et al. 2007), including considerable amounts of recommended Zinc and niacin. Rice provides almost 50–80% of daily caloric intake amongst the poor class of the society. It's a staple food and cash crop for more than three billion people in the World (Ma et al. 2007). Asian farmers constitute about 92% of the world's total rice producing group (Mitin 2009). In Asia 90% of rice is produced by smallholder farmers who are solely dependent on rice for their livelihood and food security (ANU 2006). Salinity is the second most wide spread soil problem in rice growing countries like Bangladesh after drought, and is considered as a serious limitation to increase rice production worldwide (Gregorio 1997). Approximately 7% of the world's land area, 20% of the world's cultivated land and nearly half of the irrigated land are affected by soil salinity (FAO, 2008). In the coastal area of Bangladesh, soil salinization is a major process of land degradation that decreases soil fertility and crop productivity (Kibria et al. 2015). Sea level rise occurs due to climate change and cyclonic events have already led to an increased salinity in fresh water and soil in most of the coastal areas. Soil salinization is a regionally or locally important process of soil degradation, resulting in temporal or permanent loss of arable land, and finally reduced plant growth. Accumulation of excess Na and Cl in plant body causes ionic imbalances that may impair the selectivity of root membranes and induce potassium deficiency (Gadallah, 1999). The deficiency of K initially leads to chlorosis and then causes necrosis (Gopal and Dube, 2003). Excess soluble salts reduce yields by impairing germination, or creating two-fold stress of osmotic gradients and ionic problems which interfere with the uptake of essential nutrients by plants (Stamatiadis et al., 1999, Mahajan and Tuteja, 2005). Ionic stress both leads to difficulties finding enough K, and also to Na accumulation in the leaves to toxic concentrations. It causes yield reduction and also reduces caloric and nutritional potential of agricultural products (Yokoi et al. 2002) causing leaf injury or death of plants, thus exceeding the capacity of salt compartmentalization in cytoplasm (Munns et al. 2006).

Quijano-Guerta and Kirk (2002) reported that the easiest way to address the problem of salinity is through the development of a salt tolerant rice variety. Screening of rice cultivars at field level proved to be difficult due to soil heterogeneity, climatic factors and other environmental factors which caused differentially affected plant physiology, thereby frustrating the ability to draw any sounds conclusions. Hence, screening under laboratory conditions is considered to be advantageous over field screening. In rice, screening at seedling stage is a rapid method and based on simple criteria. In the vegetative stage of rice plants, root length, shoot length and biomass have been proven as potential indicators for salt tolerance (Flowers and Yeo 1995). It has also been reported that the assessment of the actual salt tolerance of the genotypes may be determined by comparisons of their biomass production only after a long growth period (Leland et al. 1994); which therefore serves as another criterion to evaluate the salt tolerance. However, the salt tolerance at early growth stages does not always correlate with that of subsequent growth stages (Ferdose et al. 2009). In this study, therefore, we focus on evaluating the potential of salt tolerance in rice genotypes at an early growth stage i.e. at seedling stage through agar plate and hydroponics culture.

Appropriate management strategies and techniques with suitable crop genotypes having higher yield potential could contribute to the improvement of crop production in the coastal areas of Bangladesh. Various organic amendments such as farmyard manure, compost, poultry manure and mulching can be used for the amelioration of saline soils. One method for decreasing negative salinity effects is the incorporation of organic materials into soil (Wichern et al., 2006), due to the beneficial effects on soil physical, chemical, and biological properties (Iqbal et al., 2016; Chahal et al., 2017; Leogrande and Vitti, 2019). The positive biological effects are most likely caused by the positive effect of available carbon (C) derived from the added organic matter to microbial cells allowing their adjustment to osmotic stress by producing osmolytes, which counteract osmotic stress (Wichern et al., 2006). Soil salinity management did not receive proper attention in the past, but now emphasis has been given on this issue. Sustainable and profitable production of crops in salt-affected areas is possible if appropriate soil management is taking place. There is no systematic information in Bangladesh on the role of organic amendments in the mitigation of salinity in rice cultivation.

4. Project objectives:

Rice is the major food crop consumed globally. However, the production is severely affected by high salinity levels in soil, particularly at the seedling stage. In this present global perspective where increasing rice production is a necessitys to meet future demands, the proposed project objectives are:

1. To assess the performance of landraces rice genotypes in terms of seedling growth parameters at various salinity levels,
2. To study selected morphological descriptors such as salinity index (useful for screening landraces germplasms) and to identify true salinity tolerant genotypes,

3. To evaluate the role of organic soil amendments to ameliorate soil salinity;
4. To observe the effect of different salinity levels on several physiological and morphological parameters of rice;
5. To investigate the salinity and water balance in soil after addition of soil amendments.
6. To validate and upscale the nutrient management technologies to improve soil fertility, improve crop yield and enhance food security at the coastal area of Bangladesh.

5. Methodology:

A) Study area and preliminary activity: The project will be implemented at the Department of Soil Science, Patuakhali Science and Technology University (PSTU), Patuakhali. On-station research will be implemented by the project group leader and one MS student. Petri dish and Hydroponics experiments will be conducted at the net house of Soil Science department of PSTU to screen the salt tolerant rice genotypes.

B) **Screening of salt tolerant rice genotypes**

Landraces rice cultivars will be selected for screening of salinity tolerant rice genotypes.

B.1 Plant materials in petri plate culture:

Seeds of locally grown rice (*Oryza sativa* L.) genotypes will be collected from a certified seed company of the study area, from local farmers and markets, and from the Bangladesh Rice Research Institute (BRRI). After collection, all rice genotypes will be grown on petridish cultures for germination test under normal condition. Two salinity levels (4 and 12 dS m^{-1}) with three replicates per salt treatment will be added to the petri dishes. Around 25 rice seeds of each genotypes will be used in each petri dish. After germination test, maximum number of germinated seeds will be used for salinity tolerant screening.

B.2 Plant materials and growth conditions in hydroponics:

From the land races rice cultivars, seeds of high germination percentage will be used for salinity tolerance test under hydroponics. Rice seeds will be surface sterilized with bleach and sown onto 0.8% agar medium supplemented with 0.5 mM calcium chloride [$CaCl_2$, containing 2 mM 2-morpholinoethanesulfonic acid (MES) (pH 5.7)]. After germination, the seedlings will be transferred to a half-strength Kimura B solution and grown hydroponically for 14 d under natural light condition.

B.3 Treatment conditions for hydroponic experiment:

Standard rice culture solution will be used in hydroponic experiments with modified Kimura B solution. In hydroponics experiment, four levels of NaCl nutrient solution, (0, 4, 8 and 12 dS

m^{-1}) will be applied. Deionized water will be used for the nutrient solution. In the hydroponic experiment, the number of treatments from three replications after the germination tests (10 rice genotypes) will be: 10×4×3=120.

C. Data collection from petri dish and hydroponic experiments:

C.1 Germination studies: Germination percentage, coleoptile and radicle length under varying degree of salt stress for rice genotypes will be measured.

C.2 Growth and developmental parameters: Shoot growth and developmental parameters include plant height (PH) and leaf area (LA) for the selected rice genotypes. Shoot dry weight (SW), root dry weight (RW) and total dry weight (TD) will be measured.

C.3 Physiological parameters: The physiological indices like shoot length stress index (SLSI), root length stress index (RLSI), shoot dry weight stress index (SDSI) and root dry weight stress index (RDSI), chlorophyll contents (CH), anthocyanins (ANT) and nitrogen balance index (NBI), will be measured. Ion concentration such as Na, K, Ca, Cu, Zn, As, Cd, Mg, Mo, and Ni from root and shoot will be measured using standard protocols. C/N ratio of root and shoot will also be measured.

D. Pot experiment (control soil condition)

Salt tolerant rice genotypes (screening from petridish and hydroponics) will be used for a pot experiment under controlled soil conditions (sandy loam soil). The pot experiment will be carried out in the department of Soil Science, PSTU, Dumki, Patuakhali, Bangladesh. Initial soil samples will be analyzed for pH, total N, available P, exchangeable K, available S, organic carbon, CEC and EC.

Table 1. Treatment conditions for the proposed study

Treatment No.	Symbol	Treatment
1.	T0	Control (without organic amendments and saline water)
2.	T1	4 dS m^{-1} saline water
3	T2	8 dS m^{-1} saline water
4.	T3	12 dS m^{-1} saline water
5.	T4	Compost @ 5 t ha^{-1}
6.	T5	Compost @ 10 t ha^{-1}
7.	T6	Compost @ 5 t ha^{-1} with 4 dS m^{-1} saline water

8.	T7	Compost @ 10 t ha^{-1} with 4 dS m^{-1} saline water
9.	T8	Compost @ 5 t ha^{-1} with 8 dS m^{-1} saline water
10.	T9	Compost @ 10 t ha^{-1} with 8 dS m^{-1} saline water
11.	T10	Compost @ 5 t ha^{-1} with 12 dS m^{-1} saline water
12.	T11	Compost @ 10 t ha^{-1} with 12 dS m^{-1} saline water

Saline solution will be created by adding salt to tap water to make different levels of salinity. Plants will be irrigated with 4, 8 and 12 dS m^{-1} salt solution from 14 days after sowing (DAS) to maturity and control plants will be irrigated with tap water. The organic amendments will be mixed with potting soil. Soil Resources and Development Institute (SRDI) recommended doses of TSP, MoP, gypsum, and zinc sulphate will be applied to pot soil. The experiment will be laid out in a randomized complete block design (RCRD) with three replications.

Growth parameters: Shoot growth and developmental parameters including plant height (PH), tiller number (TN), and leaf area (LA), dry matter production in different plant parts, yield and yield contributing characteristics for the selected rice genotypes will be measured at harvest.

Root image acquisition analysis: After the collection of samples from the pot experiment, roots of all plants will be cut from the stems and washed on a sieve thoroughly and cautiously to avoid any destruction to the overall root structure. All the roots will be then scanned using an optical scanner linked to a computer system. Acquired images will be analyzed for different root parameters including root surface area (SA), total or cumulative root length (TRL), average root diameter (AD), root volume (RV), number of roots having laterals (RNL), number of tips (TP), number of forks (FR), and number of crossings (CR) using an image-J software.

The recorded data will be statistically analysed by the statistical program STAR (Statistical Tool for Agricultural Research)" software to examine the significant variations in the results due to different treatments. The treatment means will be compared by Least Significance Difference (LSD) test at 5% level of significance (Gomez and Gomez, 1984).

Schedule of project activity

Activity of the project (1 year_2021)	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
Collection of land races rice genotypes	■	■										
Germination test of rice genotypes (Petridish)			■	■								
Hydroponic for screening of rice genotypes				■	■	■						
Data collection from hydroponic experiment				■	■	■	■					
Pot exp. to test the yield potential						■	■	■	■	■		
Data collection from pot experiment									■	■	■	
Manuscript and final report submission									■	■	■	■

6. Expected outputs:

The following outputs will be obtained from the proposed study- i. Utilization of organic soil amendments for landraces rice cultivars of the coastal area of Bangladesh adaptive to high salinity stress will be established. ii. The study will be able to investigate the physiology of rice cultivars under salinity stress and have elucidated the complex physiological network involved in the amelioration of the detrimental effect of salinity on crops. iii. The proposed study will be helpful to screen high yielding commercial rice cultivars for salinity tolerance. iv. This study also underlines the need for future research to evaluate the role of organic soil amendments in salinity stress in rice crops and the identification of gaps in understanding of this process as a whole at a broader field level. Finally, the proposed project will be able to publish one scientific article from any high impact international journal (Biology and Fertility of Soils or Archives of Agronomy and Soil Science).

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7. Budget Summary:

Sl. No.	Name of Items	Quantity	Unit Price (Euro)	Total cost (Euro)
1	Combined soil moisture, salinity and temperature meter	1	2700	2700/-
2	Portable pH meter	1	500	500/-
3	Filter paper (Whatmann 42)	4 box	100	400/-
4	Petri plate	15 box (150 Pcs.)	20	300/-
5	3 L plastic pot for hydroponics	20	10	200/-
6	20 L plastic pot for hydroponics	10	30	300/-
7	Electronic balance	2	250	500/-
8	Pesticide sprayer and water sprayer	1/each	250	500/-
9	Printer (Color Laser Printer Lbp-7018C,	1	200	200/-
10	Mini centrifuges Thermo Scientific™	1	250	250/-
11	Different glassware's (flask, beaker, test be etc.,)	Various	Various	1,000/-
12	Chemicals and reagents (NaCl, $CaCl_2$, NH_4Cl, KH_2PO_4, NH_4NO_3, K_2SO_4, $MnSO_4$, K_2SO_4, H_2O_2, KNO_3, HNO_3, H_3BO_3, SiO_2 (Wako, chemicals), MES, Na-EDTA, Fe-citrate, Copper sulphate, Zinc sulphate, Nitric acid,	Various	Various	3,000/-

	H_2O_2, Ammonium Acetate, sodium hypochlorite, etc.,			
13	Stationaries			150/-
Total budget for research work				**10,000/-**
14	MSc student as research assistant	1	250 per month	3,000/-
Total budget for research work + MSc student				**13,000/-**