NBU Journal of
Plant Sciences

Vol. 4, March 2010 p.35-38
ISSN No. 0974 6927

Evaluation of genetic variation among fourteen rice (*Oryza sativa* L.) varieties (Landraces) of North Bengal using morphological traits

Subhas Ch. Roy*, Dipayan Rakshit and Sandeepan Sengupta
DRS Department of Botany, University of North Bengal, PO- NBU-734013, West Bengal, India

Abstract

The Northern region (North Bengal) of West Bengal possesses a rich genetic diversity of rice (*Oryza sativa* L.). This region is known for growing one important rice variety Tulaipanji with GI number in the district of North Dinajpur and many other varieties of local landraces with tolerance to biotic and abiotic stresses. A considerable range of diversity and variation exists among the genetic resources of rice landraces of North Bengal. Prominent fourteen (14) landraces such as Sadanunia, Kalonunia, Dhepi, Banni, Dudkalam, Malsira, Lalpanati, Bhadoi, Ashami, and Gobindabhog, Enda, Chenga, Katharibhog, Tulaipanji are known for their special quality attributes were collected from the different districts of North Bengal. Genetic variation among these landraces was characterized by means of morphological traits such as seed sizes, shape, volume, colour, awn character, surface texture with husk (Microscopic observation) and pericarp colour. The Karl Pearson's simple matching coefficient was used to calculate the genetic diversity among the varieties. Similarity coefficient was ranged from 0.94 to 0.64 with an average of 0.79 among the 14 varieties. Cluster analysis based on genetic similarity of these varieties gave rise to three distinct groups. The results suggested that the level of genetic diversity within this group of rice varieties of North Bengal was sufficient for breeding programs and can be used to establish genetic relationships among them on the basis of morphological traits.

Keywords: Rice, genetic variation, cluster analysis, morphology

Rice (*Oryza sativa* L.) is the principal food crop under the family Poaceae (Graminae) globally providing food and livelihood security to a large section of society at least 2/3rd of the world population. Over the last three decades in response to the declining availability of water and land, rice production has been declined at an alarming level (Childs, 2004). As a consequence, there will be serious food shortage in near future (Sakamoto and Matsuoka, 2008). Attempts are to be made to improve the existing crop production to feed the 9.0 billion world population by 2050. Due to increase in the consumption of rice along with a tremendously growing demand, there is a need to conserve the germplasm of the landraces which is expected to serve as the major way to develop the new high-yielding varieties and to raise the maximum yield potential. Collection, conservation, evaluation of diverse rice germplasm is important for the purpose. It is estimated that about 120000 varieties of rice exist in the world (Khush, 1997), consequent to selection by farmers to suit different habitats and growing conditions across the region. However, the aggressive introduction of modern verities in this region resulted on the loss of many valuable landraces from the farmer's field. Only few varieties/accessions have been employed in breeding programs, hence there is a large amount of rich diversity of rice landraces left untapped. This creates a narrow genetic base rendering the high-yielding varieties vulnerable to unpredicted biotic and abiotic stresses. Extensive screening of germplasm accessions and local landraces of rice for desirable traits can help in identifying landraces suitable for specific breeding programs (Kasem *et al.*, 2009). The genetic architecture of local landraces of rice is shaped and stabilized by natural (biotic and abiotic) and artificial (human) selection, hence they harbour variability for adoptive as well as to some extent productive characteristics. Thus, the local landraces or the wild relatives are more adaptive but less productive. And on contrary, the high-yielding varieties are more productive and less adaptive. The morphological architecture of local landraces of rice seeds is governed by and reflected in forms of their wide range of phenotypic parameters, like weight of the seed, length of the seed, volume of the seed, presence or absence of awn, colour of the pericarp, colour of the seed itself and many more. These landraces can be short, medium or long in terms of seed length. Some local varieties are found to be endowed with a pleasing aroma. Rice seeds also come in many different colours like yellow, deep brown, reddish brown, blackish brown, *etc*. Similarly, the pericarp of these varieties may exhibit colours of different types like yellowish white, light brown, deep brown, *etc*. There is no report on characterization of rice landraces of North Bengal so far carried out still to date.

In the present study, we report on the evaluation of genetic variation existing among the fourteen (14) landraces of rice varieties of North Bengal on the basis

*Corresponding author:
E-mail: subhascr@rediffmail.com

Table 1: Morphological characteristics of 14 rice (landraces) seeds are summarized in this table

Varieties (Landraces)	Weight (mg)	Volume (mm^3)	Length (mm)	Presence of awn	Awn length (mm)	Color of pericarp	Aroma	Color of husk
Sadanunia	15.8	0.159	7.7	Awned	28.6	Yellowish white	Present	Yellow
Kalonunia	12.9	0.127	6.4	Awned	9.7	Yellowish white	Present	Deep brown
Dhepi	23	0.287	7.8	Awned	12.2	Light brown	Absent	Reddish brown
Banni	26	0.273	8.9	Awned	17	Yellowish white	Absent	Blackish brown
Dudkalam	22	0.206	8.1	Awnless	-	White	Absent	Yellow
Malsira	21	0.214	8.1	Awned	12.8	Yellowish white	Absent	Reddish brown
Lalpanati	24.16	0.238	8.5	Awnless	-	Light brown	Absent	Reddish brown
Bhadoi	19.8	0.210	7.6	Awnless	-	Deep brown	Absent	Blackish yellow
Gobindabhog	10.57	0.115	5.6	Awnless	-	Yellowish white	Present	Blackish yellow
Ashami	21.63	0.217	7.7	Awnless	-	Crème color	Absent	Reddish yellow with black spots
Tulaipanji	16.28	0.138	7.9	Awned	31.9	Yellowish white	Present	Reddish yellow
Katharibhog	9.8	0.115	5.3	Awnless	-	Yellowish white	Present	Reddish yellow
Enda	24.5	0.263	7.3	Awned	13.4	Light brown	Absent	Yellowish brown
Chenga	23.7	0.233	7.5	Awnless	-	Light brown	Absent	Blackish brown

of morphological traits such as seed shape, size, colour and texture of the seeds with husk and clustering them using simple matching coefficient for their conservation purposes.

Materials and Methods

Plant materials

Fourteen varieties of rice (landraces) seeds were collected from different districts of North Bengal and maintained in the Plant Genetics & Tissue Culture Laboratory, DRS Department of Botany, University of North Bengal. These are Sadanunia, Kalonunia, Ashami, Dudkalam, Lalpanati, Banni, Malsira, Bhadoi, Dhepi, Gobindabhog, Tulaipanji, Enda, chenga and Katharibhog.

Morphological traits

Weight of single seed: Average single seed weight was measured by measuring total 5 gm of seed of each of the 14 varieties and then divided by the total numbers of rice seeds were taken. Weight of one seed was calculated in the following way- Seeds of 5gm weight/ Total No. of seeds = weight of per seed.

Volume measurement per seed: The volume of each of the seed was measured using a measuring cylinder containing 20 ml of water. The seeds were added to it one by one until the volume of the water reached 25 ml, and then the seeds were taken out and counted. The volume of each of the seeds was enumerated as follows -

Final vol. - Initial vol./Total No. of seeds = Volume of single seeds = mm^3

Measurement of length: Fourteen rice seeds from each of the varieties were taken and then the length was measured using a millimeter paper, and finally the average length was calculated for each varieties of rice.

Study of other phenotypic traits (characteristics): Other phenotypic characteristics like presence of awn, and its length, presence of aroma, color of pericarp, and color of husk were carefully observed in each variety and summarized in table 1.

Morphological data analysis: The work was based on general similarity as judged by the comparison of many characters, each given equal weight. Karl Pearson's coefficient of correlation (expressed as r), was used to determine the correlation between each pair of traits (summarized in table 2). The following formula was used to compute correlation coefficient (Sneath and Sokal, 1973) between two taxonomic units x and y:

$$r = \frac{\sum xy}{N \times SD_x \times SD_y} = \frac{\sum xy}{N \times \sqrt{\frac{\sum x^2}{N}} \times \sqrt{\frac{\sum y^2}{N}}}$$

Here r stands for coefficient of correlation, x stands for (x- arithmetic mean of X), y stands for (y- arithmetic mean of Y), S.D. stands for standard deviation of x/y series.

RESULTS AND DISCUSSION

Morphological traits such as seed texture were studied under Stereomicroscope (E330-ADUI.2x, Olympus) in all the fourteen rice varieties. Surface texture of the fourteen rice land races of North Bengal were depicted in figure 1. Each variety has distinct type of seed husk texture and specifying a unique genetic trait. Seed colour varies from one variety to other (given in table 1) ranging from yellow, brown, blackish to blackish brown. Several characters in relation to morphology of rice seeds of 14 landraces (North Bengal) were studied (given in table 1) and analyzed in detail following which an association co-efficient so called correlation coefficient was numerically calculated for every pair, thereby comparing every variety with the every other variety (Table 2). It was revealed that while the weight of each seed in mg varied from 9.80 recorded in case of Katharibhog to 24.50 recorded in Enda. Volume in mm^3 per seed was the parameter with certain variation among the landraces. Single seed weight was lowest in Katharibhog (9.8 mg) and highest in Banni (26 mg). Similarly lowest volume recorded in Katharibhog (0.115 mm^3) and highest in Dhepi (0.287 mm^3). Length was varies from 5.3 mm (in Katharibhog) to 8.9 mm (in Banni). Awn was present in the following rice varieties *viz.* Sadanunia, Kalonunia, Dhepi, Banni, Malsira, Tulaipanji, and Enda. Awn length was varies from 9.7 mm to 31.9 mm. Specific aroma was recorded in five rice varieties out of fourteen varieties, these are sadanunia, Kalonunia, Gobindobhog, Tulaipanji and Katharibhog. Seed colour varies from yellow to brown to blackish brown (Table 1). The pericarp colour was also different in different varieties. Seed size was defined by its length and the seeds were categorized as 'very long (>7.5 mm), long (6.5-7.5 mm), medium (5.5-6.5 mm), and short (<5.5 mm). Most of the rice seeds were very long, the remaining are being medium sized. Sadanunia (7.7 mm), Dhepi (7.8 mm), Banni (8.5 mm), Dudkalam (8.1 mm), Malsira (8.1 mm), Lalpanati (8.5 mm), Bhadoi (7.8 mm), Ashami (7.7 mm), Tulaipanji (7.9 mm) had length greater than 7.5 mm, Enda (7.3 mm), Chenga (7.5 mm) belonged to the category of long seeds. Gobindabhog (5.6 mm) and Kalonunia (6.4 mm) were the two medium sized varieties while Katharibhog (5.3 mm) was the only exclusively short variety. Awn was reported to be present in a very few varieties like Sadanunia, Kalonunia, Dhepi, Enda, Lalpanati, with a variation in length from 9.7 mm in Kalonunia to 31.9 mm in Tulaipanji. Only five of the fourteen rice landraces were gifted with a pleasing form of aroma. The red, black, brown, purple colour in rice pericarp due to the presence of anthocyanin pigment. Anthocyanins have antioxidant properties, which have positive human health benefits including suppression of tumor cell growth. The rice landraces having high percentage of similarity, as worked out in terms of the association co-efficient so called correlation co-efficient, were grouped together while constructing the dendrogram (Figure 2). Correlation co-efficient had values lying between 0.0 (no matches) to 1.0 (100% matches). Highest percentage of similarity worked out in terms of correlation co-efficient was recorded as 0.94 between Banni and Lalpanati and lowest in between Katharibhog and Tulaipanji (0.78) (Table 2). Dendrogram has shown three distinct clusters comprising the 14 rice varieties of North Bengal. First cluster consisting of five varieties namely Gobindabhog, Sadanunia, Kalonunia, Katharibhog and Tulaipanji. All the varieties are with unique aroma of their own and with yellow seed colour except Kalonunia. Second cluster comprises of Dudkalam, Dhepi, Lalpanati and Banni. Third cluster

Table 2: Similarity matrix was calculated based on correlation-coefficient among the varieties using morphological traits

	Enda	Banni	Malsira	Dhepi	Kalo nunia	Tulai panji	Sada nunia	Kathari bhog	Gobinda bhog	Dud kalam	Bhadoi	Ashami	Lal panati	Chenga
Enda	—													
Banni	0.765	—												
Malsira	0.941	0.706	—											
Dhepi	0.765	0.882	0.706	—										
Kalonunia	0.882	0.765	0.824	0.765	—									
Tulaipanji	0.824	0.824	0.765	0.824	0.941	—								
Sadanunia	0.706	0.824	0.647	0.824	0.824	0.882	—							
Katharibhog	0.824	0.706	0.765	0.706	0.941	0.882	0.765	—						
Gobindabhog	0.706	0.824	0.647	0.824	0.824	0.882	0.94	0.765	—					
Dudkalam	0.882	0.882	0.824	0.882	0.882	0.941	0.824	0.824	0.824	—				
Bhadoi	0.882	0.765	0.824	0.765	0.882	0.824	0.824	0.824	0.824	0.765	—			
Ashami	0.941	0.706	0.882	0.824	0.824	0.765	0.647	0.765	0.647	0.824	0.824	—		
Lalpanati	0.824	0.824	0.765	0.941	0.824	0.765	0.765	0.765	0.765	0.824	0.824	0.882	—	
Chenga	0.94	0.765	0.941	0.765	0.882	0.824	0.706	0.824	0.706	0.882	0.882	0.941	0.824	—
	Enda	Banni	Malsira	Dhepi	Kalo nunia	Tulai panji	Sada nunia	Kathari bhog	Gobinda bhog	Dud kalam	Bhados	Ashami	Lal panati	Chenga

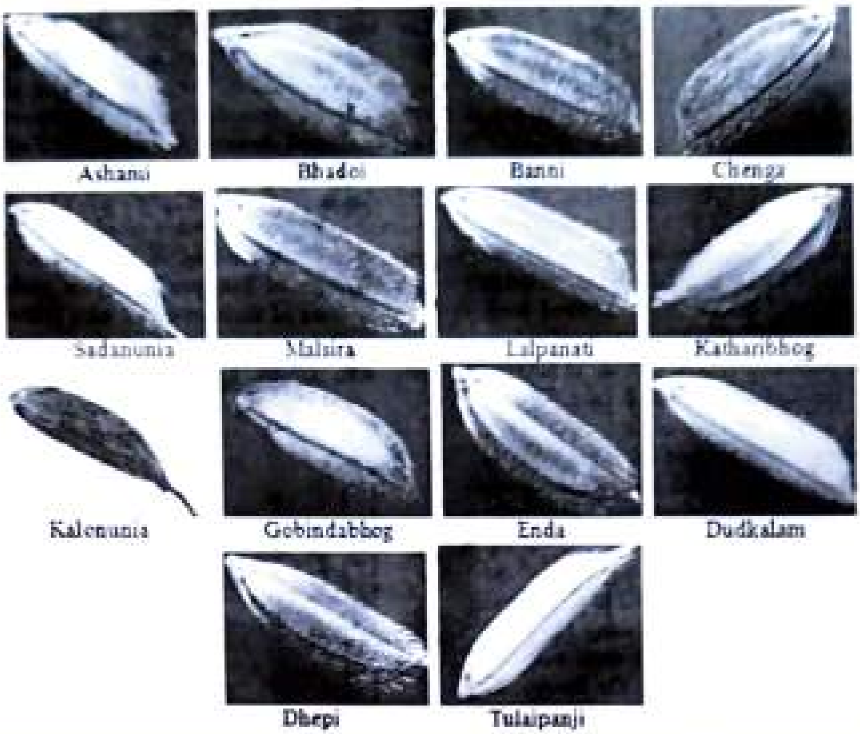

Fig1: Stereomicroscopic images of fourteen rice seeds (Landraces of North Bengal) showing different shapes, sizes and colour (Photographs were taken using Olympus Stereomicroscope E330-ADU1.2x/7c24087). Variety name was mentioned below.

consisting of the following varieties Chenga, Ashami, Bhadoi, Malsira and Enda. There are certain genetic variations existing among the 14 rice varieties under study and can be used in breeding program for crop improvement in future. Remote areas of North Bengal are yet to be assessed for their landraces. A complete picture on rice diversity will only be available when the unexplored areas of North Bengal and North-eastern regions of our country are fully explored. To safeguard the gene pool, we should let the farmers be allowed to conserve and multiply the traditional rice germplasm and hence, the introduction of high yielding varieties may be discouraged specially in the rural areas of the country where large number of rice diversities are available (Hore, 2005). All these rice landraces that are adapted under such acclimatized eco-system must be protected in *in situ* or on-farm conditions, so that their evolutionary process may be continued naturally (Rana *et al.*, 2009). It is predicted that the world population

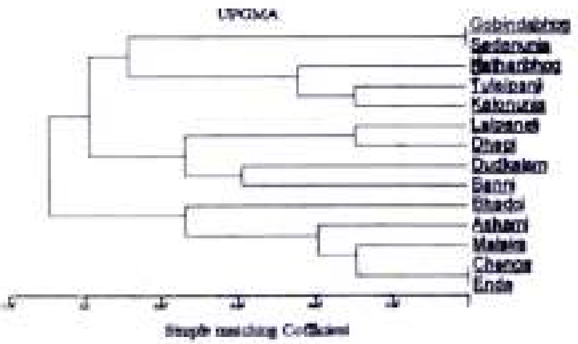

Fig 2: Dendrogram showing the three distinct clusters of 14 rice varieties (landraces) based on the morphological traits

will need an additional 50 million tones of rice annually by 2015. Under these circumstances, rice gene-pool stored in landraces and other relatives is going to play a crucial role to ensure future food security (Kumar et al., 2010). It is therefore nessecary to manage the continuing genetic erosion and address the issues of germplasm conservation of rice landraces and optimum utilization of what remains on-farm and conserved in the gene banks.

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