K- MEAN DETERMINATION OF GENETIC DIVERGENCE FOR SOME MORPHOLOGICAL AND SUBMERGENCE RELATED TRAITS IN RICE LANDRACES

[1]RAJESH KUMAR, [2]NILMANI PRAKASH*, [1]U. K. SINGH AND [1]SANTOSH KUMAR

[1]Department of Plant Breeding and Genetics

[2]Department of Agricultural Biotechnology and Molecular Biology

Rajendra Agricultural University, Pusa – 848 125, Samastipur, Bihar, India.

e-mail : niluy71@gmail.com

INTRODUCTION

India has the world's largest acreage (43.45 Mha) under rice cultivation but in terms of average productivity (2.38 tons/ha; Proceedings of 51st Annual Rice Group Meeting, 2-5 April, 2016) it ranks way behind the global leaders. One of main reasons for this is that a large part of the Indian rice cultivation is rain fed where it is subjected to the vagaries of the monsoon leading to too little or too much water for optimal rice cultivation. Rural poverty and food insecurity are especially persistent in rain fed and flood-prone rice production areas in India (Ismail *et al.*, 2013). Despite this enormous loss no consorted effort has been devoted to developing rice cultivars with in-built genetic tolerance to flood. One reason for this has been the genetically complex nature of the abiotic stress tolerance.

Submergence is a constraint to rice production in the years and areas of high rainfall. Rice is the dominant and the only suitable crop in the rain fed lowland areas of India, where flashfloods are a frequent constraint to increasing rice yields. At present, the area subject to flash flooding is thought to be more than 5 million ha. In 2007 and 2008, heavy flooding caused considerable crop losses, especially in the Eastern states of Indian. The *Sub1* gene is highly suitable for the areas subject to flash flooding of up to two weeks (Xu *et al.*, 2006, Neeraja *et al.*, 2007, and Septiningsih *et al.*, 2009). Results from farmers' field trials in 2007 indicated that variety Swarna with the *Sub1* gene for submergence tolerance can give more than 1.0 ton/ ha yield advantage in submergence-prone areas as compared to the control Swarna (Das *et al.*, 2009, Sarkar *et al.*, 2009; Singh *et al.*, 2009; Mackill *et al.*, 2012). Under severe flooding of more than 10 days, control variety Swarna without the *Sub1* gene was totally wiped out, whereas Swarna-*Sub1* provided yields of up to 2 t/ha or more.

Extensive studies indicated that there is no yield or quality penalty with the *Sub1* gene, so it should be widely deployed in rainfed lowland areas. In addition to the *Sub1* gene, there are other minor QTLs that contribute to submergence tolerance. For example, some farmers' varieties like FR13A and Khoda are more tolerant than Swarna-*Sub1*, and they also have more rapid recovery after submergence. Even the breeding line IR49830-7, which inherits the gene *Sub1* from FR13A, has tolerance to a longer duration of submergence than Swarna-Sub1 and other *Sub1* varieties. In addition, some of the improved breeding lines like IR70213-10, can tolerate moderate water stagnation in addition to submergence. While the Sub-1 gene in already available in several popular varieties for submergence tolerance but it provides the submergence tolerance for only two weeks while in most parts of the eastern states land remains submerged for more than two weeks there for it is now urgent need of hour to search for alternative and diverse source of tolerance

ABSTRACT

Genetic diversity was assessed in 184 land races including four checks using principle component analysis to assess the extent and pattern of genetic divergence based on k-mean determination. Principle component analysis (PCA) showed the first four PCs had Eigen value >1.00 and accounted 73.98% of total variation. Rotated component matrix revealed that each principle component is separately loaded with various submergence tolerant related traits. PC_1 was constituted by tiller per plant (0.47), panicle length (0.43), fertile tiller per plant (0.40) and survival % (0.37) while PC_2 was mainly composed of tolerance score (0.53) therefore intensive selection procedures can be designed to bring about rapid improvement of submergence tolerance by selecting the lines from PC_1, PC_2 and PC_3. All 184 land races were grouped into 14 clusters. Cluster II and IV showed high mean for most of the characters indicating land races from these groups can be used for further improvement of traits. 3 D plot based on three top principal components indicated that Pecalo, Houra Kani and S-177 were most divergent land races from Karabonka, Tiebimah and Urarkaruppan which might be utilized effectively in breeding programme for submergence tolerance.

to submergence which can act additively with Sub-1 gene in providing submergence tolerance for prolonged periods.

Keeping the above facts under consideration the present investigation was conducted to evaluate the local land races of rice for assessment of genetic divergence principal component analysis.

MATERIALS AND METHODS

The present investigation was conducted with 184 land races including four checks of rice collected from different countries to access the divergence using 10 quantitative and submergence tolerance related traits. The experiment was conducted at Crop Research Farm of RAU, Pusa, which is situated in district Samastipur of North Bihar, India. Field experiment was performed in Augmented Design (AD-II) during two years *i.e. Kharif* 2013-14. Each genotype was sown in four rows in plot of 5 m length with 20×15 cm plant geometry in water tank for submergence screening. Recommended fertilizer was applied at the time of crop period. Five random plants were tagged from each plot to record the data for yield and its related traits except days to 50 per cent flowering and survival percentage. Days to 50 per cent flowering was recorded on plot basis. Submergence tolerance related traits were performed as per Standard Evaluation System of Rice, IRRI, Nov. 2002 and Toojinda *et al.,* 2003 as listed below:

$$\text{Percent plant survival(PPS)} = \frac{\text{Total number of surviving seedling}}{\text{Total number of seedling counted before submergence}} \times 100$$

Total shoot elongation (TSE) = Height of shoot after de-submergence – Height of shoot before de-submergence.

$$\text{Relative shoot longation (RSE)} = \frac{\text{Elongation growth under submergence}}{\text{Elongation growth - Submergenced under non condition}} \times 100$$

Tolerance score (TS) = Scored on the basis of percent plant survival (PPS).

These data were subjected to pooled analysis for genetic divergence by using statistical package WINDOSTAT version 9.2 (INDOSTAT Service, Hyderabad).

RESULTS AND DISCUSSION

In order to maintain, evaluate and utilize the germplasm effectively, it is important to investigate the available genetic diversity (Mohammadi 2003 and Singh *et al.,* 2015). For establishing genetic relationship among the traits and their

genetic discrimination PCA was performed. Association between traits emphasized by this method may correspond to genetic linkage between loci controlling the traits or a pleotropic effect (Iezzoni and Pritts, 1991).The genetic variation present in breeding population was divided into five principle components (PCs), which explained 83.64 per cent of total variation (Table 1). The first four principle components had > 1.00 Eigen value and accounted to 73.98 per cent of total variation. The first principle component (PC1) explained 27.06 per cent of the total variation. The second PC (PC2) explained 17.10 per cent variation individually and 44.17 per cent cumulative variation. The third PC (PC 3) explained about 15.75 per cent individually and 59.92 per cent cumulatively. The fourth PC (PC 4) explained 14.06 per cent variation individually and 73.98 per cent cumulatively. Rotated component matrix revealed that each principal component is separately loaded with various submergence tolerant related traits(**Table 2**).PC_1 was constituted by tiller per plant (0.47), panicle length (0.43), fertile tiller per plant (0.40) and survival % (0.37)while PC_2 was mainly composed of tolerance score (0.53) therefore intensive selection procedures can be designed to bring about rapid improvement of submergence tolerance by selecting the lines from PC_1, PC_2 and PC_3This showed that higher loading values are attributed to submergence tolerance related traits *i.e.* Survival per cent, Tolerance score, Grain yield per plot and panicle length, indicating importance of these traits in getting better recombinants for submergence tolerance.

Cluster analysis helps to select the suitable genotype(s) for hybridization to manipulate the important traits. Choice of proper parent(s) plays a vital role for a successful plant breeding programme. Parents with more genetic distance believed to create higher variations by generating maximum recombination frequency, which increase the genetic gain in selection. The grouping of the land races was done by K-mean clustering pattern. The distribution of 184 rice land races into14 clusters and their cluster means are presented in **Table 3 and 4**, respectively. 184 land races of rice were grouped into fourteen clusters. Cluster XII comprised of only two land races (*i.e.* Tundahiya & Suraha) forming smallest and di-genotypic cluster, whereas cluster III, X comprised of twenty one, cluster IX, XI of four and cluster VI, XIII of fifteen land races each. Cluster II included maximum number of land races i.e twenty three. The four tolerant lines which had been taken for checks are SWARNA SUB-1, BPT 5204 SUB-1, FR13A and FR13B. The first two checks *viz.,* are SWARNA SUB-1 and BPT 5204 SUB-1 were clustered in cluster X and later two *viz.,* FR13A and FR13B were clustered in cluster II. The clustering of checks into particular clusters shows that the present submergence tolerant varieties are from same source while

Table 1: Eigen values and variability explained by each principle components (PCs)

	PC1	PC2	PC3	PC4	PC5
Eigen Value (Root)	2.70	1.71	1.57	1.40	0.97
% Var. Exp.	27.06	17.10	15.75	14.06	9.67
Cum. Var. Exp.	27.06	44.17	59.92	73.98	83.65

Table 2: Correlation coefficient of each submergence related trait with respect to its principle components (PC)

Traits	PC1	PC2	PC3	PC4	PC5
Days to 50 per cent flowering	0.21	0.33	0.49	0.23	0.10
Plant height (cm)	0.22	0.37	0.50	0.07	0.08
Panicle length (cm)	0.46	0.34	-0.09	0.03	0.03
Tillers per plant	0.47	0.19	-0.37	0.04	-0.11
Fertile tillers per plant	0.40	0.07	-0.43	-0.08	-0.17
Grain yield per plot (g)	-0.01	-0.01	0.25	-0.06	-0.96
Survival (%)	0.37	-0.55	0.20	0.09	0.04
Total shoot elongation (cm)	-0.17	0.08	-0.13	0.65	-0.13
Relative shoot elongation (cm)	-0.05	-0.09	-0.15	0.70	-0.02
Tolerance score	-0.39	0.53	-0.20	-0.10	-0.05

Table 3: Distribution of rice land races in various clusters

| K-Group	No. of land races	land races
I	19	KARKATI 87, KOTTAMALI, ROSA MARCHETTI, ARC 12172, MENYARHUNEI, MUYAMBA, CR 1009, ZOBGUI, LIU-TIAO-NU, MOUSSAYA, SIPULUT PANDAN, CHOW HA LA, BAGA GOHA, BARA RANGA, JAGANATHA BALLAVA, BABAI LACHHA, LUNISHREE, KOLA JOHA
II	23	AI CHIH AI K WHO 2, CODY, MAINTIMOLOTSY, LUMBINI, MOROBEREKAN, JP5, NA ALUMOLI KARUPPAN, MAPOI, GODA HENNATI, KALUWEE, NCHEL PARAGAHAKALE, NALDAK, BR 11, KHAIYAN, SOSSOKA, KAOLACK, PASAKACHUM, ROM RUNTIK, SANTI BHAN, NIPPONBARE, BAGI BARDHANA, FR-13A, FR-13B
III	21	ANH HSUNG SELN (CI 1), VENA VELLAI, KALA-RATA 1-24, JALMAGNA, PATAIM, MADAL, USA BATAPOLA AL, NIRBOI, TAH TONE, CHANGMAN, BARACHIRAMARA, HULTA BALUNGA(WILD), CHING CHANG, ASHRA LAKI, CHANGNGAT, PANIKAKUA, KHAWJII, JALKANTHI, CHUDI, BAD GEDE, SUGAPANKH
IV	13	URARKARUPPAN, TIEBIMAH, HEEN SULAI, MAHA DIKWEE, PAKRIBUNA, VADAI, CHAKIA 59, FRG 1, VAN, KARABONKA, SANG KHLA, CHUMANSAL-39,MEGHARAJ
V	16	DHOLAMON 64-3, ROSA MARCHETTI, CHANGMAKA, CHANGPHAI, S-183, JALNIDHI, KAJARAHAWA, SATHI, S-150, S-172, S-148, S-155, AUSBORO,S-138, IARI 5825, NCS 348
VI	15	TAGBA, NANG QUOT, JALDUNGI, GOTA, BUNTOK, JOHAINEANG (WET), JAHABHOG, PANIDUBI, PANIKEKEUA, GOTHAWA, SINGHARA, KARIYWA, JADHAN, DUDHA LADU, ARC 614
VII	8	PAIAM, SOSSON HONDON, IR 42, IR-73707-45-3-23, DUB GELONG, SAMBHA Sub-1, MOTI, POKKALI
VIII	9	JAYMIS, BARA SALI, ASAMIRUPA, BAGA IKRA, SAVITRI, DIHAWAN, TETEP,AZUCENA, LEMONT
IX	4	PECALO, HOURA KANI, IR-36, S-177
X	21	HSUNG TIENG, KURUWEE, BIRPALA, AUS 287, TAIPEI 309, TREMBESE, KARNAL LOCAL, SWARNA, KHARSU 80A: Oryza sp, KASALATH, HUKKUNNA KUMBALOA, CHASING, MEGHI, CHOWMAI, BAD KODI, CHANG MUI, DHUSARI AHU, CHANGPALJOM PHAI, KARAHANI, SWARNA SUB-1, BPT 5204 SUB-1
XI	4	KOSHIHIKARI, M202, SARJOO 52, JC-1
XII	2	TUNDAHIYA, SURAHA
XIII	15	LABELLE, BINULAWAN, WAHNG, KHAO NINE LAR, IR 72, JOKHRU, CHARETSUK, BAD DHANA, ASHU BHAJNA, PANKEI, BAD JHARLI, RAJBHOG, S-141, KALA-NAMAK,SATHWA
XIV	14	DULAR, KALONCHI, MARS, RETEK SIRENDAH MERA, SITA MATA, ZECHUM,LATSIKA, NANHI, IR 64,MANOHAR SALI, PADMA,LALKA DHAN, KALA BUNDE, MEGHABADALA

diverse land races other than checks have the capacity to tolerate the submergence condition and can be used as recurrent parent. The K- mean of different cluster (Table 4) indicated that land races of cluster XI possess less TPP, FTPP, SP and TS but high DFF, PH, PL, GYP, TSE, TS and moderate RSE. The land races of cluster IX possess less PL, TPP, FTPP, TSE, TS but high DFF, PH, GYP and RSE and moderate SP as

compare to Tundahiya & Suraha indicating that representative land races can be chosen from particular diverse groups based on their cluster mean and can be involved in hybridization programme for rice improvement against submergence tolerance related traits. Based on cluster mean, it was noted that the cluster II and IV has higher mean value for maximum number of traits indicated the possibility if obtaining the

Table 4: Mean characteristics (K- Mean) on various submergence tolerance related traits for each clusters in rice

Cluster	Traits/ Morphological Characters									
	DFF	PH	PL	TPP	FTPP	GYP	SP	TSE	RSE	TS
I	130.82	147.90	25.53	8.71	7.43	235.20	91.15	19.32	203.74	1.15
II	132.53	155.40	26.99	10.98	11.28	202.36	94.06	13.56	129.02	1.00
III	135.23	167.27	26.02	7.90	6.31	259.77	83.25	17.12	131.94	1.29
IV	136.03	187.47	27.46	9.37	9.97	273.38	79.08	11.25	90.65	1.73
V	100.36	107.86	17.52	5.15	6.19	233.14	69.66	27.79	372.34	2.72
VI	136.96	167.62	23.99	6.28	6.50	272.27	60.90	22.15	130.69	3.53
VII	116.50	114.98	21.90	7.52	7.45	197.05	87.01	35.88	794.64	1.56
VIII	112.65	130.82	20.06	7.08	6.71	245.56	67.54	26.39	294.65	2.95
IX	117.96	114.60	22.57	6.02	6.93	289.55	87.60	25.34	1461.30	1.00
X	107.79	130.16	25.23	10.56	8.91	265.23	92.59	22.02	210.51	1.05
XI	102.83	108.17	23.07	4.24	6.89	221.05	24.76	22.75	299.60	7.25
XII	127.77	217.17	24.48	9.02	0.71	219.96	40.08	15.31	82.46	6.00
XIII	125.88	148.73	24.39	6.87	5.71	196.74	63.26	26.74	206.11	3.50
XIV	100.30	116.16	22.87	8.41	9.21	243.21	92.00	15.58	296.27	1.14

DFF = Days to fifty percent flowering, PH = Plant height, PL= Panicle length, TPP = Tillers per plant, FTPP = Fertile Tillers per plant, GYP = Grain Yield per plot (g), SP = Survival Percentage, TSE = Total Shoot Elongation, RSE = Relative Shoot Elongation, TS = Tolerance Score.
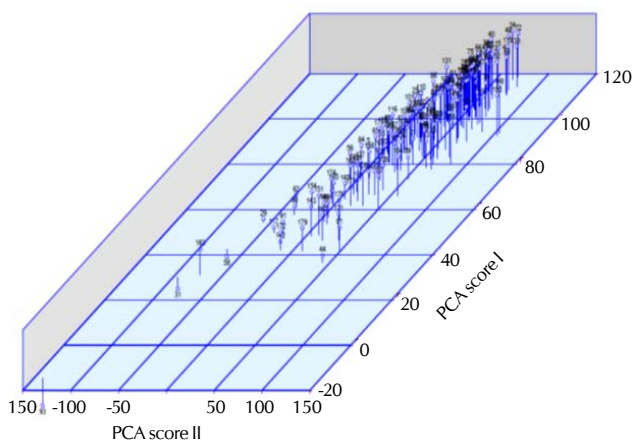
Figure 1: Distribution of 184 rice land races between first, second and third principle components (3-D Plot diagram)

transgressive segregants in early segregating and subsequent generations. Several researchers reported the significance of high survival percentage to improve the submergence duration in lowland rice (Mackill *et al.*2012; Neeraja *et al.,* 2007). 3D Plot diagram constructed on the first three principal component (**Fig: 1**) plotted Pecalo, HouraKani and S-177 at distant end whereas Karabonka, Tiebimah and Urarkaruppan were plotted at another 3D plane. These landraces can be used effectively in breeding programme for submergence tolerance trait improvement as diverse parents.Based on 3D diagram the genotype Pecalo, Houra Kani, IR-36, S-177, Koshihikari, M202, Sarjoo-52 and JC-1 also exhibited good diversity with Tundahiya & Suraha and found suitable for improvement of submergence related traits.Based on above discussion, PCA revealed the possibility for improvement of rice land races against submergence tolerance by manipulation of various traits *viz.,* TPP, FTPP, SP, TS and PH. Pecalo, Houra Kani and S-177 were found most divergent land races with Karabonka, Tiebimah and Urarkaruppan which might be utilized effectively in breeding programme for submergence tolerance. Suitable land races chosen from cluster II and cluster IV; the two diverse groups based on their cluster mean may also be involved in hybridization programs for rice improvement against submergence tolerance.

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