Original Research Article

Distinctiveness, Uniformity and Stability (DUS) characterization in Twenty-five landraces of Rice (*Oryza sativa* L.)

Abstract

For the establishment of Distinctiveness, Uniformity and Stability (DUS) characterization, Twenty five landraces of rice (*Oryza sativa* L.), collected from different parts of Tamil Nadu, were characterizsed according to the DUS guidelines of the Plant Varieties Protection and Farmers' Rights Authority, using thirty agro-morphological traits. Rice accessions were planted with three replications using the randomized block design (RBD) in 2021 at the Plant Breeding Farm, Department of Genetics and plant Breeding, Faculty of Agriculture, Annamalai University. Observations were recorded on various agro-morphological traits such as Leaf sheath, flag leaf, leaf blade, awns, panicle, ligure and clum characteristics. The results obtained this study exhibitedrevealed a wide range of variability for 25 essential characters of the 30 DUS characters studied . This The result of this study will be useful for restoration and conservation of beneficial genes. Contemporary research work will be advantageous for breeders to choose the right parent for crop improvement and the researchers for genetic diversity among landraces.

Comment [MTK1]: The results paragraph in the abstract should be measurable i.e. using numbers or % of some indicators so as to indicate the level of uniformity or distinctiveness or stability among the 25 land races. You can mention some of the results in table 2 e.g. only 3% of the sample shows very low panicle shattering etc,

Introduction

One of the most significant cereal crops worldwide is rRice (*Oryza sativa* L.) which act as a primary food source for more than half of the world's population is rice. (Sasaki and Burr, 2000). rice Rice is the second most important cereal crop after the maize in terms of world production. India has a rich and wide range of genetic wealth of rice. According to estimates from various surveys, the nation is still home to roughly 50,000 rice farm ers. (Roy JK *et al*., 1985). In India rice is cultivated in an area of about 431.94 lakh hectares with a production of about 110.15 million tonnes and productivity of 2550 kg per ha as of 2016-2017. Rice has the largest germplasm collections in the world, . landraces Landraces are traditionally

cultivated, evolved over generations with proven special features over wild relatives ~~and~~ hence serve as a treasure of useful genes. ~~and they~~ They played a very important role in the local food security and sustainable development of agriculture, in addition to their significance as genetic resource for rice genetic improvement (Tang *et al*., 2002). Characterization of germplasm eventually means recording and storing useful data that can be readily retrieved and made available to others and comfort in planning breeding programmes (Dabas *et al.*, 1994). Twenty-five landraces collected from different locations were used in the present investigation. ~~The~~ Rice descriptors of Biodiversity International ~~was~~ were used for evaluation and characterization. The genetic diversity existing among landraces will provide information on the superior parental selection for future breeding programs. Different studies suggest that India has over 70,000 germplasm accessions and also needs the selection and protection of a large number of wild species (Siddiq, 1992). The Government of India has introduced Protection of Plant Varieties and Farmers' Rights (PPV&FR) Act in 2001, for the IP protection over crop varieties including farmers' rights over traditional varieties. The concept of DUS is fundamental to the characterization of a variety as unique and provides an official description of a variety for its identity as it is globally accepted for varietal identification. In this research work, an attempt was made to characterize a set of twenty five landraces of rice germplasm for different morphological and agronomic traits and identify the variability available in the collection.

Materials and methods

Experimental area

Twenty-five landraces of rice were grown in a Randomized block design (RBD) with three replications in 2021 at the Plant Breeding farm, Department of Genetics and plant Breeding, Faculty of Agriculture, Annamalai, University. The experimental site has the co-ordinates of 11°24′ N latitude and 79°41′ E longitude at an altitude of 5.1 m above the main sea level.

Experiment materials

The present research experimental material ~~consists~~ consisted of 25 landraces of rice genotypes (*Oryza sativa* L.). Relevant common name was gathered from local farmers~~.~~ (Table 1).

Comment [MTK2]: Mention the source (s) of the 25 land races.
Like the research stations. This is unavoidable in this study because the genetic purity of the seeds for each landrace is paramount in agro-morphological trait characterization assessment.

Field plot setup

Accessions were grown in a randomized block design ~~with the~~in three replications. Each entry was sown in three rows of 25 plants, at a spacing of 25 cm between plants and 30 cm between rows and 1m~~t~~ isolation separation was maintained between two entries in each block. Crop was raised by following recommended package of practices (Manjunatha *et al*., 2018).

Recorded Observation

The observations were taken for morphological and agronomic characters. In these observations, Morphological characterization of twenty-five landraces of rice was carried out using twelve DUS characters. All the characters were studied as per procedure of biodiversity at field condition. The qualitative characters were scored visually, and the quantitative characters were measured. Visual observations were recorded based on single plant basis on ten randomly selected plants in each genotype at appropriate growth stages (Umarani *et al*., 2017).

Table 1. 25 landraces of rice used for DUS Test

Accession	Rice Landraces	Location of collection
G1	Attur Samba	Attur, Salem
G2	Arupatham Kuruvai	Maangulam, Madurai
G3	Arcode Kichili Samba	Ediyure, Kancheepuram
G4	Bhavani	Rayanathur, Thanjavur
G5	Chandikar	Vayalogam, Pudhukottai
G6	Kuruvai Kalanjium	Thiruppurambiam, Kumbakonam
G7	Kaatu Yaanam	Palamuthur, Thanjavur
G8	Kallundai	Vilangudi, Thanjavur
G9	Kichili Samba	Ediyure, Kancheepuram
G10	Karudan Samba	Ananthapuram, Tiruvannamalai
G11	Karunkuruvai	Ananthapuram, Tiruvannamalai
G12	Kallurandaikar	Parmakudi, Ramanathapuram
G13	Kandasaali	Semballur, Thanjavur
G14	Kothamalli Samba	Karungalammelpathi Thanjavur
G15	Kuzhiyadicham	Thennampalagai Thanjavur

G16	Mysore Malli	Puliyancholai, Trichy
G17	Navara	Mailaduthurai, Cuddlore
G18	Norungan	Coastal area, Nagapatiinam
G19	Sivapu Chittirai Kar	Anumantankadi, Sivagangai
G20	Salam Sannam	Mangalam, Kancheepuram
G21	Sigapu Kavuni	Krishnapuram, Thanjavur
G22	Sornamugi	Rayanathur, Thanjavur
G23	Seeraga samba	Puliyancholai, Trichy
G24	Thulasi Vasanai Seeraga samba	Kondakulan, Thanjavur
G25	Thoyamalli	Keelapalayar, Thanjavur

Results and Discussions

Thirty traits were observed in twenty-five germplasm to establish distinctiveness among germplasm and these are presented below.

Table 2. DUS Characterization of 25 landraces of rice

Sl. No.	Characters	Types	Scale	No. of Germplasms	Frequency Distribution (%)
1.	Basal leaf sheath : colour	Green	1	18	71.50
		Green with Purple lines	2	3	12.51
		Light Purple	3	3	12.0
		Purple	4	1	3.94
2.	Leaf sheath : anthocyanin colouration	Absent	0	20	79.9
		Weak	3	3	12.1
		Medium	5	2	7.80
		Strong	7	0	0.00
3.	Leaf blade : anthocyanin colouration	Absent	0	9	37.3
		Present	1	11	62.4
4.	leaf blade : distribution of anthocyanin	Tips only	1	23	91.4
		On margins	2	0	0.00
		In Blotches	3	0	0.00
		Even	4	2	8.32
5.	Leaf blade :	No green	0	0	0.00

Comment [MTK3]: This table could make more meaning if a column showing the landraces with the various characters was mentioned. It would have been easy to have an appreciation of uniformity and distinctiveness of specific landraces. As presented, readers of this article cannot identify the landraces that is completely distinct, highly uniform or stable for the 30 traits without coming back to the research team for more details

	intensity of colour	Green	3	13	53.6
		Medium	5	8	31.15
		Dark	7	4	15.23
6.	Leaf blade : attitude	Erect	1	19	75.50
		Horizontal	5	4	16.00
		Drooping	7	2	7.90
7.	Leaf blade : pubescence	Glabrous	1	3	13.1
		Intermediate	2	20	79.8
		Pubescent	3	2	8.10
8.	Auricle collar : colour	No green	0	25	100
		Green	1	0	0.00
		Light green	2	0	0.00
		Purple	3	0	0.00
		Purple lines	4	0	0.00
9.	Ligule shape	Absent	0	0	0.00
		Truncate	1	8	31.61
		Acute	2	16	63.90
		2-cleft	3	1	4.30
10.	Ligule Pubescence	Glabrous	1	15	61.07
		Partially < 50%	2	7	28.14
		Mostly >50%	3	3	10.79
11.	Ligule colour	Absent	0	0	0.00
		Whitish	1	16	63.58
		Yellowish green	2	5	21.10
		Purple	3	0	0.00
		Light Purple	4	4	15.31
		Purple lines	5	0	0.00
12.	Ligule margin hairiness	Absent	0	19	76.00
		Present	1	6	24.00
13.	Leaf blade : Length(cm)	Very short (<21cm)	1	2	7.70
		Short (22-30cm)	3	9	36.01
		Intermediate (31-50cm)	5	4	16.00
		Long (51-70)	7	6	23.98
		Very long (> 80cm)	9	4	16.28
14.	Leaf blade : width(cm)	Narrow (<1cm)	3	5	21.09
		Intermediate	5	16	63.91
		Broad (>2cm)	7	4	14.94

15.	Flag leaf : length(cm)	Short (<30cm)	3	6	23.90
		Medium (30-45cm)	5	11	44.00
		Long (>45cm)	7	8	32.07
16.	Flag leaf : width(cm)	Narrow (<1cm)	3	13	52,47
		Intermediate	5	9	35.34
		Broad (>2cm)	7	3	12.18
17.	Flag leaf : attitude(Early)	Early	1	12	48.23
		Semi- erect	3	8	30.25
		Horizontal	5	5	21.50
		Descending	7	0	0.00
18.	Culm : Habit	Erect (<15°)	1	6	23.25
		Semi-erect (-20°)	3	12	48.02
		Open (-40°)	5	3	13.41
		Spreading (>60-80°)	7	4	14.99
		Procumbent	9	0	0.00
19.	Culm : length	Very short (>50 cm)	1	2	7.20
		Very short to short(51-70 cm)	2	3	12.90
		Short(71-90cm)	3	7	28.14
		Short to intermediate(91-105cm)	4	2	7.12
		Intermediate(106-120cm)	5	9	36.07
		Intermediate to long(121-140cm)	6	1	4.31
		Long (141-155cm)	7	1	4.20
		Long to very long (156-180cm)	8	0	0.00
		Very long(>180cm)	9	0	0.00
20.	Culm : anthocyanin colour on node	Absent	0	16	64.32
		Purple	1	4	14.79
		Light purple	2	3	12.87
		Purple Lines	3	2	8.00
21.	Culm : internode anthocyanin	Absent	0	20	80.00
		Purple	1	2	7.90
		Purple Lines	2	3	12.10
22.	Culm : lodging resistance	Very weak	1	0	0.00
		Weak	3	3	11.63
		Intermediate -45°	5	15	62.07
		Strong 20°	7	7	26.14
		Very Strong	9	0	0.00
23.	Flag leaf :	Erect	1	12	48.00

	attitude(Late)	Semi-erect	3	7	28.12
		Horizontal	5	6	23.85
		Descending	7	0	0.00
24.	Lemma and palea : colour	White	1	1	3.98
		Green stripped white	2	3	12.13
		Gold and gold furrows	3	1	4.01
		Brown	4	2	8.91
		Brown spots and green	5	0	0.00
		Brown furrows on green	6	6	25.91
		Blackish brown	7	2	8.10
		Green	8	9	32.92
		Yellowish green	9	0	0.00
		Purple	10	0	0.00
		Reddish to light purple	11	0	0.00
		Purple shade	12	0	0.00
		Purple spots on green	13	0	0.00
		Purple furrows on green	14	1	4.03
		Black	15	0	0.00
25.	Awn presence	Absent	0	15	60.00
		Partly awned	1	10	40.00
		Fully awned	2	0	0.00
26.	Awn distribution	Awnless	0	15	60.00
		Tips only	1	10	40.00
		Upper quarter only	2	0	0.00
		Upper half only	3	0	0.00
		Upper three-quarters only Whole length	4	0	0.00
			5	0	0.00
27.	Awns : colour	Absent	0	15	60.00
		Whitish	1	2	7.89
		Straw	2	0	0.00
		Gold	3	1	4.01
		Brown	4	2	8.06
		Light green	5	3	12.13
		Red	6	0	0.00
		Purple	7	1	4.00
		Black	8	1	3.91
28.	Texture of	Scabrous	1	25	100

Comment [MTK4]: No need for this because the trait is awn distribution

	panicle	Smooth	2	0	0.00
29.	Panicle : shattering	Very Low (<1%) Low (~3%) Moderate (~15%) High (~35%) Very high (>50%)	1 3 5 7 9	3 17 5 0 0	12.15 68.11 19.70 0.00 0.00
30.	Panicle : threshability	Difficult (few or no grains) Intermediate (25-50%) Easy (>50%)	1 2 3	14 6 5	55.52 24.45 20.02

A total of 25 rice land races was taken for DUS characterization using 30 characters which include both qualitative and quantitative characters. The rice landraces undertaken for this study showed wide range of distinctiveness characters for all most all the morphological traits studied and similar results has been reported earlier by Rao *et al*. (2013); Tirkey *et al*. (2013); Mondal *et al*. (2014); Manjunatha *et al*. (2016); Kalyan *et al*. (2017); and Umarani *et al*. (2017). Frequency ~~distribution for all the characters under study were~~distributions for all the characters under study were computed and qualitative and quantitative characters of different agronomic and morphological parameters are given in Table 2.

Comment [MTK5]: Grouping the characters under quantitative and qualitative will give the table better understanding

For the morphological characterization or identification of land races of rice, quantitative characters are considered as morphological markers, because they are less influenced by the environmental changes. Out of 25 land races studied, 71.50% possessed green coloured basal leaf sheath, 12.51% had green with purple lines, 12% with light purple and 3.94% had purple colour. Anthocyanin colouration in leaf blade was present in 62.4% land races and absent in 37.3% of land races. All the land races showed the distribution of anthocyanin at the tip of the leaf blade.

Anthocyanin coloration on leaf sheath was absent in 79.9% landraces and has 12.1% were weak and 7.80% were medium. For the intensity of green colour in leaf blade all the accessions exhibited light to dark green in colour, in which 31.15% exhibited medium, while 53.6% and 15.23% exhibited light and dark green colour respectively. 75.50% of landraces had erect leaf blade, while 16.00% and 7.90% had horizontal and drooping attitude respectively. For

pubescence on leaf blade surface 8.10% landraces were found to be distinct for having strong pubescence, while 79.8% with intermediate pubescence and 13.1% were marked for glabrous nature. For the leaf ligule character, 63.90% possessed acute shaped ligules, 4.30% 2 cleft and 31.61% recorded truncate ligules. Among the land races 21.10% of them had yellowish green ligule, 63.58% had whitish and 15.31% had light purple colour. Ligule margin is hairy in 24% of the landraces. The length of the leaf blade was short in 36.01% and also exhibited intermediate width in 63.91% of rice landraces.

With respect to flag leaf 44% of sample size had medium length, while 32.07% and 23.90% had long and short flag leaf length respectively. Intermediate flag leaf width was observed in 35.34% landraces, 52.47% showed narrow flag leaf width. Incase of flag leaf attitude 30.25% of the types were semi erect 48.23% and 21.50% were erect and horizontal respectively. Regarding culm characters, 48.02% landraces were of semi erect type, 13.41% were open type and 23.25% were erect type. For culm length (excluding panicle), 7.02% were of very short, 12.90% were of very short to short, 28.14% were of short type, 7.12% were of short to intermediate 36.07% were of intermediate and 4.31% were of intermediate to long type and 4.20% were of long type. With respect to anthocyanin colour on node, it was absent in 64.32%; while 14.79% possessed purple colour, 12.87% had light purple and 8.0% had purple lines. 26.14% of the culm are strong and 62.07% were intermediate in culm strength. Lemma and Palea colour was Green in 32.92% and brown furrows on green in 25.91% of landraces.

Awns were absent in 60%; partly awned in 40% and no fully awned. Awns were present in the tip only at 40% and the colour was whitish in 7.89%; purple in 4.00% and gold in 4.01% landraces. The awn colour, lemma and palea colour, flower colour, colour of hair, seed colour were the most stable characters across the agro-climatic zones (Gupta *et al*., 2010; Satyavathi *et al*., 2004). Panicle shattering was low in 68.11% and moderate in 19.70% of the landraces. The threshabile was easy in 20.02%, intermediate in 24.45% and difficult in 55.52%.

Conclusion

Morphological descriptors were widely used in sequential fashion and were found convenient in discriminating different genotypes. The 25 genotypes under study exhibited wide

Comment [MTK6]: This section should be revised taking into account the following reasons;
1.As presented, this section is simply repetition of results of table 2, which is not necessary.
2. I would have rather expected you to link your results to other works highlighting the agronomic importance of some of these agromorphological traits. i.e. explain how these traits you have characterized do influence rice yield in terms of quantity and quality, as well as their importance towards tolerance to biological and environmental stresses,
3.These is because crop breeders are attracted mainly to land races that distinct themselves in agro morphological traits that can solve the problem of quantity, quality and tolerance to edaphic and climate change constraint like tolerance to drought,

range of variability for 25 essential characters of the 30 DUS characters studied. This study will be useful for restoration and conservation of beneficial genes. ~~These~~ This information on characterization will be useful for breeders, researchers and farmers to identify and also to conserve beneficial genes for crop improvement. The information generated on these landraces may also support their registration with the Plant Protection Varieties and Farmers Rights Act.

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