



Phenotypic Characterization of Diverse Rice Fertility Restorers

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/IJPSS/2021/v33i1630539

Editor(s):

(1) Prof. Marco Trevisan, Catholic University of the Sacred Heart via Emilia, Italy.

Reviewers:

(1) Reena Rani, ICAR-CAZRI, India.

(2) Raghad S. Mouhammad, Iraq.

(3) Shreenivas A. Desai, U. A. S., India.

Complete Peer review History: <https://www.sdiarticle4.com/review-history/68603>

Received 01 May 2021

Accepted 03 July 2021

Published 16 July 2021

Original Research Article

ABSTRACT

Characterization of restorer lines of rice based on seedling and plant morphology contributes in assessment of varietal identity and further its utilization in development of hybrids. With this objective the work was carried over with 90 restorer lines with three replications during Kharif season 2018. The experiment conducted under Rice Improvement Project at Seed Breeding Farm, Jawaharlal Nehru Krishi Vishwa Vidyalaya, Jabalpur (M.P.). Out of 41 morphological traits as per DUS guidelines, variability was reported for all traits except leaf sheath anthocyanin colouration, presence of leaf auricle, presence of leaf collar, presence of leaf ligules, white colour of leaf ligules, absence of stem anthocyanin colouration of internodes, male sterility and presence of secondary branching in panicle. The genotypes Mahamaya, ANP-526 and ANP-553 revealed distinctness for most of the traits.

Keywords: *Characterization; DUS guidelines; rice improvement project; rice restorer lines.*

1. INTRODUCTION

Rice is the semi-aquatic, monocot normally grown as an annual plant that belongs to family *Poaceae*, sub-family *Bumbyoidae* and Genus *Oryzae*. Among 24 known species of rice, 22 are wild and only two are cultivated species i.e., *O. sativa* (Asiatic rice) and *O. glaberima* (African rice). The species *sativa* is subdivided based on ecotype as *indica* (India), *japonica* (Japan) and *javanica* (Indonesia) [1]. Rice is a staple food for almost half of the world population and it claimed more than one-fifth of its share in calorie consumption. Globally, rice fulfils about 60% of dietary, 20% of calorie and 14% of protein requirement, which highlighted its significance in the human diet [2,3]. According to world population review 2019, India holding the second rank occupied a global share of 17.7% in terms of population, with the increasing growth rate of 1.02% yearly [4]. This called breeder's attention to increase production and productivity of rice, to meet the growing demands of the population. Thus, the exploitation of diversity is needed for the development of hybrids, which shows higher productivity per day along with good quality traits. Restorer lines male fertile lines, used as a pollen parent in commercial seed production plots serve this purpose through commercial exploitation of heterosis. Three line systems is being practiced in rice hybrid breeding programs to obtain a breakthrough in yield potential.

With the view of protecting genotypes under PPV & FR (2001), they are examined for compliance with DUS criteria needed for registration. Characterization revealed common knowledge about the uniqueness of a variety from existing variety, following standards of DUS guidelines.

Restorer line helps us to revive the male sterility of a line and high genetic variability for the specific traits characteristic of restorer parents are the prerequisites for selection and development of superior hybrid parents. However, for sustained and continuous improvement, contemporary variability ought to be created by introgressing new sources of desired genes from landraces into elite scientific discipline background. The agro-morphological characterization should eventually lead to a system of recording and storing useful data that can be readily retrieved and made available to others and help in planning breeding programs [3]. It helps in tracing traits exhibiting correlation and linkage among themselves and yield. Hence,

provided elementary information regarding plant breeding program would help in the choice of parents for hybrid production in the future. Keeping aforesaid in view, the present investigation enumerated 90 fertility restorer lines of rice to explicate the existing variability through characterization.

2. MATERIALS AND METHODS

The current research was conducted with 90 fertility restorer lines of rice with the aim to assess phenotypic variation based on morphological characterization, which could be act as a marker for identification of elite genotype. The experiment was carried out under Rice Improvement Project, at Seed Breeding Farm, Jawaharlal Nehru Krishi Vishwa Vidyalaya, Jabalpur. The seeds were sown on a raised seedbed in order to ensure the uniform germination and establishment, and subsequently 30-day-old seedlings were transplanted into the experimental field. Each genotype was grown in a single row of 4 m length with a row to row spacing of 20 cm and plant to plant spacing of 15 cm. Recommended agronomic package of practices was followed for the proper field management during the experiment. The exclusive one among all would be further utilized in hybridization programme. All the observations were recorded as per DUS guidelines. Based on botany and assessment method several morphological, quantitative and quality traits were studied.

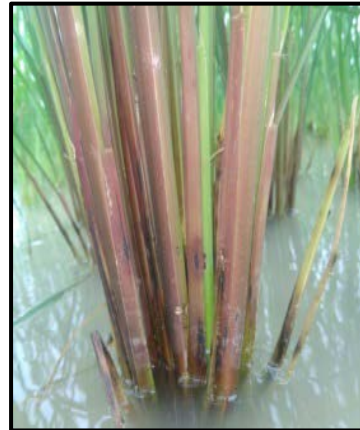
3. RESULT AND DISCUSSION

The ninety restorer lines were keenly observed because the visual assessment of variability in a population reported is necessary for a plant breeder to begin a judicious Breeding programme. The present investigation depicted no variability for eight morphological traits viz., leaf sheath anthocyanin colouration, auricle presence, leaf collar presence, leaf ligules presence, white colour of leaf ligules, absence of internodes anthocynin colouration, secondary branching presence and male sterility, as shown in Table 1 and Fig. 1. The corresponding findings were also noticed before for traits like presence of ligules, auricle and collar [5,6] and for trait panicle presence of secondary branching [7]. Therefore, the frequent occurrence of the specific trait put them in a reluctant category and could be avoided for the next selections of restorers.

Basal leaf sheath colour



Green



Purple (JR 1009)

Anthocyanin colouration of collar



Green



Purple (Mahamaya)

Anthocyanin colouration of auricles



Purple (Mahamaya)



Green

Spikelet colour of stigma



White



Purple (ANP 526)

Panicle exertion



Well Exerted



Mostly Exerted (NPT 40-018*)

Panicle curvature of main axis



Semi-straight

Deflexed

Drooping (ANP 553)

Secondary branching



Flag Leaf Attitude (early)



Sterile lemma colour
(ANP 526)

Purple colour tip of lemma
(ANP 526)

Anthocyanin keel colour
(ANP 526)



Fig 1. Morphological markers for varietal identification

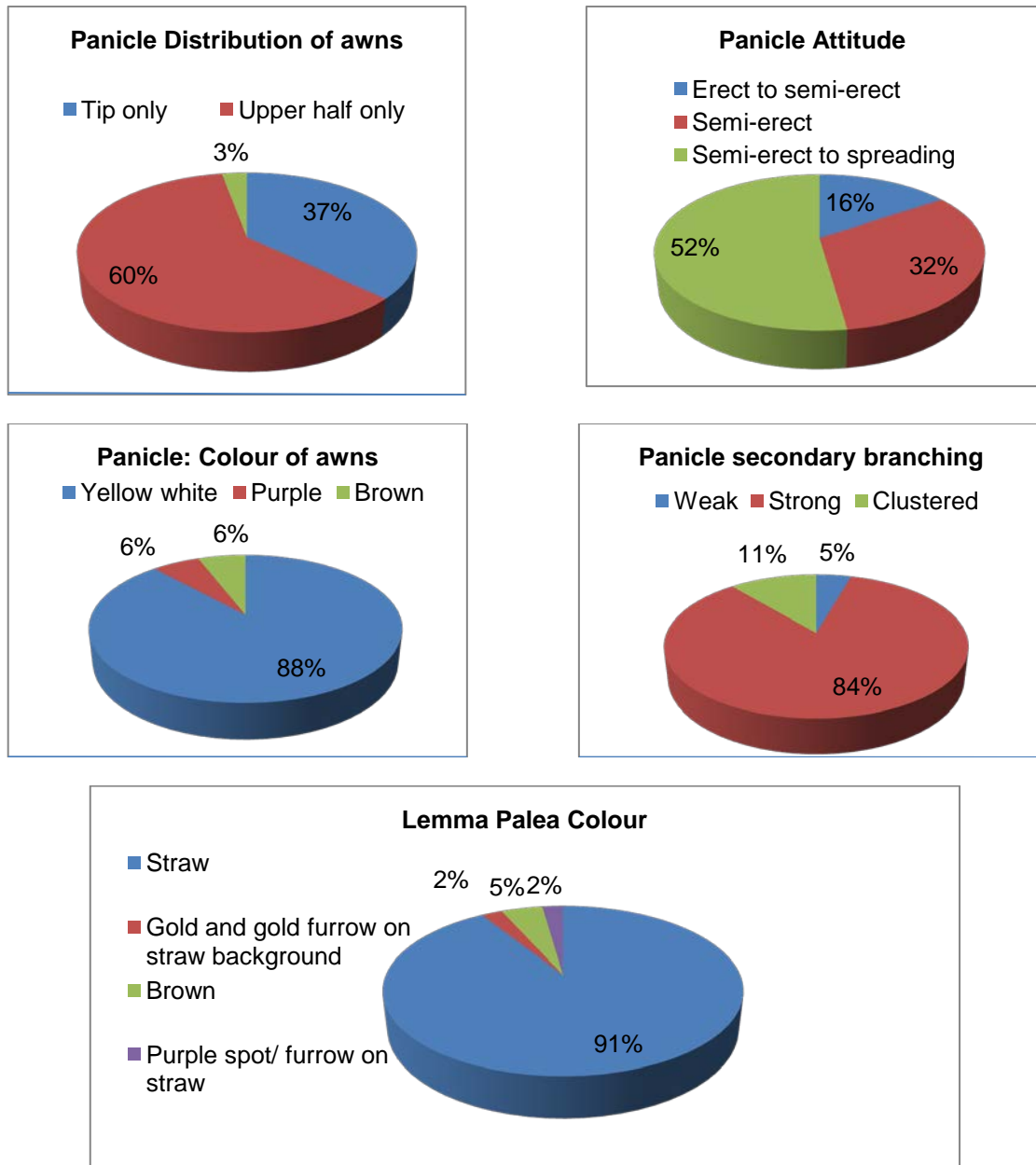


Fig 2. Frequency distribution of morphological traits showing variability

As per Tables 1 and 3, traits are classified into varied classes and depicting the phenotypic diversity among the restorers. And the frequently occurring traits clearly signified through maximum frequency per cent. Thus, genes governing such none or least variable traits are considered to be consensus with high adaptability for the Madhya Pradesh geographical location. The least variability comes with 13 traits reporting frequency under the range of 90–100% and 05 traits under the 80-90% range. Dark intensity of green colour has sufficient

frequent occurrence and this was in consonance with the findings of earlier workers [6,8,9]. Maximum frequency of leaf pubescence of blade surface reported were weak (75%). Split ligules (80%) trait exhibited commonness in comparison with acute (10%), Straw colour lemma palea colour observed in 82 genotypes, as shown in Fig. 2. The rest restores have unique index value for this particular trait, signifies its use as marker phenotypic trait in breeding programme such that ANP 526 with purple spot/ furrow on straw.

Table 1. Frequency distribution of morphological characters in fertility restorer lines

Character	Class	Frequency	Percentage
Visual Assessment by a Single observation of a group of plants or parts of plants			
LEAF			
1. Leaf: Intensity of green colour	Light	09	10.00
	Medium	77	85.56
	Dark	04	4.44
2. Leaf: Anthocyanin colouration	Absent	89	98.89
	Present	01	1.11
4 Leaf: Distribution of anthocyanin colouration	On tip only	00	00.00
	On margin only	00	00.00
	In blotches only	01	1.11
	Uniform	00	00.00
5 Leaf Sheath: Anthocyanin colouration	Absent	90	100.00
	Present	00	00.00
6 Flag leaf: Attitude of blade	Erect	52	57.70
	Semi-erect	37	41.10
	Horizontal	00	00.00
	Deflexed	01	1.11
Leaf: Senescence	Early	11	12.20
	Medium	78	86.60
	Late	01	1.11
LEMMA			
8 Lemma and Palea colour	Straw	82	91.10
	Gold and gold furrow on straw background	02	2.22
	Brown spots on straw	00	00.00
	Brown furrow on straw	00	00.00
	Brown	04	4.44
	Reddish to light purple	00	00.00
	Purple spot/ furrow on straw	02	2.22
	Purple	00	00.00
	Black	00	00.00
PANICLE			
9 Panicle: Curvature of main axis	Straight	00	00.00
	Semi-straight	18	20.00
	Deflexed	70	77.70
	Dropping	02	2.22
10 Panicle: Awns	Absent	56	62.20
	Present	34	37.70
11 Panicle: Presence of secondary branching	Absent	00	00.00
	Present	90	100.00
12 Panicle: secondary branching	Weak	04	4.44
	Strong	76	84.40
	Clustered	10	11.11
13 Panicle: Attitude of branches	Erect	00	00.00
	Erect to semi-erect	14	15.55
	Semi-erect	29	32.20
	Semi-erect to spreading	47	52.22
	Spreading	00	00.00
14 Panicle: exertion	Partly exerted	00	00.00
	Mostly exerted	04	4.44
	Well exerted	86	95.50

Visual Assessment by observation of individual plant or part of plants				
LEAF				
1	Basal Leaf : Sheath colour	Green	89	98.80
		Light Purple	01	1.11
		Purple Lines	00	00.00
		Uniform Purple	00	00.00
15	Leaf: Pubescence of blade surface	Absent	00	00.00
		Weak	75	83.33
		Medium	15	16.67
		Strong	00	00.00
		Very Strong	00	00.00
16	Leaf: Auricles	Absent	00	00.00
		Present	90	100.00
17	Leaf: Anthocyanin colorations of auricles	Colourless	89	98.80
		Light Purple	00	00.00
		Purple	01	1.11
18	Leaf: Collar	Absent	00	00.00
		Present	90	100.00
19	Leaf: Anthocyanin colouration of collar	Absent	89	98.80
		Present	01	1.11
20	Leaf: Ligule	Absent	00	00.00
		Present	90	100.00
21	Leaf: Shape of ligule	Truncate	00	00.00
		Acute	10	11.11
		Split	80	88.80
22	Leaf: Colour of ligule	White	90	100.00
		Light Purple	00	00.00
		Purple	00	00.00
STEM				
23	Culm: Attitude	Erect	18	20.00
		Semi-erect	61	67.70
		Open	09	10.00
		Spreading	02	2.22
24	Stem: Anthocyanin colouration of nodes	Absent	89	98.80
		Present	01	1.11
25	Stem: Intensity of anthocyanin coloration of nodes	Weak	00	00.00
		Medium	01	1.11
		Strong	00	00.00
26	Stem: Anthocyanin colouration of internodes	Absent	90	100.00
		Present	00	00.00
LEMMA				
27	Spikelet: Density of pubescence of lemma	Absent	00	00.00
		Weak	51	56.60
		Medium	39	43.30
		Strong	00	00.00
		Very strong	00	00.00
28	Lemma: Anthocyanin colouration of keel	Absent / very weak	88	97.70
		Weak	01	1.11
		Medium	00	00.00
		Strong	01	1.11
		Very strong	00	00.00
29	Lemma: Anthocyanin colouration of area below apex	Absent	88	97.70
		Weak	00	00.00
		Medium	01	1.11
		Strong	00	00.00

Visual Assessment by observation of individual plant or part of plants				
30	Lemma: Anthocyanin colouration of apex	Very strong	01	1.11
		Absent	87	96.60
		Weak	00	00.00
		Medium	02	2.22
		Strong	00	00.00
31	Spikelet: Colour of tip of lemma	Very strong	01	1.11
		White	00	00.00
		Yellow	85	94.44
		Brown	5	5.55
		Red	00	00.00
32	Sterile lemma colour	Purple	00	00.00
		Black	00	00.00
		Straw	89	98.80
		Gold	00	00.00
		Red	00	00.00
		Purple	01	1.11
PANICLE				
33	Panicle: Distribution of awn	Tip only	13	14.40
		Upper half only	21	23.30
		Whole length	01	1.11
34	Spikelet: Colour of stigma	White	86	95.50
		Light green	00	00.00
		Yellow	00	00.00
		Light purple	00	00.00
		Purple	04	4.44
35	Panicle: Colour of awns	Yellow white	30	33.30
		Yellow brown	00	00.00
		Brown	02	2.22
		Reddish brown	00	00.00
		Light red	00	00.00
		Red	00	00.00
		Light purple	00	00.00
		Purple	02	2.22
		Black	00	00.00
36	Panicle: Length of longest awn	Very short	00	00.00
		Short	02	2.22
		Medium	32	35.50
		Long	00	00.00
		Very long	00	00.00
37	Male Sterility	Present	90	100
		Absent	00	00

Most of fertility restorer lines fall into erect (52), followed by semi-erect (11) and deflexed (02) flag leaf attitude categories. Based on the spikelet density of pubescence of lemma entire experimental material was characterized as weak (51) and medium (39). Eighty six genotypes exhibited white stigma colour, while only four unique accessions had shown purple colour. Majority (78) genotype reported early followed by medium (11) and late (01) leaf senescence. Variation observed among ninety genotypes for traits related to panicle such as for curvature of main axis: semi-straight (18), deflexed (70) and

dropping (02); for awns: absent (56) and present (34), among these thirty four genotypes, in majority awns were found only in upper half (21) followed by only in tip (13), while yellow white colour of awn (30) were observed in majority of restorers followed by brown (02) and purple (02); for panicle exertion eighty six genotypes were well exerted and only four were mostly exerted; for secondary branching : maximum genotype exhibited strong (76) followed by clustered (10) and weak (04). In panicle attitude of branches, variability is present with semi-erect to spreading (47) followed by semi-erect (29) and erect to

semi-erect (14). The frequency distribution of some of the important traits is presented in Fig. 2 in form of pie chart.

According to above results of characterization, the traits leaf pubescence of blade, leaf anthocyanin colouration of collar, culm attitude, spikelet density of pubescence of lemma, lemma anthocyanin colouration of keel, lemma anthocyanin colouration of area below apex, lemma anthocyanin colouration of apex and spikelet colour of stigma were reported with some distinctness among genotypes [6]. While, some previous workers revealed similar findings for traits leaf anthocyanin colouration of auricle and panicle colour of awns [8,9,10], for panicle presence of awns [10,11], for lemma anthocyanin colouration of apex and spikelet colour of stigma [12]. However contradictory results were found in case of ligule shape, absences of leaf auricle and leaf collar in the landraces of rice of East India [13].

Characterization of 90 fertility restorer lines for 34 Agro-morphological traits had highlighted some genotypes with distinctness in morphology which is shown in Table 2. Among ninety genotype enquired, ANP 526 addressed specificity for traits viz, Leaf anthocyanin colouration, Leaf Distribution of anthocyanin colouration (In blotches only), Lemma: Anthocyanin colouration of Keel (strong), Lemma: Anthocyanin colouration of apex (very strong) and Spikelet colour of stigma(Purple); Mahamaya for purple colour of stigma, anthocyanin colouration of auricle and collar; JR 1009 for light purple basal leaf sheath colour; ANP 553 for deflexed flag leaf attitude of blade; NPT 37 and JR 4322-2 for short bold and Laxmi 144 for short slender decorticated grain shape. The four genotypes expressed purple colour of stigma were ANP 526, Mahamaya, NPT 14-12 and JR 1301.

Table 2. Agro-morphological characterization with unique assess ions found under index value

S.No.	Characterstics	Index value	Unique genotype
1	Leaf Intensity of green colour (GC)	Dark	JR-1004, RP 5911-52-13-3-2-2-1, ANP-553, PS-2, NPT 89*IR-36, PS-2, NPT 40-018* PUSA BASMATI
3.	Leaf Distribution of anthocyanin colouration (DAC)	In blotches only	ANP-526
4.	Flag leaf attitude (FLA)	Deflexed	ANP-553
5	Leaf: Senescence (LS)	Late	NPT 37
6	Basal leaf: Sheath colour	Light purple	JR-1009
7	Leaf Anthocyanin colouration of auricles	Purple	Mahamaya
8	Leaf Anthocyanin colouration of collar	Present	Mahamaya
9	Stem Anthocyanin colouration of nodes (ACN)	Present	JR-1021`
10	Stem Intensity of anthocyanin coloration of nodes (IAC)	Medium	JR-1021`
11	Lemma Palea Colour (LPC)	Purple spot / furrow on straw	ANP 526
12	Lemma Palea Anthocyanin colouration of keel	Strong	ANP 526
13	Lemma Palea Anthocyanin colouration of area below apex	Medium Very strong	ANP 526 RPHR 619
14	Lemma Palea Anthocyanin colouration of apex	Medium Very strong	Mahamaya, ANP526 RPHR 619
15	Sterile lemma Colour	Purple	ANP 526
16	Colour of stigma	Purple	Mahamaya, ANP 526, RPHR 619, NPT 14-12.

S.No.	Characterstics	Index value	Unique genotype
17	Panicle Curvature of main axis (PC)	Dropping	ANP 553
18	Panicle Distribution of awns (DA)	Whole length	CANP 318
19	Panicle Exertion (PE)	Mostly exerted	NPT 89*IR 64, NPT 89*IR 36, NPT 40-018* Pusa Basmati.
20	Panicle: Colour of awns	Brown Purple	CANP 318, ANP 553 ANP 526, RPHR 619

Table 3. Classification of variability on the basis of frequency range

S.NO.	Frequency range	Variability	Traits	Remark
1.	100	No Variability	08	Highly Frequent trait
2.	80-99	Least Variability	18	Relatively highly frequent
3.	60-79	Medium Variabilty	03	Medium frequent
4.	<59	High Variability	08	Least Frequent

4. CONCLUSION

From ancient times, the visual observations scoring of genotypes were used as an aid to determine genetic diversity in gene pool. The existence of distinctness, uniformity and stability is confirmed through characterization of traits under studies which were analysed as per DUS guidelines. Therefore, out of 34 morphological traits seven traits- leaf sheath anthocyanin colouration, presence of leaf auricle, presence of leaf collar, presence of leaf ligule, white colour of leaf ligule, absence of stem anthocyanin colouration of internodes and presence of secondary branching in panicle were found to be monomorphic; while fourteen traits like- basal leaf sheath colour (green, light purple), leaf anthocyanin colouration, Auricle colour (colourless, purple), anthocyanin colouration of collar, stem anthocyanin colouration of node, leaf pubescence of blade surface (weak, medium), ligule shape (acute, split), spikelet density of pubescence of lemma (weak, medium), colour of sterile lemma, stigma colour, leaf senescence, awns, awns distribution, panicle exertion were dimorphic. Sufficient amount of variability was recorded for the remaining traits. Among all traits under study, Lemma palea colour was the only tetramorphic trait showing maximum variability.

The genotype ANP 526 and Mahamaya were reported as unique genotypes in terms of many qualitative traits Thus, these restorer lines can be further utilized as morphological markers in plant breeding programme.

ACKNOWLEDGEMENT

The authors are thankful to the College of Agriculture, Jawaharlal Nehru Krishi Vishwa

Vidyalaya, Jabalpur, Madhya Pradesh, India for its support.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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Peer-review history:

The peer review history for this paper can be accessed here:
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