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Morphological Characterization of Advanced Coloured Rice Genotypes

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Abstract

Morphological characterization of 12 advanced coloured rice genotypes was carried out using 39 morphological descriptors during Kharif, 2021 at Agricultural Research Station, Bapatla and the results revealed that 20 out of 39 characters showed variation. Estimates of Shannon weaver diversity indices for the characters studied ranged from 0 to 0.81. The highest diversity index of 0.81 was exhibited by the Flag leaf: attitude of the blade (late observation). Cluster analysis revealed that BPT2858 from cluster I was unique when compared to other cultures. DUS characterization of advanced rice genotypes helps the plant breeder to maintain the cultures with 100 % genetic purity and the cultures can also be used as parents in the breeding programmes based on the objective.

Keywords: Coloured rice, Phenotypic diversity, Shannon weaver diversity index and cluster

Introduction

Most of the population in developing countries is suffering from diabetes, malnutrition, and chronic diseases where white rice is the major staple food (Dipti *et al*., 2012). Coloured rice varieties, on the other hand, are alternative healthy foods that contain high antioxidants and other nutrients (protein, vitamins, minerals, phenolic and low glycemic index) that have a significant benefit on human health. Antioxidants play an important role in reducing the risk of cancer and other diseases (Sani *et al*., 2018). Hence, a better understanding of the distribution and extent of morphological diversity among the coloured rice cultures is crucial to assist plant breeders in the selection of parents.

DUS testing is a way of determining whether a newly bred variety differs from existing varieties (the Distinctness part), whether the characteristics used to establish Distinctness are expressed uniformly (the Uniformity part), and whether these characteristics do not change over subsequent generations (the stability part). The development of a new cultivar is the result of genetic improvement and its identification should be possible through morphological traits by Distinctiveness, Uniformity, and Stability (DUS). Morphological characterization provides an identity for each genotype through the use of a series of descriptors that indicate genetic variability and distinguishability compared to the other genotypes. These traits, called morphological descriptors, must be heritable, and controlled by a few genes, and their expression should be uniform in all environments. The selection of the most discriminating traits is especially important for all the crops (Khadivi-Khub and Anjam, 2014). Morphological descriptors remain the first step in the conservation process of the plants (Podgornik *et al*., 2010) and the most suitable tool for genetic diversity (Darjazi, 2011). With the above background and scope a study was conducted to characterize 12 coloured rice advanced cultures developed at ARS, Bapatla. If genotypes with similar morphological traits are repeatedly bred together, the benefits resulting from crossing may be limited. This is because the resulting offspring will have similar genetic makeup and may not exhibit as much genetic gain (Chimello *et al*., 2017) as those produced from diverse genotypes. Therefore, it is important for breeders to select genotypes with diverse genetic backgrounds to maximize the benefits of breeding programs and achieve greater genetic gain.

Materials and Methods

The experimental material was comprised of 12 advanced cultures of coloured rice grown in *Kharif*, 2021 at Agricultural Research Station, Bapatla. Data was recorded on 39 DUS descriptors following guidelines from Protection of Plant Varieties and Farmer's Rights Authority (PPV & FRA). The characters include Coleoptile - Colour, Basal-Leaf sheath colour, Leaf-Anthocyanin colouration, Leaf sheath anthocyanin colouration, Leaf- pubescence of blade surface, Leaf-auricles, Leaf-Length of blade, Leaf- Width of blade, Time of heading, Flag leaf-Attitude of blade (early observation), Spikelet-density of pubescence of lemma, Male sterility, Lemma-Anthocyanin colouration of keel, Lemma-Anthocyanin colouration of area below apex, Lemma-Anthocyanin colouration of apex, Spikelet-colour of stigma, Stem-Length, Stem-anthocaynin colouration of nodes, Stem-anthocyanin colouration of internodes, Panicle-Length of main axis, Flag leaf-attitude of blade (late observation), Panicle curvature of main axis, Panicle-number per plant, Spikelet-colour of tip of lemma, Lemma and Palea colour, Panicle-awns, Panicle-exertion, Time of Maturity, Sterile Lemma-colour, Grain- weight of 1000 fully developed grains, Grain length, Grain width, Decorticated-grain length, Decorticated-grain width, Decorticated-grain shape, Decorticated-grain colour, Endosperm-presence of amylose, Endosperm-content of amylose and Decorticated grain-aroma.

Shannon Diversity Index: Estimates of Shannon Weaver Diversity Indices reveals the phenotypic diversity shown by the cultures. Shannon diversity indices (H^l) were calculated to study the phenotypic diversity for each character in the entire germplasm as described by Perry and McIntosh (1991) is given as:

$$H^1 = 1 - \sum_{i=1}^{n} p_i \log_e p_i$$

where p_i is the proportion of accessions in the i^{th} class of an n-class character and n is the number of phenotypic classes for a character. The indices are standardized by dividing each value of H' by $\log e_n$ to keep the value in a range of 0 to 1 in order to estimate the importance of phenotypic diversity. Analysis was done by using MS EXCEL. An arbitrary scale of diversity indices was adapted from Rabara *et al*., (2014) to categorize the computed indices into high (H' = 0.76–0.99), moderate (H' = 0.46–0.75), and low diversity (0.01–0.45). Cluster analysis was done by using Minitab software.

Results and Discussion

Phenotypic frequencies and genotypes distribution

The frequency distribution of 12 advanced coloured rice genotypes for 39 morphological descriptors are represented in **Table 1**.

Table 1. Frequency distribution of 12 coloured rice advanced cultures for 39 morphological descriptors

S. No.	Character	Phenotypic class	No. of genotypes under each class	Frequency Distribution (%)	Genotypes under each class
1	Coleoptile: Colour	Colorless	12	100	All the genotypes
2	Basal: Leaf sheath colour	Green	12	100	All the genotypes
3	Leaf: Anthocyanin colouration	Absent	12	100	All the genotypes
4	Leaf sheath anthocyanin colouration	Absent	12	100	All the genotypes
5	Leaf: pubescence of blade surface	Absent	12	100	All the genotypes
6	Leaf:auricles	Presence	12	100	All the genotypes

S. No.	Character	Phenotypic class	No. of genotypes under each class	Frequency Distribution (%)	Genotypes under each class
7	Leaf: Length of blade	Medium (30-45 cm)	4	33	BPT2848, BPT3136, BPT3143, BPT2841
		Long (> 45 cm)	8	67	BPT2858, BPT3182, BPT3137, BPT3111, BPT3141, BPT3140, BPT 3145, BPT 3178
8	Leaf: Width of blade	Medium (1-2 cm)	11	92	BPT2848, BPT3136, BPT3143, BPT3182, BPT3137, BPT3111, BPT2841, BPT3141, BPT3140, BPT 3145, BPT 3178
		Broad (> 2 cm)	1	8	BPT2858
9	Time of heading	Early (71-90 days)	6	50	BPT2848, BPT3136, BPT3137, BPT2841, BPT3141, BPT3140
		Medium (91-110 days)	3	25	BPT3111, BPT 3145, BPT 3178
		Late (111-130 days)	3	25	BPT2858, BPT3143, BPT3182
10	Flag leaf: Attitude of blade (Early observation)	Erect	12	100	All the genotypes
11	Spikelet: density of pubescence of lemma	Absent	8	67	BPT2858, BPT2848, BPT3136, BPT3143, BPT3182, BPT3137, BPT3111,BPT 3145
		Weak	1	8	BPT 3178
		Medium	3	25	BPT2841, BPT3141, BPT3140
12	Male sterility	Absent	12	100	All the genotypes
13	Lemma: Anthocyanin colouration of keel	Absent	12	100	All the genotypes
14	Lemma: anthocyanin colouration of area below apex	Absent	12	100	All the genotypes
15	Lemma: anthocyanin colouration of apex	Absent	12	100	All the genotypes
16	Spikelet: colour of stigma	White	11	92	BPT2858, BPT2848, BPT3136, BPT3143, BPT3182, BPT3111, BPT2841, BPT3141, BPT3140, BPT 3145, BPT 3178
		Purple	1	8	BPT3137
17	Stem: length	Very short (< 91 cm)	1	8	BPT2858
		Medium (111-130 cm)	11	92	BPT2848, BPT3136, BPT3143, BPT3182, BPT3137, BPT3111, BPT2841, BPT3141, BPT3140, BPT 3145, BPT 3178
18	Stem: anthocyanin colouration of nodes	Absent	12	100	All the genotypes
19	Stem: anthocyanin colouration of internodes	Absent	12	100	All the genotypes

S. No.	Character	Phenotypic class	No. of genotypes under each class	Frequency Distribution (%)	Genotypes under each class
20	Panicle; Length of main axis	Medium (21-25 cm)	3	25	BPT3136, BPT3143, BPT3140
		Long (26-30 cm)	7	58	BPT2858, BPT2848, BPT3182, BPT3137, BPT2841, BPT3141, BPT 3145
		Very long (>30 cm)	2	17	BPT3111, BPT 3178
21	Flag leaf: attitude of blade (late observation)	Erect	6	50	BPT3136, BPT3143, BPT3182, BPT3111, BPT2841,BPT 3178
		Semi-erect	1	8	BPT2848
		Horizontal	4	33	BPT2858, BPT3137, BPT3140, BPT 3145
		Deflexed	1	8	BPT3141
22	Panicle curvature of main axis	Semi-straight	1	8	BPT3182,
		Deflexed	5	42	BPT2848, BPT3143, BPT3137, BPT 3145
		Drooping	7	58	BPT2858,BPT3136, BPT3111, BPT2841, BPT3141, BPT3140, BPT 3178
23	Panicle: number per plant	Few (<11)	4	33	BPT2858, BPT3182, BPT3141, BPT 3145
		Medium (11-20)	8	67	BPT2848, BPT3136, BPT3143, BPT3137, BPT3111, BPT2841, BPT3140, BPT 3178
24	Spikelet: colour of tip of lemma	White	11	92	BPT2858, BPT2848, BPT3136, BPT3143, BPT3137, BPT3111, BPT2841, BPT3141, BPT3140, BPT 3145, BPT 3178
		Purple	1	8	BPT3182
25	Lemma and Palea colour	Straw	12	100	All the genotypes
26	Panicle: awns	Absent	12	100	All the genotypes
27	Panicle: Exertion	Mostly exerted	6	50	BPT3136, BPT3143, BPT3182, BPT3111, BPT2841, BPT 3145
		Well exerted	6	50	BPT2858, BPT2848, BPT3137, BPT3141, BPT3140, BPT 3178
28	Time of Maturity	Early (101-120)	6	50	BPT2848, BPT3136, BPT3137, BPT2841, BPT3141, BPT3140
		Medium (121-140)	3	25	BPT3111, BPT 3145, BPT 3178
		Late (141-160)	3	25	BPT2858, BPT3143, BPT3182,
29	Sterile lemma: colour	Straw	12	100	All the genotypes
30	Grain: weight of 1000 fully developed grains	Very low (< 15g)	6	50	BPT2858, BPT2848, BPT3137, BPT3111, BPT2841, BPT3141,
		Low (15-20 g)	5	42	BPT3136, BPT3143, BPT3182, BPT3140, BPT 3178
		Medium (21-25g)	1	8	BPT 3145

S. No.	Character	Phenotypic class	No. of genotypes under each class	Frequency Distribution (%)	Genotypes under each class
31	Grain Length	Medium	10	83	BPT2858, BPT2848, BPT3136, BPT3143, BPT3137, BPT3111, BPT2841, BPT3141, BPT3140, BPT 3145
		Long	2	17	BPT3182, BPT 3178
32	Grain Width	Narrow (< 1 cm)	8	67	BPT2858, BPT2848, BPT3143, BPT3182, BPT3137, BPT3111, BPT2841, BPT3141
		Medium	4	33	BPT3136, BPT3140, BPT 3145, BPT 3178
33	Decorticated: grain length	Medium	10	83	BPT2858, BPT2848, BPT3136, BPT3143, BPT3137, BPT3111, BPT2841, BPT3141, BPT3140, BPT 3145
		Long	2	17	BPT3182, BPT 3178
34	Decorticated: grain width	Narrow (< 2mm)	8	67	BPT2858, BPT2848, BPT3143, BPT3182, BPT3137, BPT3111, BPT2841, BPT3141
		Medium (2-2.5mm)	4	33	BPT3136,BPT3140, BPT 3145, BPT 3178
35	Decorticated grain: shape	Short slender	1	8	BPT2858
		Short bold	1	8	BPT3140
		Medium slender	8	67	BPT2848, BPT3136, BPT3143, BPT3137, BPT3111, BPT2841, BPT3141, BPT 3145
		Long bold	1	8	BPT3182
		Long slender	1	8	BPT 3178
36	Decorticated grain colour	Light red	1	8	BPT3141
		Red	6	50	BPT2858, BPT3143, BPT3182, BPT3111, BPT3140, BPT 3178
		Variegated purple	2	17	BPT3136, BPT 3145
		Purple	1	8	BPT2848
		Dark purple	2	17	BPT3137, BPT2841
37	Endosperm: presence of Amylose	Present	12	100	All the genotypes
38	Endosperm: content of Amylose	Medium (20-25%)	12	100	All the genotypes
39	Decorticated grain: Aroma	Absent	12	100	All the genotypes

Leaf characters

Among the leaf traits, all the genotypes exhibited colour-less coleoptiles colour, green basal leaf sheath colour, lack of anthocyanin colour in both leaf and leaf sheath, absence of leaf pubescence of blade surface, presence of leaf auricles and erect flag leaf attitude of blade (early observation). For the length of the leaf blade, 33 % of genotypes exhibited medium length (30-45 cm) and 67 % exhibited long (> 45 cm) length of the blade. Medium width (1-2 cm) of the leaf blade was exhibited by 92 % of genotypes and 8 % of genotypes exhibited a broad (> 2 cm) width of the leaf blade. In the case of Flag leaf attitude of the leaf blade, all the genotypes exhibited erect flag leaf in early observation but in the case of late observation, 50 % of genotypes exhibited erect flag leaf attitude, 33% horizontal, 8% semi-erect and remaining 8 % exhibited deflexed flag leaf attitude.

Spikelet characters

Awns were absent in all the genotypes and exhibited straw coloured lemma and palea. 67 % of genotypes exhibited absence of spikelet density of pubescence of lemma, 7 % showed weak pubescence, and the remaining 25 % medium density of pubescence of lemma. Though all the genotypes evaluated in the present study are coloured genotypes, absence of anthocyanin colour in three parts of the lemma *viz.*, Lemma anthocyanin colouration of the keel, lemma anthocyanin colouration of the area below the apex and anthocyanin colouration of the apex was observed. White-coloured stigma was observed in almost all the genotypes except BPT3137 where purple-coloured stigma was seen.

Stem characters

The stem anthocyanin colouration of nodes and internodes was absent in all the genotypes. For stem length, almost all the genotypes exhibited medium stem length ranging from 111 to 130 cm except BPT2858 which exhibited very short stem length of less than 91 cm.

Panicle characters

Long panicle length of the main axis was observed in 58 % of genotypes, whereas 25 % of genotypes exhibited medium and 17 % of genotypes showed very long panicle length of the main axis. For panicle curvature deflexed type was observed in 42 % of genotypes, drooping in 58 % and 8 % of genotypes showed a semi-straight pattern. Medium panicle numbers range from 11-20 in 67 % of genotypes, whereas 33 % of the genotypes showed few panicle numbers per plant. Mostly exertion type of panicle was seen in 50 % of genotypes and the remaining 50 % showed a well-exerted type of panicle.

Grain characters

The grain weight of 1000 fully developed grains was found to be very low (< 15 g) in 50 % of genotypes, 42 % of genotypes exhibited low grain weight and 8 % showed medium grain weight. The grain length was found to be medium in 83 % of genotypes and long grain length was observed in BPT 3182 and BPT3178 genotypes. Narrow grain width was seen in 67 % of genotypes and 33 % showed medium grain width. Medium slender type of decorticated grain shape was observed in 67 % of genotypes and the remaining genotypes showed short slender, short bold, long bold, and long slender types of grain shape. A lot of variation was observed in the character decorticated grain colour, where 50 % of genotypes (BPT2858, BPT3143, BPT3182, BPT3111, BPT3140, and BPT3178) exhibited red colour, 17 % of genotypes (BPT3136 and BPT3145) showed variegated purple colour, 17 % of genotypes (BPT3137 and BPT3841) showed dark purple colour and the remaining 8 % (BPT2848) showed purple coloured grain colour (**Figure 1**). All the genotypes contain medium amylose content in endosperm, while grain aroma was absent in all the genotypes.

Figure 1: Variation in decorticated grain colour among 12 coloured rice advanced cultures

Shannon weaver diversity analysis

The estimates of Shannon weaver diversity indices (**Table 2**) revealed the extent of phenotypic diversity showed by 12 rice advanced cultures in a particular trait. Shannon weaver diversity indices in the present study for 39 morphological descriptors ranged from 0 to 0.81. The traits were grouped into three categories based on the index values *viz*., high diversity (0.76-0.99), Moderate diversity (0.46 to 0.75), and Low diversity (0.01 – 0.45). Out of 39 characters studied only 20 characters exhibited phenotypic diversity. The Shannon weaver diversity index values ranged from 0.81 to 0.16. The highest diversity index value of 0.81 was observed in Flag leaf: attitude of the blade (late observation). Similar results of high diversity index value for the flag leaf attitude of the blade were reported by Rawte and Saxena, 2018. Moderate index values were exhibited by spikelet: density of pubescence of lemma(0.51), decorticated grain shape (0.56), grain: weight of 1000 fully developed grains (0.57), decorticated grain width (0.58), panicle number per plant (0.58), leaf: length of the blade (0.58), panicle: length of the main axis (0.6), decorticate grain colour (0.62), panicle exertion 0.63), panicle curvature of the main axis (0.64), time of maturity (0.65) and time of heading (0.65). Similar results of moderate index value for panicle curvature of the main axis were reported by Rao *et al*., (2021) and Rabara *et al.*, 2014 reported for Panicle exertion, panicle length of the main axis, and length of the leaf blade. Low diversity index values were exhibited by the spikelet colour of the tip of the lemma (0.16), stem length (0.18), spikelet colour of stigma (0.18), leaf width of the blade (0.26), decorticated grain length (0.28), grain length (0.28) and grain width (0.4). Lemma and Palea colour exhibited a 0 index value, similar results were also reported by Hein *et al*., (2007).

The genotypes were grouped into five major clusters based on similarity index (**Figure 2**) cluster I consists of only one genotype i.e., BPT 2858 which has unique characters than other genotypes *viz*., broad leaf width, very short stem length and short slender grain shape. Cluster II consists of three genotypes, BPT2848, BPT3137 & BPT3145 with 94 % similarity. Cluster III consists of four genotypes BPT 3136, BPT3111, BPT2841, and BPT3178 with 95 % similarity. Cluster IV consists of two genotypes BPT 3143 and BPT3182 with 96 % similarity. Cluster V consists of two genotypes that are 97 % similar to each other.

Table 2. Estimates of Shannon weaver diversity indices for 39 morphological descriptors studied

S.No.	Character	Shannon Weaver Diversity Index
High Diversity (0.76-0.99)		
1	Flag leaf: attitude of blade (late observation)	0.81
Moderate Diversity (0.46-0.75)		
2	Time of heading	0.65
3	Time of Maturity	0.65
4	Panicle: curvature of main axis	0.64
5	Panicle: Exertion	0.63
6	Decorticated grain colour	0.62
7	Panicle: length of main axis	0.6
8	Leaf: length of blade	0.58
9	Panicle: number per plant	0.58
10	Decorticated: grain width	0.58
11	Grain: weight of 1000 fully developed grains	0.57
12	Decorticated grain shape	0.56
13	Spikelet: density of pubescence of lemma	0.51
Low Diversity (0.01-0.45)		
14	Grain width	0.4
15	Grain Length	0.28
16	Decorticated: grain length	0.28
17	Leaf: width of blade	0.26
18	Spikelet :colour of stigma	0.18
19	Stem: length	0.18
20	Spikelet:colour of tip of lemma	0.16
In variant		
21	Coleoptile: Colour	0
22	Basal: Leaf sheath colour	0
23	Leaf: Anthocyanin colouration	0
24	Leaf sheath anthocyanin colouration	0
25	Leaf: pubescence of blade surface	0
26	Leaf: auricles	0
27	Flag leaf: Attitude of blade (Early observation)	0
28	Male sterility	0
29	Lemma: Anthocyanin colouration of keel	0
30	Lemma: anthocyanin colouration of area below apex	0
31	Lemma: anthocyanin colouration of apex	0
32	Stem: anthocyanin colouration of nodes	0
33	Stem: anthocyanin colouration of internodes	0
34	Lemma and Palea colour	0
35	Panicle: awns	0
36	Sterile lemma: colour	0
37	Endosperm: presence of amylose	0
38	Endosperm: content of amylose	0
39	Decorticated grain: Aroma	0

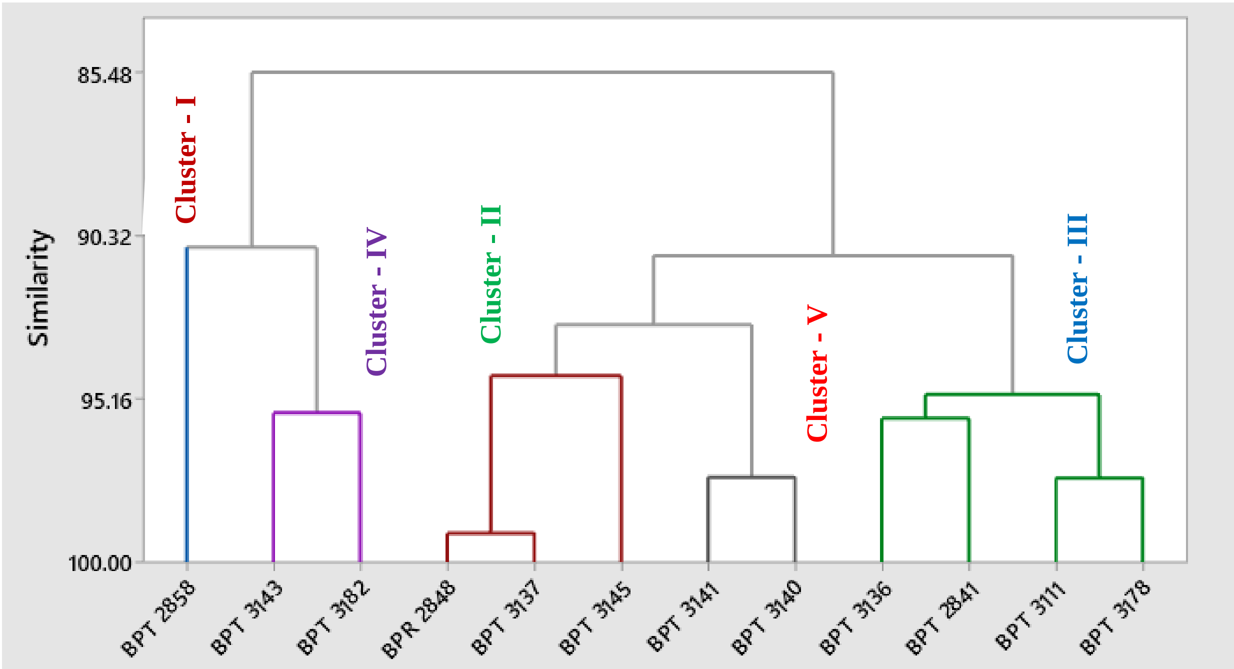

Figure 2: Dendrogram of 12 advanced coloured rice genotypes based on 39 morphological descriptors

Conclusion

The present study revealed a significant amount of variation in the decorticated grain colour trait. Rice genotypes possessing purple or red grain colour are known to be abundant in antioxidants, which have been shown to lower the risk of cancer and other diseases. Till now coloured varieties are rarely developed but coloured landraces are being cultivated in some areas with problems like lodging and not having consumer preference due to bold grains. The colored rice advanced cultures analyzed in this study exhibited leaf characteristics similar to those of currently available white rice cultivars, with no anthocyanin coloration, having a non-lodging growth habit, and mostly slender to medium-slender grains. Thus, by evaluating the yield-related parameters of these advanced cultures, they can be released directly as new varieties or used as parental stock in breeding programs aimed at developing colored rice varieties.

References

Chimello, Antonio Marcos, Jesus JG, Teodoro Paulo, Rossi Ana, Araújo KL, Marostega TN, Neves LG and Barelli Marco. 2017. Morphological descriptors and ISSR molecular markers in the evaluation of genetic variability of *Tectona grandis* genotypes. *Genetics and Molecular Research*, 16. 10.4238/gmr16029665.

Darjazi BB. 2011. Morphological and pomological characteristics of fig (*Ficus carica* L.) cultivars from Varamin, Iran. *African Journal of Biotechnology*, 10: 19096-19105.

Dipti SS, Bergman C and Indrasari SD. 2012. The potential of rice to offer solutions for malnutrition and chronic diseases. *Rice,* 5:161-18.

Hein NL, Sarhadi WA, Oikawa Y and Hirata Y. 2007. Genetic diversity of morphological responses and the relationships among Asia aromatic rice (*Oryza sativa* L.) cultivars. *Tropics,* 16: 343-355.

Khadivi-Khub A and Anjam K. 2016. The relationship of fruit size and light condition with number, activity and price of *Blastophaga psenes* wasp in caprifigs**.** *Trees,* 30: 1855-1862

Podgornik M, Vul I, Vrhovnik I and Mavsar DB. 2010. A survey and morphological evaluation of fig (*Ficus carica* L.) genetic resources from Slovenia. *Scientia Horticulturae*,125: 380-389.

Naga Durga Rao N, Roja V, Tushara M, Satyanarayana Rao V and Srinivasa Rao V. 2021.Characterization and diversity analysis of Rice germplasm. *Biological Forum – An International Journal*, 13: 170-179.

Perry MC and McIntosh MS. 1991. Geographical patterns of variation in the USDA soybean germplasm collection: International Morphological traits. *Crop Science*, 31: 1350–1355.

Rabara RC, Ferrer M, Diaz CL, Newingham MCV and Romero GO. 2014. Phenotypic diversity of farmers' traditional rice varieties in the Philippines. *Agronomy,* 4: 217-241.

Rawte S and Saxena R. 2018. Phenotypic diversity and correlation analysis for agro morphological traits in 100 landraces of rice from Chhattisgarh. *International Journal of Pure and Applied Biosciences,* 6: 345 – 353.

Sani NA, Sawei J and Ratnam W. 2018. Physical, antioxidant and antibacterial properties of rice (*Oryza sativa* L.) and glutinous rice (*Oryza sativa* var. glutinosa) from local cultivators and markets of Peninsular, Malaysia. *International Food Research Journal*, 25: 2328–2336.