ISSN NO. 2320-5407

Journal homepage: http://www.journalijar.com

INTERNATIONAL JOURNAL OF ADVANCED RESEARCH

RESEARCH ARTICLE

CHARACTERIZATION OF TRADITIONAL AROMATIC RICE CULTIVARS BY CHEMICAL MARKERS

Dileepkumar Masuthi[1]*, Vyakarnahal B. S.[2] , Deshpande V. K, [3] R.C. Jagadeesha[4]., Mukesh L. Chavan[5] and Khaja Rubina S,

[1]. Assistant Professor of Seed Science and Technology, [4] Professor of Genetics and Plant breeding, [5]Associate Professor of Crop Physiology. Department of Biotechnology and Crop Improvement KRC College of Horticulture,Arabhavi-591218, Gokak Tq, Belgaum Dist. Karnataka, India

[2,3] Professor of Seed Science and Technology. Department of Seed Science and Technology, College of Agriculture, University of Agricultural Sciences, Dharwad-580005.Kranataka, India

Manuscript Info

Manuscript History:

Received: 18 March 2015
Final Accepted: 15 April 2015
Published Online: May 2015

Key words:

Traditional varieties, scented rice, chemical markers, Characterization

Corresponding Author

Dr. Dileepkumar Masuthi

Abstract

The phenol colour test, out of 32 genotypes, 12 genotypes showed no colour change, 15 genotypes were light brown, 8 genotypes were brown and six genotypes were dark brown in colour. For the modified phenol test with $CuSO_4$, 10 genotypes were found with no colour change, 9 genotypes were light brown, 9 genotypes were brown, 9 genotypes were dark brown in colour and four genotypes were Black (IET-18393, Kalanamak, Kagi sali and Huggi bhatta) in colour. Based on the colour development of the decanted solution by Sodium hydroxide, 3 genotypes showed no colour change (Sindagi local, IET-21046 and Huggi bhatta), 14 genotypes were yellow and 24 genotypes were light yellow in colour. In case of potasium hydroxide 8 genotypes were shown no colour change, 14 genotypes were light yellow, 13 genotypes were dark yellow and six genotypes were having reddish brown (Sindagi Local, Parimala Kalavi, Kalanamak, Kagi sali, Huggi bhatta and Kari basmati) in colour. In response to KI test, 16 genotypes were bluish brown colour and remaining 25 genotypes were reddish brown in colour group.

INTRODUCTION

Rice has a renowned relationship with the humans since ages. Presently, more than half of the world's population depends on rice as a staple food. Asia can be considered as 'Rice Basket' of the world, as more than 90 per cent of the rice is produced and consumed in Asia, a region with high population density. The improvement of indigenous small and medium grained aromatic rice, which possesses outstanding quality like aroma, kernel elongation after cooking, fluffiness and taste were somewhat neglected as they lacked export value. Aroma, elongation short grained aromatic rice is known to be superior to Basmati types. Domestic market exists for the indigenous aromatic rice which is popular in their native areas of cultivation. Further, management of the indigenous aromatic rice genetic resources by way of characterization and documentation helps in protection of these unique bio resources in accordance with the provision laid out in Protection of Plant Varieties and Farmers Right (PPV and FR) Act (Anon., 2001). The variety characterization and identification have attained much importance in almost all major agricultural crops to ensure the quality of seeds marketed to the farmers. Finally, the ultimate consumers of the harvested seed also often need to be certain that they are purchasing the variety ought to be, particularly if the

seed is to be used for mechanized processing. Thus, it is clearly important from many points of view to be able to distinguish between and identify crop varieties.

Although characterization by visual observation offers the greatest advantages of requiring no equipment, it is subjective requiring great experience and the potential for discrimination is poor. Moreover high number of new cultivars and their mutual similarity do not allow their differentiation based on phenotype. It is less suitable when the results are required rapidly and is influenced by environment because of its mutagenic and continuous expression. For these reasons varietal identification generally require additional observations by analyzing chemical of seed in the laboratory. Various biochemical tests are being used to reveal chemical differences among the seeds or seedlings of different genotypes. These tests have several important characteristics in common. They require virtually no technical expertise or training and can be completed in a relatively short time. The results of these tests are usually distinct, easily interpreted and help in grouping of rice genotypes.

Materials and Method

Districts of Transitional belt and the western ghat of Northen Karnataka (Uttara Kannada, Belgaum, Dharwad, Haveri, Shimoga and Davanageri districts), which majorly contribute largely for the rice germplasm. local farmers from these areas were enquired primarily about the native aromatic landraces. And these seeds were taken for the chemical tests that include Phenol test, modied Phenol test, KOH test, NaOH test and KI test.

Phenol test -Two hundred (50 x 4) seeds were presoaked in distilled water for 24 hours at $25^0 \pm 1$°C. Then they were transferred on to two layers of Whatman No.1 filter paper saturated with four per cent phenol solution. The petri dishes were covered and incubated at 25 ± 1°C and the colour reaction was noted after 24 hours. Based on the development of seed coat colour, the selected genotypes were classified into four categories (Jaiswal and Agarwal, 1995) as No change in colour, Light brown and Dark brown

Modified phenol test-Modified phenol test was conducted similar to standard phenol test except that seeds were soaked in 0.5 per cent $CuSO_4$ solution for 24 hours instead of distilled water. Colour reaction was noted after 48 hours of incubation and the genotypes were classified into five categories (Jaiswal and Agarwal, l995) as No change in colour, Light brown, Brown, Dark brown and Black

NaOH test-Seeds were soaked in 2 per cent NaOH solution for one hour and change in colour of the solution and seeds were observed after one hour. Based on the intensity of the colour reaction the genotypes were classified into three groups *viz*., no change in colour, yellow and light yellow (Chakrabaurty and Agarwal, 1989).

KOH test-Seeds were soaked in 4 per cent KOH solution for three hours and change in colour of the solution were observed. Based on the intensity of the colour reaction the genotypes were classified into four groups *viz*., no change in colour, light yellow, dark yellow and reddish brown types.

KI test-The dehusked seeds were cut at the center to expose the endosperm. Immerse the cut surface of the endosperm to weak KI solution (0.2% Iodine + 2 % KI) for five minutes and observed for the change in colour. The glutinous endosperm stains brown and non-glutinous endosperm stains bluish brown.

Result and Discussion

Identification of traditionally varietal has involved visual inspection of morphological characters of plants with the help of a reference manual, listing systematic description of national set of varieties. Based on the colour development of the decanted solution for the Phenol test, the 41 genotypes selected, the response of 12 genotypes was no colour change, 15 genotypes were light brown, 8 genotypes were brown (Table:1) and six genotypes were dark brown in colour. The phenol colour test, which is an index of polyphenol oxidase activity, is a simple method for grouping the rice varieties (Vanangamudi *et al.,* 1988). During phenol colour test, phenol gets oxidized into dark colour melanin catalysed primarily by the enzyme tyrosinase in the seed coat and is under simple genetic control (Bhowal *et al.,* 1969).

All the selected genotypes responded positively to the modified phenol test with $CuSO_4$. Among 41 genotypes, the response of 10 genotypes were no colour change, 9 genotypes were light brown, 9 genotypes were brown, 9 genotypes were dark brown in colour, and four genotypes were Black (IET-18393, Kalanamak, Kagi sali and Huggi bhatta) in colour. Hence, based on the colour reaction with phenol and modified phenol tests the genotypes can be classified into different groups and the standard phenol test with $CuSO_4$ found to be better in distinguishing cultivars. The presence of metallic ions Cu++ in modified test enhances the phenol colour reaction

since it is an enzymatic reaction and these ions act as catalyst (Banerjee and Chandra, 1977).The difference in the phenol colour reaction of hulls seem to be due to the differences in the genetic background, presumably concerning the enzyme system. The genotypes were grouped based on phenol colour reaction test in rice by Vanangamudi *et al*. (1988), Devi Singh *et al*. (2011) and Jaiswal and Agarwal (1995).The present findings revealed that phenol and modified phenol tests with $CuSO_4$ could be used as simple, quick and cheap method for grouping of the rice genotypes.

Based on the colour development of the decanted solution for sodium hydroxide (Chakrabarty and Agarwal, 1989),out of 41 genotypes the response of 3 genotypes showed no colour change (Sindagi local, IET-21046 and Huggi bhatta), 14 genotypes were yellow and 14 genotypes showed light yellow in colour. In case of potassium hydroxide the cultivars of present study showed positive response to this test. They turned yellow colour solution. However, the intensity of colour varied among the varieties. Eight genotypes have shown no colour change, 14 genotypes were light yellow, 13 genotypes were dark yellow and six genotypes showed reddish brown in colour.

Potassium hydroxide and sodium hydroxide tests are useful to distinguish red seed varieties from white seed varieties, if the seed coat colour of red seeded varieties vanished due to unfavorable climate condition. Vanagamudi *et al*. (1988) reported that among 85 rice varieties, 71 varieties showed negative reaction to these chemicals and remaining varieties showed deep-wine red staining. Based on the change in colour of solution the genotypes were grouped into two groups *viz*., brown and bluish brown response to KI test. Among 41 genotypes, 16 genotypes were bluish brown group and 25 genotypes were reddish brown in colour.

Various biochemical tests are being used to reveal chemical differences among the seeds or seedlings of different genotypes. These tests have several important characteristics in common. They require virtually no technical expertise or training and can be completed in a relatively in short time. The results of these tests are usually distinct, easily interpreted and help in grouping of rice genotypes.

Table1: Grouping of the scented rice varieties based on biochemical tests

Sl. No.	Genotypes	Phenol test	Modified phenol test	NaOH	KOH Test	KI Test
1.	Ambemore	Brown	Brown	Yellow	Light Yellow	Bluish Brown
2.	Andra basmathi	No colour change	No colour change	Light Yellow	No colour change	Brown
3.	Badsha bhog	No colour change	No colour change	Yellow	Light Yellow	Brown
4.	Beeraga	No colour change	No colour change	Yellow	Dark Yellow	Brown
5.	Belgaum basmathi	Brown	Dark Brown	Light Yellow	Light Yellow	Bluish Brown
6.	Deharadhun basmathi	No colour change	No colour change	Light Yellow	Light Yellow	Bluish Brown
7.	Delhi basmathi	No colour change	Light Brown	Light Yellow	Dark Yellow	Brown
8.	Gandha sali	Light Brown	Brown	Light Yellow	No colour change	Brown
9.	Geregi sanna	No colour change	No colour change	Yellow	Light Yellow	Brown
10.	Huggi bhatta	Light Brown	No colour change	Light Yellow	No colour change	Brown
11.	Kagi sali	Dark Brown	Black	No colour change	Reddish Brown	Bluish Brown
12.	Kagi sanna	Dark Brown	Black	Light Yellow	Reddish Brown	Brown
13.	Kalanamak	No colour	Light Brown	Light Yellow	Dark Yellow	Bluish Brown

		change				
14.	Kari basmathi	Dark Brown	Black	Light Yellow	Reddish Brown	Brown
15.	Karigajivile	Brown	Dark Brown	Yellow	Reddish Brown	Brown
16.	Kolpa lathi	Dark Brown	Dark Brown	Yellow	Dark Yellow	Bluish Brown
17.	Kumuda	Light Brown	Light Brown	Light Yellow	Light Yellow	Brown
18.	Local ambemore	Light Brown	Brown	Light Yellow	Light Yellow	Brown
19.	Malgudi sanna	Light Brown	No colour change	Yellow	Light Yellow	Bluish Brown
20.	Mugadh sugandha	No colour change	Light Brown	Yellow	No colour change	Brown
21.	Parimala kalavi	Dark Brown	Brown	Light Yellow	Reddish Brown	Brown
22.	Sindagi local	Brown	Dark Brown	No colour change	Reddish Brown	Brown
23.	Sugandhi	Light Brown	Brown	Light Yellow	No colour change	Bluish Brown
24.	Vasane bhatta	No colour change	No colour change	Light Yellow	Light Yellow	Bluish Brown
25.	Yalakki sali	Brown	Dark Brown	Light Yellow	Light Yellow	Brown
26.	Pusa 1460	Brown	Light Brown	Yellow	Dark Yellow	Brown
27.	Pusa 44	Light Brown	Dark Brown	Light Yellow	Light Yellow	Bluish Brown
28.	Pusa basmathi-1	Light Brown	Light Brown	Light Yellow	Light Yellow	Brown
29.	Pusa sugandha-2	Light Brown	Light Brown	Yellow	Dark Yellow	Brown
30.	Pusa sugandha-3	Brown	Brown	Light Yellow	Light Yellow	Brown
31.	Pusa sugandha-4	Light Brown	Dark Brown	Yellow	Dark Yellow	Brown
32.	Pusa sugandha-5	Light Brown	Brown	Light Yellow	Dark Yellow	Bluish Brown
33.	IET-18393	Dark Brown	Black	Light Yellow	Dark Yellow	Brown
34.	IET-19228	Light Brown	Light Brown	Yellow	Dark Yellow	Brown
35.	IET-19236	No colour change	No colour change	Yellow	Dark Yellow	Bluish Brown
36.	IET-19713	Brown	Dark Brown	Light Yellow	No colour change	Bluish Brown
37.	IET-20472	Light Brown	Dark Brown	Yellow	No colour change	Bluish Brown
38.	IET-21042	No colour change	No colour change	Light Yellow	No colour change	Brown
39.	IET-21046	No colour change	Brown	No colour change	Dark Yellow	Bluish Brown
40.	IET-21053	Light Brown	Light Brown	Light Yellow	Dark Yellow	Bluish Brown
41.	IET-21058	Light Brown	Brown	Light Yellow	Light Yellow	Brown

References

Banerjee, S. K. and Chandra, R., (1977), Modified phenol test for the varietal identification of wheat seed. *Seed Sci. & Technol*., **5:** 53-60.

Bhowal, J.G., Banerjee, S. K. and Joshi, M.G., (1969), The evolution of phenol colour reaction gene in wheat. *Japan J. of Genet*., **44:** 123-128.

Chakrabarty, S.K. and Agrawal, R. L., (1989), Identification of blackgram varieties II. Utilization of morphological characteristics of seedlings. *Seed Res*., **17(2):**139- 142.

Devi Singh, Anuj Kumar, Navneet. Kumar, Vipin Kumar, Sachin Kumar, Amal Bhardwaj, Preeti Sirohi and S. Chand. (2011). Comparison of traditional physico-chemical methods and molecular marker assays for characterization of Basmati rice (Oryza sativa L.). African J. of Biotechnology. **10(62),** pp. 13390-13398.

Jaiswal, J.P. and Agrawal, R. L., (1995), Varietal purity determination in rice: modification of the phenol test. *Seed Sci. & Technol*., **23**: 33-42.

Vanangamudi, K., Palanisamy, V. and Natesan, P., (1988), Variety determination in rice by phenol and potassium hydroxide tests. *Seed Sci. & Technol.*, **16**: 465-470.