RESEARCH PAPER | Chemistry | Volume : 5 | Issue : 4 | April 2015 | ISSN - 2249-555X

Electrophoretic Patterns of Seed Proteins From Different Varieties of Rice As Influenced by Various Combinations of Plant Nutrients

KEYWORDS: Banding patterns; Electrophoresis; Fly ash; Protein contents; Rm values.

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ABSTRACT *Protein samples extracted from two varieties of rice namely Pooja and MTU-1001, grown in Kharif and Rabi seasons respectively, which were given nine different but identical fly ash, nitrogen, phosphorous and potassium (NPK) fertilizer and farm yard manure treatments were subjected to disc polyacrylamide gel electrophoresis (PAGE). It was observed that those treatments, which changed the protein contents of the varieties, also changed the banding patterns. Therefore, in case of both the varieties grown in different seasons the banding patterns of all the nine treatments were different. However, the increase or decrease in protein contents due to such treatments did not show any correlation with those.*

Introduction

Rice protein is a complex mixture of various types of proteins with different molecular weights. Through PAGE they can be separated to appear as different bands depending upon their mobilities, from which their molecular weights can be approximately ascertained. The band can be either major or minor depending upon the concentration of the protein of a given molecular weight forming it. Various research workers [1-5] have been able to ascertain the quality of protein and its soluble fractions of different varieties of rice by studying these bands. However, in our work we aimed at determining the changes, if any in the electrophoretic patterns of rice protein due to the effect of the application of various sources of soil nutrients like fly ash, fertilizer (NPK) and farm yard manure, known to influence the protein contents.

Materials and Methods

Rice varieties cv. Pooja and cv. MTU-1001 were grown in two different seasons Kharif and Rabi respectively in 2012 in the Regional Research and Technology Transfer Station of Orissa University of Agriculture and Technology, Bhubaneswar. Both of those were given nine different treatments consisting of various doses of fly ash (FA), NPK fertilizer and farm yard manure (FYM) with only NPK serving as the control. The treatments were: (I) Recommended Dose (RD) of N, P (P_2O_5), K (K_2O) @ 80, 40, 40 $kgha^{-1}$ (II) FA @ 20 tha^{-1} (III) FA @ 40 tha^{-1} (IV) RD + FA @ 20 tha^{-1} (V) RD +FA @ 40 tha^{-1} (VI) 50 % RD + FA @ 20 tha^{-1} (VII) 50 % RD+ FA @ 40 tha^{-1} (VIII) 50 % RD + FA @ 20 tha^{-1} + FYM @ 10 tha^{-1} (IX) 50 % RD + FA @ 40 tha^{-1} + FYM @ 10 tha^{-1}.

Extraction of protein and preparation of samples for gel electrophoresis were done essentially according to [6-7]. The solutions for the gel were also prepared as per the above authors. Then the gels were prepared as per Davis [8] and the electrophoresis, staining, destaining as well as determination of Rm values were made according to Parida [7]. Disc gel electrophoresis in polyacrylamide gel was carried out for all the above 18 samples.

Results and Discussion

A large number of bands often with very close Rm values were observed for different treatments (Fig. 1a and 1b). The bands were numbered from the slowest moving to the fastest moving arbitrarily taking into account the proximity of their Rm values as done by [9-10]. The Rm values of the major and minor bands found in different treatments are given in table-1. Table-2 shows the occurrence of different bands in each of the treatments. It is observed from these tables that nine treatments of the two varieties of rice grown in two different seasons that were tested had 3 to 7 major bands and 4 to 8 minor bands.

The distribution of major and minor bands in those are shown in Table-1. It suggests that the largest number of major bands (Sl. No. 21 to 31) have an intermediate mobility. There was a sharp decrease in the number of major bands when the Rm values were more than 0.59 (i.e., higher than Sl. No.31). The number of minor bands, however, increased when the Rm values were 0.67 or more.

A comparison of the banding patterns of the nine treatments of cv. Pooja grown in Kharif season shows that they are all different from one another. Similar is also the case with the cv. MTU-1001 grown in Rabi season. The changes are more glaring when the major bands were taken into consideration. As for example in Pooja variety grown in Kharif season treatment-1 (K-I) which received only N, P, K fertilizer had four major bands while treatment-2 (K-II) which received only 20 t/ha fly ash had only two. On the other hand treatment-3 (K-III) receiving 40 tha^{-1} fly ash had five major bands while treatment-4 (K-IV) and treatment-5 (K-V) had only four each. Treatment-6 (K-VI) had three closely spaced major bands while treatment-7 (K-VII) had the maximum number of eight major bands. Treatment-8 (K-VIII) had four and treatment-9 (K-IX) had five major bands. Although it seems that the treatments receiving more fly ash had more major bands, the relation is not quite true always. The minor bands in all the treatments were between ten to twelve which is only less in case of treatment-7 (K-VII).

In case of cv. MTU-1001 grown in Rabi season all the treat-

Volume : 5 | Issue : 4 | April 2015 | ISSN - 2249-555X

ments had four to five major bands and eight to ten minor bands. The effect of soil nutrients on the banding patterns does not appear to be significant in any way.

Varieties Pooja and MTU-1001 are not genetically related. Many workers [6-9] have suggested that the banding patterns have a genetic basis. Therefore, genetically related varieties show more or less similar banding patterns. However, our observations indicated that treatments of different soil nutrients, which changed the protein contents of the rice grains also change the banding patterns, but those were different for different varieties, which are genetically unrelated. Therefore, the increase or decrease in protein content did not appear to have any correlation with the banding patterns. Even the treatments in which there were considerably significant increase in protein contents were found to have less number of major bands in comparison to those in which it was not so significant viz., treatment-VIII and treatment-VII of both the varieties.

Table-1
Rm Values of Different Major and Minor Bands

Sl. No	Rm value	Major Band	Minor Band	Sl. No	Rm value	Major Band	Minor Band	Sl. No	Rm value	Major Band	Minor Band
1	2	3	4	1	2	3	4	1	2	3	4
1	0.05, 0.06	-	K-I, K-IV, R-VIII, R-IX	20	0.40	K-II, K-IV, K-VII	K-IX	39	0.70, 0.71	K-VII, R-II	K-VIII, R-VIII, K-IV, K-V, K-IX, R-III, R-VII
2	0.07, 0.08	-	K-V, K-VII, K-VIII, K-IX, R-II, R-V	21	0.42	R-VIII	-	40	0.72, 0.73, 0.74	R-VII	R-I, R-IV, K-II, R-VI, R-IX, K-III, K-V
3	0.10,0.11, 0.12	R-I, R-IV	K-II, K-III, K-VI, R-VI, K-I	22	0.43	K-V, K-VI, K-VIII, R-II, R-V, K-IX	K-I, R-III	41	0.76, 0.77, 0.78	-	K-II, K-III, K-IV, K-VI, K-VIII, R-I, R-V, K-I
4	0.13, 0.14	K-III, K-VII	K-VIII, R-III	23	0.44	R-IV	K-VII	42	0.80, 0.81	-	K-V, K-IX, R-III
5	0.15	R-VIII	R-I, R-VII	24	0.46, 0.47	K-III, K-VII	R-IV	43	0.82	-	R-I, R-VII, R-IX
6	0.16, 0.17	K-V, R-V, R-VI	K-VI, R-II	25	0.48	R-VII, R-VIII	-	44	0.83	R-II	R-III, R-IX, R-VII,
7	0.19, 0.20	K-IV	K-III, R-IV	26	0.49, 0.50	K-IV, K-V, K-VI, R-V	K-IX, R-II, R-III	45	0.86	-	K-I, K-II, K-VI, K-IX, R-IV, R-VI, R-VIII
8	0.21	R-VII, R-VIII, R-IX	-	27	0.51	K-VII, R-IX	K-I, K-II, K-V, R-VIII	46	0.89	-	K-VI
9	0.22, 0.23	K-III, K-V, K-VIII, R-III, R-IX	R-VI	28	0.52	R-I	-	47	0.91	-	R-II, R-VII
10	0.24	R-IX	R-III	29	0.53, 0.54	R-VI, K-III, K-VIII	K-IV, R-VII	48	0.92, 0.93	-	R-V
11	0.25	R-I	K-VIII	30	0.57	K-I, K-II, K-IX, R-II, R-III	K-V, K-VIII, R-V, R-VI, R-IX	49	0.94	-	R-II, R-IV, R-IX
12	0.27, 0.28	K-I, K-II, K-IV, R-IV, K-VII, R-VI, K-III, K-VIII, R-III	R-V, R-VIII	31	0.59	K-II, K-VII	K-III	50	0.97	-	R-V, R-VIII, R-IX
13	0.29, 0.30, 0.31	R-IV	K-V, K-VI, K-VII, K-IX, R-II	32	0.60	R-I	R-II				
14	0.32, 0.33	R-I, R-V	-	33	0.61	R-I	R-II				
15	0.34, 0.35	K-IX	K-VIII, K-I, K-II, K-III, R-I	34	0.62, 0.63	R-V	K-IV, K-VII, K-VIII, R-III, R-VI				

Volume : 5 | Issue : 4 | April 2015 | ISSN - 2249-555X

16	0.36	R-VII, R-VIII	-	35	0.64	-	R-VII				
17	0.37	R-III	K-VI, R-IV	36	0.65, 0.66	K-I, K-IX	K-V				
18	0.38	K-I, K-VI	-	37	0.67	R-IX	K-IX, K-I, K-II, K-IV				
19	0.39	R-IX		38	0.68, 0.69	-	K-I, K-II, K-IV, K-III, K-VI				

N.B: K –Kharif, R- Rabi, I to IX – Different treatments

Fig 1(a)
Bands of Kharif rice receiving nine treatments

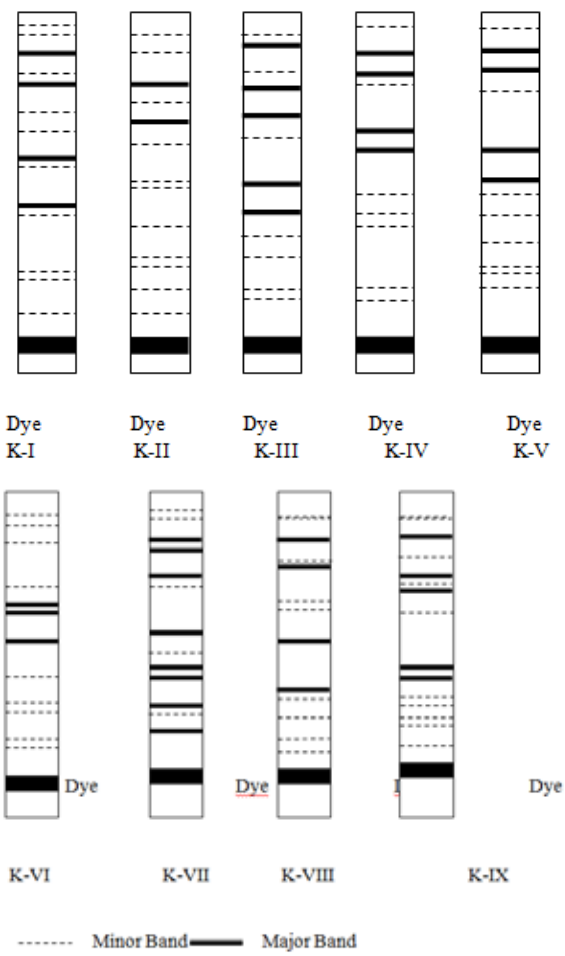
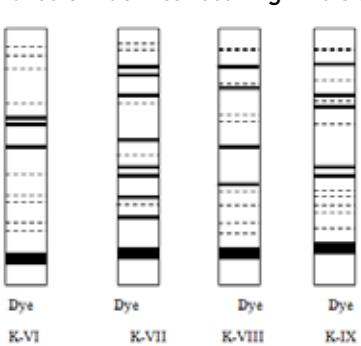


Fig 1(b)
Bands of Rabi rice receiving nine treatments



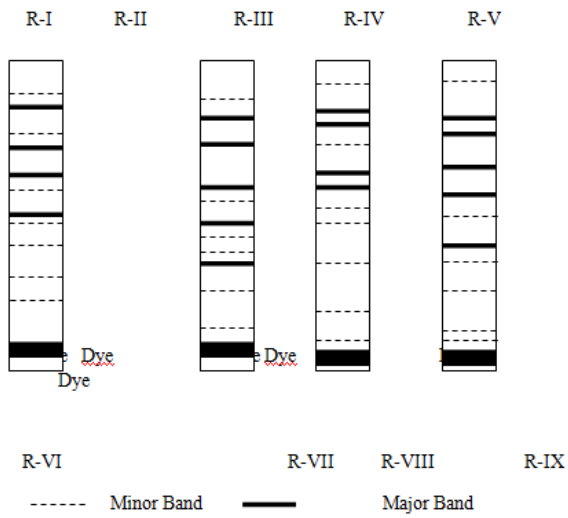


Table-2
The bands contained in different treatments

Treatments	Serial numbers of the major bands	Serial numbers of the minor bands
K-I	12, 18, 30, 36	1, 3, 15, 37, 38, 41, 42, 45
K-II	12, 20, 30, 31	3, 15, 37, 38, 40, 41, 42, 45
K-III	2, 4, 5, 7	1, 3, 6, 8, 9, 10, 11
K-IV	7, 12, 20, 26	1, 29, 34, 37, 38, 39, 41
K-V	6, 9, 22, 26	2, 13, 30, 36, 39, 40, 42
K-VI	18, 22, 26	3, 6, 13, 17, 38, 41, 45, 46
K-VII	4, 12, 20, 24, 27, 31, 39	2, 13, 23, 34
K-VIII	9, 12, 22, 29	2, 4, 11, 15, 30, 34, 39, 41
K-IX	9, 15, 22, 30, 36	2, 13, 20, 37, 39, 42, 45, 48
R-I	3, 11, 14, 28, 32	5, 15, 40, 41, 43
R-II	22, 30, 39, 44	2, 6, 13, 32, 47, 49
R-III	9, 12, 17, 30	4, 10, 34, 39, 41, 44
R-IV	3, 12, 13, 23	7, 17, 40, 45, 49
R-V	6, 14, 22, 26, 34	2, 12, 30, 41, 48, 50
R-VI	6, 12, 29	3, 9, 30, 34, 40, 45
R-VII	8, 16, 25, 33, 40	5, 29, 37, 39, 43, 44, 47
R-VIII	5, 8, 16, 21, 25	1, 12, 39, 45, 50
R-IX	8, 10, 19, 27, 37	1, 30, 40, 43, 44, 49, 50

Volume : 5 | Issue : 4 | April 2015 | ISSN - 2249-555X

N.B: K –Kharif, R- Rabi, I to IX – Different treatments

Summary and Conclusion

When the protein samples of two different rice varieties Pooja and MTU-1001 grown in Kharif and Rabi seasons respectively, to which nine different treatments of fly ash, fertilizer and farm yard manure were applied, were subjected to polyacrylamide gel electrophoresis, it was found that the banding patterns were different not only in the two genetically different varieties but also in the same variety. Although it appeared that the banding patterns are influenced by the treatments of various combinations of plant nutrients, those are not common for the similar treatments in different varieties. No correlations could also be found between those and increase or decrease in protein contents of the rice varieties.

Acknowledgement

The authors are obliged to the Orissa University of Agriculture and Technology, Bhubaneswar, India and the S'O'A University, Bhubaneswar, India for providing the necessary facilities to carry out the research work.

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