Short Communication



Detection of a protein marker for screening of MYMV resistant mungbean genotypes

Jagruti Pattnaik and C. Kole*

Laboratory of Molecular Biology & Biotechnology, O.U.A.T., Bhubaneswar 751 003

(Received: August, 2000; accepted: November 2001)

Mungbean Yellow Mosaic Virus (MYMV) is the most destructive disease in mungbean, *Vigna radiata* (L.) Wilczek, in the Indian subcontinent and adjacent countries of South East Asia [1]. It may cause yield losses even to the tune of 100%. Use of resistant genotypes is the most effective alternative to mitigate this yield loss.

Legume seeds contain 70% globulin, 15-20% albumin and 15-20% glutelin [2]. Association of seed proteins with host response to biotic stress in crop plants has already been reported [3, 4, 5]. Analysis of seed proteins using sodium dodecyl sulfatepolyacrylamide gel electrophoresis (SDS-PAGE) in mungbean has been employed for depiction of the seed protein profile [6] and characterization and identification of varieties [6, 7] and elucidation of evolution and phylogentic relationship [6, 8]. This approach could also be used for screening of MYMV resistant genotypes in this crop.

We report here on the detection of a seed protein marker to distinguish between MYMV resistant and susceptible genotypes in mungbean by electrophoretic analysis of seed albumins.

Materials and methods

Eleven genotypes of mungbean were used in this study. These included six MYMV resistant genotypes: LGG 460, Jhainmung, COGG 901, MGG 332, WGG 37 and PDM 84-146 and five MYMV susceptible genotypes: COBG 2, BSN 1, T 44, ML 5 and Kalamung. Their interaction phenotypes for MYMV infection were studied previously (data not shown).

Albumins were extracted from seed flour by suspending in prechilled distilled water for 4h at 0° C followed by centrifugation at 12000g at 0° C for 5 min and denatured with an equal volume of cracking buffer

[0.125M Tris. HCl (pH 6.8), 4% SDS, 20% Glycerol, 10% 2-Mercaptoethanol and 0.01% Bromophenol Blue] by boiling at 100^oC in a waterbath for 1 min [7]. Electrophoretic analysis was done in a 12% SDS-PAGE [9] at 20mA.

SDS-PAGE of seed albumins of the 11 mungbean genotypes resulted in 12 polypeptide bands of diverse molecular weights, Rm values ranging from 0.043 to 0.507 (Table 1). Out of these, six bands (Rm 0.043, 0.130, 0.159, 0.202, 0.289 and 0.434) varied for their expression and one of these with Rm value 0.202 (Fig. 1) was present only in the susceptible genotypes.



Fig. 1. Differences in seed albumin banding patterns of MYMV resistant (Lane 1, var. COGG 901) and MYMV susceptible (Lane 2, var. T 44) genotypes of mungbean; Rm value of the polypeptide band expressed only in the susceptible genotypes on the right

^{*}Present address: Department of Genetics and Biotechnology, Allahabad Agricultural Institute, Allahabad 211 007

Band #	Rm value	MYMV resistant genotypes						MYMV susceptible genotypes				
		LGG 460	Jhainmung	COGG 901	MGG 332	WGG 37	PDM 84-146	COBG 2	BSN 1	T 44	ML 5	Kalamung
1	0.043	+	+	-	-	-	-	-	+	+	+	-
2	0.086	+	+	+	+	+	+	-	+	+	+	+
3	0.101	+	+	+	+	+	+	+	+	+	+	+
4	0.130	+	-	+	+	+	+	+	+	+	+	+
5	0.159	-	-	-	-	+	+	+	+	+	+	+
6	0.202	-	-	-	-	-	-	+	+	+	+	+
7	0.260	+	+	+	+	+	+	+	+	+	+	+
8	0.289	+	-	+	-	-	-	+	+	+	+	-
9	0.376	+	+	+	+	+	+	+	+	+	+	+
10	0.434	+	+	+	-	-	- :	+	-	-	-	-
11	0.463	+	+	+	+	+	+	+	+	+	+	+
12	0.507	+	+	+	· +	+	+	+	+	+	+	+

Table 1. Polypeptide banding patterns of 12 mungbean genotypes derived from SDS-PAGE of seed albumins

Seed proteins are known to be controlled by multigene families [10]. Their expression, however, is monogenically controlled with codominance of molecular weight variants and presence of a band being dominant over absence [6]. Deletion or mutation in regulatory and/or structural genes may lead to failure of protein expression [11]. In the present study, a polypeptide band was not expressed in the resistant genotypes. This band can be used in indirect screening of MYMV resistant genotypes. Such differentially expressed polypeptides present only in the susceptible genotypes were detected for indirect screening of rice genotypes resistant to green leafhopper also [5].

The physiological implication of the polypeptide expressed only in the susceptible genotypes in the present study and the strength of linkage of the loci controlling this polypeptide and MYMV susceptibility is not known. Works on linkage analysis in F_2 population(s) segrergating for expression of the polypeptide and host response to MYMV infection, and biochemical characterization of the differentially expressed polypeptide are in progress.

Acknowledgements

Authors are thankful to the Indian Council of Agricultural Research, New Delhi for financial assistance in the form of an ad-hoc project.

References

- 1. **Poehlman J. M.** 1991. The Mungbean. Oxford & IBH Publ. Co., New Delhi, India.
- 2. Aykroyd W. R., Doughty J. and Walker A. 1982. Legumes in Human Nutrition, FAO, Rome, Italy.

- Osborn T. C., Blake T., Gepts P. and Bliss F. A. 1986. Bean arcelin 2: Genetic variation, inheritance and linkage relationship of a novel seed protein of *Phaseolus vulgaris*. Theor. Appl. Genet., **71**: 847-855.
- Prasad D. T., Umapathy N. S. and Veeranna R. 1996. Genotypic variation in cowpea (*Vigna unguiculata*) cultivars in relation to insect resistance. J. Plant Biochem. Biotech., 5: 47-49.
- Padmavathi G., Kole C. and Siddiq E. A. 1999. Detection of protein markers for identification of rice genotypes resistant to green leafhopper. Indian J. Genet., 59: 417-421.
- Naik B. S. 1998. Genetic Characterization of Cultivars and Seed Protein in Mungbean, Unpub. Ph.D. Thesis, Utkal University, Bhubaneswar.
- Mohanty J. B. 1997. Electrophoretic Approach of Genotype Characterization of Mungbean, Unpub. M.Sc. (Ag.) Thesis, Orissa University of Agriculture and Technology, Bhubaneswar.
- Kole C., Panigrahi J. and Patnaik A. 2000. Vigna glabrescens is a natural polyploid of V. radiata and V. umbellata: An evidence from seed albumin electrophoresis (Abstract). In: National Symposium on Biotechnology for Sustainability in Agriculture, G. B. Pant University of Agriculture and Technology, Pantnagar, India, pp. 129.
- Laemmli U. K. 1970. Cleavage of structural protein during the assembly of the head of bacteriophage T4. Nature, 227: 680-685.
- de Lumen B. O. 1990. Molecular approaches to improving the nutritional and functional properties of plant seeds as food sources: Development and comments. J. Agril. Food Chem., 38: 1779-1788.
- Brown J. B. S., Osborn T. C., Bliss F. A. and Hall T. C. 1981. Genetic variation in the subunits of globulin-2 and albumin seed proteins of French bean. Theor. Appl. Genet., 60: 245-250.