

Molecular Breeding for Yellow Mosaic Disease in Mungbean



Biotechnology

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Introduction

Mungbean is the third largest pulse crop in India occupying 3.55 million hectare with a production of 1.40 million tons (Anonymous, 2013). One of the major destructive diseases in mungbean causing great yield loss is Yellow mosaic disease (Yellow Mosaic Disease) caused by different species of white-fly transmitted geminivirus belonging to genus begomovirus and family geminiviridae such as Mungbean yellow mosaic virus (MYMV), Mungbean yellow mosaic India virus (MYMIV), Horsegram yellow mosaic virus and Dolichos yellow mosaic virus. First reported in mungbean in India by Nariani, 1960. Main pathogens causing YMD in mungbean in India are MYMV and MYMIV. Host species and susceptibility of each plant affect the development of disease symptoms after virus infection. The considerable and significant reduction in yield due to infestation of this disease necessitates us to develop MYMV tolerant mungbean cultivar. Now a day's molecular markers are playing very important role in isolation of disease resistant gene and development of resistant cultivar. Conventional breeding is time consuming and less efficient in compare to marker assisted selection. Thus identification of MYMV disease resistance gene and molecular markers linked with MYMV tolerance will contribute a lot to achieve success in dealing with MYMV disease in mungbean.

Yellow mosaic disease

YMD is one of the major diseases of mungbean in India caused by mungbean yellow mosaic virus resulting irregular green and yellow patches in older leaves and complete yellowing of younger leaves. Higher incidence of disease have been observed during spring and rainy season due to favorable condition for multiplication of the vector *Bemisia tabaci* in spring and rainy seasons (Singh and Gurha, 1994). Reports on losses due to disease revealed reduction of 9.6 to 38.2 % in height, 7 to 28.5 % in fresh weight of shoot and 4.3 to 22.1 % in dry weight, 25.7 % in 1000 seed weight of susceptible cultivar (Premchand and Varma, 1983) and yield loss of 83.9 % and a maximum growth reduction of 62.94% in MYMV infected cultivar (Quaiser Ahmed, 1991). Diseased plant usually matures late. There is a positive correlation of whitefly population on 20 and 30 day crop and disease incidence at 45 days old crop with maximum temperatures (Murgesan and Chelliah, 1977).

Mungbean Yellow Mosaic Virus

The causal agent of yellow mosaic disease belongs to the largest and the most important genus begomovirus within the family Geminiviridae (Bos, 1999). The virus has geminate particle morphology (20 x 30 nm) and the coat protein encapsulates spherical, single stranded DNA genome of approximately 2.8 Kb have either a monopartite (a single DNA) or a bipartite (with two DNA

components: DNA-A and DNA-B) genome Organization (Hull, 2004). More ever the YMV disease in legumes is caused by begomoviruses with bipartite genomes (Karthikeyan *et al.* 2004). Successful infection and transmission of this virus occurs by a whitefly *Bemisia tabaci* (Nariani, 1960). Successful transmission of MYMV by grafting was reported by several researchers Nariani (1960); Ahmed and Harwood (1973); Debrot and Ordosgoiti, 1975; Muniyappa, 1980; Varma, 1963; Sudhakar Rao *et al.*, 1980; Singh *et al.*, 1971; Suteri, 1974.

Mode of inheritance of resistance to mungbean yellow mosaic virus (MYMV) in mungbean

Knowledge of the mode of inheritance to MYMV in mungbean is useful for incorporation of resistance into agronomically poor, but desirable genetic resources. There are only a few contrasting published reports on the inheritance of resistance to MYMV in mungbean. The monogenic recessive inheritance for MYMV has been reported by Singh and Patel (1977), Malik *et al.* (1986 & 1988). Resistance to YMD in mungbean was reported to be controlled by a single major recessive gene with modifiers (Malik *et al.* 1986, Thakur *et al.* 1997), or two recessive genes (Dhole and Reddy 2012) or complementary recessive genes (Shukla and Pandya, 1985).

Development of YMV-tolerant mungbean variety

Development of YMV-tolerant variety is of prime importance for stabilizing the yield levels. Introduction of YMV-tolerant pulses in the farmer's field will circumvent the problems associated with the residual effects of the insecticides/ pesticides in the soil. Identification of resistant lines through conventional breeding is time consuming and requires evaluation at 'hot spot' area (Selvi *et al.* 2006). Thus, indirect selection using molecular markers linked to MYMV resistant genes would facilitate precision plant breeding and high-throughput marker assisted selection (MAS) of resistant genotypes. An attempt should made to develop molecular marker(s) linked with the YMV-tolerance in *Vigna radiata*. The marker will be of use in marker-assisted selection and will hopefully aid in the development of resistant cultivars in relatively shorter time spans. In a study by Chattopadhyay *et al.* (2008), mungbean genotypes were screened for MYMV resistance using specific primer sequence derived from conserved regions of resistance (R) gene. Utilisazation of in silico methods for the development of trait linked molecular markers in mungbean has been done in a study by Singh *et al.* (2013). Hence, an ideal strategy would be to search for the DNA marker *per se* linked with the resistance to all the above said diseases, unlike the selection for resistance genes on phenotypic scale which is also a time consuming process. Screening of resistance genes with molecular markers is important for identification of gene-based markers which can further fasten the breeding programme. Basak *et al.*

(2004) screened the *Vigna mungo* genotypes showing segregation for YMV symptoms with 24 sets of soybean 'R' gene-based RGA markers and found only one pair which differentiated the resistant from the susceptible ones.

Molecular Markers and QTL linked with MYMV in mungbean

Inheritance study MYMV resistance gene has revealed that different sources may have different MYMV resistance gene, thus markers identified using one source of MYMV resistance gene may not work for another source. Resistance gene analog (RGA) markers YR4 and CYR1, were associated with resistance to MYMV in blackgram (*Vigna mungo* (L.) Hepper) reported by Maiti *et al.* (2011). CYR1 is proposed as part of the candidate disease resistance (R) gene (Maiti *et al.* 2011). Recently, the R gene CYR1 was fully isolated from blackgram (Maiti *et al.* 2012). Dhole and Reddy (2013) reported that SCAR marker MYMVR-583 is associated with MYMV resistance in mungbean. This marker is 6.8 cM from the resistance gene. Gupta *et al.* (2013) reported that CEDG180 is linked to MYMV resistance in blackgram at a distance of 12.9 cM. Chen *et al.* (2013) identified four major QTLs on three different linkage groups for MYMV resistance using AFLP and SSR markers. Kitsanachandee *et al.* 2013, identified three QTLs for the MYMV in India, the QTLs were each on LGs 2, 4 and 9A and were designated as qMYMIV1, qMYMIV2 and qMYMIV3, respectively and QTL analysis by single marker analysis revealed that only one marker cp02662 on LG2 was associated with MYMV resistance in India. qMYMIV1 was located between markers CEDG100 and cp02662, showing an LOD score of 2.62, which was significant at $P = 0.05$. qMYMIV2 was localized between markers DMB-SSR008 and VR113. qMYMIV3 was located between markers CEDG166 and CEDG304. They also reported detection of Two QTLs, qMYMIV4 and qMYMIV5 by CIM(composite interval mapping) for disease resistance in Pakistan. qMYMIV5 was detected on LG6 between markers CEDG121 and CEDG191. Chen *et al.* (2013) identified SSR marker DMB-SSR158 linked to major QTL controlling MYMV resistance MYMIVr9_25 and according to the mungbean genetic map reported by Kajonphol *et al.* (2012), it was on LG2 between markers CEDG100 and CEDG108. Therefore,

MYMIVr9_25, qMYMIV1 and qMYMIV4 are common QTLs conferring MYMV resistance. This locus was detected in different locations and years, with different sources of resistance and scoring systems; thus, it is highly robust and can be used in marker-assisted selection. DMBSSR151, which is linked to MYMIVr8_48.8, a QTL controlling MYMV identified by Chen *et al.* (2013), was mapped to LG5 of map reported by Kitsanachandee 2013, but no QTL was detected in this genome region in their report. Based on the linkage map of mungbean reported by Isemura *et al.* (2012), CEDG180 is mapped to LG10. Basak *et al.* 2004 reported development of a RGA-based polymorphic marker, 'VMYRI', linked with YMV- resistance, the sequence of the marker and the predicted amino acid sequence indicate that 'VMYRI' is a part of a candidate gene for YMV-resistance and its sequence of 445bp DNA fragment produced by RGA 1F-CG/RGA 1R only from homozygous tolerant and the heterozygous lines.

Constraints & Conclusion

Lack of enough genetic diversity in the parental material used and narrow genetic base resulting in inherently low yield potential. The disease incidence is seasonal and cannot be created as and when desired by artificial means. Mungbean is one of the important pulse crops of India, and YMD is major production constraint limiting mungbean yield considerably. Therefore, there is an urgent need to develop high-yielding varieties with resistance to diseases with greater yield stability. Using conventional methods, selection of the YMD-resistant genotypes depend on the field screening for more than one year or season. Further, the inconsistencies in scoring disease reaction may affect the introgression of YMD resistance into elite mungbean genotypes. Hence, the use of molecular markers for identifying resistance gene is particularly important as it fastens breeding programme aimed at introgression of resistance gene associated with phenotypic analysis. Development of tightly linked markers endowed with the features of resistance gene candidates, may be useful for generating superior genotypes with durable YMV-resistance

REFERENCE

- Anonymous. (2013). Foodgrains Production in 2013-14. <http://www.icar.org.in/>.
- Ahmed, M. & Harwood, R. F. (1973). Studies on whitefly transmitted yellow mosaic of urd bean (*Phaseolus mungo*). *Plant Disease Reporter*, 57: 800-802.
- Basak, J., Kundagrami, S., Ghose, T. K & Pal, A. (2004). Development of Yellow Mosaic Virus (YMV) resistance linked DNA marker in *Vigna mungo* from populations segregating for YMV-reaction. *Molecular Breeding* 14:375-383.
- Bos, L. (1999). *Plant Viruses: Unique and Intriguing Pathogens: A Text Book of Plant Virology*. Backhuys Publishers, the Netherlands, 305-306.
- Chattopadhyay, K., Bhattacharyya, S., Mandal, N. & Sarkar, H. K. (2008). PCR-Based Characterization of Mungbean (*Vigna radiata*) genotypes from Indian Subcontinent in Intra- and Inter-Specific Level. *Indian Journal of Plant Biochemistry and Biotechnology* 17: 141-148.
- Chen, H. M., Ku, H. S., Schafleitner, R., Bains, T. S., Kuo, G. C., Liu, C. A. & Nair, R.M. (2013). The major quantitative trait locus for mungbean yellow mosaic Indian virus resistance is tightly linked in repulsion phase to the major bruchid resistance locus in a cross between mungbean [*Vigna radiata* (L.) Wilczek] and its wild relative *Vigna radiata* sub sp. *sublobata*. *Euphytica* 192: 205-216.
- Debrot, C.E.A. & Ordosgoitti, F.A. (1975). Studies on a yellow mosaic of soybean in Venezuela. *Agronomy Tropics*, 25: 435-449.
- Dhole, V. J., Reddy, K. (2012). Genetic analysis of resistance to mungbean yellow mosaic virus in mungbean (*Vigna radiata*). *Plant Breeding* 131:414-417.
- Dhole, V. J. & Reddy, K. S. (2013). Development of a SCAR marker linked with a MYMV resistance gene in mungbean (*Vigna radiata*). *Plant Breeding* 132: 127-132.
- Gupta, S., Gupta, D. S., Anjum, T. K., Pratap, A. & Kumar, J. (2013). Inheritance and molecular tagging of MYMV resistance gene in blackgram (*Vigna mungo* L. Hepper). *Euphytica* 193: 27-37.
- Hull, R. (2004). *Mathew's Plant Virology*, Fourth Edition. Elsevier Publishers, India. pp.180-182.
- Isemura, T., Kaga, A., Tabata, S., Somta, P, Srinives P, Shimizu T, Jo U, Vaughan DA and Tomooka N (2012) Construction of a genetic linkage map and genetic analysis of domestication related traits in mungbean (*Vigna radiata*). *PLoS One* 7(8): e41304.
- Kajonphol, T., Sangsiri, C., Somta, P., Toojinda, T. & Srinives, P. (2012). SSR map construction and quantitative trait loci (QTL) identification of major agronomic traits in mungbean (*Vigna radiata* (L.) Wilczek). *SABRAO Journal of Breeding Genetics* 44: 71-86.
- Karthikeyan, A. S., Vanitharani, R., Balaji, V., Anuradha S., Thillaichidambaram, P., Shivaprasad, P. V., Parameswari, C., Balamani, V., Saminathan, M. & Veluthambi, K. (2004). Analysis of an isolate of Mungbean yellow mosaic virus (MYMV) with a highly variable DNA B component. *Arch. Virol.* 149: 1643-1652.
- Kitsanachandee, R., Somta, P., Chatchawankanphanich, O., Akhtar, K. P., Shah, T. M., Nair, R. M., Bains, T. S., Sirari, A., Kaur, L. & Srinives, P. (2013). Detection of quantitative trait loci for mungbean yellow mosaic India virus (MYMIV) resistance in mungbean (*Vigna radiata* (L.) Wilczek) in India and Pakistan. *Breeding Science* 63: 367-373.
- Maiti, S., Basak, J., Kundagrami, S., Kundu, A. & Pal, A. (2011). Molecular marker-assisted genotyping of mungbean yellow mosaic India virus resistant germplasms of mungbean and urdbean. *Molecular Biotechnology* 47:95-104.
- Maiti, S., Paul, S. & Pal, A. (2012). Isolation, characterization, and structure analysis of a non-TIR-NBS-LRR encoding candidate gene from MYMV-resistant *Vigna mungo*. *Molecular Biotechnology* 52: 217-233.
- Malik, I. A., Sarwar, G. & Ali, Y. (1986). Genetic studies in mungbean (*Vigna radiata* (L.) Wilczek). *Inheritance of tolerance to mungbean Yellow Mosaic Virus and some morphological characters*. *Pak. J. Botany* 18(2): 189 - 198.
- Malik, I. A., Ali, Y. & Saleem, M. (1988). Incorporation of tolerance to Mungbean Yellow Mosaic Virus from local germplasm into exotic large seeded mungbean. *PP* 297 - 307. In: S. Shanmugasundaram (ed.). *Proc. Second Int'l. Symp. on mungbean*. AVRDC Bangkok, Thailand. | 20. Muniyappa, V. (1980). Whiteflies. In: *Vectors of Plant Pathogens*, Eds. Harris K. F. and Maramorosch K., Academic Press, New York, pp.39-85.
- Murugesan, S. & Chelliah, S. (1977). Influence of sowing time on the incidence of the vector *Bemisia tabaci* (Genn.) and the yellow mosaic disease of greengram. *Mad. Agri. Jour.* 64(2): 128-130.
- Nariani, T.K. (1960). Yellow Mosaic of mung (*Phaseolus aureus* L.). *Ind. Phytopathol.* 13: 24 - 29.
- Premchand and Varma. J. P. (1983). Effect of yellow mosaic on growth components and yield of mungbean and urdbean. *Haryana Agricultural University Journal of Research* 13(1): 98-102.
- Quaiser Ahmed. (1991). Growth attributes and grain yield of mungbean plants affected by mungbean yellow mosaic virus in field. *Ind. Phytopathol.*, 43(4): 559-560.
- Singh, R.A. & Gurha, S.N. (1994). Influence of cropping seasons on the incidence of yellow mosaic disease in mungbean genotypes. *Indian Journal of Pulses Research*, 7(12): 206-208.
- Selvi, R., Muthiah, A. R., Manivannan, N. (2006). Tagging of RAPD marker for MYMV resistance in mungbean (*Vigna radiata* (L.) Wilczek). *Asian J Plant Sci* 5:277-280.
- Shukla, G. P. & Pandya, B. P. (1985). Resistance to Yellow Mosaic in Greengram. *SABRAO Journal*, Vol. 17, No. 3, pp. 165-171.
- Singh, H., Sandhu, G. S., & Mavi, G. S. (1971). Control of yellow mosaic virus in soybean *Glycine max* (L.) Merrill by the use of granular insecticides. *Indian Journal of Entomology*, 33: 272-278.
- Singh, N., Singh, H., & Nagarajan, P. (2013). Development of SSR markers in mungbean, *Vigna radiata* (L.) Wilczek using in silico methods. *Journal of Crop and Weed* 9(1):69-74.
- Sudhakar, R. A., Prasad, R., R. D. V. J. & Reddy, P. S. (1980). Whitefly transmitted yellow mosaic disease of groundnut (*Arachis hypogaea* L.). *Current Science* 49: 160.
- Suteri, B. D. (1974). Occurrence of soybean yellow mosaic in Uttar Pradesh. *Current science*, 43: 689-690.
- Thakur, R. P., Patel P. N. & Verma, J. P. (1997). Genetical Relationship between Reaction to Bacterial Leaf Spot, Yellow Mosaic and Cercospora Leaf Spot Diseases in Mungbean (*Vigna radiata* L. Wilczek). *Euphytica*, Vol. 26, No. 3, 1977, pp. 765-774.
- Varma, P. M. (1963). Transmission of plant viruses by whiteflies. *Bulletin of National Institute of Science, India*, 24: 11-23.