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RESEARCH PAPER

Genetic resource development for mungbean yellow mosaic virus (MYMV) resistance in mungbean [*Vigna radiata* (L.) Wilczek]

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Abstract: Mungbean [*Vigna radiata* (L.) Wilczek] is an important short duration grain legume which is grown in different parts of the country, for grain and green manure. It is an outstanding source of easily digestible proteins with low flatulence, which balances the staple rice diet in Asia. Mungbean yellow mosaic disease (MYMD) caused by whitefly (Bemisia tabaci) transmitted mungbean yellow mosaic virus (MYMV) is an important constraint of mungbean cultivation in India. The current investigation was taken up to develop MYMV resistant genotypes through pedigree method of breeding. Eighty-eight F_3 progenies were evaluated for their reaction to MYMV under natural field conditions during summer 2018-2019 with infector rows grown after every ten F_3 progenies of test entries. On screening the F_3 progenies for reaction to MYMV the percent disease incidence varied from 5 to 92.3%. The lines DGGV-200, DGGV-218, DGGV-281 and DGGV-284 were observed to be resistant and DGGV-198, DGGV-206, DGGV-212, DGGV-215, DGGV-226,DGGV-268 and DGGV-282 were moderately resistant.

Key words: Disease, Mungbean, Resistance, Screening

Introduction

Green gram [*Vigna radiata* (L.) Wilczek], also known as mungbean, mungu, moong, pachai payaru, golden bean and green soy. It is an important pulse crop belong to family Fabaceae, sub family papilionoideae, genus Vigna. It has diploid chromosome number 2n=2x=22 (Karpenchenko,1925). It is from Indo-Burma region of Hindustan (Vavilov, 1926). In India, green gram is extensively cultivated in *kharif* season in Madhya Pradesh, Uttar Pradesh, Andhra Pradesh, Odisha, Punjab, Maharashtra, Rajasthan, Karnataka, and Tamil Nadu. Whereas, in Tamil Nadu, Bihar, Orissa, Andhra Pradesh, Uttar Pradesh, West Bengal, Maharashtra, Karnataka states it is grown as rabi crop.In India total area sown under *kharif* Mungbean in 2017-18 is 31.81 lakh ha and it shows reduction of 7.90% area as compare to 2016-17, and area sown under *rabi* greengram in 2017-18 is 2.97 lakh ha and it shows reduction of 18.17% area as compared to 2016- 17 (Anon., 2016). Production is 19.01 lakh tonnes and productivity is 467 kg ha^{-1} (Anon., 2019). The six states of Madhya Pradesh, Rajasthan, Maharashtra, Karnataka, Uttar Pradesh and Andhra Pradesh together account for more than 80 per cent of the total area under pulses. Only 16 per cent of the total area under pulses was irrigated. The states with higher percentage of irrigated area under pulses were Andhra Pradesh, Madhya Pradesh, Uttar Pradesh and Karnataka. Coincidentally, these three states have the highest share of area and production under pulses.

In Karnataka the total greengram area is 4 lakh hectares, production is 1.1 lakh tonnes and the productivity is 389 kg ha^{-1} (Anon., 2019). The major pulse growing districts in Karnataka are Gadag, Kalaburagi, Vijayapura, Bagalakot, Belagavi, Bidar, Raichur, Dharwad and Mysuru (Shiraganvi, 2019).

In India green gram is grown in poor fertile soils where other crops cannot be grown and growing in soils with less water profile, but it needs high soil moisture (Kumar *et al.,* 2010). The constraints for achieving higher yield are inherently low genetic yield potentiality, pod shattering, indeterminate growth, unproductive plant types, asynchronous maturity, low harvest index, low partitioning efficiency, small seed size and susceptibility to biotic and abiotic stresses (Mogali and Hegde, 2020). Since, green gram is being grown under marginal conditions of moisture stress and less fertile lands, natural selection played a major role in determining the plant type and other characteristics of this crop than human selection even long after the crop domestication.

Diseases are the major problem for green gram cultivation which has adverse effect on both quality and quantity of product. It suffers from many diseases caused by fungi, bacteria, viruses, nematodes, and also abiotic stresses. Among these, foliar diseases such as powdery mildew, anthracnose, cercospora leafspot and MYMV are more prevalent.

Among the major diseases that cause significant yield loss in mungbean, yellow mosaic disease is the most devastating disease especially in south Asian countries and seeks the attention of the scientists towards it. Within a short interval of time it has the ability to spread over a large crop field. This is one of the major constraints in summer green gram production. MYMV infects majority of leguminous crops *viz.,* cowpea, black gram and soybean *etc.* This was found to be most severe disease in the countries *viz.,* Nepal, Srilanka, India, Pakistan and some parts of south-eastern areas (Subedi *et al*., 2014). Due to reduced rainfall, the number of transmitters is very high, hence the incidence of MYMV is very high in summer season. The MYMV infestation will increase as crop grows to different developmental stages *viz.*, first trifoliate leaf stage (08-12%), second trifoliate leaf stage (16-20 %), flowering stage (41-55 %) and maturity stage (65-76 %) (Pawar and Mahatma, 2013).

The virus, Mungbean Yellow Mosaic Virus (MYMV) is the cause for the disease termed Mungbean Yellow vein Mosaic disease. It belongs to the family of *Geminiviridae* and genus *Begomovirus*. MYMV was identified for the first time in 1955 in 1955 (Karthikeyan *et al.,* 2014). It is having single stranded DNA with the genome size of 2.8 kb (Hull, 2004). By using degenerate primers, through PCR and gel-electrophoresis study, 700 bp of DNA-A molecule of Mungbean Yellow Mosaic Virus can be observed. This group of viruses was also termed as "Legumoviruses" (Nair *et al*., 2017). MYMV virus particles were first observed in plant cells of green gram in 1981 by Thongmeearkom and co-workers.

The symptoms first appear as minute golden yellow patches on affected trifoliate and is concentrated severely to the periphery of the leaf veins. Infected leaves of green gram showed irregular and mixed pattern of green and yellow colors, even the leave veins turn into green and yellow patches. The entire leaves turn into yellow colour as the plant get older and it looks like a senesced leaf. There are a fewer number of flowers and pods in the infected plant. In case of severe infestation, the entire pod turns yellow and the seeds in the pods get reduced. Seeds showed shriveled coat texture with mosaic yellow and green patches (Subedi *et al*., 2014). The symptoms are recorded on almost all aerial parts of an affected plant *viz*., stem, petiole, seeds and pods.

The bipartite begomaviruses are transmitted by special group of sucking insects called white flies (*Bemisia tabaci*, Gennadius), belongs to the older hemiptera. They showed circulative peristant manner of virus transmission, where viruses are transmitted through insect stylets (Czosnek *et al*., 2017). Some strains of MYMV showed mechanical transmission in Thailand (Ahmad *et al*., 2017).

Green gram is grown in all the seasons of the year (*kharif, rabi* and *summer*). So, the cropping season highly influences the pest population. Whitefly population density is high during summer season due to high temperature. They survive better in hot and humid weather conditions. After 20 to 30 days of sowing, the insect population is observed to be very high (Karthikeyan *et al*., 2014). *Kharif* and *rabi* seasons are unfavourable periods for pest multiplication.

When MYMV infestation occurs early in the plant population, there is higher yield reduction. If the population gets infested three weeks after seeding, then leads to yield loss up to 100 per cent (Karthikeyan *et al*., 2014). If the infestation occurs eight weeks after seeding, then the yield loss would be lesser. In a study conducted by Sudha *et al*. (2013) under field condition during *rabi*-2006 among 78 green gram genotypes, 28 genotypes have shown resistance response against MYMV disease. For the resistance confirmation, 28 genotypes were again sown during *summer*-2007 and agro-innoculation of MYMV was done by using 2 MYMV strains. Results revealed that only 3 genotypes showed resistance to the VA 221 strain while, 77 genotypes were indicated as susceptible to strain VA 239. One genotype recorded resistance reaction to strain VA 239, but it was susceptible to another strain VA 221.

Material and methods

DGGV-2 is a high yielding green gram varity, susceptible to MYMV was crossed with WGG-42 which is resistant to MYMV. The cross was generated during 2017 and the F_1 was advanced during 2018. True F_1 plants were identified and F_2 seeds were harvested in bulk. Each of the F_2 seeds were sown and advanced to F_3 generation. The present investigation was carried out with eighty-eight F_3 families derived by selfing F_2 plants (DGGV-2 x WGG-42) were sown in summer 2019-20 and were evaluated for yield and reaction to MYMV in comparison with checks *viz*. DGGV-2, WGG-42, IPM-2-14, DGG-7 under natural field condition. The screening was conducted in Main Agricultural Research Station (MARS), AICRP on MULLaRP, UAS, Dharwad under natural field condition. The experiment was laid out in augmented design. Each row length was 4m with plant to plant spacing 10 cm and row to row spacing 30 cm. Each F_3 family was sown along with their resistant and susceptible checks. The observations were recorded at regular intervals. All the agronomical practices were followed to raise a good crop except spraying of insecticides to foster whitefly population.

The field was regularly monitored to observe the symptoms of MYMV under natural field condition. After first and second week of sowing there was no symptom of MYMV. After third week onwards the symptoms started in some susceptible lines and then it was spread very quickly. The per cent disease incidence was calculated by using the formulae given by Basir *et al.* (2005). The disease incidence was observed on 7th, 8th and 9th week after sowing. Based on the per cent disease incidence, the populations were categorized into different groups based on 0-5 scale given by Basir *et al*. (2005).

Per cent disease incidence

The Per cent Disease Incidence was calculated by using the following formula given by Basir *et al*. (2005).

$$\text{Per cent disease incidence} = \frac{\text{Total number of plants infected in a row}}{\text{Total number of plants in a row}}$$

Results and discussion

The eighty-eight breeding lines (F_3) of green gram were screened for MYMV under natural field condition during *summer*-2019 season. Different breeding lines had shown differential reaction with their varied disease symptoms. No disease symptoms were observed on any of the breeding lines till the crop was three weeks old. The symptoms of MYMV disease started appearing on the leaves of young plants of susceptible breeding lines which became more prominent with time. After three weeks of sowing, the susceptible genotypes started showing symptoms of MYMV, which progressed with time up to seventh week. Most of the genotypes exhibited these symptoms with relatively similar intensity up to ninth week. Hence recording per cent disease incidence value between seventh to ninth weeks after sowing will be more reliable than computing mean per cent disease incidence value at intervals of V, VII and IX week.

However, the disease incidence was recorded on ninth week after sowing to determine the per cent disease incidence based on the disease scoring scale given by Basir *et al*. (2005). Hence, precise disease reaction was attributed to different breeding lines.

Among eighty-eight genotypes derived from the cross DGGV-2 × WGG-42 (F_3) some progenies have shown highly resistant, resistant and moderately resistant reaction to MYMV (Table 2).

The advanced breeding line DGGV-227 showed highly resistant reaction with per cent disease incidence value of 4 per cent. The lines DGGV-200, DGGV-218, DGGV-281 and DGGV-284 were observed to be resistant with the least per cent disease incidence values 7.6 %, 7.6%, 5% and 7.69%, respectively. The progeny lines DGGV-198 (Per cent disease incidence - 18.18%), DGGV-206 (Per cent disease incidence - 19.04%), DGGV-212 (Per cent disease incidence-11.2%), DGGV-215 (Per cent disease incidence-14.2%), DGGV-226 (Per cent disease incidence-15%), DGGV-268 (Per cent disease incidence-13.3%) and DGGV-282 (Per cent disease incidence-11.1%) have recorded moderately resistant reaction to MYMV.

In the present investigation it was observed that the disease incidence was very high in *summer* as compared to the other season i.e 0-100 per cent. The presence of mere highly resistant lines from the test germplasm population of green gram highlights the need for extensive work for discovering new sources of germplasm collection. Dearth of resistant varieties imposes the development of virus resistant varieties through inter-specific hybridization and by use of tools of biotechnology in the future (Table 2).

Among the eighty-eight advanced breeding lines derived from the cross DGGV-2 × WGG-42 in F_3 generation, four breeding line showed resistance reaction (DGGV-200, DGGV-218, DGGV-281 and DGGV-284) with the per plant yield of 1.64 g, 3.65 g, 3.94 g and 4.08 g, respectively and twenty-three breeding lines showed moderately resistant reaction to MYMV, when we considered mean per cent disease incidence values of breeding lines from a particular cross as a whole but some of the breeding lines showed resistance response to the MYMV, when we considered per cent disease incidence value of each progeny row separately. But none of the genotypes are comparable to the desirable pod features and high yield potential of the released agronomically superior and popular variety DGGV-2. Hence, all these traits can be introgressed and back crossed to DGGV-2 to breed high yielding genotypes conferring tolerance to the major biotic stresses prevalent in the area. Similar results were observed by Kingsly *et al.* (2015); Sudha *et al*. (2013); Kabi *et al*. (2017) and Dharajiya *et al.* (2018).

Conclusion

The lines DGGV-200, DGGV-218, DGGV-281 and DGGV-284 were observed to be resistant. They can be used for further confirmation of reaction and utilization in further breeding programme to develop resistant variety for Mungbean Yellow Mosaic virus (MYMV) along with the good yield.

Table 1. Disease scoring scale (0-5) for Mung bean Yellow Mosaic Virus (MYMV) based on the percentage of disease incidence (PDI)

Disease Scale	Percent infection	Visual symptoms	Category	Reaction group
0	All plants free of virus symptoms	Complete absence of symptoms	Highly resistant	HR
1	1-10% infection	Small yellowish spots scattered on some leaves	Resistant	R
2	11-20% infection	Yellowish bright spots common on leaves, easy to observe	Moderately resistant	MR
3	21-30% infection	Yellowish bright specks on leaves, easy to observe with larger patches of symptoms	Moderately Susceptible	MS
4	31-50% infection	Bright yellow specks or spots on all leaves, minor stunting of plants and less number of pods	Susceptible	S
5	50% and more infection	Yellowing or chlorosis of all leaves on whole plants, Shortening of internode, severe stunting of plants with no yield or few flowers and deformed pods produced with small, immature and shriveled seeds	Highly Susceptible	HS

Table 2. Response of superior F_3 families to MYMV incidence under natural field conditions

Sl. No.	Pedigree	Genotypes	Per cent disease incidence	Category
1		DGGV-200	7.6	Resistant
2		DGGV-218	7.6	Resistant
3		DGGV-281	5.0	Resistant
4		DGGV-284	7.69	Resistant
6	DGGV-2 × WGG-42	DGGV-198	18.18	Moderately Resistant
7		DGGV-206	19.04	Moderately Resistant
8		DGGV-212	11.2	Moderately Resistant
9		DGGV-215	14.20	Moderately Resistant
10		DGGV-226	15.0	Moderately Resistant
11		DGGV-268	13.3	Moderately Resistant
12		DGGV-282	11.1	Moderately Resistant

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