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RESEARCH ARTICLE

Screening of blackgram genotypes for molecular variability in reaction to *Mungbean yellow mosaic disease*

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ABSTRACT: Sixteen blackgram genotypes were tested for yellow mosaic disease under field conditions and identified nine genotypes as resistant, one as moderately resistant, four as susceptible and two as highly susceptible. No genotypes were found to be immune or highly resistant against yellow mosaic disease. Polymerase Chain reaction (PCR) of DNAs from sixteen genotypes using Sequence Characterized Amplified Region (SCAR) markers namely, SCAR 20F/20R resulted in specific amplification in nine genotypes indicating resistant nature of these genotypes. There was no amplification in six genotypes while faint amplification could be observed in the genotype LBG-752 that recorded 14.47% incidence of the disease along with moderately phenotypic reaction.

Keywords: Blackgram, Yellow mosaic disease. *Mungbean yellow mosaic virus*, polymerase chain reaction, SCAR marker

Blackgram [*Vigna mungo* (L.) Hepper] is an excellent source of easily digestible protein with low flatulence. It supplies 26% protein, 57% carbohydrate, 1.2% fat and is a good source of phosphoric acid, calcium, thiamine (B1), riboflavin (B2) and niacin (B3) (Singh and Awasthi, 2004). Yellow mosaic disease is caused by *Mungbean yellow mosaic virus* (MYMV), *Mungbean yellow mosaic India virus* (MYMIV), *Dolichos yellow mosaic virus* (DoYMV) and *Horsegram yellow mosaic virus* (HgYMV). In south India the usually disease is being caused by MYMV, a whitefly (*Bemisia tabaci*) transmitted geminivirus. It is a serious constraint in blackgram cultivation and could result up to 100% yield losses due to yellowing of leaves (Biswas *et al*., 2009). As the disease could not be managed satisfactorily by insecticides or any chemical applications, other alternatives of controlling the disease should be explored.

Therefore, it was considered important to screen and identify the sources of resistance against yellow mosaic disease in blackgram using genetically linked molecular markers that could facilitate marker assisted selection for rapid evaluation. The development of PCR based Sequence characterized amplified region (SCAR) developed from RAPD markers is a method of choice to test MYMV resistance in blackgram because it is simple, rapid, time saving and more authentic than field screening results. The marker has been consistently associated with the genotypes resistant to MYMV but susceptible genotypes without the resistance gene lack the marker (Prasanthi *et al*., 2013). In the present study phenotypic screening was followed by molecular screening using SCAR 20F/20R marker designed from DNA nucleotide sequence amplified with OPQ1 RAPD marker closely linked to the gene of resistance against yellow mosaic disease in resistance source PU-31 (Prasanthi *et al*., 2011)

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MATERIAL AND METHODS

The experiment was conducted during *kharif* 2014-15 at the Regional Agricultural Research Station (RARS), Lam, Guntur using 16 genotypes of blackgram namely KPU-1, KPU-9, KPU-6, KPU-29, KPU-21, KPU-22, KPU 12-133, KPU 12-1731, OBG-32, LBG-752, DKU-87, DKU-102, UG-281, PU 12-11, Co5 and LBG-623 (susceptible check) obtained from RARS, Lam. A Randomised Block Design with two replications in a microplot of 5 x 4 m with spacing of 30 x 10 cm was followed and percent disease incidence was recorded weekly using the formula

$$\text{Per cent MYMV disease incidence} = \frac{\text{Number of plants infected in a micro plot}}{\text{Total number of plants in a micro plot}} \times 100$$

MYMV disease severity was recorded weekly by using 0-9 modified scale of All India Coordinated Research Project on MULLaRP (Alice and Nadarajan, 2007) and per cent disease index (PDI) was computed using the formula given by Wheeler (1969).

$$\text{PDI} = \frac{\text{Sum of all the numerical ratings}}{\text{Number of observations} \times \text{Maximum disease rating}} \times 100$$

Table 1. Categorization of blackgram genotypes based on yellow mosaic disease disease severity

PDI	Rating	Reaction
0.1-5	1.0 to 2.0	Resistant (R)
5.1-15	2.1 to 4	Moderately resistant (MR)
15.1-30	4.1 to 5	Moderately susceptible (MS)
30.1-75	5.1 to 7	Susceptible (S)
75.1-100	7.1 to 9	Highly susceptible (HS)

The genotypes were assigned different disease reactions based on the categorization given by Gantait and Kantidas (2009) (Table 1).

The cumulative progress of disease was estimated by using PDI recorded at weekly interval in different blackgram genotypes and Area under Disease Progress Curve (AUDPC) values were calculated (Wilcoxson *et al.,* 1975).

$$\text{AUDPC} = \sum_{i=1}^{K} \frac{1}{2}(S_i + S_{i-1}) \times \text{d}$$

Where,

S_i = Disease incidence at i^{th} day or evaluation

K= Number of successive evaluation of the disease

d= Interval between i and i-1evaluation of disease

For conducting molecular characterization of genotypes, 16 blackgram genotypes were raised in pots during rabi 2014 at Genomics Laboratory, Institute of Frontier Technologies, Regional Agricultural Research Station (RARS), Tirupati and were used for DNA isolation five days after sowing (DAS).

DNA isolation

DNA of blackgram samples was extracted from leaves using the modified CTAB method (Murray and Thompson, 1980). Plant material (500 mg) was ground in a pre-sterilized pestle and mortar with liquid nitrogen until a fine powder was obtained and transferred to a sterile 2 ml eppendorf tube. One ml of pre-heated (65°C) extraction buffer (100 mM Tris (pH 8.0), 1.4 M NaCl, 20 mM EDTA, 2% CTAB, 1% PVP, 0.1 % Mercaptoethanal) was added to this and incubated for one h in water bath at 65°C. Tubes were centrifuged (Refrigerated Eppendorf centrifuge) at 10,000 rpm for 10 min at room temperature and the supernatant was collected into eppendorf tubes. The collected supernatant was mixed with equal volume of chloroform and isoamyl alcohol (24:1). 1 µl of RNase (100 µg/µl) was added to this mixture and incubated at room temperature for 10-20 min. The tubes were centrifuged at 10,000 rpm for 10 min and the supernatant was separated. 0.1 volume of 3 M sodium acetate (pH 4.8) and 0.6 volume of ice cold isopropanol were added to the supernatant and incubated at -20°C for overnight. After incubation, the tubes were centrifuged at 13,000 rpm for 20 min at 4°C. The supernatant was discarded and the pellet was washed with 70% ethyl alcohol and again centrifuged at 13,000 rpm at 4°C for 10 min, supernatant was discarded and pellet was air dried, dissolved in 100 µl of sterile distilled water. The DNA obtained was quantified using Nanodrop spectrophotometer and the samples were stored at -20°C for further use.

SCAR marker analysis

Plant genomic DNA was used as template for PCR amplification as described by Williams *et al*. (1990). Amplification was carried out with 20 µl reaction mixture containing 1X assay buffer, 2 mM $MgCl_2$, 0.2 mM dNTP, 10 µ mole SCAR primer 20F/20R designed from the sequence of cloned RAPD product for specific amplification of the loci identified by selected RAPD marker, OPQ1 (BG-YMV-Q1), 50-100 ng of genomic DNA and 1 U of Taq DNA polymerase (Prasanthi *et al.,* 2011). Amplification was performed in 0.2 ml thin walled tubes using a thermocycler (Corbett, Australia) programmed for initial denaturation at 94°C for 2 min, followed by 35 cycles of denaturation at 94°C for 1 min, annealing at 67.4°C for 1 min, primer extension at 72°C for 2 min and a final extension at 72°C for 7 min.

Analysis of PCR products by agarose gel electrophoresis

Amplified product was analysed by Agarose gel electrophoresis as described by Sambrook and Russell (2001). Agarose gel of 1% (w/v) was prepared by dissolving 500 mg of agarose (Axygen, USA) in 50 ml of 1 X TBE buffer. At lukewarm temperature, two µl of ethidium bromide (10 mg/ml) was added and poured into gel casting tray of mini horizontal electrophoresis unit (Hoefer, USA). The DNA samples were loaded after mixing it with loading dye (Fermentas, USA) and the electrophoresis was carried in 1 X TBE buffer at 50 V (Labemate Power Pack 300, USA) till the dye front reached the lower part of the agarose gel. The migration pattern of the DNA fragments in the gel was recorded using gel documentation system (Alpha Innotech, USA) in an auto exposure mode.

RESULTS AND DISCUSSION

Yellow mosaic disease (YMD) incidence ranged from 3.73 (DKU-87) to 96.15% (LBG-623) in the genotypes screened at 91 DAS. Significantly low YMD incidence was recorded in nine genotypes *viz.,* DKU-87 (3.73%), DKU-102 (4.19%), KPU-29 (5.57%), KPU-21 (5.79%), KPU-6 (5.97%), PU 12-11 (6.63%), KPU 12-133 (6.78%), KPU 12-1731 (6.97%) and UG-218 (7.01%) while 14.47% incidence was observed in LBG-752 genotype. Significantly high YMD incidence was recorded in genotypes Co5 (93.53%) and LBG-623 (96.15%) and in other four genotypes *viz*., OBG-32, KPU-1, KPU-22 and KPU-9 disease incidence was 67.38, 64.37, 56.68 and 49.21% respectively (Table 2). After three weeks of sowing, yellow mosaic disease was recorded in LBG-623 and Co5 with disease severity (PDI) of 0.56 and

Table 2. Yellow mosaic disease incidence in blackgram genotypes during kharif 2014-15

Genotypes	Per cent disease incidence (%)										
	21 DAS	28 DAS	35 DAS	42 DAS	49 DAS	56 DAS	63 DAS	70 DAS	77 DAS	84 DAS	91 DAS
KPU-1	0.00 *(0.00)	0.31 (3.13)	0.92 (5.49)	2.03 (7.92)	11.50 (19.78)	28.30 (32.10)	33.61 (35.42)	36.46 (37.12)	53.07 (46.77)	63.04 (52.56)	64.37 (53.36)
KPU-6	0.00 (0.00)	0.00 (0.00)	0.23 (2.71)	0.97 (5.64)	1.45 (6.91)	2.16 (8.45)	2.87 (9.74)	3.61 (10.95)	4.81 (12.66)	5.03 (12.95)	5.97 (14.11)
KPU-29	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	1.02 (5.79)	1.50 (6.96)	2.58 (9.16)	3.06 (10.06)	4.08 (11.64)	4.55 (12.31)	5.10 (13.04)	5.57 (13.65)
KPU-9	0.00 (0.00)	0.49 (4.00)	1.19 (6.24)	2.47 (8.87)	5.33 (13.20)	7.58 (15.95)	10.94 (19.31)	27.52 (31.61)	42.56 (40.69)	48.18 (43.93)	49.21 (44.53)
KPU-21	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.43 (3.74)	2.12 (8.36)	2.52 (9.09)	3.09 (10.12)	3.67 (10.99)	4.15 (11.72)	4.73 (12.53)	5.79 (13.59)
KPU-22	0.00 (0.00)	0.67 (4.51)	1.00 (5.74)	2.00 (7.60)	6.65 (14.87)	13.73 (21.70)	18.47 (25.44)	26.24 (30.68)	44.94 (42.02)	54.64 (47.66)	56.68 (48.84)
KPU 12-133	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.55 (4.23)	1.67 (7.33)	2.23 (8.33)	3.36 (10.45)	3.96 (11.46)	5.10 (13.04)	6.24 (14.45)	6.78 (15.06)
KPU 12-1731	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.96 (5.61)	2.49 (8.92)	3.29 (10.44)	4.25 (11.88)	5.21 (13.18)	6.01 (14.18)	6.57 (14.85)	6.97 (15.30)
OBG-32	0.00 (0.00)	0.88 (5.35)	1.76 (7.61)	4.76 (12.59)	10.64 (18.96)	26.25 (29.90)	34.76 (35.77)	36.29 (37.03)	51.32 (45.74)	59.32 (50.36)	67.38 (55.15)
LBG-752	0.00 (0.00)	0.96 (5.62)	2.77 (9.39)	3.62 (10.96)	6.27 (14.49)	8.08 (16.51)	8.93 (17.34)	9.89 (18.32)	11.70 (19.99)	12.66 (20.83)	14.47 (22.34)
DKU-87	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.60 (4.41)	1.06 (4.18)	1.73 (7.41)	2.32 (8.42)	2.86 (9.66)	3.16 (10.07)	3.54 (10.82)	3.73 (11.13)
DKU-102	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.49 (2.83)	1.19 (5.71)	2.17 (8.16)	3.09 (10.04)	3.22 (10.32)	3.45 (10.69)	3.73 (11.12)	4.19 (11.80)
UG-218	0.00 (0.00)	0.00 (0.00)	0.52 (4.11)	1.47 (6.80)	2.85 (9.71)	3.29 (10.43)	4.24 (11.87)	5.11 (12.99)	6.06 (14.20)	6.49 (14.68)	7.01 (15.32)
PU 12-11	0.00 (0.00)	0.00 (0.00)	0.74 (4.91)	1.40 (6.73)	2.80 (9.54)	3.23 (10.19)	4.19 (11.72)	4.71 (12.52)	5.67 (13.76)	6.11 (14.27)	6.63 (14.91)
Co5	1.02 (5.79)	1.96 (8.03)	3.82 (11.26)	9.39 (17.82)	23.38 (32.15)	50.74 (45.41)	60.25 (51.31)	73.03 (59.02)	79.21 (63.22)	88.45 (70.47)	93.53 (76.90)
LBG-623	0.46 (3.86)	3.53 (10.82)	5.76 (13.88)	12.05 (20.30)	30.36 (33.36)	54.16 (47.37)	64.97 (53.71)	77.49 (61.67)	82.36 (65.14)	91.35 (73.00)	96.15 (79.08)
SEm±	0.04	0.37	0.37	0.97	1.14	0.92	2.35	1.90	1.81	1.21	1.58
CD (P ≤ 0.05%)	0.11	1.11	1.12	2.92	3.44	2.77	7.09	5.74	5.46	3.64	4.76
CV %	11.93	29.43	16.60	23.48	17.04	9.88	23.02	16.13	13.30	8.10	10.01

Figures in parentheses are arcsine transformed values, *Mean of two replications, DOS: 18-07-2014

1.11 in the respective genotypes. With the advancement of age of the crop all genotypes happen to encounter the disease and till harvest there was significant progress in occurrence of the disease. PDI ranged from 5.00 (DKU-87) to 79.98 (LBG-623) (Table 3). Based on 0-9 score and their corresponding disease reactions, nine out of sixteen genotypes *i.e.*, DKU-87, KPU 12-133, DKU-102, UG-281, KPU-21, KPU-6, KPU-29, KPU 12-1731 and PU 12-11 were resistant with 0.85 to 1.50 disease scores. In LBG-752, disease score of 2.75 was recorded and categorized as moderately resistant. The genotypes KPU-1, KPU-22, KPU-9 and OBG-32 were categorized as susceptible (5.65 to 6.65 rating) and genotypes Co5 and LBG-623 as highly susceptible (7.60 to 7.85) (Table 4). Yellow mosaic disease incidence in genotype Co5 was reported as 68% (Khattak *et al*., 2004) and 100% (Sahoo and Sahu, 1991). Obaiah *et al*. (2013) and Prasanthi *et al*. (2013) reported LBG-752 as moderately resistant and LBG-623 as highly susceptible genotypes to YMD infection. Sowmini and Palaniappan (2014) reported Co5 as YMD susceptible genotype. Obaiah *et al*. (2013) evaluated 56 blackgram genotypes for YMD infection and reported 22 genotypes as resistant, 11 as moderately resistant and the remaining 23 as susceptible. Kumar *et al*. (2014) recorded genotypes of blackgram *i.e.*, Azad U-2, KU 96-3, LBG-645, IPU 2-43 and NDU 5-7 as MYMV disease resistant. Genotype evaluation was documented by several workers earlier (Biswas *et al*., 2009; Bag *et al*., 2014; Gopi *et al*., 2016). Shad *et al*. (2006) and Prasanthi *et al*. (2013) observed that blackgram genotypes with MYMV symptoms at early age of the crop were found to be susceptible with high PDI than resistant genotypes and the present results are in accordance with the earlier reports.

The highest Area under Disease Progress Curve (AUDPC) value (2130.64) was recorded in the genotype

Table 3. Yellow mosaic disease severity (PDI) in blackgram genotypes during kharif 2014-15

Genotypes	Per cent Disease Index (PDI)										
	21 DAS	28 DAS	35 DAS	42 DAS	49 DAS	56 DAS	63 DAS	70 DAS	77 DAS	84 DAS	91 DAS
KPU-1	0.00 *(0.00)	0.56 (4.27)	2.78 (9.57)	6.11 (14.17)	14.44 (22.19)	20.55 (26.90)	27.22 (31.41)	36.66 (37.25)	48.33 (44.02)	53.89 (47.21)	60.33 (50.94)
KPU-6	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.56 (3.02)	1.11 (6.04)	2.22 (8.28)	3.33 (10.36)	5.00 (12.89)	7.22 (15.48)	8.33 (16.75)	9.44 (17.88)
KPU-29	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.57 (4.30)	1.11 (6.04)	2.78 (9.54)	3.89 (11.33)	5.00 (12.89)	7.78 (16.02)	8.33 (16.54)	10.00 (18.32)
KPU-9	0.00 (0.00)	1.11 (6.05)	3.89 (11.33)	6.11 (14.17)	10.55 (18.89)	16.66 (24.07)	23.89 (29.23)	37.22 (37.57)	49.44 (44.66)	52.78 (46.57)	62.94 (52.49)
KPU-21	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.56 (3.02)	1.67 (5.26)	2.22 (6.08)	2.78 (6.81)	4.44 (11.11)	6.11 (13.50)	7.22 (14.99)	7.78 (15.81)
KPU-22	0.00 (0.00)	0.56 (4.27)	1.67 (7.31)	5.00 (12.89)	6.66 (14.90)	15.00 (22.74)	22.78 (28.49)	32.78 (34.89)	44.44 (41.79)	51.67 (45.94)	62.15 (52.08)
KPU 12-133	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.56 (3.02)	1.11 (4.28)	1.67 (5.26)	3.33 (9.83)	5.55 (13.34)	7.78 (16.02)	10.00 (18.32)	11.10 (19.43)
KPU 12-1731	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.56 (3.02)	2.78 (9.54)	4.44 (12.07)	5.00 (12.89)	7.22 (15.56)	8.89 (17.30)	10.00 (18.40)
OBG-32	0.00 (0.00)	1.11 (6.05)	3.89 (11.33)	7.77 (16.14)	12.24 (20.14)	21.11 (27.33)	32.78 (34.89)	38.89 (38.55)	50.55 (45.30)	58.89 (50.11)	58.48 (49.87)
LBG-752	0.00 (0.00)	0.00 (0.00)	0.56 (4.27)	2.22 (8.28)	5.00 (12.37)	7.22 (15.29)	10.00 (18.32)	12.78 (20.82)	15.00 (22.69)	17.78 (24.88)	21.11 (27.22)
DKU-87	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.31 (2.24)	1.11 (4.28)	1.67 (5.26)	2.22 (8.28)	2.78 (9.54)	4.44 (12.07)	5.00 (12.89)
DKU-102	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.56 (3.02)	1.67 (7.31)	2.78 (9.54)	3.89 (11.33)	5.00 (12.89)	5.55 (13.55)	6.66 (14.90)
UG-218	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.56 (4.27)	1.67 (7.31)	2.78 (9.54)	4.44 (12.07)	7.22 (15.48)	8.89 (17.30)	10.56 (18.94)	8.89 (17.04)
PU 12-11	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.56 (4.27)	1.11 (4.28)	2.78 (9.10)	5.55 (13.34)	7.22 (15.48)	8.89 (17.21)	10.55 (18.89)	11.67 (19.92)
Co5	0.56 (4.27)	2.22 (8.56)	5.00 (12.89)	8.88 (17.33)	13.33 (21.34)	19.44 (26.10)	32.22 (34.54)	40.55 (39.53)	54.44 (47.53)	62.78 (52.39)	75.55 (60.55)
LBG-623	1.11 (6.05)	3.89 (11.32)	6.11 (14.17)	9.44 (17.70)	17.22 (24.48)	22.77 (28.41)	33.88 (35.53)	46.11 (42.75)	58.89 (50.10)	65.55 (54.06)	79.98 (63.53)
SEm ±	0.04	0.18	0.49	0.99	1.39	1.80	1.92	1.27	1.27	1.14	1.78
CD (P ≤ 0.05%)	0.11	0.53	1.48	2.97	4.19	5.43	5.79	3.84	3.84	3.45	5.38
CV %	11.65	13.99	22.22	25.73	25.31	24.05	20.30	11.15	9.48	7.82	11.16

Figures in parentheses are arcsine transformed values, *Mean of two replications, DOS: 18-07-2014

LBG-623 corresponding to its highly susceptible nature to YMD infection and the lowest AUDPC value (105.09) was observed in DKU-87 (Table 4). Lowest value indicated small amount of cumulative disease progress that resulted in delay in the onset and severity of symptom development (Raju and Anilkumar, 1990). The highest AUDPC value (2402.7) was reported in blackgram susceptible check Barabanki Local (Bag *et al*., 2014) which corroborate with the present results. In mungbean the highest AUDPC value was reported in susceptible genotype, HUM-12 compared to resistant genotypes (Biswas *et al*., 2009).

Validated SCAR marker BG-YMV-Q1 (OPQ1) linked to yellow mosaic disease resistance showed variability among the 16 genotypes screened for the desired amplicon of 532 bp. The genotypes KPU-21, PU 12-11, KPU-29, KPU 12-133, KPU-6, KPU 12-1731, DKU-102, UG-218 and DKU-87 exhibited strong amplification while the genotype LBG-752 showed faint amplification. The amplicons obtained with the said SCAR marker were compared with PU-31 that was validated earlier as a YMD resistant parent using the same SCAR marker (Prasanthi *et al*., 2013). As the marker is linked with YMD resistance genes, a clear indication of variability among the tested genotypes was observed and the genotypes that responded positively to the marker were in confirmation with the results of phenotypic observation and thus these genotypes can be treated as resistant ones. Genotype LBG-752 that recorded 14.47% YMD incidence with moderately resistant phenotypic reaction was found positive to the SCAR marker with a faint amplicon of the desired size. Phenotypically categorized YMD susceptible genotypes KPU-1, KPU-9, KPU-22, OBG-32, Co5 and LBG-623 with high percent disease incidence (49.21-

Table 4. Reaction of blackgram genotypes to MYMV using SCAR marker

Genotypes	SCAR Marker Status	Score	Disease incidence	AUDPC	Disease reaction
KPU-1	A	5.65	64.37	1684.80	S
KPU-6	P	1.13	5.97	227.29	R
KPU-29	P	1.30	5.57	241.03	R
KPU-9	A	5.90	49.21	1631.70	S
KPU-21	P	1.05	5.79	202.11	R
KPU-22	A	5.70	56.68	1481.20	S
KPU 12-133	P	0.90	6.78	248.71	R
KPU 12-1731	P	1.30	6.97	237.04	R
OBG-32	A	6.65	67.38	1795.12	S
LBG-752	P	2.75	14.47	567.58	MR
DKU-87	P	0.85	3.73	105.09	R
DKU-102	P	0.95	4.19	159.15	R
UG-218	P	1.00	7.01	283.73	R
PU12-11	P	1.50	6.63	297.31	R
Co5	A	7.60	93.53	1938.28	HS
LBG-623	A	7.85	96.15	2130.64	HS

A: Absent; P: Present; R: Resistant; MR: Moderate resistant S: Susceptible; HS: Highly susceptible

96.15%) responded negatively to SCAR marker amplification thus confirming the results of Prasanthi *et al*. (2013) (Table 4, Fig 1a,b).

High level of selection accuracy for resistance has been reported with DNA markers (Hittalmani *et al*., 1995). The presence of SCAR marker was reported to be consistently associated with all genotypes resistant to MYMV while susceptible genotypes without resistance gene lacked the marker (Souframanien and Gopalakrishna, 2006). MYMVR-583 SCAR marker was reported to produce amplification in twelve blackgram

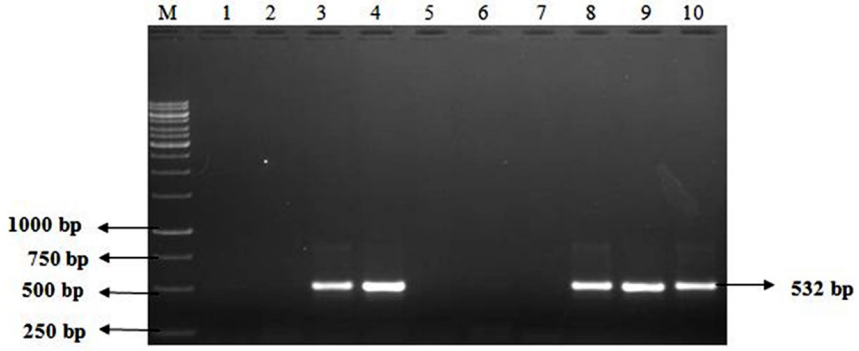

Fig. 1a. Amplification profile of the SCAR marker BG-YMV-Q1, used to screen YMD resistance and susceptible genotypes of blackgram. Lane M: 1 KB Ladder, Lane 1: LBG-623 (Susceptible check), Lane 2: KPU-1, Lane 3: KPU-21, Lane 4: 12-11, Lane 5: Co5, Lane, 6: KPU-9, Lane 7: KPU-22, Lane 8: KPU-29, Lane 9: KPU 12-133, Lane 10: KPU-6

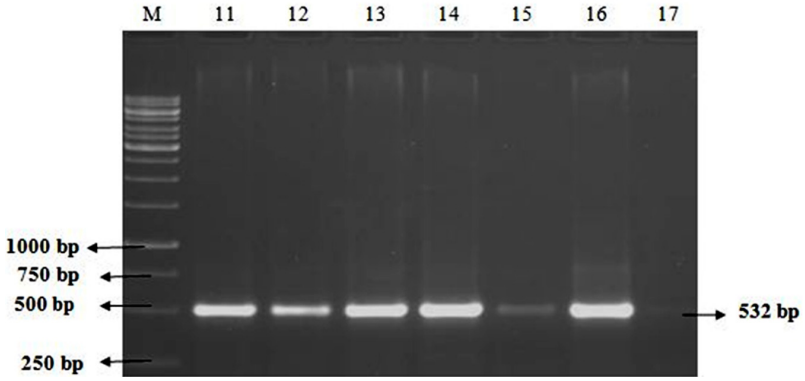

Fig. 1b. Amplification profile of the SCAR marker BG-YMV-Q1, used to screen YMD resistance and susceptible genotypes of blackgram. Lane M: 1 KB Ladder, Lane 11: PU-31 (Resistant check), Lane 12: KPU-12-1731, Lane 13: DKU-102, Lane 14: UG-218, Lane 15: LBG-752, Lane, 16: DKU-87, Lane 17: OBG-32

genotypes that were phenotypically reported as resistant and moderately resistant under field conditions while absence of the desired fragment was reported in highly susceptible genotypes (Dhole and Reddy, 2013). Molecular markers based studies facilitate plant breeders to carry out repeated genotyping throughout the growing season in the absence of any disease incidence. This method of genotyping would save time and labour during the introgression of YMD resistance through molecular breeding. A resistance gene that can be transferred to cultivars with excellent yield potential by using the marker-assisted breeding would be beneficial. The present investigation paves the way to the plant breeders for developing YMD resistant varieties of blackgram.

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