Field screening of greengram (Vigna radiata L.) genotypes for resistance against Urdbean Leaf Crinkle Virus

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ABSTRACT
In the current study one hundred and seven greengram genotypes screened at Department of Pulses, TNAU, Coimbatore using AICRP pest susceptibility scale. It was observed that RME-16-3, RME-16-12, MLT-GG R-16-007, MLT-GG R-16-009, COGG 1319 (rabi-greengram) were highly resistant (HR) to Urdbean Leaf Crinkle Virus (ULCV). None of the greengram genotype is highly resistant in kharif season. Resistance in each case were associated with lower incidence, lower spread and milder symptoms relative to the susceptible cultivars. KEM 16-1, KEM 16-8, KEM 16-20, KME -33, MLT- GG K-16-01, MLT- GG K-16-05 (kharif-greengram) and RME-16-10, MLT-GG R-16-008 (rabi-greengram) were highly susceptible (HS). In susceptible cultivars of greengram, the disease is characterized by enlargement of trifoliate leaves and crinkling of the leaf lamina. However, severity of the symptoms varies in different cultivars. Early infected greengram plants showed complete sterility. The affected plants become stunted and gave a bushy appearance and no pods were formed in severely affected plants. Better understanding of resistance of genotypes will help in improvement of varieties resistant to viral diseases will certainly brighten the prospects of plant virus control in the coming decades.

Key words: Greengram, Genotypes, Leaf crinkle virus, PDI, PSP, Resistance, Screening.

INTRODUCTION
Greengram [Vigna radiata (Linn.)] also known as mungbean belong to the family leguminosae is native to India and nutritious grain legume crops grown in 23 countries of the world. In India greengram occupies an area of about 3.51 million hectare, producing 1.80 million tonnes with the productivity of 511 kg (Anonymous, 2012). Among several viruses reported to naturally infect the mungbean the urdbean leaf crinkle virus (ULCV) is considered to be economically important and most serious disease. Leaf crinkle disease that infects the crops at various stages of its growth, which reduce both quantity and quality of seed. This disease has become one of the major production constraints in both blackgram and greengram especially rabi and summer under uplands and rice fallow situations. The disease manifests itself at the second or third trifoliate leaf stage with the development of crinkling, curling symptoms and malformation of flowers. The disease can cause crop losses to an extent of 100 per cent depending on the variety and stage of infection. Studies regarding availability of resistant genotypes accessions are very limited. The aim of our study to exploit the available resistant greengram genotypes for resistance against the leaf crinkle virus.

MATERIALS AND METHODS
Field screening: Field trials were conducted at the Department of Pulses, TNAU, Coimbatore during kharif 2016 and rabi 2016. About 107 greengram genotypes were screened against Urdbean Leaf Crinkle Virus. For every five test genotypes two rows of a susceptible check, SL 1082, ML-5 (as per AICRP recommendation) and one row of resistant check CO8 (as per the variety release report) were sown. Each test entry was sown in two rows with row length 3 m, row spacing 30 cm and plant to plant spacing 10 cm. The experiment was laid out in randomized complete block design (RCBD) with three replications. The standard agronomic practices were applied to all the treatments throughout the crop growing season but no insecticide was applied. The field was subjected to natural invasion and build-up of population of aphid species and whiteflies, the vector of urdbean leaf crinkle virus and consequently to infection of greengram plants by the virus. The disease incidence of each of the test lines was assessed in fifteen days interval by following disease rating scale of 0-9 grades (AICRP) by calculating the Pest Susceptibility per cent (PSP) and consequently level of resistance/susceptibility of test lines was determined the virus.

PSP= Damage in check entry- Damage in test entry/ Damage in check entry × 100

Assessment of per cent disease incidence (PDI): To work out the per cent disease incidence, total number of plants and number of plants infected with leaf crinkle virus were counted leaving the out of two rows on all the four sides in each field. PDI was calculated by adopting the following formula (Salam et al., 2011).

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Per cent disease incidence =
Number of infected plants / Total number of plants×100

RESULTS AND DISCUSSION

In the present study it was observed that RME-16-3, RME-16-12, MLT-GG R-16-007, MLT-GG R-16-009, COGG 1319 were highly resistant (HR) to ULCV (Table 2). None of the genotypes was found to be highly resistant in kharif season (Table 1).

Twenty, fifty, twenty genotypes of greengram fell under resistant (R), moderately resistant (MR) and susceptible respectively in kharif and rabi seasons (Table 3 and 4). Resistance in each case was associated with lower incidence, lower spread and milder symptoms relative to the susceptible cultivars. Kadian (1982) reported nine mungbean genotypes viz., 15176, 15225, 15227, 15229, 15227, L-24-2, ML-5, T-44, and T-51 as resistant to ULCV when they evaluated 390 mungbean germplasm/varieties by sap-inoculation method. Nene and Kolte (1972) identified five mungbean cultivars viz., 24-3, Baisakhi, T-2, T-44 and T-51 as resistant to ULCV by following sap inoculation method.

In the present study the genotypes KEM 16-1, KEM 16-8, KEM 16-20, KME -33, MLT- GG K-16-01, MLT- GG K-16-05 (Table 1) and RME-16-10, MLT-GG R-16-008 (Table 2) were highly susceptible (HS). In susceptible cultivars of greengram, the disease is characterized by enlargement of trifoliate leaves and crinkling of the leaf lamina. However, severity of the symptoms varies in different cultivars. Early infected blackgram plants showed complete sterility. The affected plants become stunted and gave a bushy appearance and no pods were formed in severely affected plants. In inoculation method, Iqbal et al. (1991) reported that S 332 and S 539 were found to be highly resistant and cv. S 433 and S 275 behaved as susceptible and highly susceptible, respectively. Vijay Kumar (1993) reported that cultivars like LBG 402 and 693 were susceptible and LBG 17 and 695 are highly susceptible under field conditions.

Table 1: Screening of greengram accessions during kharif 2016 against ULCV.

| Accessions | Total plants | % Damage in test entry | PDI | PSP | PSI | Resistance level |
|------------|--------------|------------------------|-----|-----|-----|-----------------|
| KEM 16-1   | 64.50        | 11.67                  | 18.09 | -31.72 | 8 | HS |
| KEM 16-2   | 66.50        | 3.29                   | 4.94 | 62.89 | 3 | MR |
| KEM 16-3   | 89.00        | 4.04                   | 4.54 | 54.40 | 3 | MR |
| KEM 16-4   | 90.50        | 3.24                   | 3.58 | 63.43 | 2 | R |
| KEM 16-5   | 83.00        | 2.53                   | 3.05 | 71.45 | 2 | R |
| KEM 16-6   | 85.00        | 7.13                   | 8.39 | 19.50 | 5 | S |
| KEM 16-7   | 80.00        | 0.82                   | 1.02 | 90.75 | 2 | R |
| KEM 16-8   | 49.00        | 20.48                  | 41.80 | -131.18 | 9 | HS |
| KEM 16-9   | 106.50       | 3.12                   | 2.93 | 64.76 | 3 | MR |
| KEM 16-10  | 111.50       | 3.52                   | 3.16 | 60.26 | 3 | MR |
| KEM 16-11  | 116.00       | 1.83                   | 1.58 | 79.31 | 2 | R |
| KEM 16-12  | 127.50       | 3.17                   | 2.49 | 64.23 | 3 | MR |
| KEM 16-13  | 88.50        | 8.86                   | 10.02 | -0.04 | 6 | S |
| KEM 16-14  | 109.00       | 2.15                   | 1.97 | 75.77 | 2 | R |
| KEM 16-15  | 116.50       | 2.49                   | 2.14 | 71.85 | 3 | MR |
| KEM 16-16  | 125.00       | 5.51                   | 4.41 | 37.84 | 4 | MR |
| KEM 16-17  | 117.50       | 6.87                   | 5.85 | 22.46 | 5 | S |
| KEM 16-18  | 103.50       | 2.90                   | 2.80 | 67.23 | 3 | MR |
| KEM 16-19  | 71.00        | 4.85                   | 6.82 | 45.31 | 4 | MR |
| KEM 16-20  | 69.00        | 15.91                  | 23.06 | -79.57 | 9 | HS |
| KEM 16-21  | 70.50        | 2.73                   | 3.87 | 69.22 | 3 | MR |
| KEM 16-22  | 90.00        | 3.88                   | 4.32 | 56.16 | 3 | MR |

Table 1: Continue............
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| Code     | PDI | PSP | PSI | CHECK |
|----------|-----|-----|-----|-------|
| KEM 16-23| 100.00| 3.13 | 3.13 | 64.67 | MR |
| KEM 16-24| 84.00 | 7.22 | 8.60 | 18.49 | 5   | S   |
| KEM 16-25| 92.00 | 0.27 | 0.30 | 96.90 | 2   | R   |
| KEM 16-26| 87.00 | 3.35 | 3.85 | 62.15 | 3   | MR  |
| KEM 16-27| 111.00 | 7.73 | 6.97 | 12.71 | 5   | S   |
| KEM 16-28| 92.00 | 3.06 | 3.33 | 65.44 | 3   | MR  |
| KEM 16-30| 106.00 | 5.73 | 5.40 | 35.34 | 4   | MR  |
| KEM 16-31| 106.50 | 1.41 | 1.33 | 84.04 | 2   | R   |
| KEM 16-32| 103.50 | 4.25 | 4.11 | 52.01 | 3   | MR  |
| KEM 16-33| 111.00 | 7.73 | 6.97 | 12.71 | 5   | S   |
| KEM 16-34| 87.50 | 3.35 | 3.85 | 62.15 | 3   | MR  |
| KEM 16-35| 92.00 | 7.82 | 8.69 | 11.70 | 4   | MR  |
| KEM 16-36| 119.50 | 1.39 | 1.16 | 84.32 | 2   | R   |
| KEM 16-37| 92.00 | 3.06 | 3.33 | 65.44 | 3   | MR  |
| KEM 16-38| 106.00 | 5.73 | 5.40 | 35.34 | 4   | MR  |
| KEM 16-39| 111.00 | 7.73 | 6.97 | 12.71 | 5   | S   |
| KEM 16-40| 92.00 | 3.06 | 3.33 | 65.44 | 3   | MR  |
| KEM 16-41| 106.50 | 1.41 | 1.33 | 84.04 | 2   | R   |
| KEM 16-42| 103.50 | 4.25 | 4.11 | 52.01 | 3   | MR  |
| KME-1    | 54.00 | 4.05 | 7.50 | 54.31 | 3   | MR  |
| KME-4    | 36.50 | 4.63 | 12.70 | 47.69 | 4   | MR  |
| KME-15   | 96.50 | 5.77 | 5.98 | 34.82 | 4   | MR  |
| KME-19   | 91.00 | 3.05 | 3.35 | 65.55 | 3   | MR  |
| KME-31   | 62.00 | 8.25 | 13.00 | 6.90  | 6   | S   |
| KME -33  | 38.00 | 17.58 | 46.25 | -98.38 | 9   | HS  |
| KME 15.1 | 86.50 | 7.32 | 8.46 | 17.40 | 5   | S   |
| LGG 15.6 | 105.50 | 6.74 | 6.39 | 23.90 | 3   | S   |
| SME 16-2 | 78.50 | 4.58 | 5.84 | 48.29 | 4   | MR  |
| SME 16-8 | 107.50 | 4.70 | 4.37 | 46.95 | 4   | MR  |
| SME 16-29| 97.00 | 0.25 | 0.26 | 97.21 | 2   | R   |
| SME 16-34| 58.50 | 10.24 | 17.51 | -15.59 | 7   | S   |
| SME 16-37| 45.50 | 4.05 | 8.90 | 54.31 | 3   | MR  |
| SME 16-38| 85.00 | 4.05 | 4.76 | 54.31 | 3   | MR  |
| SME 16-39| 58.50 | 10.24 | 17.51 | -15.59 | 7   | S   |
| SME 16-40| 45.50 | 4.05 | 8.90 | 54.31 | 3   | MR  |
| SME 16-29| 97.00 | 0.25 | 0.26 | 97.21 | 2   | R   |
| SME 16-34| 58.50 | 10.24 | 17.51 | -15.59 | 7   | S   |
| SME 16-37| 45.50 | 4.05 | 8.90 | 54.31 | 3   | MR  |
| SME 16-38| 85.00 | 4.05 | 4.76 | 54.31 | 3   | MR  |
| SME 16-39| 58.50 | 10.24 | 17.51 | -15.59 | 7   | S   |
| SME 16-29| 97.00 | 0.25 | 0.26 | 97.21 | 2   | R   |
| SME 16-34| 58.50 | 10.24 | 17.51 | -15.59 | 7   | S   |
| SME 16-37| 45.50 | 4.05 | 8.90 | 54.31 | 3   | MR  |
| SME 16-38| 85.00 | 4.05 | 4.76 | 54.31 | 3   | MR  |
| SME 16-39| 58.50 | 10.24 | 17.51 | -15.59 | 7   | S   |
| SME 16-29| 97.00 | 0.25 | 0.26 | 97.21 | 2   | R   |
| SME 16-34| 58.50 | 10.24 | 17.51 | -15.59 | 7   | S   |
| SME 16-37| 45.50 | 4.05 | 8.90 | 54.31 | 3   | MR  |
| SME 16-38| 85.00 | 4.05 | 4.76 | 54.31 | 3   | MR  |
| SME 16-39| 58.50 | 10.24 | 17.51 | -15.59 | 7   | S   |
| SME 16-29| 97.00 | 0.25 | 0.26 | 97.21 | 2   | R   |
| SME 16-34| 58.50 | 10.24 | 17.51 | -15.59 | 7   | S   |
| SME 16-37| 45.50 | 4.05 | 8.90 | 54.31 | 3   | MR  |
| SME 16-38| 85.00 | 4.05 | 4.76 | 54.31 | 3   | MR  |
| SME 16-39| 58.50 | 10.24 | 17.51 | -15.59 | 7   | S   |
| SME 16-29| 97.00 | 0.25 | 0.26 | 97.21 | 2   | R   |
| SME 16-34| 58.50 | 10.24 | 17.51 | -15.59 | 7   | S   |
| SME 16-37| 45.50 | 4.05 | 8.90 | 54.31 | 3   | MR  |
| SME 16-38| 85.00 | 4.05 | 4.76 | 54.31 | 3   | MR  |
| SME 16-39| 58.50 | 10.24 | 17.51 | -15.59 | 7   | S   |
| SME 16-29| 97.00 | 0.25 | 0.26 | 97.21 | 2   | R   |
| SME 16-34| 58.50 | 10.24 | 17.51 | -15.59 | 7   | S   |

PDI- Per cent Disease Incidence, PSP- Pest Susceptibility Per cent, PSI- Pest Susceptibility Per cent.
Table 2: Screening of greengram accessions during *rabi* 2016-2017 against ULCV.

| Accessions   | Total plants | % Damage in test entry | PDI | PSP | PSI | Resistance level |
|--------------|--------------|-------------------------|-----|-----|-----|------------------|
| RME-16-1     | 56.00        | 0.50                    | 0.89| 50.00| 3   | MR               |
| RME-16-2     | 84.00        | 0.75                    | 0.89| 25.00| 4   | MR               |
| RME-16-3     | 33.00        | 0.00                    | 0.00| 100.00| 1 | HR               |
| RME-16-4     | 34.00        | 1.00                    | 2.94| 0.00 | 6   | S                |
| RME-16-5     | 81.00        | 0.50                    | 0.62| 50.00| 3   | MR               |
| RME-16-6     | 104.00       | 1.25                    | 1.20| -25.00| 7 | S                |
| RME-16-7     | 24.00        | 0.25                    | 1.04| 75.00| 2   | R                |
| RME-16-8     | 16.00        | 0.25                    | 1.56| 75.00| 2   | R                |
| RME-16-9     | 78.00        | 0.50                    | 0.64| 50.00| 3   | MR               |
| RME-16-10    | 49.00        | 1.75                    | 3.57| -75.00| 9 | HS               |
| RME-16-11    | 22.00        | 0.25                    | 1.14| 75.00| 2   | R                |
| RME-16-12    | 83.00        | 0.00                    | 0.00| 100.00| 1 | HR               |
| RME-16-13    | 35.00        | 0.75                    | 2.14| 25.00| 4   | MR               |
| MLT-GG R-16-001 | 68.00      | 1.00                    | 1.47| 0.00 | 6   | S                |
| MLT-GG R-16-002 | 35.00      | 0.25                    | 0.71| 75.00| 2   | R                |
| MLT-GG R-16-003 | 65.00      | 1.00                    | 1.54| 0.00 | 6   | S                |
| MLT-GG R-16-004 | 40.00      | 1.00                    | 0.00| 0.00 | 1   | S                |
| MLT-GG R-16-005 | 19.00      | 1.00                    | 0.00| 0.00 | 1   | S                |
| MLT-GG R-16-006 | 19.00      | 1.00                    | 0.00| 0.00 | 6   | S                |
| MLT-GG R-16-007 | 56.00      | 0.00                    | 0.00| 100.00| 1 | HR               |
| MLT-GG R-16-008 | 17.00      | 2.00                    | 0.00| -100.00| 9 | HS               |
| MLT-GG R-16-009 | 22.00      | 0.00                    | 0.00| 100.00| 1 | HR               |
| MLT-GG R-16-010 | 14.00      | 0.75                    | 5.36| 25.00| 4   | MR               |
| COGG 1319    | 17.00        | 0.00                    | 0.00| 100.00| 1 | HR               |
| COGG 1332    | 12.00        | 0.25                    | 2.08| 75.00| 2   | R                |
| COGG 1339    | 29.00        | 0.25                    | 0.86| 75.00| 2   | R                |
| MLT-GG 11-2  | 10.00        | 0.50                    | 5.00| 50.00| 3   | MR               |
| ML 5         | 5.75         | 1.25                    | 39.74|     |     | CHECK            |
| SL 1082      | 22           | 1.25                    | 5.68|     |     | CHECK            |
| C08           | 12.5         | 1                       | 8.00|     |     | CHECK            |

PDI: Per cent Disease Incidence, PSP: Pest Susceptibility Per cent, PSI: Pest Susceptibility Per cent.

Table 3: Reaction of greengram accessions against urdbean leaf crinkle disease during *kharif* 2016.

| Resistance level | Accessions                                                                 |
|------------------|---------------------------------------------------------------------------|
| HR               | KEM 16-4, KEM 16-5, KEM 16-7, KEM 16-11, KEM 16-14, KEM 16-25, KEM 16-28, KEM 16-31, KEM 16-33, KEM 16-37, KEM 16-40, SME 16-29, COGG 13-19, COGG 13-32, KEM 16-2, KEM 16-3, KEM 16-9, KEM 16-10, KEM 16-12, KEM 16-15, KEM 16-16, KEM 16-18, KEM 16-19, KEM 16-21, KEM 16-22, KEM 16-23, KEM 16-26, KEM 16-29, KEM 16-30, KEM 16-32, KEM 16-34, KEM 16-35, KEM 16-36, KEM 16-38, KEM 16-39, KEM 16-41, KEM 16-42, KEM 16-43, KEM 16-45, KEM 16-46, KEM 16-48, KME-1, KME-4, KME-15, KME-19, SME 16-2, SME 16-8, SME 16-37, SME 16-38, COGG 10-10, COGG 10-02, MLT-GG K-16-02, MLT-GG K-16-03, MLT-GG K-16-04, MT-LG K-16-06, MLT-GG K-16-07, MLT-GG K-16-09 |
| R                | KEM 16-6, KEM 16-13, KEM 16-17, KEM 16-24, KEM 16-27, KEM 16-44, KEM 16-47, KME 15 1, LGG-486, SME 16-34, MLT-GG K-16-08, MLT-GG K-16-10 |
| MR               | KEM 16-1, KEM 16-8, KEM 16-20, KME-33, MLT-GG K-16-01, MLT-GG K-16-05 |
| S                | KEM 16-2, KEM 16-12, KEM-GG R-16-007, MLT-GG R-16-009, COGG 1319, RME-16-3, RME-16-12, MLT-GG R-16-007, MLT-GG R-16-009, COGG 1319 |
| HS               | KEM 16-1, KEM 16-8, KEM 16-20, KME-33, MLT-GG K-16-01, MLT-GG K-16-05 |

Table 4: Reaction of greengram accessions against urdbean leaf crinkle disease during *rabi* 2016-2017.

| Resistance level | Accessions                                                                 |
|------------------|---------------------------------------------------------------------------|
| HR               | RME-16-3, RME-16-12, MLT-GG R-16-007, MLT-GG R-16-009, COGG 1319, RME-16-7, RME-16-8, RME-16-11, MLT-GG R-16-002, COGG 1332, COGG 1339 |
| R                | RME-16-4, RME-16-6, MLT-GG R-16-001, MLT-GG R-16-003, MLT-GG R-16-004, MLT-GG R-16-005, MLT-GG R-16-006 |
| MR               | RME-16-1, RME-16-2, RME-16-5, RME-16-9, RME-16-13, MLT-GG R-16-010, COGG 11-2 |
| S                | RME-16-10, MLT-GG R-16-008, |
Seventy seven blackgram and ten greengram cultivars/germplasm lines were evaluated against BLCD both under natural and artificial conditions using a six point scale. No cultivar/germplasm line of blackgram and greengram was found to be resistant/highly resistant to the disease. None of the popular cultivars (LBG 17, 20, 648 and T9) grown in these areas were free from the disease (Nageswara Rao, 2002). Five mungbean genotypes; VC-3960 (A-89), NCM209, 98-CMH-016, NM-2 and BRM-195 were found free of disease symptoms (Bashir et al., 2005). In the present study there was difference in the reaction of different cultivars/ lines against ULCV in different seasons. There is variability in susceptibility of genotypes in different seasons. ML T-GG R-16-001 and MLT-GG R-16-005 germplasm lines of greengram are susceptible during rabi season and the same genotypes became more susceptible during kharif season.

**CONCLUSION**

Since ULCV is seed-borne, the initial source of infection under field conditions comes from seed. The virus infection at an early stage of the plants was known to cause heavy losses. Therefore use of virus-free seed and development of virus-resistant mungbean cultivars using the present source of resistance would help to control the disease and its further spread in new localities. Use of resistant varieties is regarded as an economical and durable method for controlling plant diseases, especially those caused by viruses. Better understanding the resistant sources will help in the improvement of varieties that will certainly brighten the prospects of plant virus control in the coming decades. Now the disease is becoming a great threat to the Vigna species in order to manage the disease very effectively the casual organism of the virus should be identified and characterized and the mechanism of vector-virus interaction should be explored for thorough understanding of disease epidemiology.

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