



# Screening of Cluster Bean Genotypes/ Varieties for Resistance to *Xanthomonas axonopodis* pv. *cyamopsidis* Using Two Methods of Inoculation

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## Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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## ABSTRACT

Clusterbean [*Cyamopsis tetragonaloba* (L.) Taub] is an arid and semi-arid legume crop belonging to the family *leguminaceae*. It is a drought enduring leguminous crop because of its profound tap rooting system and has high capability to get well from water stress. Total fifty-nine varieties/genotypes of clusterbean were screened under artificial Inoculation field conditions at College Research Farm, College of Agriculture, Bikaner against bacterial blight disease during the

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*Kharif* season of 2018 and 2019. Two alternative inoculation techniques-seed inoculation and foliar sprays of bacterial suspension were employed to test for resistance to *Xanthomonas axonopodis* pv. *cyamopsidis* bacterial blight infection. ( $2.5 \times 10^8$ cfu/ml). Out of 59 varieties, only one variety RGC-1066 found resistant against bacterial blight disease, 17 were found moderately resistant, 17 were moderately susceptible and 24 varieties were found susceptible. No one variety found highly susceptible against bacterial blight.

**Keywords:** Clusterbean; bacterial blight; resistant; *Xanthomonas axonopodis* pv. *Cyamopsidis*.

## 1. INTRODUCTION

Clusterbean [*Cyamopsis tetragonaloba* (L.) Taub.] ( $2n=14$ ) commonly known as "guar" means "cow food" is an arid and semi-arid legume crop belonging to the family *leguminaceae*. It is a drought enduring leguminous crop because of its profound tap rooting system and has high capability to get well from water stress. The green pods are a healthy vegetable with 82.5% water, 3.7% protein, 9.9% carbohydrates, 0.2% fat, 2.3% fiber, and 1.4% other minerals, including 0.13 % calcium, 0.25 % phosphorus, 5.8 mg/100 g iron, and 49 mg/100 g vitamin. In India, there are 39.36 million hectares of clusterbean crops, which produce 16.24 million tons, with a yield of 428 kg per hectare. Clusterbean cultivation spans a total of 28.41 lakh hectares in Rajasthan, producing 12.84 lakh tons and yielding 452 kg per hectare. Rajasthan is the top-producing state for cluster beans, with more than 70% of the country's total production.

Bacterial blight of clusterbean caused by *Xanthomonas axonopodis* pv. *cyamopsidis* has been reported from Arizona [1], Madison [2] and Brazil [3]. In India, disease has been reported in Rajasthan [4], Haryana [5] and Karnataka [6-8]. The leaf blight pathogen *Xanthomonas a.* pv. *cyamopsidis* causes drastic reduction in plant stand and yield as high as 58% in cultivar. The pathogen is seed-borne and has a one-year lifespan in seeds. Pathogen is seed transmitted; thus, it can cause infection from pre appearance phase to growing stages of the plant in favourable environmental situation for the duration of the crop season. The disease appears both as leaf spot and blight simultaneously [9]. Cultural, chemical or biological strategies for disease management are not adequate to control the efficient or economic. Identification of germplasm with resistance to pathogen appears promising for reducing damage and yield losses caused by pathogen [10-12]. In this study, two inoculation procedures were used to assess how fifty-nine cultivars of

cluster beans responded to *Xanthomonas a.* pv. *cyamopsidis* infection.

## 2. MATERIALS AND METHODS

Fifty-nine clusterbean germplasms/varieties of indigenous as well as exotic origin cultivars were screened for identification of resistance sources against artificially inoculated *X. a.* pv. *cyamopsidis*. The experiments were conducted at Research farm, College of Agriculture, Bikaner during the *Kharif* seasons of 2018 and 2019. The test entries were planted during mid-July and harvested during the November.

Seeds of different cultivars were artificially inoculated with *X. a.* pv. *cyamopsidis* by soaking in bacterial cell suspension ( $2.5 \times 10^8$ cfu/ml) [13]. Seeds were sown in rows, each of 5 m length and maintaining row to row and plant to plant distance as 30 x 10 cm, with three replications in randomized block design (R.B.D.). After every two test entries, one row of susceptible check (local) was planted, as well as around the entire experiment. A fresh 72 hrs old bacterial culture, grown on Nutrient Agar media was used for inoculations on the plants. The culture was harvested in 10 ml sterile water diluted to a concentration of  $2.5 \times 10^8$ cfu /ml and used immediately. The suspension was sprayed on plants with hand atomizer twice at 24 hrs interval. After the 7 days of inoculation when symptoms appear, the disease intensity was quite serious during the season due to favourable conditions for the development of the disease.

Observations for disease severity were recorded by visual scoring as per the standard continuous rating 0-5 scale [13]. Disease severity on each genotype was recorded at pre-flowering (30 DAS) and maturity stage using a 0-5 disease rating scale. On the basis of disease per cent disease index was calculated using formula described earlier and varieties were categorized on basis of per cent disease index range.

**Table 1. List of clusterbean germplasms / varieties**

| S.NO. | Germplasms / varieties  | Source   |
|-------|---|--|
| 40    | RGr-16-3, RGr-16-3-1, RGr-16-3-2, RGr-16-3-3, RGr-16-3-4, RGr-16-3-5, RGr-16-3-6, RGr-16-3-7, RGr-16-4, RGr-16-5, RGr-16-5-1, RGr-16-5-2, RGr-16-5-3, RGr-16-5-4, RGr-16-5-5, RGr-16-8, RGr-16-9, RGr-16-9-1, RGr-16-9-2, RGr-16-9-3, RGr-16-9-4, RGr-16-9-5, RGr-16-9-6, RGr-16-11, RGr-16-11-1, RGr-16-11-2, RGr-16-11-3, RGr-17-1, RGr-17-2, RGr-17-4-7-2, RGr-17-4-3-2, RGr-17-4-4-1, RGr-19-1-1, RGr-19-12-1, RGr-19-12-2, RGr-19-11-1, RGr-19-16-1, RGr-19-18-1, RGr-19-19-1, RGr-19-19-2 | RARI, Rajasthan Agricultural Research Institute, Durgapura, Jaipur |
| 19    | RGC-563, RGC-936, RGC-963, RGC-986, RGC-1002, RGC-1003, RGC-1017, RGC-1031, GAUG 1104, CAZG 15-2, GAUG-1502, CAZG- 15-5, GAUG-1501, GAUG 1304, CAZG-15-7, RGC-1033 (ch), HG 2-20(ch), X-10, RGC-1066, HG-563(ch)  | Agricultural Research Station, Bikaner                             |

**Table 2. Category of varieties/lines based on per cent disease index**

| Rating | PDI                | Category                    |
|--------|--------------------|-----------------------------|
| 0      | 0 or less than 1.0 | Free from disease (Immune)  |
| 1      | 1-10%              | Resistant (R)               |
| 2      | 10 - 25%           | Moderately resistant (MR)   |
| 3      | 25 -50%            | Moderately susceptible (MS) |
| 4      | 50 - 75%           | Susceptible (S)             |
| 5      | More than 75%      | Highly susceptible (HS)     |

Disease index on foliage was calculated using the formula of McKinney [14]:

$$PDI = \frac{\text{Sum of all numerical ratings}}{\text{Total number of leaves observed} \times \text{Maximum rating scale}} \times 100$$

### 3. RESULTS AND DISCUSSION

Total fifty-nine varieties/genotypes of clusterbean were screened under artificial Inoculation field conditions at College Research Farm, College of Agriculture, Bikaner against bacterial blight disease during the *Kharif* season of 2018 and 2019. The observations on disease intensity on various genotypes/varieties were recorded and categorized as per their disease reaction. The rating scale 0-5 was used for recording the observation.

The results on pooled basis of both the *kharif* seasons 2018 and 2019 one variety RGC-1066 was found resistant against bacterial blight disease. Fifteen genotypes / varieties viz., RGC-563, RGC-1002, RGC-1017, RGC-1031, GAUG-1304, CAZG-15-7, RGr-16-3-3, RGr-16-3-4, RGr-16-5-1, RGr-16-5-2, RGr-16-9-3, RGr-16-9-4, RGr-17-4-7-2, RGr-17-4-3-2, RGr-19-19-1 were moderately resistant (MR), while nineteen genotypes / varieties namely RGC-936, RGC-1003, RGC-1055, RGC-1033(ch), CAZG-15-5, RGr-16-3-1, RGr-16-3-2, RGr-16-4, RGr-16-5-3, RGr-16-5-5, RGr-16-9, RGr-16-9-1, RGr-16-9-2, RGr-16-9-5, RGr-16-11-1, RGr-19-11-1, RGr-19-16-1, RGr-19-18-1, RGr-19-12-1 were observed moderately susceptible (MS) reaction against

bacterial blight of clusterbean. Twenty four genotypes / varieties i.e. RGC-963, RGC-986, GAUG-1502, GAUG-1501, HG-2-20(ch), X-10, HG-563(ch), RGr-16-3 RGr-16-3-5, RGr-16-3-6, RGr-16-3-7, RGr-16-5, RGr-16-5-4, RGr-16-8, RGr-16-9-6, RGr-16-11, RGr-16-11-2, RGr-16-11-3, RGr-17-1, RGr-17-2, RGr-17-4-4-1, RGr-19-1-1, RGr-19-12-2, RGr-19-19-2 were found reaction susceptible (S) and no single genotypes / varieties was found highly susceptible. Similar finding reported by Karwasra and Chand [15] they conducted a field experiment at Hisar on 590 cultivars and strains, under conditions of natural infection during 1976, none was immune, 35 were graded as resistant, 41 moderately resistant and remaining were susceptible to bacterial blight. When the resistant lines were further tested by inoculation in 1977, only GP-380, GP-508B and GP-590B were still resistant. Gandhi and Chand [16] revealed the absence of immune of resistance source against bacterial blight in clusterbean lines with moderate degree of resistance identified from genetic stock include. GP-380, GP-508 B and GP-590 HG-75, HG-258, RGC-990, HGC-365, HGS-502, HGS-504 D-39-1, HG-75, HG-441, HG-182, PLG-851C-9065, HG-765, HG-464, HD-312 and HG-513.

**Table 3. Disease reaction of clusterbean varieties/genotypes to *X. a. pv. cyamopsidis* under artificial inoculation conditions**

| S.N. | Germplasm/varieties | Kharif 2018           |                  | Kharif 2019           |                  | Pooled                |                  |
|------|---------------------|-----------------------|------------------|-----------------------|------------------|-----------------------|------------------|
|      |                     | Disease Intensity (%) | Disease reaction | Disease intensity (%) | Disease reaction | Disease intensity (%) | Disease reaction |
| 1    | RGC-563             | 13.25                 | MR               | 17.43                 | MR               | 15.34                 | MR               |
| 2    | RGC-936             | 37.56                 | MS               | 29.13                 | MS               | 33.34                 | MS               |
| 3    | RGC-963             | 52.55                 | S                | 47.10                 | MS               | 49.82                 | MS               |
| 4    | RGC-986             | 58.50                 | S                | 64.33                 | S                | 61.41                 | S                |
| 5    | RGC-1002            | 12.40                 | MR               | 19.56                 | MR               | 15.98                 | MR               |
| 6    | RGC-1003            | 36.80                 | MS               | 29.45                 | MS               | 33.12                 | MS               |
| 7    | RGC-1017            | 17.15                 | MR               | 13.67                 | MR               | 15.41                 | MR               |
| 8    | RGC-1031            | 19.70                 | MR               | 24.54                 | MR               | 22.12                 | MR               |
| 9    | RGC-1055            | 41.67                 | MS               | 36.50                 | MS               | 39.08                 | MS               |
| 10   | GAUG-1502           | 53.70                 | S                | 55.45                 | S                | 54.57                 | S                |
| 11   | CAZG- 15-5          | 65.30                 | S                | 48.30                 | MS               | 56.80                 | MS               |
| 12   | GAUG-1501           | 56.84                 | S                | 61.45                 | S                | 59.14                 | S                |
| 13   | GAUG 1304           | 17.50                 | MR               | 21.67                 | MR               | 19.58                 | MR               |
| 14   | CAZG-15-7           | 13.67                 | MR               | 16.33                 | MR               | 15.00                 | MR               |
| 15   | RGC-1033 (ch)       | 23.34                 | MR               | 27.33                 | MS               | 25.33                 | MS               |
| 16   | HG 2-20(ch)         | 67.84                 | S                | 61.67                 | S                | 64.75                 | S                |
| 17   | X-10                | 53.50                 | S                | 51.30                 | S                | 52.40                 | S                |
| 18   | RGC-1066(ch)        | 6.40                  | R                | 7.50                  | R                | 6.95                  | R                |
| 19   | HG-563(ch)          | 63.27                 | S                | 52.50                 | S                | 57.88                 | S                |
| 20   | RGr-16-3            | 47.44                 | MS               | 53.25                 | S                | 50.34                 | S                |
| 21   | RGr-16-3-1          | 27.50                 | MS               | 34.67                 | MS               | 31.08                 | MS               |
| 22   | RGr-16-3-2          | 34.37                 | MS               | 29.50                 | MS               | 31.93                 | MS               |
| 23   | RGr-16-3-3          | 23.67                 | MR               | 18.45                 | MR               | 21.06                 | MR               |
| 24   | RGr-16-3-4          | 16.90                 | MR               | 23.25                 | MR               | 20.07                 | MR               |
| 25   | RGr-16-3-5          | 68.70                 | S                | 61.30                 | S                | 65.00                 | S                |
| 26   | RGr-16-3-6          | 53.32                 | S                | 57.45                 | S                | 55.38                 | S                |
| 27   | RGr-16-3-7          | 69.67                 | S                | 64.50                 | S                | 67.08                 | S                |
| 28   | RGr-16-4            | 52.50                 | S                | 46.45                 | MS               | 49.47                 | MS               |
| 29   | RGr-16-5            | 56.00                 | S                | 52.25                 | S                | 54.12                 | S                |
| 30   | RGr-16-5-1          | 24.30                 | MR               | 13.67                 | MR               | 18.98                 | MR               |
| 31   | RGr-16-5-2          | 21.10                 | MR               | 24.40                 | MR               | 22.75                 | MR               |
| 32   | RGr-16-5-3          | 29.45                 | MS               | 36.45                 | MS               | 32.95                 | MS               |
| 33   | RGr-16-5-4          | 54.17                 | S                | 57.20                 | S                | 55.68                 | S                |
| 34   | RGr-16-5-5          | 46.12                 | MS               | 39.50                 | MS               | 42.81                 | MS               |
| 35   | RGr-16-8            | 67.55                 | S                | 64.33                 | S                | 65.94                 | S                |
| 36   | RGr-16-9            | 45.17                 | MS               | 36.40                 | MS               | 41.78                 | MS               |
| 37   | RGr-16-9-1          | 42.84                 | MS               | 33.45                 | MS               | 38.14                 | MS               |
| 38   | RGr-16-9-2          | 29.60                 | MS               | 31.00                 | MS               | 30.30                 | MS               |
| 39   | RGr-16-9-3          | 19.78                 | MR               | 24.40                 | MR               | 22.09                 | MR               |
| 40   | RGr-16-9-4          | 22.50                 | MR               | 17.30                 | MR               | 19.90                 | MR               |
| 41   | RGr-16-9-5          | 38.17                 | MS               | 31.50                 | MS               | 34.83                 | MS               |
| 42   | RGr-16-9-6          | 52.50                 | S                | 58.60                 | S                | 55.55                 | S                |
| 43   | RGr-16-11           | 63.50                 | S                | 53.30                 | S                | 58.40                 | S                |
| 44   | RGr-16-11-1         | 33.78                 | MS               | 27.50                 | MS               | 30.64                 | MS               |
| 45   | RGr-16-11-2         | 47.00                 | MS               | 51.67                 | S                | 49.33                 | S                |
| 46   | RGr-16-11-3         | 62.83                 | S                | 59.33                 | S                | 61.08                 | S                |
| 47   | RGr-17-1            | 57.61                 | S                | 61.45                 | S                | 59.53                 | S                |
| 48   | RGr-17-2            | 64.50                 | S                | 57.25                 | S                | 60.87                 | S                |
| 49   | RGr-17-4-7-2        | 21.06                 | MR               | 15.67                 | MR               | 18.36                 | MR               |
| 50   | RGr-17-4-3-2        | 18.06                 | MR               | 24.67                 | MR               | 21.36                 | MR               |
| 51   | RGr-17-4-4-1        | 65.29                 | S                | 61.30                 | S                | 63.29                 | S                |
| 52   | RGr-19-1-1          | 54.50                 | S                | 57.50                 | S                | 56.00                 | S                |
| 53   | RGr-19-12-1         | 17.83                 | MR               | 26.30                 | MS               | 22.06                 | MS               |
| 54   | RGr-19-12-2         | 70.50                 | S                | 66.45                 | S                | 68.47                 | S                |
| 55   | RGr-19-11-1         | 38.44                 | MS               | 43.25                 | MS               | 40.84                 | MS               |
| 56   | RGr-19-16-1         | 47.00                 | MS               | 46.45                 | MS               | 46.72                 | MS               |

| S.N. | Germplasm/varieties | Kharif 2018           |                  | Kharif 2019           |                  | Pooled                |                  |
|------|---------------------|-----------------------|------------------|-----------------------|------------------|-----------------------|------------------|
|      |                     | Disease Intensity (%) | Disease reaction | Disease intensity (%) | Disease reaction | Disease intensity (%) | Disease reaction |
| 57   | RGr-19-18-1         | 30.67                 | MS               | 37.50                 | MS               | 34.08                 | MS               |
| 58   | RGr-19-19-1         | 18.32                 | MR               | 24.30                 | MR               | 21.31                 | MR               |
| 59   | RGr-19-19-2         | 55.33                 | S                | 52.40                 | S                | 53.86                 | S                |
|      | <b>C.D (P=0.05)</b> | <b>7.41</b>           |                  | <b>7.61</b>           |                  | <b>8.69</b>           |                  |
|      | <b>SEm(±)</b>       | <b>2.61</b>           |                  | <b>2.68</b>           |                  | <b>3.06</b>           |                  |
|      | <b>C.V.</b>         | <b>9.00</b>           |                  | <b>9.47</b>           |                  | <b>10.90</b>          |                  |

**Table 4. Categorization of clusterbean genotypes/varieties according to disease reaction against bacterial blight (pooled) under artificial conditions**

| Genotypes/varieties  | Category of infection | Per cent leaf area affected | Host reaction               |
|--|-----------------------|-----------------------------|-----------------------------|
| Nil  | 0                     | 0 or less than 1.0          | Immune (I)                  |
| RGC-1066 (01)  | 1                     | 1-10%                       | Resistant (R)               |
| RGC-563, RGC-1002, RGC-1017, RGC-1031, GAUG-1304, CAZG-15-7, RGr-16-3-3, RGr-16-3-4, RGr-16-5-1, RGr-16-5-2, RGr-16-9-3, RGr-16-9-4, RGr-17-4-7-2, RGr-17-4-3-2, RGr-19-19-1 (15)  | 2                     | 10 - 25%                    | Moderately Resistant (MR)   |
| RGC-936, RGC-1003, RGC-1055, RGC-1033(ch), CAZG-15-5, , RGr-16-3-1, RGr-16-3-2, RGr-16-4, RGr-16-5-3, RGr-16-5-5, RGr-16-9, RGr-16-9-1, RGr-16-9-2, RGr-16-9-5, RGr-16-11-1, RGr-19-11-1, RGr-19-16-1, RGr-19-18-1, RGr-19-12-1 (19)   | 3                     | 25 -50%                     | Moderately Susceptible (MS) |
| RGC-963, RGC-986, GAUG-1502, GAUG-1501, HG-2-20(ch), X-10, HG-563(ch), RGr-16-3 RGr-16-3-5, RGr-16-3-6, RGr-16-3-7, RGr-16-5, RGr-16-5-4, RGr-16-8, RGr-16-9-6, RGr-16-11, RGr-16-11-2, RGr-16-11-3, RGr-17-1, RGr-17-2, RGr-17-4-4-1, RGr-19-1-1, RGr-19-12-2, RGr-19-19-2 (24) | 4                     | 50 - 75%                    | Susceptible (S)             |
| Nil  | 5                     | More than 75%               | Highly Susceptible (HS)     |

Similarly results also reported at S K Nagar, Gujarat several promising lines with enhanced disease resistance against bacterial leaf blight (GAUG 9406, GG 1, RGC 1027), *Alternaria* blight (GAUG 9406, GAUG 9005, GAUG 9003 and GC 1) and root rot (GAUG 9406, GG 1 and HGS 844) have been identified [17].

Similarly finding also reported by Lesly [18] screened 169 genotypes to bacterial blight during Kharif 2004 in Dharwad with 56, 27, 47, 22 and 17 resistant, moderately resistant, moderately susceptible, susceptible and highly susceptible reaction, respectively. Resistant genotypes such as IC202823, IC257420 and IC97767 showed high seed yield, harvest index and number of pods per plant.

#### 4. CONCLUSION

This study represents of two inoculation methods and fifty-nine clusterbean cultivars. further testing of these inoculation methods as well as

comparison with others on more clusterbean cultivars will be necessary to identify clusterbean cultivars with resistance to *X. a. pv. cyamopsidis* inoculation load and both methods combine gave highest disease severity index. In screening out of fifty-nine varieties, only 1 variety were found completely resistant, 17 moderately resistant, 17 moderately susceptible, 24 susceptible against bacterial blight disease.

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#### COMPETING INTERESTS

Authors have declared that no competing interests exist.

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