

Effect of Black Gram Leaf Crinkle Virus Infection on Seed Set and Distribution of Virus in the Seeds

By

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ABSTRACT

Black gram leaf crinkle virus induced pollen sterility which ranged from 13.64 to 72.09 per cent depending on the variety of black gram. The significance of pollen sterility as a limiting factor of yield is discussed. The study on distribution of BLCV in the seeds of black gram revealed that the virus was present in the plumule, radicle and cotyledons of the germinating seed but not in the seed coat. The virus was found at a higher frequency in the plumule.

INTRODUCTION

The transmissibility of black gram leaf crinkle virus (BLCV) through seeds (Kolte and Nene, 1972) and the factors affecting seed transmissibility of BLCV (Narayanasamy and Jaganathan, 1974) have been reported. The present study was taken up to find out the effect of virus infection on seed set and the distribution of BLCV in black gram seeds and the results are presented here.

MATERIALS AND METHODS

The fertility status of pollen from healthy and BLCV - infected plants of black gram (*Phaseolus mungo* L.) varieties CO 1, CO 2, PLS 364 and *Urd Ujjaini* was assessed to find out the possible effect of virus infection. The pollen grains were stained with acetocarmine-glycerine mixture and the percentage of stainable pollen was calculated.

The seeds from BLCV - infected CO 1 black gram plants were placed on moist filter paper kept in sterilized petri-dishes at the rate of 5 seeds per petri-dish. The samples were drawn after 72, 96 and 120 hours after soaking the seeds. The seed coat, plumule, radicle and cotyledons from each seed were separated and crushed separately with two drops of 0.1 M phosphate buffer at pH 7.0. The inoculum thus prepared from different tissues was inoculated on separate 10-day old healthy test plants of CO 1 black gram. The presence of BLCV in different tissues was detected, when the inoculated test plants developed symptoms of infection.

RESULTS AND DISCUSSION

Among the various factors responsible for sterility of plants, the pollen sterility is considered to be important. The stainability of pollen with acetocar-

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mine-glycerine mixture was taken as the basis of fertility of pollen from different varieties of black gram and the data are presented in Table 1. The percentages

TABLE 1. Effect of BLCV on pollen fertility

| Variety | | No. of stained pollen/100 | | Percentage of pollen sterility |
|-------------|------|---------------------------|----------|--------------------------------|
| | | Healthy | Diseased | |
| CO 1 | 1 | 95 | 19 | 80.00 |
| | 2 | 92 | 29 | 68.48 |
| | 3 | 98 | 33 | 66.66 |
| | Mean | | | 71.60 |
| Urd Ujjaini | 1 | 93 | 24 | 74.19 |
| | 2 | 96 | 25 | 73.96 |
| | 3 | 91 | 29 | 68.13 |
| | Mean | | | 72.09 |
| CO 2 | 1 | 95 | 81 | 14.74 |
| | 2 | 96 | 84 | 12.50 |
| | 3 | 95 | 82 | 13.68 |
| | Mean | | | 13.64 |
| PLS 364 | 1 | 91 | 72 | 20.88 |
| | 2 | 94 | 80 | 14.89 |
| | 3 | 90 | 75 | 16.67 |
| | Mean | | | 17.48 |

of pollen sterility was very high in the case of CO 1 (71.6) and Urd Ujjaini (72.09), while in CO 2 and PLS 364 the percentage of sterile pollen was considerably low.

The BLCV has been observed to cause different degrees of sterility depending on the age of plants at the time of infection. The earlier the plants is infected the more complete will be the sterility of the plant. Even late infection by BLCV has been noticed to cause

high percentage of pollen sterility. The transmission of plant viruses through pollen has been reported by many workers (Crispin *et al.*, 1961; Ryder, 1964; Gilmer, 1965; Williams and Smith, 1967). However, in all these cases the fertility of the pollen was not presumably affected by the viruses. The fact that BLCV could cause considerable sterility of pollen in certain varieties of black gram, assumes greater significance in self-pollinated crops like black gram in which pollen sterility leads to heavy reduction in seed set and consequent loss in yield.

The data presented in Table 2 show that 40 per cent of the seeds tested carried BLCV. The presence of BLCV in any one or more tissues of the seeds could be detected when the test plant inoculated with the inoculum prepared from those tissues, developed symptoms of the disease. None of the plants inoculated with the inoculum from the seed coat developed the symptoms indicating the complete absence or undetectable titre of BLCV in the seed coat.

Among the other tissues namely plumule, radicle and cotyledons, the plumule contained BLCV in 8 of 12 seeds and of these the presence of virus only in plumule was detected in 4 seeds, while in other 4 seeds the virus was present either in the radicle or cotyledons in addition to the plumule. The presence of BLVC in radicle was detectable in 7 of 30 seeds tested and its presence only in radicle was observed in one seed. The cotyledons of the

TABLE 2. Distribution of BLCV in black gram

| Period after soaking | Seed No. | Plumule | Radicle | Cotyledons |
|----------------------|----------|---------|---------|------------|
| 72 hours | 1. | * | — | — |
| | 2. | * | — | — |
| | 3. | — | — | — |
| | 4. | — | — | — |
| | 5. | * | — | — |
| | 6. | — | — | — |
| | 7. | — | — | — |
| | 8. | * | * | — |
| | 9. | — | * | — |
| | 10. | — | — | — |
| 96 hours | 1. | — | — | — |
| | 2. | — | — | — |
| | 3. | * | — | — |
| | 4. | — | — | — |
| | 5. | — | * | — |
| | 6. | — | — | — |
| | 7. | * | * | — |
| | 8. | — | — | — |
| | 9. | — | * | * |
| | 10. | — | — | — |
| 120 hours | 1. | — | — | — |
| | 2. | — | — | — |
| | 3. | * | * | — |
| | 4. | — | — | — |
| | 5. | * | — | * |
| | 6. | — | — | — |
| | 7. | — | * | * |
| | 8. | — | — | — |
| | 9. | — | — | — |
| | 10. | — | — | — |

* BLCV present — BLCV absent

seeds yielded BLCV in 4 out of 30 seeds and it is significant that none of the seeds showed the presence of BLCV only in the cotyledons.

When the distribution of BLCV in the seeds examined after different periods of soaking is considered, 4 out of 5, 2 out of 4 and 2 out of 3 seeds showed the presence of BLCV in the plumules of seeds soaked 72, 96 and 120 hours respectively. As the period after soaking increased, the percentages of seeds in which the tissues, in addition to plumule, yielding the virus increased. Forty per cent of infected seeds showed the presence of BLCV in the radicle of the seeds tested 72 hours after soaking. The percentage increased to 75 and 66.66 in the case of seeds tested after 96 and 120 hours after soaking.

Plant viruses may be carried by the seeds externally like tobacco mosaic virus in tomato seeds (Taylor *et al.*, 1961; Broadbent, 1965) or internally as in the case of other viruses (Fulton, 1964). In the present study the evidences that BLCV was carried only internally, as the inoculum prepared from the seed coat did not give any positive transmission. The embryo and the endosperm appear to carry BLCV as evidenced from the positive transmission obtained using the inoculum from plumule and cotyledons. In the germinating seeds there may be a quick downward movement of BLCV from the plumule to other plant parts. This may be the probable reason for the observed increase in the percentage of BLCV yielding radicle, as the period after soaking the seeds increased.

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