

## Effect of Leaf Crinkle Virus infection on Nodulation, Productivity and Yield of Black Gram *Vigna mungo*

\*Sanjay Srivastava, D.K.Srivastava<sup>1</sup> and Alok K. Singh<sup>2</sup>

Botany Department,  
Harish Chandra P.G. College,  
VARANASI-221001 (U.P.) INDIA

<sup>1</sup>Botany Department,  
D.C.S.Khandelwal P.G. College,  
MAU-275101 (U.P.) INDIA

<sup>2</sup>Botany Department,  
C.M.P. Degree College,  
PRAYAGRAJ-211002 (U.P.) INDIA

\*Corresponding Author

E-mail: sanjayhpcpg@rediffmail.com

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### ABSTRACT

In the present study, effect of Urd bean leaf crinkle virus (ULCV) infection on the nodulation, productivity and yield of black gram (*Vigna mungo*) was investigated. Virus infection was shown to reduce the number, size and weight of nodules in both the cultivars of black gram viz., T-9 and IPU 94-1. Primary productivity was also adversely affected due to virus infection. Gross and net production rates decreased whereas respiratory loss increased in infected leaves. Virus infection also reduced the grain yield which ranged from 6.81 to 66 percent over healthy plants when inoculated at different growth stages. Decrease in the number of pods per plant and 100 grain weight was recorded. Early inoculated plants showed higher yield loss than the mid and late inoculated ones.

Figures : 07

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KEY WORDS : Black gram, leaf crinkle virus, Nodules, Primary productivity, Yield

### Introduction

Black gram (*Vigna mungo*) is an important pulse crop of many Asian countries including India. In India, it is mainly consumed in form of 'daal' (whole or split, husked or unhusked). It is also used as a nutritive fodder especially for milch animals and also as green manure. Uttar Pradesh stands first in the production of urd bean (16.98 %) followed by Andhra Pradesh (16.75 %) and Madhya Pradesh (15.07 %). During a survey of village areas of four districts of eastern Uttar Pradesh, viz., Azamgarh, Gorakhpur, Maharajganj and Varanasi, black gram crops were found to be infected with several viral diseases. These included, leaf crinkle, leaf curl and mosaic. In the terai zone of district Gorakhpur and Maharajganj, the predominant and the most widespread was the leaf crinkle disease. The percent disease

incidence ranged from 28 to 85 although in Tamil Nadu, recent studies<sup>9</sup> have revealed a disease incidence of only 11.4 to 28.3%. The infected plants stunted, showed severe crinkling and distortion of leaves and bore only a few and smaller pods. The disease also damaged the root system and nodulation upto a great extent. Present investigation was carried out to generate information on nodulation, productivity and yield of black gram plants infected with leaf crinkle virus.

### Materials and Methods

#### Nodulation studies

For nodulation studies, 60 seedlings of each cultivar of Black gram viz., T-9 and IPU 94-1 were raised in earthen pots. Both the lots were inoculated with virus isolate. Inoculations were performed at 10 days after

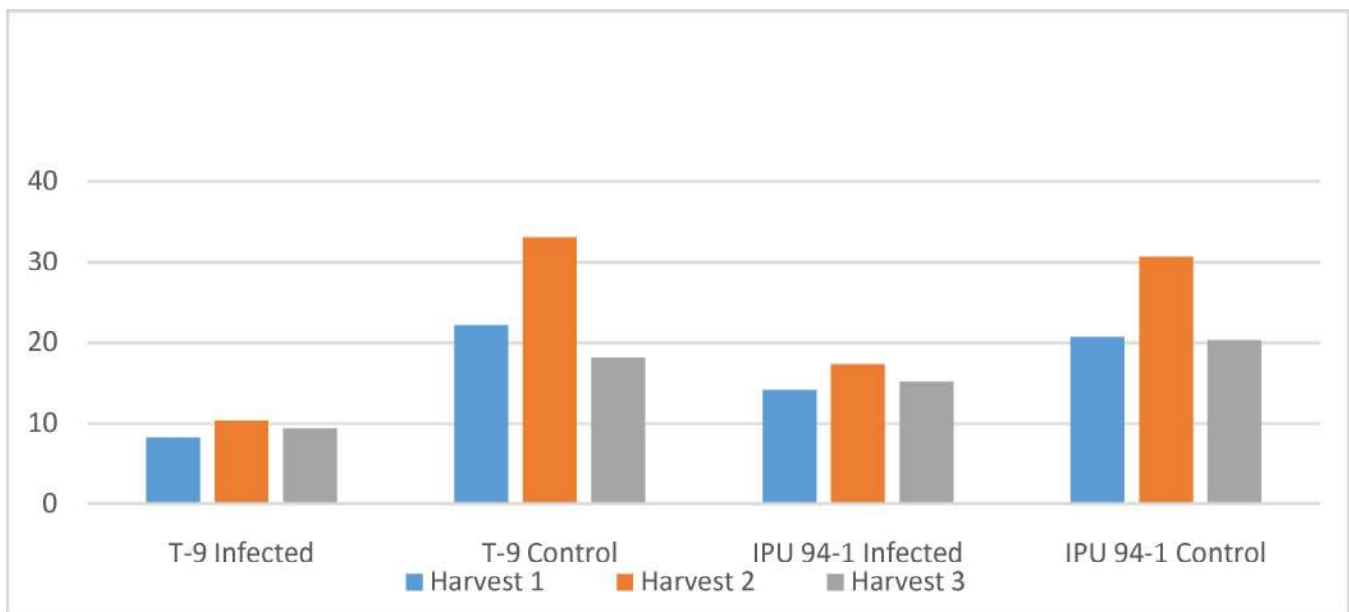
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**TABLE -1 : Primary productivity of black gram cv. T-9 at different periods of leaf crinkle virus inoculation (H = Healthy; D = Diseased)**

Initial and final weight of leaf discs (g <sup>-2</sup> )	Days after inoculation (DAI)									
	10		20		30		40		50	
	H	D	H	D	H	D	H	D	H	D
Initial	26.05	30.84	26.98	32.00	28.34	35.15	32.42	40.21	30.97	38.36
5 hrs. in sun	28.51	32.77	29.72	34.05	31.15	37.54	35.89	42.93	33.89	41.00
5 hrs. in dark	23.88	28.28	24.38	29.27	25.58	32.25	29.54	37.21	28.07	35.29

**Productivity (mg m<sup>-2</sup> h<sup>-1</sup>)**

Net Production	246	193	274	205	281	239	347	272	292	264
Respiratory loss	217	256	260	273	276	290	288	300	290	307
Gross Production	463	449	534	478	557	529	635	572	582	571



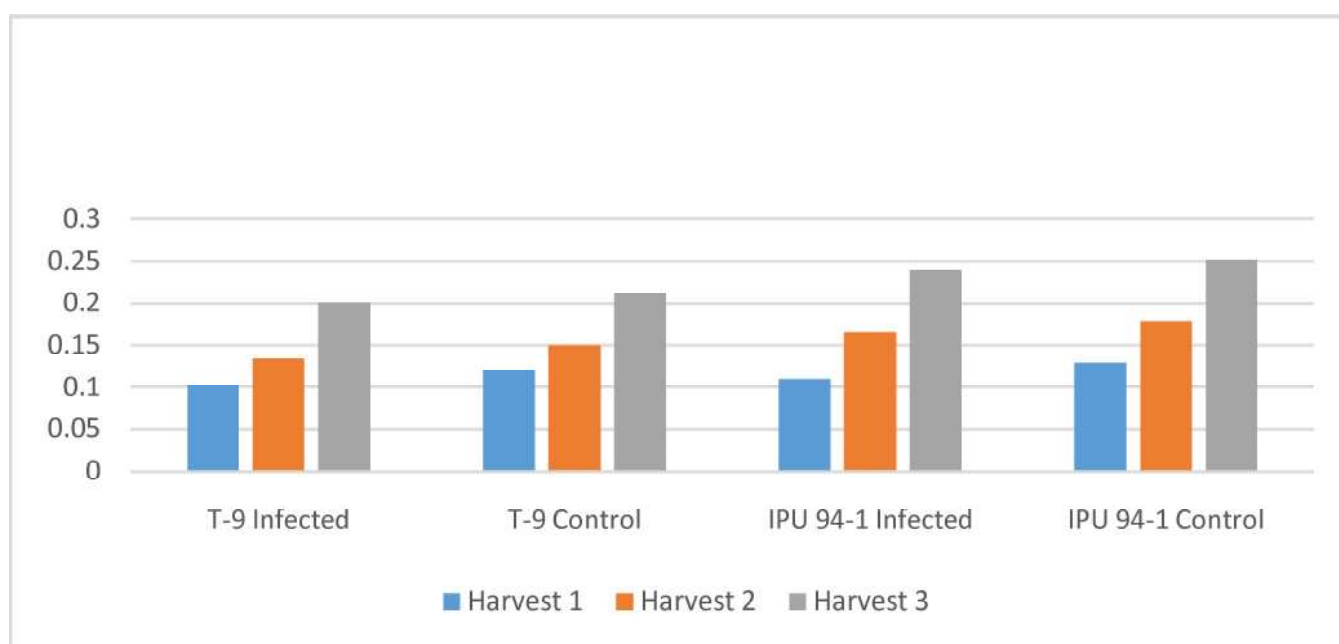
**Fig. 1 : Effect of leaf crinkle virus infection on nodule number/plant in black gram cultivars at different periods of harvest.**

sowing (DAS). Control plants of each cultivar were also raised simultaneously. Root nodules of the inoculated and control plants were harvested at periodic intervals.

Twenty plants of each cultivar were dug out at 35 days after inoculation (DAI). The harvesting of the next lot of 20 plants of both the cultivars was performed at 55 days

**TABLE -2 : Effect of ULCV infection on the yield of Urd bean cv. T-9 H/C = Healthy/Control; EI = Early Inoculation; MI = Mid Inoculation; LI = Late Inoculation**

Category	Av. No. of pods/plant	Av. No. of seeds/pod	Av. Length of pod/plant (cm)	100 grain weight (g)	Yield/plant	% Decrease over healthy
H/C	15.6	7.6	4.1	4.95	5.87	
EI	6.5	7.3	3.15	4.2	1.99	66
MI	9.7	7.4	3.96	4.75	3.4	42.07
LI	15.0	7.6	3.99	4.8	5.47	6.81

**Fig. 2 : Effect of virus infection on nodule size (average diameter in cm) in black gram cultivars at different periods of harvest**

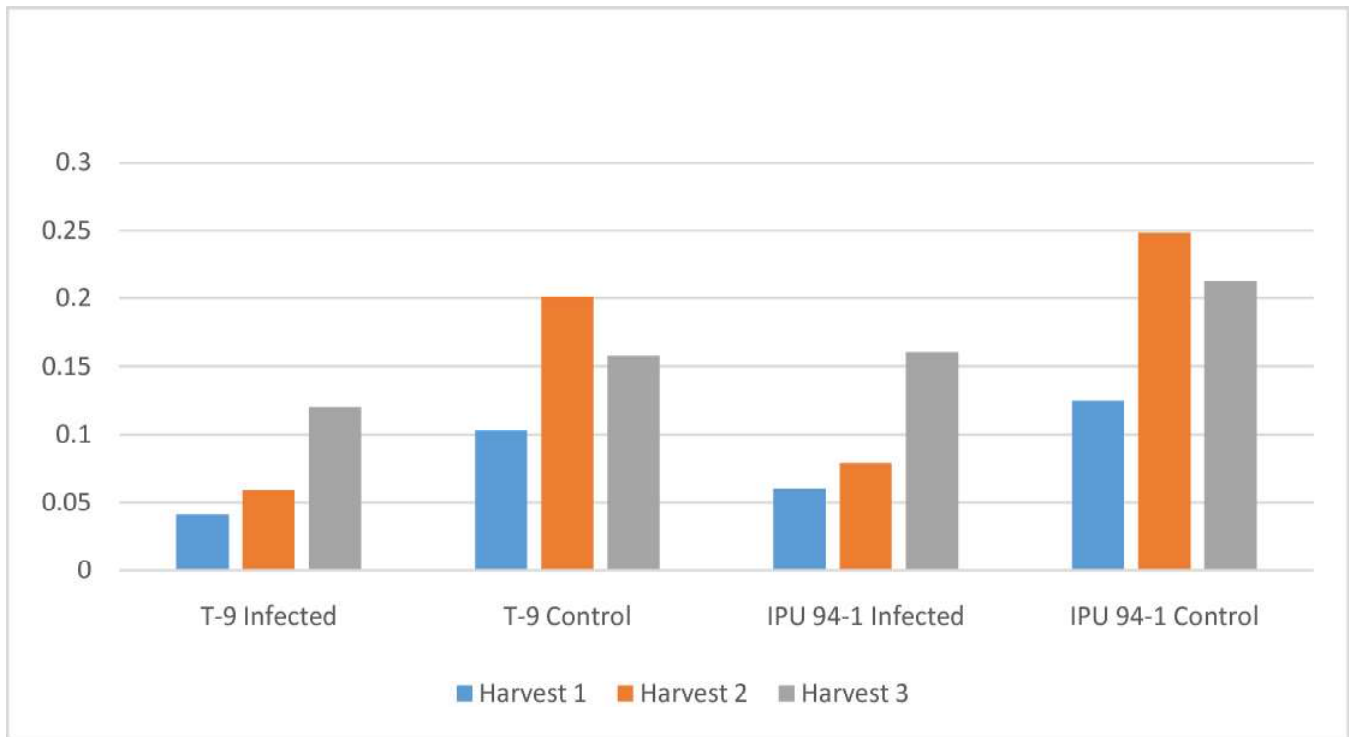
after inoculation.

Plants were dug out carefully to avoid damage of the root system. The initiation of root nodules on primary and secondary roots of urd bean was recorded 3 to 30 days after inoculation (DAI). The nodules were carefully separated from the roots after removing the soil in running water. Nodules obtained from each group of plants were counted, weighed and their diameters measured. All the experiments were performed in insect-proof conditions.

### Primary productivity

Throughout the experiment, for systemic multiplication, *Phaseolus mungo* cv. T-9 was used as host plant and urd bean leaf crinkle virus (ULCV) as the virus. Clay pots (30 cm diameter) containing a mixture of loam

and compost (1:1) were used to grow the test plants. These plants were watered on a regular basis. The seedlings were raised in insect-proof chamber. One hundred seedlings of black gram (each one 10 days old) were inoculated with ULCV. An equal number of seedlings of the same age were inoculated with phosphate buffer (pH 7; 0.1 M) to serve as healthy control. Leaf samples from healthy and virus infected urd bean plants were collected separately on the 10<sup>th</sup>, 20<sup>th</sup>, 30<sup>th</sup>, 40<sup>th</sup> and 50<sup>th</sup> day after inoculation (DAI). Primary productivity was measured in accordance with the method developed by earlier workers<sup>11</sup>. Intact leaves from both healthy and virus infected urd bean plants were collected between 8 a.m. to 10 a.m. in the morning. The sampled leaves were ensured to be of approximately uniform size. In case of virus infected leaves, only those



**Fig. 3 : Effect of virus infection on fresh nodule weight (g) / plant in black gram cultivars at different periods of harvest**

having well marked and uniform disease symptoms were collected. In order to maintain their turgid state, leaves were immersed in water for 10 to 15 minutes. Using a cork-borer, 150 discs of 1 cm diameter each, were cut both from the leaves of healthy and virus infected plants. These discs were spread out in three petri dishes. Each petri dish contained 50 discs placed on moist cotton pad to prevent injury due to high temperature. Out of these three petri dishes, the first one was kept in normal sunlight for 5 hrs. The second was placed in dark chamber with 40% KOH kept in a beaker, for 5 hrs. The contents of the third petri dish were oven dried at 80°C immediately and their weights recorded. After 5 hrs. contents of the first petri dish (kept in sunlight) and

second (kept in dark chamber) were oven dried at 80°C and their weights were recorded. The difference between the dry weights of the leaf discs of the first and third petri dishes gave increase in photosynthetic dry matter (net production) and that of the second and third petri dishes gave decrease in dry matter due to respiratory loss. The sum total of net production and respiratory loss represented gross production.

Gross Production (GP) = Net Production (NP) + Respiratory Loss (R)

### Yield

For estimating the effect of virus infection on the yield of black gram, plants of variety, cv. T-9 were grown in experimental fields having sandy loam soil. The experiment was performed in randomized block<sup>6</sup> with three replications. Three plots (3x6 meters) were laid out and 40 seeds were sown in each plot in 8 rows, 75 cm apart. Each row had 5 plants. The plants were subjected to following treatments. Ten plants were used per treatment being replicated thrice.

1. Control-healthy plants : treated with phosphate buffer only
2. Early inoculation : 10 days old plants inoculated with virus
3. Mid-term inoculation : 20 days old plants inoculated with virus
4. Late inoculation : 30 days old plants inoculated with virus



**Fig. 4 : Seeds of Black gram**



**Fig. 5 : ULCV infected leaf of black gram**

In order to control insect infection, 0.1 percent Malathion solution was sprayed once a week. Watering of the plants was done on a regular basis, once a week.

Following observations were made regularly:

1. Number of fruits/plant
2. Fruit (pod) size/plant
3. Number of seeds/pod
4. Seed weight (100 grains)
5. Yield/plant
6. Percent decrease over healthy

## Results and Discussion

### Effect on nodulation

In the present investigation, leaf crinkle virus infection was observed to reduce the number, size and fresh weight of nodules in black gram cultivars (Figs. 1, 2 and 3). Nodules appeared approximately at the same time in healthy (control) and inoculated plants. In ULCV infected urd bean cultivars, maximum reduction in nodule number was observed in T-9 followed by IPU 94-1. In both the cultivars, decrease in nodule number in 1<sup>st</sup>, 2<sup>nd</sup> and 3<sup>rd</sup> harvest ranged between 25.6 to 68.7 percent. In present study, number of nodules increased upto the second harvest and then decreased in the third harvest (Fig. 1). Size and weight of fresh nodules had a steady increase upto the third harvest (Figs. 2, 3). In earlier study<sup>3</sup> nodule number and their fresh weights were reported to have been reduced by 53.7 and 55.4 percent respectively. These results are in conformity with earlier reports on nodulation in other host-virus combination studies<sup>4,16</sup>. Although study of nitrogen fixing efficiency in virus infected plants was not part of this study but earlier studies particularly in soybean mosaic virus disease<sup>16</sup> have shown that reduced nodule number and size is usually associated with increased nitrogen fixing



**Fig. 6 : Nodulation in healthy/control plant**

efficiency in virus infected plants.

Nodule reductions (Figs. 6 & 7) were presumably the result of virus multiplication which has brought about physiological changes such as reduced photosynthesis or increased respiration<sup>1,2,5</sup>, auxin imbalance<sup>15</sup> and disturbed enzyme levels *etc.* These changes had directly or indirectly affected the symbiotic relationship between *Rhizobium* and black gram.

### Effect on Productivity and Yield

The findings as specified in (Table-1), suggest that the rate of net production (NP) and gross production (GP) decreased and that of respiratory loss (L) increased in virus infected black gram leaves. It was observed that in healthy and virus infected plants, respiratory loss increased with the age of the plant, net production and gross production on the other hand increased upto 40<sup>th</sup> day of inoculation and then decreased.

The results are similar to findings of earlier



**Fig. 7 : Nodulation in virus inoculated plant**

studies<sup>7,8,12,13</sup> in virus infected crops of barley, frenchbean, *Nicotiana* and cowpea respectively. It was observed that the leaf crinkle infection had significantly reduced the number of pods per plant, seeds per pod, 100 grain weight and yield per plant (Table-2). The virus infected plants produced small, malformed and shrivelled seeds.

Early inoculated plants, showed a greater reduction in yield parameters as compared to mid and late inoculated ones. Early inoculation resulted in a yield loss of upto 66% whereas mid and late inoculations had reduced the yield by 42.07% and 6.81% respectively in virus infected plants. These findings are very much in coherence with findings of other workers in various other host-virus combinations<sup>10,14</sup>.

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Virus infection affects the yield by reducing the photosynthetic rate or by enhancing respiration. But a cumulative effect of both these factors seems to be the most plausible cause.

### Conclusion

This study reveals the enormous devastation caused by leaf crinkle virus in urd bean plants. Not only it results in poor nodulation (reduced number, size and weight of nodules) but the productivity and yield are also significantly reduced. Seeds, which form the edible part of the plant are shrivelled and malformed with very poor market value. In view of such a great loss due to ULCV infection, effective control measures involving both cultural practices and resistant varieties are required.

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