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Urdbean Leaf Crinkle Disease- Assessment of Seed Transmissibility and Its Effect on Yield and Seed Quality in Urdbean [*Vigna mungo* (L.) Hepper]

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ABSTRACT

Background: Urdbean leaf crinkle disease (ULCD) is one of the most devastating biotic stress factors which is responsible for drastic yield loss in urdbean [*Vigna mungo* (L.) Hepper]. The etiology of ULCD is still unknown and there are conflicting reports on its transmission through seed, insect vectors, grafting and infected sap.

Methods: Per cent seed transmission of ULCD had been studied during *kharif*, *rabi* and summer during the year 2019-2020 under natural and insect proof conditions. The effect on yield and seed quality parameters due to ULCD infection was studied. Urdbean cultivar VBN 8 was used in the present study.

Result: The per cent seed transmission of ULCD was recorded ranging from 52.88 to 71.64% under natural conditions. In the insect proof glasshouse condition, the seed transmission was 83.62%. All the yield and seed quality parameters were adversely affected upon ULCD infection. The reduction in yield was recorded as 64.36% over the uninfected healthy plants. In ULCD infected seeds, the germination was reduced (84%) as compared to healthy seeds (92%).

Key words: Insect proof condition, Natural conditions, Seed quality parameters, Seed transmission, ULCD, Urdbean, Yield parameters.

INTRODUCTION

Vigna mungo (L.) Hepper, commonly known as urdbean or blackgram is an important legume crop grown in all the pulse growing districts of Tamil Nadu. The crop is affected by important virus diseases *viz*., mungbean yellow mosaic, leaf curl, stem necrosis and leaf crinkle apart from few fungal and bacterial diseases. Among the virus diseases, urdbean leaf crinkle disease (ULCD) is considered as the most devastating one since it drastically reduces seed quality and quantity. Though ULCD had been first reported from Delhi by Nariani (1960), the etiology is still unknown.

The disease causes symptoms *viz*., crinkling, puckering, curling, rugosity of leaves, enlargement of leaf lamina, stunting and malformation of floral parts (Kanimozhi *et al*., 2009). The infected plants produce sterile flowers and few pods which lead to decrease in grain yield from 35 to 81% depending upon the genotypes, time of infection, growing season and environmental conditions (Ashfaq *et al*., 2008; Sravika *et al*., 2018; Sravika *et al*., 2019; Sathya *et al*., 2022; Latake *et al*., 2020). The ULCD is mainly transmitted through infected seeds as reported by several workers (Narayanasamy and Jaganathan, 1975; Beniwal *et al*., 1983b; Beniwal and Chaubey. 1984; Dubey and Sharma, 1985; Mishra *et al*., 1994; Ahmad *et al*., 1997; Reddy *et al*., 2005; Kanimozhi *et al*., 2009). It is also transmitted through insects (Narayanasamy and Jaganathan, 1973; Brar and Ratual, 1987; Bashir *et al*., 2005b; Sravika *et al*., 2018), grafting (Bashir *et al*., 2005b) and sap inoculation (Biswas *et al*., 2012; Dubey *et al*., 2020).

Depending upon the genotypes and time of infection of ULCD has been reported to decrease grain yield from 35 to 81% (Bashir *et al*., 1991). The disease affects both vegetative growth and yield attributes of urdbean plants (Kadian, 1982; Kadian, 1994; Perane *et al*., 1997; Sharma *et al*., 2007; Kanimozhi *et al*., 2009, Sravika *et al*., 2018). Since the incidence of leaf crinkle disease is increasing in recent years, it was considered as important to understand the role of seed transmission in the yield loss caused by this dreadful disease. With this background, a systematic study was undertaken to assess percent seed transmission of urdbean leaf crinkle disease (ULCD) and its effect on yield and seed quality parameters were studied.

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MATERIALS AND METHODS

Collection of ULCD infected seed samples

The symptomatic leaf crinkle infected and asymptomatic,

apparently healthy plants of cultivar Cv. VBN8 were tagged with labels during summer 2019 in research farms of Tamil Nadu Agricultural University (TNAU), Coimbatore (Fig 1a). At the stage of maturity, the seeds from infected and healthy plants were collected, cleaned and used for further study.

Seed transmission under natural conditions

To assess seed transmission of ULCD, the collected seeds were sown in successive season *kharif*, *rabi* and summer of the year 2019-2020 in the research farm of Department of Agronomy, TNAU, Coimbatore. The seeds were sown with plant to plant and row to row spacing of 30cm and 10cm respectively in field with three replications. The development of ULCD symptoms *viz*., crinkling, malformation of floral buds was observed periodically. The transmission data was subjected to statistical analysis by CRD Design. The percent seed transmission based on the visual symptom was calculated 45 days after sowing (DAS) using the following formula:

$$\text{Seed transmission (\%)} = \frac{\text{Number of plants showing ULCD symptoms}}{\text{Total number of plants}} \times 100$$

Seed transmission under glasshouse conditions

The collected seeds from both ULCD infected and asymptomatic healthy plants were sown in insect proof chambers of PL 480 glasshouse in Department of Plant Pathology, TNAU, Coimbatore with three replications. In each replication, 100 seeds were sown in pots @ 10 seeds/ pot with sterilized soil and the plants were kept under vector proof conditions. The plants were observed periodically for symptom development. The data was subjected to statistical analysis by CRD design.

Effect of ULCD incidence on yield attributes of urdbean

An experimental field trial was conducted at the research farm of Department of Agronomy, TNAU, Coimbatore to study influence of ULCD incidence on yield parameters. The collected seeds of summer 2019 were sown on *kharif* 2019 with plant to plant and row to row spacing of 30cm and 10cm respectively. The crop was raised as per the standard agronomic practices. Twenty five diseased plants showing distinct crinkling and healthy plants with no signs of infection were randomly selected, tagged and harvested individually at maturity. The data were recorded on various yields contributing factors *viz*., plant height (cm), internode length (cm), number of branches/plant, number of leaves/plant, number of pods/ plant, pod length (cm), seeds/pod, yield/ plant and 100 seed weight/plant (g). For scanning electron microscopy (SEM) studies, image of healthy and infected seeds were captured with Scanning Electron Microscope (Quanta 250) available in the Department of Nano Science and Technology, TNAU, Coimbatore. The yield loss was calculated by the following formula and result were compared by t-test.

$$\text{Per cent yield loss} = \frac{\text{Yield per healthy plant - Yield per infected plant}}{\text{Yield per healthy plant}} \times 100$$

Effect of ULCD infection on seed quality of urdbean

The germination percentage and vigour index of seeds were collected from healthy and leaf crinkle infected plants was conducted by using paper roll towel method (Agarwal and Dadlani, 1992). Twenty five seeds placed on double layer of germination paper were rolled carefully and allowed to germinate at 25±1°C for 7 days and the germination percentage was recorded. The vigour index of the seedlings was calculated by the following formula given by Abdul-Baki and Andersan (1973). The data was subjected to statistical analysis by CRD design.

$$\text{Vigour index} = [\text{Root length (cm)} + \text{Shoot length (cm)}] \times \text{Germination percentage (\%)}$$

RESULTS AND DISCUSSION

Assessment of seed transmission of ULCV under natural conditions

Seed transmission of ULCD was evaluated under field conditions at Department of Agronomy, TNAU, Coimbatore during *kharif*, *rabi* and summer season of the year 2019-2020 (Fig 1b). The symptoms were observed first on the third trifoliate leaves (35 DAS) as crinkling of leaves, stunting



Fig 1a: Tagging of ULCD infected plants for collection of infected seeds.



Fig 1b: Urdbean plants infected by ULCD under field condition.

of plants, malformation of floral parts and which lead to poor or no pod formation (Fig 1c). Table 1 shows the percent seed transmission of seeds collected from ULCD infected plants was upto 71.64, 52.88 and 68.23% in *kharif*, *rabi* and summer respectively. The per cent seed transmission of ULCD in urdbean was 83.62% and there was no symptom development in plants raised from seeds of asymptomatic plants under insect proof conditions (Table 2). The results confirmed with several workers on seed transmission of ULCV (Mishra *et al*., 1994; Rao and Reddy, 2005, Himanshu and Vishunavat, 2006; Sharma *et al*., 2014; Latake *et al*., 2020; Dubey *et al*., 2021).

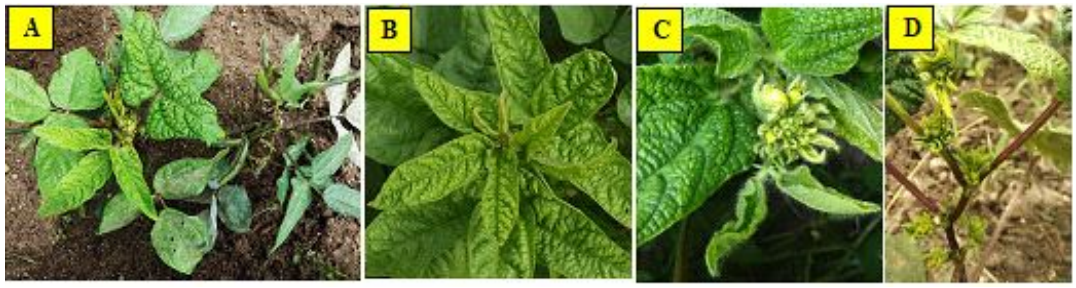
The seed transmission of ULCV from naturally infected plants was 17.6% (Dubey and Sharma, 1985). ULCD is transmitted through seeds and the rate of transmission was 2.7 to 46% (Ahmad *et al*., 1997). According to Reddy *et al*. (2005), the seed lots infected by ULCV showing 2.0 to 3.6% of seed borne infection recorded 45.2 to 86.5 per cent disease incidence under field conditions. The percentage of seed transmission of ULCV in urdbean and mungbean was 34.50 and 26.25% respectively (Kanimozhi *et al*., 2009).

Effect of ULCD infection on yield attributes of urdbean

Under natural conditions, yield parameters of all the components were adversely affected due to ULCD infection (Fig 2). Since the leaf crinkle symptoms ended up with severe malformations of inflorescence and poor pod set. The loss in yield was mainly due to decrease in number of pods/plants, pod length, number of seeds/pod and 100 seed weight (test weight) in ULCD infected plants compared to healthy plants. The reductions in various yield parameters are presented in (Table 3 and Fig 3). The reduction in plant height (23.67%), internode length (66.89%), branches/plant (37%), leaves/plant (19.92%), pod/plant (69.77%), pod length (20.48%), seeds/pod (37.50%), 100 seed weight (30.12%) of ULCD infected plants over healthy plants.

The ULCD infected plants of urdbean recorded with reduced yield/plant than healthy one (Table 2). The reduction in yield was 64.36%. The results were similarity with several workers (Kolte, 1971; Nene, 1973; Beniwal and Chaubey, 1979; Singh, 1980, Himanshu and Vishunavat, 2006; Mandhare *et al*., 2007).

Kadian (1982) reported a loss in yield which ranged from 2.82 to 95.17% due to leaf crinkle disease in blackgram cv. T9. Chattopadhay *et al*. (1986) reported the yield loss which ranged from 14.6 to 55.8% in 14 cultivars of greengram. Depending upon genotype and time of infection, the losses of grain yield due to urdbean leaf crinkle virus was 35 to 81% (Bashir *et al*., 1991). Perane *et al*. (1997) reported in grain yield loss due to LCV ranged from 20.43 to 24.21%. Sharma *et al*. (2002) reported a yield reduction ranged from 78 to 81% in ULCV infected blackgram plants.



A. Plants infected by ULCD (Right: Infected; Left: Healthy); B. Crinkling of leaves; C. malformation of floral buds; D. Stunting of plants.

Fig 1c: Symptoms of ULCD in blackgram.

Table 1: Assessment of seed transmission of ULCD during 2019-2020 under natural conditions.

Season	Total number of plants	No. of infected plants	% of seed transmission*	SEm ±	CD at 5%
Kharif 2019	2405	1723	71.64	0.67	4.08
Rabi 2019-20	1108	586	52.88	0.46	2.81
Summer 2020	1668	1138	68.23	0.52	3.18

*Mean of three replications.

Table 2: Assessment of seed transmission under insect proof conditions.

Particulars*	No. of seeds sown	No. of seeds germinated	No. of ULCD plants	% seed transmission**	SEm ±	CD at 5%
I	100	94	78	83.62	1.49	4.77
H	100	99	0	0.00	-	-

*H: Seeds collected from asymptomatic plants; I: Seeds collected from ULCD infected plants.

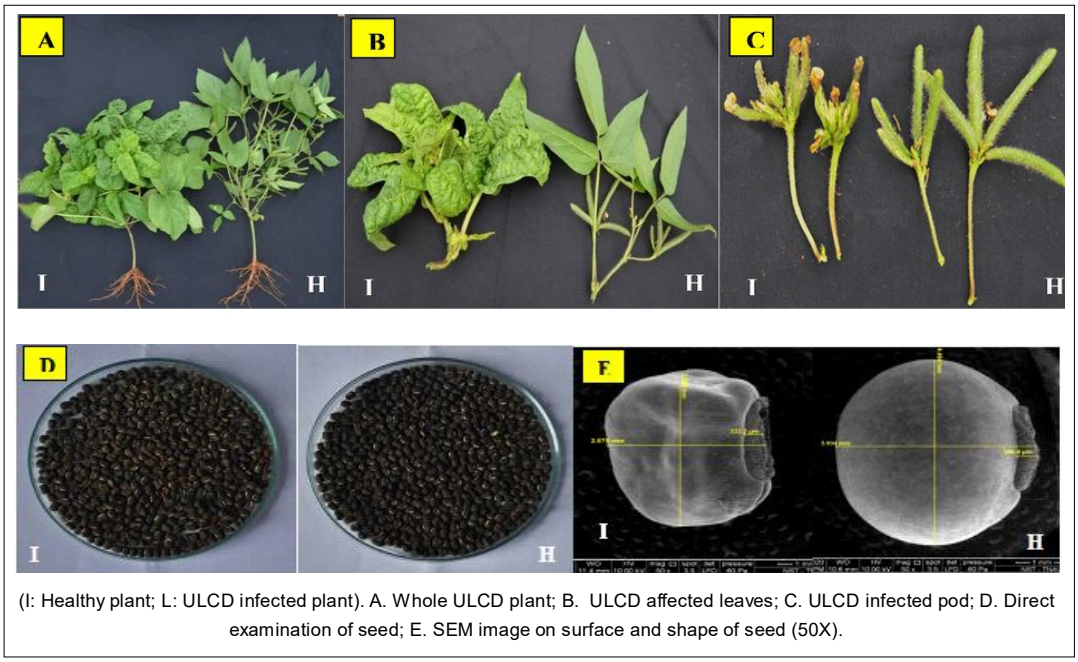
**Mean of three replications.

Sravika *et al*. (2018) reported a grain yield reduction of 85.67% upon ULCD infection.

Effect of ULCD infection on seed quality of urdbean

The results of the paper roll towel method carried out to assess the germination percentage and vigour index of healthy and ULCD seed were given in Table 4. The seeds from ULCD infected plants showed lower germination of 84.00% compared to healthy seeds (92.00%). The root length and shoot length were also found reduced in ULCD infected plant was 12.86% and 22.89% respectively compared to un-infected healthy plants.

The vigour index was reduced in ULCD infected seedling (1576.96) as compared to healthy seedling (2762.76). The results were agreed with findings of Mandhare *et al*. (1999) and Mandhare *et al*. (2007). The seed germination (45%)



(I: Healthy plant; L: ULCD infected plant). A. Whole ULCD plant; B. ULCD affected leaves; C. ULCD infected pod; D. Direct examination of seed; E. SEM image on surface and shape of seed (50X).

Fig 2: Effect of ULCD infection on growth parameters in urdbean.

Table 3: Effect of ULCD infection on yield attributes of urdbean under natural conditions.

Yield attributing parameters	Healthy	ULCD infected	% Reduction	t value*
Height/plant (cm)	22.53	17.20	23.67	4.86
Internode length (cm)	2.01	0.67	66.89	15.84
Branches/plant	6.67	4.20	37.00	5.98
Leaves/plant	16.4	13.13	19.92	2.75
Pods/plant	11.47	3.47	69.77	8.79
Pod length (cm)	5.53	4.40	20.48	5.26
Seeds/pod	5.87	3.67	37.50	7.43
100 seed weight (g)	5.27	3.68	30.12	13.08
Seed yield (g/plant)	26.70	9.50	64.36	10.88

*Significant at P value above 0.05.

Table 4: Effect of ULCD infection on seed quality parameters of urdbean.

Variety	Germination (%)		Shoot length (cm)		Root length (cm)		Vigour index (cm)	
	H	I	H	I	H	I	H	I
Vamban-8	92 (73.57)	64 (53.13)	14.78	12.88	15.25	11.76	2762.76	1576.96
% reduction	30.43		12.86		22.89		42.92	
SEm ±	1.78	0.74	1.69	1.62	1.83	1.56	57.48	25.31
CD at 5%	5.81	2.41	4.96	4.85	5.48	4.56	187.43	82.54

Figures in the parentheses are arcsine transformed values. Where, H: Healthy; I: ULCD Infected.

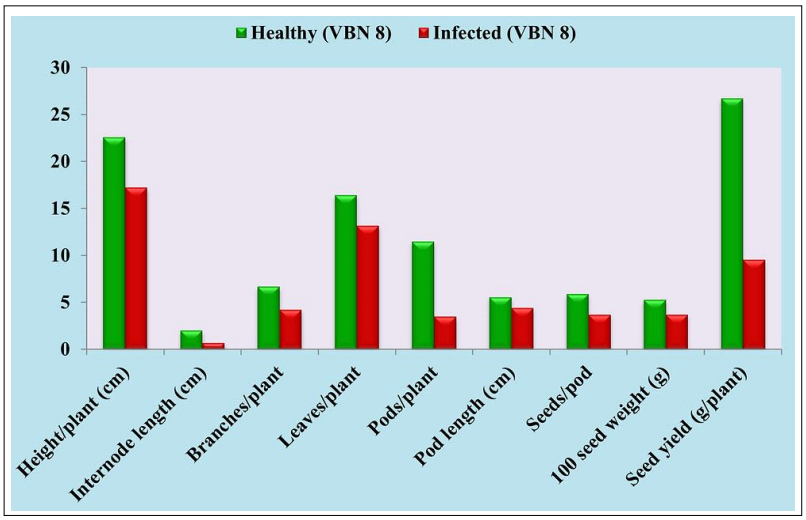


Fig 3: Effect of ULCD infection on yield parameters of urdbean.

and seed vigour (0.74OD) was reduced in ULCV infected seeds (Sharma *et al*., 2015). Kanimozhi *et al*. (2009) also reported significant reduction in seed germination (75%) and seed vigour (2376) in ULCV infected seeds compared to healthy seed.

CONCLUSION

The leaf crinkle disease is transmitted mainly through infected seeds which is responsible for wide spread occurrence of leaf crinkle disease. Currently, this dreadful disease highly influences the grain yield loss due to malformation of flowers and reduction of pod number. The seed borne inoculum was found to be responsible for adversely affecting the various yield parameters as well as yield and seed quality of urdbean. Therefore, the yield losses could be minimized by planting of healthy seed material and eliminating ULCD infected plants under field condition in order to maintain the healthy seeds in seed chain. Further work on the transmission of various agents *viz*., graft, sap, insect vector need to be carried out and influences of environmental factors *viz*., temperature, relative humidity are to be correlated with leaf crinkle disease development for devising effective management strategies.

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Conflict of interest

All authors declared that there is no conflict of interest.

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