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Screening of lentil germplasms for *Stemphylium* blight resistance in West Bengal condition

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ABSTRACT

Lentil (Lens culinaris Medik.) is one of the most nutritious and protein rich legume grown in a large scale in India. Stagnation in productivity of this legumehas been observed as it suffers from many diseases, Stemphylium blight being one of them can reduce the crop yield upto 62-100 per cent. An experiment was conducted in District Seed Farm(AB Block) of Bidhan Chandra KrishiViswavidyalaya, Kalyani, West Bengal to search resistant sources of lentil which will help in stress breeding programme in future. Seventy seven accessions obtained from AICRP-MULLaRP(BCKV centre) were screened under natural field condition along with artificial disease inoculation technique during rabi 2015-16 and 2016-17. As a result eleven genotypes namely VL151, RLG195, L4769, LL1397, DL14-2, VL126, RKL14-20, IPL334, L4710, PL210 and Precozwere identified as moderately resistant against the disease.These genotypes may be further utilized in Stemphylium blight resistance breeding programme in lentil.

Keywords **:** Lentil, resistant source, screening and *Stemphylium* blight

Lentil is an important rainfed *rabi* legume which is originated from the region between Afganistan, India and Turkistan (Barulina, 1930). It was cultivated in 52 countries on 30.5 million ha area with an annual production of 19.9 million tones and had a productivity of 654.44 kg ha^{-1} in 2014 (FAOSTAT, 2016). The yield of lentil is not very encouraging in India as it is lower than the world average. The reasons for low yield can be the occurrence of various biotic and abiotic stress factors at different growth stages. Lentil is reported to be suffered from many diseases like Sclerotinia white mold, Botrytis grey mold, Anthracnose, Ascochyta blight, *Stemphylium* blight, wilt, root rot *etc*., which reduce the productivity of lentil by 20–25 per cent (Sharma, and Shukla, 2014). Among these diseases, *Stemphylium* blight (*Stemphylium botryosum* Wallr.) is of major importance as it tends to poses potential threat of total yield loss. The disease was recorded in India by Nene *et al*. (1984); in Bangladesh by Bakr and Zahid (1987), in Iran by Kaiser (1972) and in Hungary by Simay (1990). In India, the intensity of the disease was 82.55per cent and the loss was recorded as 93.4per cent (Singh *et al.*, 1999) and in Bangladesh recorded crop loss was 80 to 92.35per cent (Bakr and Ahmed., 1992). *Stemphylium* blight is a seed borne disease which initially appears during flowering as small, light beige to brown lesions often with angular light and dark brown areas on leaves and leaflets of both above and under the canopy which causes severe leaf drop resulting in defoliated plants and which serves as a source of spores for future infection. Small lesions may join eventually and produce irregularly shaped spots which can kill the entire branch. Researches have shown that the disease severity increased under 18-22 ^{0}C temperature with 85per cent relative humidity (Sinha and Singh, 1993). *Stemphylium* produces airborne spores which on windy days can spread and attack healthy lentil plants as well as other crops like onion.

Many fungicides have been found to control the fungal blight disease effectively with dissimilar cost-benefit ratio (Das, 2015). The ideal and most economical way to protect the crop from *Stemphylium* blight is to cultivate resistant varieties. With this aim several lentil genotypes were screened to identify resistant sources which can be exploited in breeding programmes in future.

MATERIALS AND METHODS

The experiment was conducted with seventy seven accessions of lentil which were obtained from AICRP-MULLaRP (BCKV centre, Mohanpur) and screened against *Stemphylium* blight under field condition in District Seed Farm (AB block), Bidhan Chandra Krishi Viswavidyalaya, Kalyani, Nadia, West Bengal. The experiment was carried out during both the *rabi* season of 2015-16 and 2016-17. Seeds of lentil were sown in randomized block design (RBD) in two replications. A spacing of 30 cm between the rows was maintained while plants were sown 4 cm apart from each other. K75 was considered as susceptible check variety and Precoz was considered as resistant check. Variety K75 was sown after every four rows of tested accessions to increase the disease pressure naturally in field. Initially nitrogen (N), phosphorus (P_2O_5), potash (K_2O) fertilizers were applied at a rate of 20:40:0 kg ha^{-1}. Irrigation was

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provided as required. As the disease severity vary greatly on the prevailing atmospheric temperature and humidity just to ensure enhanced and equal disease pressure artificial inoculum of the fungus was done under the same field condition both the years.To prepare the inoculum, lesion was cut from the infected leaves and placed on the moist chamber for 2-3 days to allow the sporulation. Single spores were picked up from lesion and transferred on PDA (Potato Dextrose Agar) plate and incubated in BOD (Biological Oxygen Demand) incubator at 25^0C. Around about one weeks later of incubation, the culture was transferred to fresh plates of PDA for multiplication. One litre of stock suspension of fungal culture was then prepared by blending and straining the culture and diluted in a compressed air sprayer @ 1 litre per 12 litres of water and sprayed in field in the evening hours. Observations were recorded on randomly selected ten plants for each accession after three weeks of disease onset. Disease severity percent of the fungal blight was assessed using 0-9 scale given by Hashemi *et al*., 2005, where, 0= No infection (Immune), 1 = below 10% of foliage affected (Resistant or R), 3= 30% of foliage affected (Moderately Resistant or MR), 5=50% of foliage affected (Moderately Susceptible or MS), 7=70% of foliage affected (Susceptible or S), 9= above 70% of foliage affected (Highly Susceptible or HS). Per cent Disease Index (PDI) was worked out using the formula, PDI = [Sum of numerical rating/total number of observations taken x maximum disease score] x 100. On the basis of disease severity, genotypes were classified into different groups viz., immune, resistant, moderately resistant, moderately susceptible, susceptible and highly susceptible as disease severity percent is calculated by multiplying the mean disease severity of ten plants of each accession with 100. Disease grade for each genotype was also calculated by using the following formula-

Disease Grade = [(Maximum Disease Scoring × Actual Disease Percent) ÷ 100].

RESULTS AND DISCUSSION

In order to identify resistant sources of lentil genotypes against *Stemphylium* blight disease screening under field condition was done. For greater accuracy the entire screening programme was performed on the basis of artificial inoculum. Percent disease severity ranged from 21.21 to 78.52per cent in 2016-17 (Table4) which was higher than the percent disease severity from 18.89% to 77.78%in 2015-16(Table2).Out of seventy seven genotypes, none of the genotype was found under immune (Disease grade- 0), resistant (Disease grade-1) and highly susceptible (Disease grade-9) category during 2015-16, but fifteen genotypes *viz.* LL1370, VL151, LL1375, RLG195, L4727, L4769, LL1397, DL14-2, VL526, VL126, RKL14-20, IPL334, L4710, PL210 and Precoz showed moderately resistance (Disease grade-3) disease reaction and fifty one genotypes *viz*. VL148, IPL333, PL4, PL213, L4737, L4147, WBL77, LL1320, L4751, VL525, LL1374, PL194, LL1373, RVL14-5, L4717, L4771, BPL15, L4076, PL024, RLG191, VL150, TRCL-1, RKL1003-24C, L4726, PL220, L4764, L4735, IPL534, KLS14-23, PL406, LH84-8, DPL15, RVL13-5, LL1404, KLB1442, PL063, DPL62, IPL316, PL175, PL218, BPL14, RKL24C-59, PL221, LL1318, HUL57, VL507, NDL14-22, IPL225, RVL-13-7, Moitree and K-75 showed moderately susceptible disease reaction (Disease grade-5) against the *Stemphylium* blight (Table1). Rest of the genotypes showed susceptible reaction. So, during *rabi,* 2015-16 only 20 and 66 per cent of tested genotypes revealed moderately resistant and moderately susceptible disease reaction accordingly against this fungal blight (Fig.1).

During *rabi,* 2016-17, none of the genotype was found either to be immune (Disease grade-0) or resistant (Disease grade-1). But eleven entries *viz*. VL151, RLG195, L4769, LL1397, DL14-2, VL126, RKL14-20, IPL334, L4710, PL210 and Precoz were observed as moderately resistant (Disease grade-3) against *Stemphylium* blight and forty seven entries *i.e.* VL148, LL1370, VL526, IPL333, PL4, LL1375, L4727, PL213, L4147, LL1320, L4751, VL525, LL1374, PL194, LL1373, RVL14-5, L4717, L4771, BPL15, L4076, PL024, RLG191, VL150, TRCL-1, RKL1003-24C, L4726, PL220, L4735, IPL534, KLS14-23, LH84-8, DPL15, RVL13-5, LL1404, KLB1442, PL063, DPL62, IPL316, BPL14, RKL24C-59, PL221, LL1318, K-75, IPL225, RVL-13-7, VL507 and NDL14-22 were found moderately susceptible (Disease grade-5) (Table 3). Thirteen entries (L4737, RKL607-1, WBL77, L4764, PL406, PL175, PL218, KLS218, HUL57, KLS1445, IPL406, IPL227, Moitree) were observed as susceptible (Disease grade-7). Only six genotypes *viz.*, RVL14-4, L4755, RL3-5, L4762, KLS14-1, JL3 were found highly susceptible (Disease grade-9). So, 14 per cent of genotypes were found to be moderately resistant and 61 per cent were of moderately susceptible type, whereas 17 per cent genotypes showed susceptibility towards the disease and only 8 per cent were found to be highly susceptible in *rabi,*2016-17 (Fig. 2). Podder (2012) conducted an experiments with seventy accessions selected from 7 wild species of the genus *Lens* in growth chamber, greenhouse as well as in the fields at Saskatoon and Bangladesh and found out that 20, 66 and 14per cent of tested accessions were of moderately resistant, moderately susceptible and susceptible against *Stemphylium* blight respectively. Whereas, Rashid *et al*.

Table 1: Disease scale and grouping of genotypes on the basis of reactions to *Stemphylium* blight (*rabi*, 2015-16)

Scale	Disease severity percent	Disease reaction	Number of genotypes	Name of genotypes
0	No infection.	Immune	0	—
1	Below 10% of foliage affected	Resistant Moderately	0	—
3	30% of foliage affected	resistant	15	LL 1370, VL 151, LL 1375, RLG 195, L 4727, L 4769, LL 1397, DL 14-2, VL 526, VL 126, RKL 14-20, IPL 334, L 4710, PL 210, Precoz
5	50% of foliage affected	Moderately susceptible	51	VL 148, IPL 333, PL 4, PL 213, L 4737, L 4147, WBL 77,LL 1320, L 4751, VL 525, LL 1374, PL 194, LL 1373, RVL 14-5, L 4717, L 4771, BPL 15, L 4076, PL 024, RLG 191, VL 150, TRCL-1, RKL 1003-24C, L 4726, PL 220, L 4764, L 4735, IPL 534, KLS 14-23, PL 406, LH 84-8, DPL 15, RVL 13-5, LL 1404, KLB 1442, PL 063, DPL 62, IPL 316, PL 175, PL 218, BPL 14, RKL 24C-59, PL 221, LL 1318, HUL 57, VL 507, NDL 14-22, IPL 225, RVL-13-7, Moitree, K-75
7	70% of foliage affected	Susceptible	11	RVL 14-4, RKL 607-1, L 4755, RL 3-5, L 4762, KLS 14-1, KLS 218, JL 3, KLS 1445, IPL 406, IPL 227
9	Above 70% of foliage affected	Highly susceptible	0	—

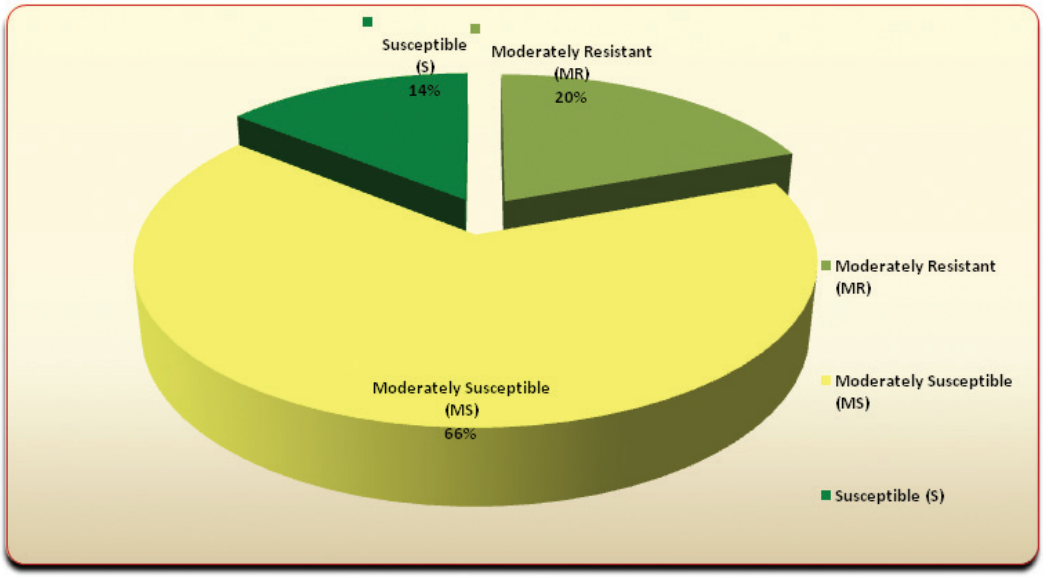

Fig. 1: Percentage of screened genotypes on the basis of reactions to *Stemphylium* blight (*rabi*,2015-16)

Table 2: Percent disease index (PDI), disease grade and disease reaction of lentil genotypes against *Stemphylium* blight during *rabi*, 2015-16:

Sl. No.	Genotypes	Average percent disease index(%) *		Disease grade	Disease reaction
1	VL 148	40.74	(39.66)	3.67	MS
2	LL 1370	28.15	(32.04)	2.53	MR
3	VL 151	26.67	(31.09)	2.40	MR
4	IPL 333	37.78	(37.93)	3.40	MS
5	PL 4	35.56	(36.60)	3.20	MS
6	LL 1375	31.81	(34.33)	2.86	MR
7	RLG 195	25.93	(30.61)	2.33	MR
8	L 4727	31.85	(34.36)	2.87	MR
9	PL 213	40.74	(39.66)	3.67	MS
10	RVL 14-4	77.78	(61.87)	7.00	S
11	L 4737	53.92	(47.25)	4.85	MS
12	RKL 607-1	59.26	(50.34)	5.33	S
13	L 4769	25.19	(30.12)	2.27	MR
14	L 4147	45.19	(42.24)	4.07	MS
15	WBL 77	51.11	(45.64)	4.60	MS
16	LL 1320	42.22	(40.53)	3.80	MS
17	LL 1397	27.41	(31.57)	2.47	MR
18	L 4751	42.22	(40.53)	3.80	MS
19	VL 525	45.19	(42.24)	4.07	MS
20	LL 1374	43.70	(41.38)	3.93	MS
21	PL 194	43.53	(41.28)	3.92	MS
22	DL 14-2	20.00	(26.57)	1.80	MR
23	LL 1373	42.22	(40.53)	3.80	MS
24	VL 526	26.85	(31.21)	2.42	MR
25	RVL 14-5	35.50	(36.57)	3.20	MS
26	L 4717	46.67	(43.09)	4.20	MS
27	L 4771	48.15	(43.94)	4.33	MS
28	BPL 15	40.00	(39.23)	3.60	MS
29	VL 126	24.44	(29.63)	2.20	MR
30	L 4076	39.26	(38.80)	3.53	MS
31	PL 024	35.56	(36.60)	3.20	MS
32	RLG 191	35.74	(36.72)	3.22	MS
33	VL 150	41.11	(39.88)	3.70	MS
34	TRCL-1	43.70	(41.38)	3.93	MS
35	RKL 14-20	25.93	(30.61)	2.33	MR
36	RKL 1003-24C	42.22	(40.53)	3.80	MS
37	L 4726	48.15	(43.94)	4.33	MS
38	PL 220	38.15	(38.14)	3.43	MS
39	L 4764	49.63	(44.79)	4.47	MS
40	L 4735	43.70	(41.38)	3.93	MS
41	IPL 534	35.64	(36.66)	3.21	MS
42	KLS 14-23	39.26	(38.80)	3.53	MS
43	PL 406	48.15	(43.94)	4.33	MS
44	LH 84-8	46.67	(43.09)	4.20	MS
45	DPL 15	34.90	(36.21)	3.14	MS
46	L 4755	74.81	(59.88)	6.73	S
47	RL 3-5	72.59	(58.43)	6.53	S
48	IPL 334	27.41	(31.57)	2.47	MR
49	RVL 13-5	45.93	(42.66)	4.13	MS

Contd.

Table 2 Contd.

Sl. No.	Genotypes	Average percent disease index(%) *		Disease grade	Disease Reaction
50	LL 1404	35.40	(36.51)	3.19	MS
51	L 4762	71.85	(57.96)	6.47	S
52	KLB 1442	33.38	(35.29)	3.00	MS
53	KLS 14-1	68.89	(56.10)	6.20	S
54	PL 063	51.11	(45.64)	4.60	MS
55	DPL 62	41.01	(39.82)	3.69	MS
56	IPL 316	48.15	(43.94)	4.33	MS
57	PL 175	49.63	(44.79)	4.47	MS
58	PL 218	51.11	(45.64)	4.60	MS
59	BPL 14	46.67	(43.09)	4.20	MS
60	RKL 24C-59	42.18	(40.50)	3.80	MS
61	PL 221	42.22	(40.53)	3.80	MS
62	L 4710	26.67	(31.09)	2.40	MR
63	KLS 218	62.22	(52.07)	5.60	S
64	JL 3	71.85	(57.96)	6.47	S
65	PL 210	25.93	(30.61)	2.33	MR
66	LL 1318	42.22	(40.53)	3.80	MS
67	HUL 57	51.11	(45.64)	4.60	MS
68	VL 507	43.01	(40.98)	3.87	MS
69	NDL 14-22	37.04	(37.49)	3.33	MS
70	KLS 1445	56.30	(48.62)	5.07	S
71	IPL 406	61.48	(51.64)	5.53	S
72	IPL 227	57.78	(49.47)	5.20	S
73	IPL 225	39.74	(39.08)	3.58	MS
74	RVL-13-7	38.52	(38.36)	3.47	MS
75	Moitree (LC)	44.44	(41.81)	4.00	MS
76	PRECOZ (RC)	18.89	(25.76)	1.70	MR
77	K-75 (SC)	50.56	(45.32)	4.55	MS
SEm(±)		**1.70**			
LSD (0.05)		**4.80**			

**Figure in parentheses are angular transformed values*

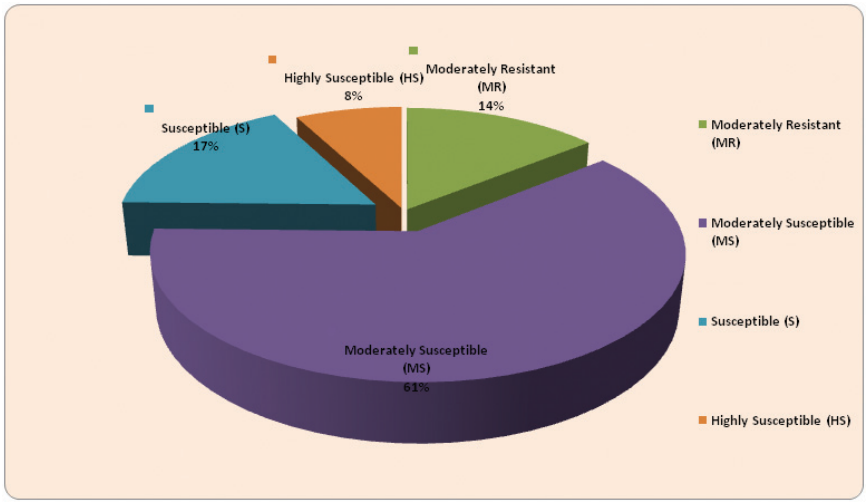

Fig. 2: Percentage of screened genotypes on the basis of reaction to *Stemphylium* blight (*rabi*, 2016-17)

Table 3: Disease scale and grouping of lentil genotypes on the basis of reactions to *Stemphylium* blight (*rabi*, 2016-17):

Scale	Disease severity percent	Disease reaction	Number of genotypes	Name of genotypes
0	No infection.	Immune	0	—
1	Below 10% of foliage affected	Resistant	0	—
3	30% of foliage affected	Moderately resistant	11	RLG 195, L 4769, LL 1397, DL 14-2, VL 526, VL 126, RKL 14-20, IPL 334L 4710, PL 210, Precoz
5	50% of foliage affected	Moderately susceptible	47	VL 148, LL 1370, VL 151, IPL 333, PL 4, LL 1375, L 4727, PL 213, L 4147, LL 1320, L 4751, VL 525, LL 1374, PL 194, LL 1373, RVL 14-5, L 4717, L 4771, BPL 15, L 4076, PL 024, RLG 191, VL 150, TRCL-1, RKL 1003-24C, L 4726, PL 220, L 4735, IPL 534, KLS 14-23, LH 84-8, DPL 15, RVL 13-5, LL 1404, KLB 1442, PL 063, DPL 62, IPL 316, BPL 14, RKL 24C-59, PL 221, LL 1318, K-75, IPL 225, RVL-13-7, VL 507, NDL 14-22
7	70% of foliage affected	Susceptible	13	L 4737, RKL 607-1, WBL 77, L 4764, PL 406, PL 175, PL 218, KLS 218, HUL 57, KLS 1445, IPL 406, IPL 227, Moitree
9	Above 70% of foliage affected	Highly susceptible	6	RVL 14-4, L 4755, RL 3-5, L 4762, KLS 14-1, JL 3

Table 4: Percent disease index (PDI), disease grade and disease reaction of lentil genotypes against *Stemphylium* blight during *rabi*, 2016-17

Sl. No.	Genotypes	Average percent disease index (%) *		Disease grade	Disease reaction
1.	VL 148	43.06	(41.01)	3.88	MS
2.	LL 1370	32.68	(34.87)	2.94	MS
3.	VL 151	31.37	(34.06)	2.82	MS
4.	IPL 333	40.28	(39.39)	3.63	MS
5.	PL 4	38.19	(38.17)	3.44	MS
6.	LL 1375	35.95	(36.84)	3.24	MS
7.	RLG 195	29.17	(32.69)	2.63	MR
8.	L 4727	36.07	(36.91)	3.25	MS
9.	PL 213	44.77	(42.00)	4.03	MS
10.	RVL 14-4	78.52	(62.39)	7.07	HS
11.	L 4737	55.46	(48.14)	4.99	S
12.	RKL 607-1	63.61	(52.90)	5.73	S
13.	L 4769	29.98	(33.20)	2.70	MR
14.	L 4147	47.22	(43.41)	4.25	MS
15.	WBL 77	52.78	(46.59)	4.75	S
16.	LL 1320	44.44	(41.81)	4.00	MS
17.	LL 1397	30.56	(33.56)	2.75	MR
18.	L 4751	44.44	(41.81)	4.00	MS
19.	VL 525	47.22	(43.41)	4.25	MS
20.	LL 1374	45.83	(42.61)	4.13	MS
21.	PL 194	45.83	(42.61)	4.13	MS

Contd.

Table 4 Contd.

Sl. No.	Genotypes	Average percent disease index (%) *		Disease grade	Disease reaction
22.	DL 14-2	22.22	(28.13)	2.00	MR
23.	LL 1373	44.44	(41.81)	4.00	MS
24.	VL 526	29.25	(32.74)	2.63	MR
25.	RVL 14-5	39.58	(38.99)	3.56	MS
26.	L 4717	48.61	(44.20)	4.38	MS
27.	L 4771	50.00	(45.00)	4.50	MS
28.	BPL 15	42.36	(40.61)	3.81	MS
29.	VL 126	27.08	(31.36)	2.44	MR
30.	L 4076	41.67	(40.20)	3.75	MS
31.	PL 024	39.26	(38.80)	3.53	MS
32.	RLG 191	38.03	(38.07)	3.42	MS
33.	VL 150	40.93	(39.78)	3.68	MS
34.	TRCL-1	45.83	(42.61)	4.13	MS
35.	RKL 14-20	26.85	(31.21)	2.42	MR
36.	RKL 1003-24C	44.44	(41.81)	4.00	MS
37.	L 4726	50.00	(45.00)	4.50	MS
38.	PL 220	38.15	(38.15)	3.43	MS
39.	L 4764	51.39	(45.80)	4.63	S
40.	L 4735	45.83	(42.61)	4.13	MS
41.	IPL 534	36.85	(37.37)	3.32	MS
42.	KLS 14-23	41.67	(40.20)	3.75	MS
43.	PL 406	50.00	(45.00)	4.50	S
44.	LH 84-8	48.61	(44.20)	4.38	MS
45.	DPL 15	36.15	(36.96)	3.25	MS
46.	L 4755	76.94	(61.30)	6.93	HS
47.	RL 3-5	75.57	(60.38)	6.80	HS
48.	IPL 334	29.17	(32.69)	2.63	MR
49.	RVL 13-5	47.92	(43.81)	4.31	MS
50.	LL 1404	41.43	(40.07)	3.73	MS
51.	L 4762	73.41	(58.96)	6.61	HS
52.	KLB 1442	33.33	(35.26)	3.00	MS
53.	KLS 14-1	70.44	(57.06)	6.34	HS
54.	PL 063	52.78	(46.59)	4.75	MS
55.	DPL 62	39.62	(39.01)	3.57	MS
56.	IPL 316	50.00	(45.00)	4.50	MS
57.	PL 175	51.39	(45.80)	4.63	S
58.	PL 218	52.78	(46.59)	4.75	S
59.	BPL 14	48.61	(44.20)	4.38	MS
60.	RKL 24C-59	46.58	(43.04)	4.19	MS
61.	PL 221	44.44	(41.81)	4.00	MS
62.	L 4710	28.47	(32.25)	2.56	MR
63.	KLS 218	63.35	(52.74)	5.70	S
64.	JL 3	74.16	(59.45)	6.67	HS
65.	PL 210	28.47	(32.25)	2.56	MR
66.	LL 1318	44.44	(41.81)	4.00	MS
67.	HUL 57	52.78	(46.59)	4.75	S
68.	VL 507	45.06	(42.17)	4.06	MS
69.	NDL 14-22	40.93	(39.78)	3.68	MS
70.	KLS 1445	61.31	(51.54)	5.52	S
71.	IPL 406	63.19	(52.65)	5.69	S
72.	IPL 227	55.40	(48.10)	4.99	S
73.	IPL 225	42.96	(40.95)	3.87	MS

Contd.

Table 4 Contd.

Sl. No.	Genotypes	Average percent disease index (%) *	Disease grade	Disease reaction
74.	RVL-13-7	40.97 (39.80)	3.69	MS
75.	Moitree (LC)	49.96 (44.98)	4.50	S
76.	PRECOZ (RC)	21.2 (27.42)	1.91	MR
77.	K-75 (SC)	49.29 (44.59)	4.44	MS
SEM(±)	**1.61**	–	–	
LSD (0.05)	**4.53**	–	–	

**Figure in parentheses are angular transformed values*

(2009) screened and found twenty one lentil entries *viz.* 10/P8406-122, FLIP-92-52LX, LR-9-135, LR-9-130, LR-9-179, LR-9-69, LR-9-100, LR-9-118, LR-9-28, LR-9-25, ILL-4605, Precoz, LR-9-57, LR-9-107, LR-9-105, LR-9-48, LR-9-62, LR-9-25, 10/P11X955-135, 10/P2 FLIP-92-52LX955-167(4) and 10/P8405-23 which were resistant to *Stemphylium* blight.

Based on disease severity index, eleven genotypes (VL151, RLG195, L4769, LL1397, DL14-2, VL126, RKL14-20, IPL334, L4710, PL210 and Precoz) showed moderately resistant disease reaction against *Stemphylium* blight in both the years. No resistant and immune lentil genotypes were found against *Stemphylium* blight. These identified moderately resistant genotypes can be used in further cross-breeding programme.

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