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Field Screening and Marker Assisted Selection of Late Blight Resistant Potato Lines

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Authors' contributions

This work was contributed by all authors. Author SI conducted field experiment, marker assisted selection and prepared final draft of the manuscript. Author AR supported with marker assisted selection of hybrids, literature review and editing. Author ASMN did laboratory work and provided P. infestans sample for artificial inoculation. Author MAS designed and supervised the study and did final editing of the manuscript. Author LR helped in designing the study. All authors approved the final manuscript.

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ABSTRACT

Late blight (LB), caused by *Phytophthora infestans* (Mont.) de Bary, is the most destructive disease of potato. The main objective of this study was to evaluate 72 potato lines, derived from crosses of the recipient cv. ACI Pakri-1 (female) and a LB resistant donor variety (male), against LB disease. Parent materials, LB susceptible varieties Diamant and BARI Alu-40 and LB resistant variety BARI Alu-77 were used as check varieties. The experiment was conducted with three levels of inoculums pressure (i) LB inoculation & no fungicide, (ii) No LB inoculation & no fungicide and (iii) No LB inoculation & fungicide. LB infection was assessed at 10 day intervals by scoring the percentage of foliage destruction. Subsequently, the area under the disease progress curve (AUDPC), relative AUDPC and susceptible scale value were estimated. Three categories of potato lines were selected considering level of LB infection and tuber appearance - (a) LB resistant - 8 lines, (b) LB tolerant - 14 lines, and (c) LB susceptible - 14 lines. The foliage destruction of selected LB resistant

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lines was considered to be between 1 to 25%, in LB tolerant lines between 25 to 50%, and in LB susceptible lines between 51 to 90% at 85 DAP. The recipient ACI Pakri-1 had 100% foliage destruction at 63-65 DAP. In LB resistant lines low AUDPC, rAUDPC and susceptible scale value were found. The highest susceptible scale value 9 was recorded mostly in LB susceptible cv.ACI Pakri-1 and BARI Alu-40; where the range of susceptible scale value of LB resistant lines was 0.1 to 2.13. LB resistance genes Rpi-abpt and Rpi-blb1, amplified by PCR using primers R2-F1/R2-R3 and 1521/518 respectively, were identified through marker assisted selection (MAS) in the donor variety and crossed LB resistant line 13, 41, 61, 72 and 54 but absent in susceptible ACI Pakri-1.Round tubers, mostly of uniform size, deep to light red and white skin, red and deep eye were found in 8 resistant lines. The highest number of tubers/ hill (20) was recorded in line 8 under natural inoculums pressure, and the highest weight of tuber/hill (460 g) was found in line 61 projecting a yield of 38.3 Mt/ha, which was higher than the donor variety (31.5 Mt/ha) and the recipient ACI Pakri-1 (29.0 Mt/ha) under fungicide application treatment. However, resistance breeding for resistance against potato diseases is developing day by day in Bangladesh. In this regard, our study might contribute in resistance breeding to LB resistance potato variety development.

Keywords: Potato; LB resistance; inoculums pressure; AUDPC; rAUDPC; susceptible scale value; MAS.

1. INTRODUCTION

Potato (*Solanum tuberosum)* is the third most important food crop in Bangladesh, next to rice and wheat [1]. It can play a very important role in alleviating nutrition deficiency and ensuring food security for the people of this country. The crop is known as one of the most sensitive crops in respect to disease infestation, resulting in a large negative impact on yield and tuber quality. Among all potato diseases, late blight (LB) has the most negative impact on yield [8]. Potato late blight causes serious crop damage, yield reduction and profit loss due to fungicide application. The disease is caused by the pathogen *Phytophthora infestans* (Mont.) de Bary. *P. infestans* can be cultured on artificial media, and can survive for indefinite period in the laboratory.

In countries like Bangladesh, the yield loss per year and cost of fungicide use in potato cultivation were reported to be high. In addition to financial loss, LB disease poses a threat for food security, human health and environment. Some strategies like host resistance, fungicide application, disease forecasting, and sanitation have been deployed for LB disease management in the past. However, the most effective and efficient ways to control any plant disease is with host plant resistance. Host resistance could break down in oomycete in several ways *viz*., mixed reproduction systems that allow rapid pathogen propagation and promote, gene flow, large effective population size and high mutation rates [3]. In case of potato, efforts to control LB disease with host resistance have been limited by many factors. However, most Bangladeshi farmers still cultivate varieties susceptible to LB, and resistance levels of the most commonly used potato varieties are not adequate.

According to the gene for gene concept, host resistance against any disease requires a specific resistance gene (R gene) that interacts with a parallel avirulence allele of a gene in the pathogen [4,5,6,7]. These R genes in potato are effective in preventing the development of late blight if the invading *P. infestans* race contains the corresponding a virulence gene. In *S. demissum*, 11 R genes have been characterized in potato [8,9]. In this present study, Marker Assisted Selection (MAS) was conducted with 8 selected resistant lines.

A total of 72 potato lines, obtained through field selection of hybrid materials, were evaluated along with two parents, two LB susceptible varieties, and one LB resistant variety under artificial as well as normal inoculum pressure in field conditions during the 2017-18 season. Marker assisted selection (MAS) was used to confirm the presence of known resistance genes in the selected late blight resistant lines. The original F_1 seed materials were the products of crosses between the susceptible recipient cv. ACI Pakri-1 (female) and an LB resistant variety (male). The main objective of the study was to improve the popular indigenous potato variety ACI Pakri-1, through incorporation of LB resistance.

2. MATERIALS AND METHODS

2.1 Setting of the Experiment

The research was carried out at Debiganj ASRBC Station (Panchagarh). One successful cross, ACI Pakri-1 (recipient) x LB Resistant donor variety, was obtained in 2015-16 season. Out of the plants raised from F_1 seeds in 2016-17 season, 72 individual plants were selected. The tubers obtained from the selected 72 plants were considered as the planting materials for planting in 72 lines during the 2017-18 season. In addition, tubers of 5 potato varieties, namely, ACI Pakri-1 (female parent), donor parent (male), Diamant (LB susceptible check), BARI Alu-40 (LB susceptible check) and BARI Alu-77 (LB resistant check), were considered as planting materials. Thus, there were 77 materials in total for planting and evaluation during the 2017-18 season.

Five tubers of each of 77 entries were planted for each level of LB inoculation treatment. A plant spacing of 60cm X 20cm was maintained. Normal cultural practices were applied. Well-sprouted tubers were planted on 14 November 2017. Each of the 77 planting materials were subjected to the following 3 levels of LB inoculation; (i) LB inoculation & No fungicide applied, (ii) No LB inoculation & No fungicide applied, (iii) No LB inoculation but Fungicide applied. Pencozeb and Acrobate MZ were used for the Fungicide application treatment. The fungicides were applied at 5 day intervals; starting at 32 days after planting (DAP) and continuing up to 85 DAP.

Phytophthora infestans cultures were collected from ASRBC Laboratory, ACI Limited, Dhaka. The following steps were taken for LB inoculation: (i) Stage of application: 30 days after planting, (ii) Preparation: Each plant was covered with a transparent plastic bag before 24 hours of inoculation, and kept covered up to 48 hours after inoculation with *Phytophthora*, (iii) Inoculation: Inoculums was applied to cover all expanded leaves of test plants using a hand sprayer. Normal water was applied on check plants. Three plants from each line were inoculated, (iv) Maintenance after inoculation: After establishment of infection, the plastic covers were removed, and the plants were misted 3 times a day at 10 a.m., 2 p.m. and 6 p.m. for 30 days, (v) Late blight severity was recorded as percentage (%) foliage area damaged on each plant at 30 days after inoculation. The stage of plant growth at which inoculums was applied, and plants covered with transparent plastic bag before 24 hours of inoculation are shown in Fig. 1(a) and 1(b), respectively.

2.2 LB Gene Confirmation through PCR

In this present study Marker Assisted Selection (MAS) was conducted in selected 8 resistant lines along with parental materials. Five *P. infestans* resistance genes named Rpi-blb1 [10], Rpi-bt1 [11], Rpi-abpt [12], Rpi-ber1 [13] and Rpi-sto1 [14] were used based on prior publications and total 5 primers were used for MAS (Table 1). Sprouting was initiated in freshly harvested tuber by 50mg/l GA_3 solution treatment. DNA was isolated from sprout by following CTAB method [15]. PCR was performed with selected markers [16]. Amplified PCR products were detected on a 3% agarose gel stained in 1 x Tris-base EDTA buffer and visualised on a UV trans-illuminator.

Fig. 1. (a) Potato plants ready for LB inoculation at 30 DAP, (b) plants covered with transparent polythene bag before 24 hours of inoculation

Table 1. LB resistant genes, respective markers, sequences and their annealing temperature for PCR amplification used in this study

Gene name	Marker	Sequence	Annealing T^0C
Rpi-ber1	Q133	F:ATCATCTCCTCAAAGAATCAAG R:ATCTCCCCATTGACAACCAA	56.5
Rpi-sto1	Ssto-448	F:GTGGAACGCCGTCCATCCTTAG R:TGCATAGGTGGTTAGATGTATGTTTGATTA	65.6
Rpi-abpt	R2-F1/R2-R3	F:GCTCCTGATACGATCCATG R:ACGGCTTCTTGAATGAA	52.5
Rpi-blb1	1521/518	F: GAAAGTCTAGAGTTACACTGG R: CAATCACAATGGCAGGAACC	58.0
Rpi-bt1	BT1F/BT1R	F: CTACATGGCTGTCATTCACT R: CATAGGGCAACATTTAATCTC	56.0

2.3 Recording of Data

The method for collection of data on LB infection and yield components & yield are presented below.

2.3.1 Data on LB infection

Data were recorded at 10 days interval, starting from 1st inoculation or 1st visible lesion [17].

1. Days to 1st lesion
2. Degree of sporulation (0-3 scale)
   - 0- Corresponds to no visible sporulation
   - 1- Represents very sparse sporulation
   - 2- Represents sporulation on few leaves
   - 3- Many sporangiophores being visible
3. Level of Infection
   a) 0 - No signs of infection.
   b) 0.1 - First single spore-bearing spots.
   c) 1.0 - Weak level of infection (5-10 lesions per a plant).
   d) 5.0 - About 50 lesions per plant; 1 of 10 leaf lobes is infected.
   e) 25 - Almost all leaves infected; plants are still in normal form. Field looks green.
   f) 50 - All plants infected; 50% of leaf area dead. Field still green with brown spots.
   g) 75 - Infection spread over 75% of leaf area. Field looks brown and green.
   h) 95 - Plants have only single leaves, but the stems are green.
   i) 100 - All leaves dead, and stems are dead or dry.

2.3.2 Data on tuber morphological characteristics

Data on tuber color, tuber shape, eye color, eye deep/shallow and uniformity of the tuber were recorded.

2.3.3 Data on yield components and yield

Data on days to 1st emergence, days to 80% emergence, number and weight of tubers per plant at harvest and yield of tubers per hectare were recorded.

2.4 Analysis of Data

Foliage destruction data was used for calculating Area Under Disease Progression Curve (AUDPC) followed by relative AUDPC (rAUDPC).

$$AUDPC = \sum_{i=0}^{n-1}[(X_{i+1}+X_i)/2]\ (T_{i+1} - T_i)$$

Where, "T" is the time of each reading, "X" is the percentage of affected foliage at each reading and "n" is the number of readings. The variable "T" can represent Julian days, days after planting or days after emergence [18].

Then, the relative area under the disease progress curve (rAUDPC) was calculated. This value was obtained by dividing the AUDPC by the total number of days elapsed between the first and last evaluation of the foliage destruction [19].

rAUDPC= AUDPC/ (Difference between last evaluation and the first evaluation X 100)

Again, susceptible level was calculated from the rAUDPC value and scoring highest value 9 for highly susceptible [10].

$$S_X = S_y\ (D_x/D_y)$$

Sy and Dy represent, respectively, the assigned susceptibility scale value and observed disease measure (AUDPC or rAUDPC) for the standard genotype. Sx and Dx represent, respectively, the calculated susceptibility scale value and

observed disease measurement for the genotype in question.

Finally, yield was calculated by the equation recommended by Tuber Crop Research Centre (TCRC), Bangladesh Agricultural Research Institute (BARI).

3. RESULTS AND DISCUSSION

3.1 LB Infection

There was no LB infection in the treatment where fungicides were applied at regular intervals. Data on LB infection were collected from the treatments - LB inoculation & No fungicide applied and No LB inoculation & No fungicide applied. In general, similar results were obtained from both LB treatments. The 1st lesion was observed in all lines at 29 to 40 DAP and 80% or more infected plants were recorded at 32 to 46 DAP which was similar to a susceptible check, resistant check and parents.

On the basis of field evaluation, potato lines were selected under 3 categories, as follows, taken under consideration the level of LB infection and appearance of tubers: (a) LB resistant lines, (b) LB tolerant lines, and (c) LB susceptible, but with attractive appearance of tuber and high yield. A total of 36 lines were selected in these three categories. Rest of the 36 lines were highly susceptible to LB (90 to 100% foliage destruction) and tuber appearance and yield were not at a satisfactory level.

Category-1 (LB resistant lines) included lines showing high tolerance to LB under both levels of inoculums pressure. The foliage destruction in the category was considered to be in the range of 1 to 25% at 85 DAP. In some lines, foliage destruction at 85 DAP was as low as in the LB resistant check variety BARI Alu-77. On the other hand, the recipient ACI Pakri-1 had 100% foliage destruction at 63-65 DAP (Fig. 2). The donor had 58-65% foliage destruction at 85 DAP. The foliage destruction dramatically increased from 10% to 90% at 40 to 50 DAP in LB susceptible check varieties, Diamant and BARI Alu-40 and the recipient (female) variety ACI Pakri-1 under both artificial and natural inoculum pressure. Slower rate of foliage destruction was observed in most of the selected resistant lines till 85 DAP since 1st infection. It may be due to host resistance of the selected lines, which reduced pathogen virulence as defined by decreased infection efficiency, diminished sporangia production, and a reduction in the size of necrotic lesions [20]. Eight lines were selected in LB resistant Category-1 (Fig. 2). Degree of sporulation recorded in most of the lines was 1(representing very sparse sporulation).

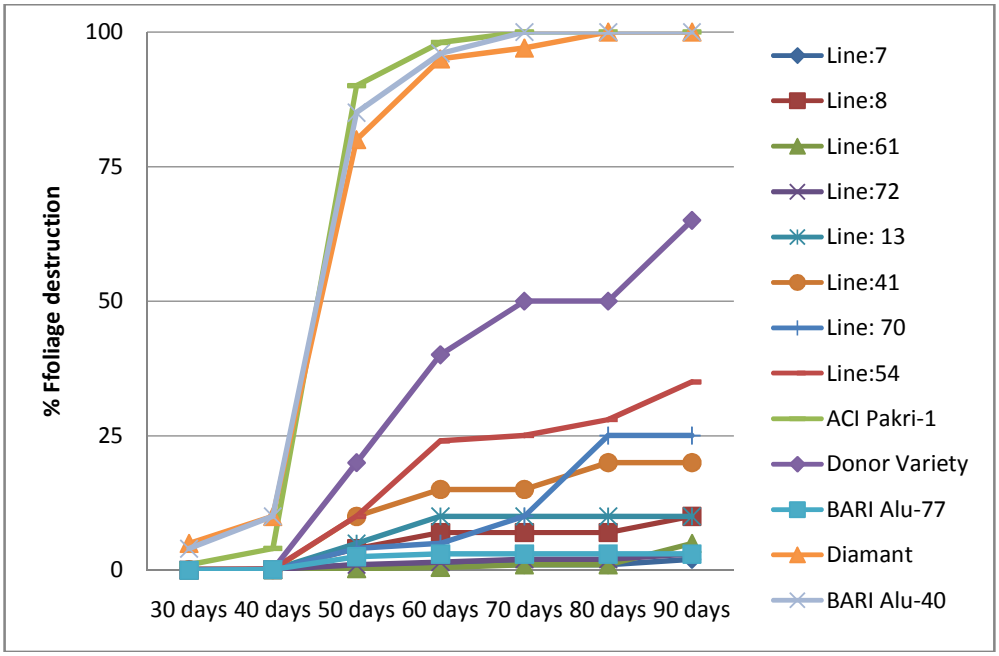

Fig. 2. Foliage destruction due to LB in 8 selected resistant potato lines and parents

Table 2. AUDPC, rAUDPC and Susceptible Scale Value of 8 selected LB resistant lines and check varieties

Lines/ Varieties	AUDPC	rAUDPC	Susceptible scale value
Line:7	51.5	0.009	0.10
Line:8	301	0.050	0.61
Line:61	54.5	0.009	0.11
Line:72	81	0.014	0.16
Line: 13	401	0.067	0.82
Line:41	701.5	0.117	1.43
Line: 70	566	0.094	1.15
Line:54	1047.5	0.175	2.13
ACI Pakri-1	4425	0.738	9.00
Donor Variety	1926.5	0.321	3.92
BARI Alu-77	131	0.022	0.27
Diamant	4345	0.724	8.84
Bari Alu-40	4430	0.738	9.00

By using foliage destruction data, AUDPC was calculated, followed by relative AUDPC (rAUDPC) for 8 selected LB resistant lines along with check varieties. The value of AUDPC and rAUDPC represented the severity of disease. The AUDPC values ranged from 51.5 to 1047.5, and rAUDPC values ranged from 0.009 to 0.175 (Table 2). On the other hand, values of AUDPC and rAUDPC of parent materials were higher (ACI Pakri-1 = 4425 & Donor LB resistant variety = 1926.5) than the selected LB resistant lines.

Some lines had even lower AUDPC and rAUDPC than the resistant check variety BARI Alu-77, indicating that the lines were resistant to LB disease. Low rAUDPC values indicated low levels of infection during the evaluation period, corresponding to more resistant genotypes [21]. However, susceptible scale value for 8 selected LB resistant lines and parent materials and check varieties were calculated by using rAUDPC value. The highest susceptible scale value 9, was set by allowing for highest value of rAUDPC, 0.738 in BARI Alu-40 and ACI Pakri-1. The range of susceptible scale value in selected LB resistant lines was 0.10 to 2.13, where donor variety had the value 3.92.

On the other hand, a total of 14 lines were selected in the LB tolerant category (Category-2), which had medium level of foliage destruction by LB (Table 3). Among the selected 14 LB tolerant lines, foliage destruction due to LB ranged from 25 to 50% at 85 DAP, which was lower than in the donor variety under both artificial and natural inoculums pressure. Higher degree of sporulation, 2 (representing sporulation on few leaves) to 3 (many sporangiophores being visible) was recorded in the lines of this category than LB resistant lines.

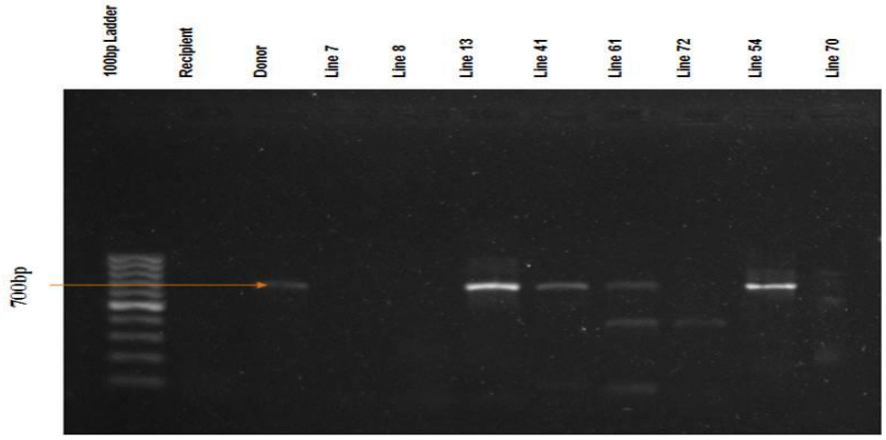

Fig. 3. Confirmation of LB gene Rpi-abpt, using marker R2-F1/R2-R3, in the donor variety and in 13, 41, 61, 72 and 54

Table 3. Infection, foliage destruction and sporulation in selected 8 LB resistant and 14 LB tolerant lines

Lines/ varieties	Days to 1st lesion		Days to 80% or more infected plants		% Foliage destruction at 85 DAP		Degree of sporulation (0-3 scale)	
	T^1	T^2	T^1	T^2	T^1	T^2	T^1	T^2
Cross-derived selected LB resistant lines								
Line:7	33	30	34	32	1	1	1	1
Line:8	35	34	37	35	10	7	1	1
Line: 61	36	30	38	32	4	5	1	1
Line: 72	39	33	42	35	2	3	1	1
Line: 13	31	34	35	36	9	10	1	1
Line: 41	34	29	36	32	19	20	2	2
Line: 70	38	41	42	46	26	25	2	2
Line: 54	30	30	32	34	28	25	2	2
Cross-derived selected LB tolerant lines								
Line:1	31	30	33	33	25	35	2	2
Line:2	37	38	40	40	25	30	2	2
Line: 6	30	30	34	32	25	25	2	2
Line: 11	34	33	36	34	50	53	3	3
Line: 18	29	29	31	30	40	45	3	3
Line: 32	31	33	33	35	40	40	2	2
Line: 44	34	32	37	35	25	25	2	2
Line: 40	35	34	38	36	35	30	2	2
Line: 53	34	30	39	33	50	40	2	2
Line: 68	33	32	35	35	25	25	2	2
Line: 71	40	38	44	44	45	30	2	2
Line: 29	33	33	35	36	48	43	3	3
Line: 56	33	33	34	35	40	45	3	3
Line: 45	35	34	37	35	50	40	3	3
Parents/ varieties (Check)								
ACI Pakri-1	29	28	29	29	100*	100*	3	3
Donor Variety	30	30	32	34	65	58	3	3
BARI Alu-77	41	38	45	47	2	3	1	1
Diamant	33	30	33	31	100**	100**	3	3
BARI Alu-40	29	29	31	31	100***	100***	3	3

*T^1= LB inoculation & No fungicide; T^2=No LB inoculation & No fungicide ; *at 65; **at 78; *** at 83 DAP*

Lastly, another 14 lines were selected in category-3 (LB susceptible). These lines were selected on the basis of their attractive color, shape and uniformity of tuber and high yield. The lines were susceptible to late blight. The level of foliage destruction in the lines, at 85 days, ranged between 50 to 90%.

3.2 Late Blight Gene Confirmation

Five gene specific primers were selected to see whether the late blight resistant genes were present in selected 8 LB resistant lines. PCR product for Rpi-abpt amplified using marker R2-F1/R2-R3 showed presence of resistant gene in the donor variety as well as in five lines, namely, lines 13, 41, 61, 72 and 54 among 8 selected LB resistant lines (Fig. 3). It was reported that R2 gene was found in the some potato varieties/lines which had resistance against late blight in field screening, indicated that R2 gene was responsible for resistance against late blight [12]. On the other hand, Rpi-abpt gene in the recipient variety, ACI Pakri-1 was absent. The late blight resistant gene, Rpi-blb1, was also absent in the recipient variety, ACI Pakri-1; but was present in the donor variety, when DNA amplified using 1521/518 marker. Presence of Rpi-blb1 late blight resistant gene was shown in lines 13, 41, 61, 72, 54 and 70. Similar findings were reported by Chen et al. [16], who stated that genotypic and phenotypic data were related in case of late blight disease when DNA markers derived from the Rpi-blb1 gene were used for

marker assisted selection. However, from these findings it is revealed that, late blight resistant gene had been successfully transferred to the recipient variety from the donor, in most of the selected LB resistant lines. The other three late blight resistant genes, Rpi-ber1, Rpi-sto1 and Rpi-bt1 were present in all selected lines as well as parental materials. Lines 7 and 8 showed high level of resistance against late blight in field screening, but none of the two resistant genes, Rpi-abpt and Rpi-blb1, was found in these two lines. Resistance in line 7 and 8, observed in field screening, was possibly due to the newly formed resistant gene or gene combination through hybridisation between the donor and recipient varieties. More field screening and MAS would be needed to find precise findings. However, based on these results, it could be concluded that the Rpi-abpt and Rpi-blb1 genes are responsible for resistance to *P. infestans* in the donor variety as well as in some selected LB resistant lines. This resistant gene confirmation in different potato hybrid lines would help us to go through with those lines for further screening.

3.3 Morphological Features of Tubers

Tubers with deep to light red and white skin were found in LB resistant category (Fig. 4; Table 4). All tubers of cross-derived selected 8 lines were round shaped, as in the recipient parent ACI Pakri-1. In case of uniformity, uniform sized tubers were found in most lines in the same way less uniform sized were observed in few lines. Eye color and eye depth of the selected lines were also recorded. Mostly red colored and deep in depth eyes were observed which was similar to parents.

Most of the lines under LB tolerant category had red to light red skinned and round tubers. Two lines having oval shaped tubers and one line with oblong shaped tubers were also found.

On the other hand, mostly light red to red skinned tubers, and 3 white skinned tubers were found in Category-3 (LB susceptible, but with attractive tubers and high yield). All round, but 1 oval shaped tuber line was recorded; and commonly, deep eye with red color was observed. Uniformity was average. The morphological characteristics of the tubers of selected lines are presented in Table 4.

3.4 Yield Component and Yield

Data on yield component and yield were recorded at harvest after 90 DAP. Although there was wide variation in number of tubers per hill among the selected LB resistant lines (Category-1), the number was not significantly affected by the LB inoculums treatments (Table 4). But the weight of tuber per hill and estimated yield of tubers per hectare were noticeably influenced by LB inoculums treatment. The weight of tubers per hill ranged between 120 and 460 g among the selected 8 LB resistant lines under the no LB inoculums & no fungicide treatment. Application of LB inoculums did not show any positive response to yield of tubers per hill. Application of fungicide was effective on selected lines. The weight of tubers ranged between 120-420 g/hill under the inoculation & no fungicide treatment, 120-460 g/hill under the no inoculation & no fungicide treatment, and 240-480 g/hill under the no inoculation & fungicide treatment. The corresponding estimated yield of tubers per unit area ranged between 10.0 -35.0, 10.0 - 38.3 and 20.0 - 40.0 Mt/ha, respectively (Table 5).

In case of the recipient variety ACI Pakri-1, the weight of tubers was only 31 g/hill when no inoculums or fungicide was applied, and only 20 g/hill when only inoculums was applied.

Fig. 4. Tubers of LB resistant category having different skin color

Table 4. Morphological characteristics of the tubers of selected lines

Lines/ Varieties	Skin color	Shape of tuber	Color of eye	Depth of eye	Tuber uniformity (1-5 scale)
Cross derived selected LB resistant lines					
Line:7	White	Round	Red	Shallow	5
Line:8	Red	Round	Red	Deep	4
Line: 61	White (Slightly red)	Round	White	Deep	4
Line: 72	Red	Round	Red	Deep	4
Line: 13	Light red	Round	Red	Deep	3
Line: 41	White (Slightly red)	Round	White	Shallow	4
Line: 70	Deep Red	Round	Red	Deep	3
Line: 54	White	Round	White	Shallow	5
Cross-derived selected LB tolerant lines					
Line:1	Light red	Round	Red	Deep	4
Line:2	Red	Round	Red	Deep	4
Line: 6	Light red	Round	Red	Deep	4
Line: 11	Red	Round	Red	Shallow	3
Line: 18	Light red	Round	Red	Deep	2
Line: 32	Light red	Round	Red	Deep	4
Line: 44	Light red	Round	Red	Shallow	4
Line: 40	Light red	Round	Red	Shallow	3
Line: 53	Light red	Round	Red	Deep	3
Line: 68	White	Oval	White	Shallow	4
Line: 71	White	Oblong	Red	Shallow	3
Line: 29	Light red	Oval	Red	Shallow	4
Line: 56	Red	Round	Red	Shallow	4
Line: 45	White (slightly red)	Round	Red	Shallow	4
Parents/ varieties (Check)					
ACI Pakri-1	Red	Round	Red	Deep	4
Donor Variety	Red	Round	Red	Deep	4
BARI Alu-77	Red	Long	Red	Shallow	4
Diamant	White	Long	White	Shallow	3
BARI Alu-40	White	Long	White	Shallow	4

But the weight of tubers was 348 g/hill when fungicide was applied. The weight of tubers per hill of the LB resistant check variety BARI Alu-77 was significantly higher under all levels of inoculums treatment, demonstrating 540 g/hill under fungicide application and 427 g/hill under no fungicide & no inoculums treatment. Here also, inoculums did not show any significant positive response. The LB susceptible check variety Diamant demonstrated weight of tubers per hill similar to ACI Pakri-1 as influenced by treatments receiving only inoculums or no inoculums & no fungicide. Among all treatments, the check variety BARI Alu-40 gave the highest yield of tubers/hill (610 g) when fungicide was applied.

As mentioned earlier, 14 potato lines were selected as LB tolerant materials (Category-2). The weight of tubers ranged between 60-300 g/hill under inoculation & no fungicide treatment, 60-300 g/hill under no inoculation & no fungicide treatment, and 280-460 g/hill under the no inoculation & fungicide treatment. Corresponding yields were, 5-25, 5-25 and 23-38 Mt/ha respectively (Table 5).

Another set of 14 potato lines was selected as LB susceptible, but had attractive appearance of tuber and high yield (category-3). In this category, the weight of tubers ranged between 17-45 g/hill under inoculation & no fungicide treatment, 20-47 g/hill under no inoculation & no fungicide treatment, and 320-490 g/hill under the no inoculation & fungicide treatment. Corresponding yields were, 1.4-3.4, 1.7-3.9 and 26.7-40.8 Mt/ha respectively.

Table 5. Yield components and yield of selected lines

Lines/ Varieties	No. of tubers/hill			Wt. of tubers/hill (g)			Yield of tubers (Mt/ha)		
	T_1	T_2	T_3	T_1	T_2	T_3	T_1	T_2	T_3
Cross derived selected LB resistant lines									
Line:7	17.4	16.0	18.6	380	360	400	31.7	30.0	33.3
Line:8	18.0	20.0	17.4	320	360	370	26.7	30.0	30.8
Line: 61	11.4	16.2	15.0	420	460	480	35.0	38.3	40.0
Line: 72	13.6	12.2	17.0	300	280	320	25.0	23.3	26.8
Line: 13	11.6	10.8	11.8	220	200	320	18.3	16.7	26.7
Line: 41	7.6	9.6	10.0	120	120	240	10.0	10.0	20.0
Line: 70	14.0	14.3	9.0	150	167	267	12.5	13.9	22.3
Line: 54	13.7	14.8	16.0	167	160	340	13.9	13.3	28.3
Cross-derived selected LB tolerant lines									
Line:1	13.4	14.8	16.2	300	180	420	25.0	15.0	35.0
Line:2	5.0	8.0	15.0	145	180	400	12.0	15.0	33.3
Line: 6	7.2	10.4	10.2	140	140	320	11.7	11.7	26.7
Line: 11	8.2	12.2	15.8	120	160	420	12.5	13.3	35.2
Line: 18	6.0	4.4	14.4	60	60	360	5.0	5.0	30.0
Line: 32	8.6	11.6	16.4	220	220	420	18.3	18.3	35.0
Line: 44	10.0	11.0	12.0	240	300	440	17.5	25.0	36.7
Line: 40	14	11	20	160	140	320	13.3	10.8	26.7
Line: 53	9.8	10.4	18.8	220	260	460	18.3	21.8	38.3
Line: 68	5.4	7.6	13.0	160	260	380	13.3	21.8	31.7
Line: 71	4.6	6.6	9.2	140	260	400	11.7	21.7	33.3
Line: 29	9.6	7.8	16.6	120	160	360	10.0	13.3	30.0
Line: 56	6.6	7.2	18	70	80	400	5.8	6.7	33.3
Line: 45	6.2	8.0	9.0	100	120	280	8.3	10.0	23.3
Parents/ varieties (Check)									
ACI Pakri-1	6.0	9.6	29.8	20	31	348	1.7	2.6	29.0
Donor Variety	9.6	10.0	14.8	223	252	378	18.6	21.0	31.5
BARI Alu-77	9.6	8.0	9.8	468	427	540	39.0	35.6	45.0
Diamant	4.4	3.0	7.8	27	47	322	2.3	3.9	26.8
Bari Alu-40	10	10.4	14	156	96	610	13.0	8.0	50.8

T^1- LB inoculation & No fungicide; T^2=No LB inoculation & No fungicide; T^3=No LB inoculation & Fungicide

It has been reported that, potato yield loss primarily due to late blight is dependent on variety susceptibility or tolerance/ resistance, and disease management practices [22,23]. The findings of the present study agree with the opinion of Kankwatst et al. [24], who reported that integration of host resistance and fungicide application reduced late blight severity by more than 50%, and increased yield by more than 30%.

Although the number of tubers per hill was similar among the LB tolerance lines and parent materials, some lines showed significantly higher weight of tuber per hill than ACI Pakri-1 (Table 5). All LB tolerance lines showed higher yield per hectare than ACI Pakri-1 when treated with artificial inoculums pressure and without any fungicide application. The highest yield among LB tolerant lines was 25.0 Mt/ha (in Line: 1), when LB inoculation was done but no fungicide was applied. The yield of the line recorded was 35.0 Mt/ha when fungicide was applied. Some research results indicated that, 50 to 70% yield loss might occur due to late blight, depending on degree of resistance of the cultivar [25,26,27].

The lines under Category-3 were selected not on the basis of their resistance against LB, but on the basis of their attractive appearance of tubers and high yield. Among the selected 14 susceptible lines, the highest number of tubers per hill was found in Line-17. Some lines had higher weight of tubers per hill than the parent materials. The estimated yields were 40.8 Mt/ha in Line-35 and Line-36, and 39.7 Mt/ha in Line-65.

4. CONCLUSION

The present experiment was conducted in one season for screening of hybrid-derived potato

materials against LB. Preliminary LB resistant and tolerant lines were found but it would not be reasonable to make concrete suggestion and recommendation for late blight resistant potato variety. However, this study could be helpful for potato resistant breeding especially in screening methods of potato germplasms/lines against LB. Further screening against late blight under late blight inoculums pressure is suggested.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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