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RESEARCH ARTICLE

Mapping of quantitative trait loci for resistance to turcicum leaf blight in maize (*Zea mays* L.)

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ABSTRACT

Turcicum Leaf Blight (TLB) is a common foliar disease of maize. The aim of this study was to find quantitative trait loci (QTL) for TLB resistance in maize. By crossing two inbred lines, CM 212 (susceptible) and CM 145 (resistant), a mapping population was developed, which was evaluated in two environments namely E_1 (Varanasi, U.P., India; 25^0 N, 83^0 E) and E_2 (Nagenahalli, Karnataka, India; 12^0 N, 76^0 E). Data on four disease severity traits viz., Percent Disease Index (PDI), Area Under Disease Progress Curve based on PDI (AUDPC-PDI), Lesion Area (LA), and Area Under Disease Progress Curve based on LA (AUDPC-LA) were generated for locating QTLs. Fifteen QTL intervals for resistance to TLB were located on chromosomes 1, 2, 4, 5, 6 and 9. Out of these fifteen QTLs, two QTLs were reported for trait PDI on chromosomes 1 and 5 for TLB resistance at E_1; seven QTLs were identified at E_2, where two QTLs were reported for PDI on chromosome 4, four QTLs for trait area under disease progress curve based on PDI and one QTL was reported for trait lesion area. Additionally, identified at pooled environment, six QTLs exhibited the LOD values in the range of 2.64 and 7.77, and corresponding R^2 in the range of 10.80 and 17.74 in the individual environments and across the environments.

Keywords: Area under disease progress curve; Lesion area; Percent diseases index; Quantitative trait loci; Turcicum leaf blight

INTRODUCTION

Turcicum leaf blight (TLB) commonly known as northern corn leaf blight (NCLB) is incited by the ascomycete fungus *Setosphaeria turcica* (Luttrell) Leonard and Suggs, with its conidial state *Exserohilum turcicum* (Passerini) Leonard and Suggs (Perkins and Pederson, 1987; Chung et al., 2010). TLB causes significant losses (28 to 91 percent) to yield and grain quality, where the level of genetic resistance of the genotype, climatic conditions during the growth cycle, and the production system largely decides its severity (Singh et al., 2004; 2014).

Screening of molecular markers for polymorphism among the cultivars of maize (*Zea mays* L.) has been argued as the basis for constructing high-density genetic linkage map and eventually for the identification of new quantitative trait loci (QTL) responsible for TLB-resistance (Li et al., 2018). QTLs identification in return helps in marker-assisted selection (MAS) process for improving genetic traits in crop plants. Genomic regions associated with quantitative resistance to TLB have been identified in several earlier studies using different populations and environments aiming at eventually improving host resistance (Brewster et al., 1992; Dingerdissen et al., 1996; Schechert et al., 1999; Welz et al., 1999; Welz and Geiger, 2000; Wisser et al., 2006; Asea et al., 2009, 2012; Balint-Kurti et al., 2010; Xia et al., 2020; Ranganatha et al., 2021).

Improper estimates of QTLs location are common due to lower power of resolution. Some QTLs in these studies were consistent across the experiments and were agreed to be the "consensus" QTL conditioning partial resistance (rQTL) (Asea et al., 2009). However, many of these reported rQTLs have not been precisely mapped and therefore display consistent conflicts (Wisser et al., 2006). So far, only few QTLs conferring resistance to pathogens have been validated (Abalo et al., 2009; Asea et al., 2009, 2012). Hence, an attempt was made to identify SSR markers co-segregating with the resistant TLB genotypes, identify QTLs for TLB in the Indian *Z. mays* germplasm pool, and validate some of the previously reported QTLs for TLB resistance in maize.

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MATERIAL AND METHODS

Plant material

A recombinant inbred line (RIL) population consisting of 155 F_6 lines was developed by single-seed descent from a cross between the inbred lines CM 212 and CM 145 of maize (*Zea mays* L.). The RILs and both parents were planted in the field at Varanasi (Uttar Pradesh, India)-E_1 (25^0 N, 83^0 E) and Nagenahalli (Karnataka, India)-E_2 (12^0 N, 76^0 E). The field experiments were carried out during *Kharif* (Rainy Season) of 2017 in both the environments. The resistant line CM 145 was derived from the Pop 31 breeding material at Almora (Uttarakhand), is a TLB resistant line in early maturity group. An Indian *Z. mays* inbred line developed at Almora (Uttarakhand), the susceptible line CM 212 is early duration *Z. mays* inbred with a high degree of susceptibility to TLB. CM 212 has been developed into an elite inbred line that has been used widely in *Z. mays* breeding programs in India. All of the RILs were arranged in the field in a randomized block design with two replications. CM 145 and CM 212 were planted in each block as the resistant and susceptible controls, respectively. Lines were planted into two rows with 25 cm plant to plant distance and 70 cm row to row distance at two plants per hill. Standard agricultural management practices were employed throughout each growing season in each location.

Disease development and assessment

Artificial inoculation was performed using crushed leaf material infected with the fungus *Setosphaeria turcica* (Luttrell) Leonard and Suggs as inoculum, belonging to the dominant race 0 through the identification of physiological races, collected previously at the same location. Experimental plots were inoculated at the V10–V12 (5 to 6 weeks after sowing) growth stages by placing 20–30 pathogen-colonized sorghum seeds and crushing the diseased leaves into the leaf whorl of each plant (Carson, 1995).

Four disease traits of TLB *viz.,* Percentage Disease Index (PDI), Area Under Disease Progress Curve based on PDI (AUDPC-PDI), Lesion area (LA) and Area Under Disease Progress Curve based on Lesion area (AUDPC-LA), were recorded in both the environments. In Varanasi (E_1), data was recorded at three different growth stages of *Z. mays viz.*, flowering stage 50 days after sowing (50 DAS), dough stage, (60 DAS) and brown husk stage (70 DAS). In Nagenahalli (E_2), data was recorded at five different growth stages *viz.,* pre-flowering stages (30 DAS and 40 DAS), flowering stage (50 DAS), dough stage (60 DAS) and brown husk stage (70 DAS). PDI was calculated using the 1-5 scale (Payak and Sharma, 1985). LA was calculated according to the formula given by Leath and Pederson (1986):, where L is lesion length and W is lesion width. It was taken from infected leaves of each entry and mean was calculated by sum and averaging of all infected leaves.

AUDPC was estimated using the formula given by Campbell and Madden (1991):

$$AUDPC = \sum_{i=1}^{n-1}\left[\frac{(X_{i+1} + X_i)}{2}\right] + t_{i+1} + X_i$$

Where X_i is the disease index expressed as a proportion at the i^{th} observation, t_i is the time (days after planting) at the i^{th} observations and n is the total number of observations. AUDPC-PDI and AUDPC-LA were also calculated at the same growth stages in the two environments.

Parental polymorphism assay

A set of 500 simple sequence repeat (SSR) markers obtained from different sources [Applied Biotechnology Centre, CIMMYT and Asian Maize Biotechnology Network (AMBIONET)] were used to screen the *Z. mays* inbreds to identify polymorphic SSR markers. The parental polymorphism survey was conducted at Molecular Breeding Lab, BHU, Varanasi (Uttar Pradesh). Parental polymorphism survey revealed the parents CM 212 and CM 145 as the best combination to raise as the mapping population. The CM 212 and CM 145 pair was differentiated by 103 SSR polymorphic primers.

DNA isolation

Genomic DNA was isolated from 21-24 days-old seedlings according to the modified method based on Saghai-Maroof et al. (1994).

Map construction and QTLs detection

For each segregating marker, a Chi-square analysis {$\chi^2=\Sigma$ (Observed-Expected)/Expected} was performed to test for deviation from the expected segregation ratio (1:2:1). Linkage analysis of SSR markers was conducted using the Kosambi (1944) mapping functions with a minimum log10 odds ratio (LOD) of 2.5 and maximum recombination frequency of 0.4 performed by QTL IciMapping 4.1 software. Quantitative trait loci (QTL) analysis for each individual environment and a combined one, across all environments were performed by composite interval mapping using ICIM 4.1.

Statistical analysis

Statistical analysis of all four characters [PDI, AUDPC (PDI), LA and AUDPC (LA)] for ANOVA and traits correlation was performed by PROC GLM procedure using SAS (V 9.2) software package (SAS Institute Inc., 2004). ANOVA was calculated for all four disease parameters in each environment as well as combined/pooled over environments using SAS (V 9.2) software package (SAS Institute Inc., 2004). In context with testing the heritability and traits correlation, estimates of broad sense heritability

(h^2) was calculated from ANOVA over environments using PROC GLM procedure of SAS software according to the formula suggested by Burton and De-Vane (1953) for each disease character:

$$h^2 = \frac{\sigma^2 g}{\sigma^2 g + \sigma^2 \epsilon}$$

Where, $\sigma^2 g$ = genotypic variance and $\sigma^2 e$ = environmental variance. Correlation was estimated for all four-disease parameters of each environment as well as over the environments by PROC GLM procedure of SAS software.

RESULTS

Four disease traits *viz*., PDI, AUDPC-PDI, LA and AUDPC-LA were considered to generate phenotypic data for the purpose of locating and mapping QTLs for TLB in *Z. mays*. Major results pertaining to the mentioned above four traits are summarized here under.

Percent disease index and AUDPC-PDI

Mean PDI at 70 DAS for TLB in the resistant (CM 145) and susceptible (CM 212) parents ranged from 31.61 (E_1- BHU Varanasi- The host University) to 35.95 (E_2- Nagenahalli) and 55.17 (E_1) to 78.26 (E_2), respectively (Table 1). The disease progress curve based on PDI of RILs indicated a continuous increase in disease severity from flowering to brown husk stage in both the environments (Fig. 1). The resistant and susceptible parental lines exhibited contrasting phenotypes for TLB in both the environments. The AUDPC-PDI of RILs ranged from 564.89 - 790.34 in E_1 to 1611.44 - 2323.58 in E_2, and indicated large phenotypic variations within as well as between environments (Table 1). The disease was found less severe at flowering stage (30 DAS), whereas, the severity of diseases was highest at brown husk (70 DAS) stage that indicated a continuous disease development throughout different plant growth stages.

The ANOVA revealed significant differences among treatments, environments and treatment x environment for both the traits (Table 2). Keeping in view the differences in disease pressure in two environments, it was decided to analyse data of the two environments separately as well as the pooled data. The highest value of heritability estimates in broad sense (Table 2) was recorded for AUDPC based on percent disease index (0.74). However, low value of heritability was observed for PDI (0.39). The phenotypic correlation among four disease traits in two environments (4 × 2) and pooled analysis over environment are presented in Table 3. Further, the correlation studies indicated positive

Table 1: Range and mean value of Percent Disease Index (PDI), AUDPC-PDI, Lesion Area (LA) and AUDPC-LA and coefficients of variation for 155 $F_{2:6}$ lines from the cross of CM 212 x CM 145 from individual environments and across environments

Genotype	Percent disease index (PDI)			AUDPC-PDI			Lesion Area			AUDPC-LA		
	E_1*	E_2**	Pooled	E_1*	E_2**	Pooled	E_1*	E_2**	Pooled	E_1*	E_2**	Pooled
CM 212	55.17	78.26	66.72	835.73	2159.48	1497.60	13.61	27.46	20.54	179.63	369.00	274.32
CM 145	31.61	35.95	33.78	565.45	1292.15	928.80	5.40	5.75	5.58	71.85	78.18	75.02
$F_{2:6}$ range	33.58-48.31	57.50-82.21	47.85-60.58	564.89-790.34	1611.44-2323.58	1112.34-1468.74	4.08-23.77	6.85-33.09	6.95-25.28	50.67-242.62	79.67-318.4	84.66-260.82
$F_{2:6}$ mean	38.10	72.65	55.37	646.03	1856.58	1251.31	10.81	19.49	15.16	105.13	211.09	158.11
CV#	7.87	5.96	6.76	3.24	3.78	4.47	11.37	12.13	13.10	12.52	15.92	17.99

CV was estimated from 155 entries including parents in RBD; *Agricultural Research Farm, BHU, Varanasi; ** Agricultural Research Station, Nagenahalli, Karnataka

and highly significant correlations between the two traits whether considered individually in E_1 or E_2 or pooled analysis. A significant correlation (0.6530, 0.6309 and 0.6484) was observed also between PDI and AUDPC-PDI for E_1, E_2 and pooled environments, respectively (Table 3).

Lesion area and AUDPC-LA

Mean LA at 70 DAS of 155 F_6 lines ranged from 10.81 cm^2 (E_1-Varanasi) to 19.49 cm^2 (E_2-Nagenahalli). In general disease severity was greater in E_2 but contrary to PDI and AUDPC-PDI values, the disease progress curve for lesion area indicated that 155 $F_{2:6}$ lines ranged from 4.08 to 23.77 in E_1, while it was 6.85 to 33.09 in E_2, but in general cases it was lying between resistant and susceptible parents (Fig. 2). The AUDPC-LA of the RILs ranged from 50.67 to 242.62 in E_1 and 79.67 to 318.4 in E_2. These values indicated phenotypic variation in both environments with high degree of severity in E_2. The ANOVA exhibited highly significant differences among treatments, environments as well as among environment × treatment for both the traits (Table 2). The heritability estimates in broad sense (Table 2) were high for the trait AUDPC based on lesion area (0.73). However, low value of heritability was observed for lesion area (0.37). Further, the phenotypic correlation between all four disease parameters is presented in Table 3. LA was significantly correlated (0.8440, 0.7484 and 0.7610) with AUDPC-LA in E_1, E_2 and across environments, respectively.

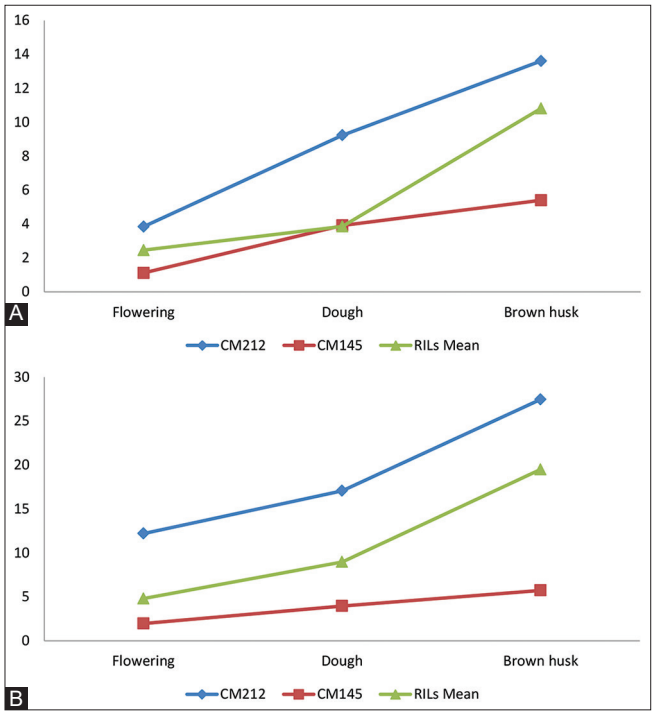

Fig 1. Disease progress curve based on mean PDI of P1, P2 and RILs of cross CM 212× CM 145 at E_1 (Varanasi, Uttar Pradesh, India; 25° N, 83° E) (A) and E2 (Nagenahalli, Karnataka, India; 12° N, 76° E) (B).

SSR linkage map and analysis of QTLs

An analysis was performed of 500 microsatellite markers covering the whole genome for polymorphism between CM 212 and CM 145 inbred lines. The QTL analysis was conducted for E_1 and E_2 separately as well as for the pooled environments for the four disease traits (PDI, AUDPC-PDI, LA and AUDPC-LA). Identified herein 103 (24.41%) markers were found polymorphic. The construction of genetic map with these markers covered about 3485.05 cM with 83 markers distributed across *Z. mays* genome. The average distance between adjacent marker loci was found about 41.99 cM. However, 20 markers remained ungrouped since they were genetically unlinked. Notably, 15 QTL intervals for resistance to TLB were located on chromosomes 1, 2, 4, 5, 6 and 9. Out of 15 QTLs, 2 QTLs were reported for PDI on chromosomes 1 and 5 flanking with umc1064-bnlg 1057 and dupssr1-bnlg1208 markers, respectively for TLB resistance at BHU, Varanasi (U.P.). On the other, 7 QTLs were identified at Nagenahalli (Karnataka), where 2 QTLs was identified for PDI on chromosome 4, four QTLs for AUDPC-PDI on chromosomes 2, 4 and 6, and 1 QTL was found for trait LA on chromosome 1. At pooled environment, 6 QTLs were identified, where 1 QTL was identified each for PDI (on chromosome 4), LA (on chromosome 9 flanking with phi061-bnlg2122 markers), and AUDPC (LA) (on chromosome 4 flanking with nc004-nc005 markers); and 3 QTLs for found AUDPC- PDI on chromosomes 2 and

Table 2: Pooled analysis of variance of four disease traits (PDI, AUDPC-PDI, LA and AUDPC-LA) involving 155 $F_{2:6}$ lines with parents across the environment

Source of variation	df	Mean Sum of Square			
		PDI	AUDPC-PDI	LA	AUDPC-LA
Treatment	156	35.35**	19134.50**	41.59**	3603.46**
Environment	1	184603.30**	228992298.90**	11772.84**	1764619.94**
Replication	1	118.00	33893.70	20.37	2411.59
Treatment*Environment	156	25.76**	15068.20**	37.15**	3355.37**
Error		13.97	3119.60	22.12	810.30
CV% h^2 (Broad Sense)		6.76 0.39	4.47 0.74	13.10 0.37	17.99 0.73

**significant at 0.01 probability level

4. For these QTLs the log10 odds ratio values ranged from 2.64 to 7.77 and corresponding R^2 ranged from 10.80 to 17.74 in the individual environments and over the environments. The gene action of all QTLs showed over dominance at their respective chromosome (Table 4).

DISCUSSION

Identification and mapping of QTLs are important for studying genetically complex forms of plant disease resistance, and also for facilitating the studies on interactions between resistance genes, pathogens and environment. The present study reported 15 QTLs for TLB resistance in *Z. mays*. Moderate to high incidence of disease for all 4 traits namely PDI, AUDPC-PDI, LA and AUDPC-LA was observed in both the environments (E_1: BHU, Varanasi; E_2: Nagenahalli, Karnataka). However, the disease incidence was found to be higher in E_2 compared to E_1, despite similar epiphytotic conditions prevailing in both environments. Nagenahalli (E_2) is considered as a hotspot for TLB in India (Srivastava et al., 2015; Singh and Srivastava 2017). High natural incidence of TLB in E_2 was also reported earlier by several workers (Singh et al., 2014). These authors argued that a long duration of high humidity and slightly cooler temperature (15-25 °C) are important for ideal TLB disease development. In the present study, both environments exhibited adequate levels of humidity. High humidity, optimal for TLB disease development was present in both E_1 (Varanasi) and E_2 (Nagenahalli); however, average temperature range varied at E_1 (32 to 35 °C) and E_2 (20-25 °C).

Table 3: Correlation of four disease traits (PDI, AUDPC-PDI, LA and AUDPC-LA) involving 155 $F_{2:6}$ lines with parents on the basis of individual as well as across environments

Characters	Environments	PDI	AUDPC-PDI	LA
AUDPC-PDI	Env-1	0.6530**		
	Env-2	0.6309**		
	Pooled (Env-1 & Env-2)	0.6484**		
LA	Env-1	0.0999	0.0927	
	Env-2	0.1111	0.0573	
	Pooled (Env-1 & Env-2)	0.1247	0.0478	
AUDPC-LA	Env-1	0.1178	0.1430	0.8440**
	Env-2	0.1628**	0.1969**	0.7484**
	Pooled (Env-1 & Env-2)	0.1889**	0.1915**	0.7610**

**significant at 0.01 probability level.

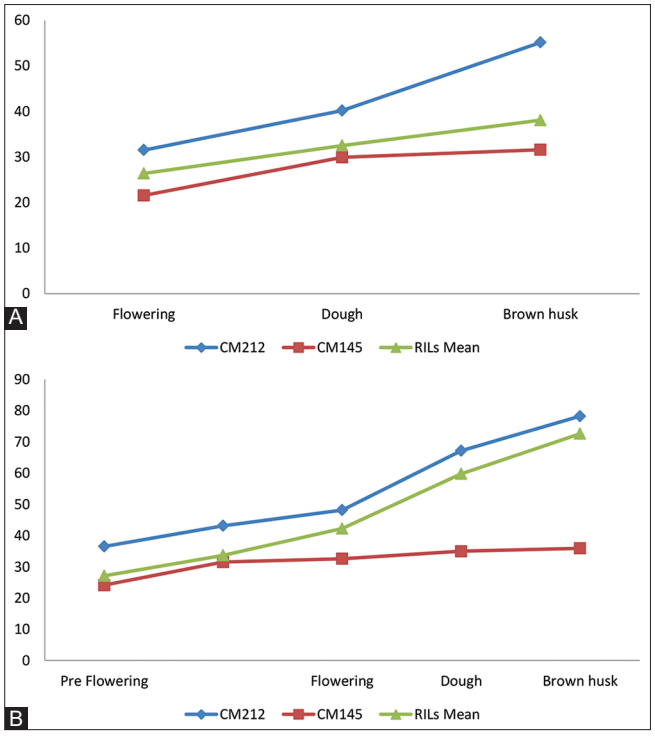

Fig 2. Disease progress curve based on means lesion area of P1, P2 and RILs of cross CM212 × CM145 at E_1 (Varanasi, Uttar Pradesh, India; 25° N, 83° E) (A) and E2 (Nagenahalli, Karnataka, India; 12° N, 76° E) (B).

The TLB disease development and identification of QTLs for diseases resistance have been reported earlier (Balint-Kurti et al., 2010; Singh and Srivastava 2017; Xia et al., 2020; Ranganatha et al., 2021). While mapping QTLs for TLB in two environments, Balint-Kurti et al. (2010) observed a lower disease pressure in Clayton (NC) with slightly high temperatures during the growing season when compared to Aurora (NY) with cooler temperatures. So, the trend observed in the present study supports adoption of different inoculation techniques owing to local practices for creating epiphytotic conditions for TLB.

Notably, the mapping of QTLs in $F_{2:6}$ lines have also been reported by several studies including Schechert et al. (1999) and Welz et al. (1999). In view of these reports, the heavy disease pressure maintained in field plots with artificial epiphytotic conditions combined with replicated disease evaluations in two different environments confirmed the assay employed in this study as sufficiently sensitive in the detection of QTLs effects on TLB resistance. The moderate estimates of heritability indicated that resistance to pathogens was heritable and early generation selection could result in improved germplasm under high disease pressure evaluations. Moderate to high heritability (0.58 to 0.90) has also been reported for TLB in *Z. mays* (Asea et al., 2012). The authors also suggest that reasonable progress in selection is possible for TLB in *Z. mays*. Broad-sense heritability estimates based on variance components analysis of $F_{2:6}$ lines used for QTLs analysis were 0.70 for number of lesions and disease severity (Freymark et al., 1994). Comparatively high to medium estimates of heritability indicated a better expression in

Table 4: QTL identified for Percent disease index (PDI), Area under Disease Progress Curve based on PDI (AUDPC-PDI), Lesion Area (LA) and Area under Disease Progress Curve based Lesion area (AUDPC-LA) in $F_{2:6}$ populations of cross CM 212 × CM 145

Trait	Bin	Flanking Markers	LOD	PVE (%)	Genetic effects			Gene action
					Add	Dom	d/a	
			Environment 1					
PDI	1.11/1.06	umc1064-bnlg1057	4.00	12.02	5.09	-0.16	-31.61	OD
PDI	5.02/5.04	dupssr1- bnlg1208	2.75	12.10	3.94	0.12	33.62	OD
			Environment 2					
PDI	4.05/4.11	nc005- phi019	2.81	15.15	-9.01	-0.38	23.65	OD
PDI	4.11/4.08	phi019- umc1051	2.94	15.25	-9.03	-0.41	22.24	OD
AUDPC (PDI)	2.01/2.04	bnlg1338-bnlg1175	7.12	11.16	262.32	10.56	24.83	OD
AUDPC (PDI)	4.08/4.08	bnlg2162-umc1086	2.91	11.17	244.97	17.42	14.06	OD
AUDPC (PDI)	4.08/4.04	umc1086- phi074	5.19	11.21	240.05	11.04	21.74	OD
AUDPC (PDI)	6.05/6.04	bnlg1922-umc1014	7.77	11.06	282.83	3.47	81.50	OD
LA	1.08/1.11	phi002- bnlg2123	2.64	17.74	-4.23	-0.31	13.87	OD
			Pooled					
PDI	4.11/4.08	phi019- umc1051	2.84	16.49	-3.39	-0.53	6.40	OD
AUDPC (PDI)	2.01/2.04	bnlg1338-bnlg1175	5.41	10.91	136.72	6.97	19.62	OD
AUDPC (PDI	4.08/4.08	bnlg2162-umc1086	2.75	10.80	101.55	10.18	9.97	OD
AUDPC (PDI)	4.08/4.04	umc1086- Phi074	4.50	10.95	113.12	6.99	16.18	OD
LA	9.03/9.01	phi061- bnlg2122	2.88	11.32	-4.03	0.36	-11.14	OD
AUDPC (LA)	4.03/4.05	nc004- nc005	2.66	14.81	-42.20	-7.60	5.55	OD

segregating generations and appropriate disease traits for studying TLB.

AUDPC has been argued as the most appropriate trait for QTLs analysis (Leaths and Pederson, 1986; Welz and Geiger, 2000). Here, high correlation coefficients were observed between PDI and AUDPC-PDI and LA and AUDPC-LA values within individual environments and across environments with a range from 0.6309 to 0.8440 ($P<0.0001$). However, the correlation coefficient between PDI and LA; PDI and AUDPC-LA (E_1); AUDPC-PDI and LA; AUDPC-PDI and AUDPC-LA (E_1) were low and not statistically significant, when calculated for individual and across environments. These correlations coincide with earlier studies including that of Balint-Kurti et al. (2010), where the authors reported a moderate Pearson correlation coefficient 0.49 to 0.67. Additionally, the authors also reported significant correlation between the two phenotypes related to disease resistance namely, weight mean disease (WMD) and incubation period (IP) within and between environments.

Due to variations in major components in environments at E_1 and E_2, this study chose to analyze each environment with respect to four disease traits (PDI, AUDPC-PDI, LA and AUDPC-LA) separately as well as pooled analysis over environments. The observed herein 15 QTLs, located on chromosome 1, 2, 4, 5, 6 and 9 and all QTLs identified were found effective and environment-specific. Earlier, Balint-Kurti et al. (2010) reported many QTLs for NCLB resistance, where 6 QTLs were present on chromosome arm 4 at bins 4.06/4.08. This study reported 8 QTLs on the 4th chromosome, located somewhere between bins 4.03 to 4.11. Environment specific TLB resistant QTLs have been observed earlier in a number of previous studies including Asea et al. (2009) and (2012). In the study of Balint-Kurti et al. (2010), 2 WMD QTLs were reported at bins 2.00/2.01 and 4.08; and only one IP QTL in bin 2.02. These authors further reported the detection of QTL in bin 4.08 in all three environments analyzed separately. In this study also, 3 QTLs associated with AUDPC based on PDI on chromosomes 2 and 4 were reported in the environment, E_2.

In *Z. mays*, resistance to TLB is a complex quantitatively inherited trait. Comparisons were made in QTLs for TLB from other studies of Dingerdissen et al. (1996), and Welz and Geiger (2000). These authors reported AUDPC as more appropriate trait for QTL study in *Z. mays*. In particular, Dingerdissen et al. (1996) identified QTLs for AUDPC on chromosome 1 and on 2S, 3L, 5S, 6L, 7L, 8L and 9S. On the other, Welz and Geiger (2000) discovered QTLs for AUDPC located on chromosomes 1 to 9 in three different mapping populations. Notably, all 3 populations carried QTLs in identical genomic regions on chromosomes *viz*., chromosome-3 (bin 3.06/07), chromosome-5 (bin 5.04) and chromosome 8 (bin 8.05/06). In the reported herein study also, QTL for AUDPC has been identified on chromosome 2 (bin 2.01/2.04) and chromosome 4 (bin 4.08/4.04) in individual environment. Gene action was mostly over dominant or recessive.

CONCLUSIONS

The mapping of quantitative trait loci has been one of the primary goals for locating markers that can be widely used for MAS in a breeding program. However, due to the lack of consistency of QTLs across environments, the use of MAS is lagging behind. To this end, 15 QTLs identified in this study were found significant in the environments E_1, E_2 and in both environments pooled together. The outcomes of this study indicated the role of genotype x environment interaction in low disease appearance in the first environment (E_1). It would also be helpful to initiate pyramiding program for multiple genes by MAS that in turn may control different mechanisms of disease resistance in crops such as *Z. mays*.

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Authors' contributions

Dan Singh Jakhar, conducted experiments for QTL mapping for TLB in *Z. mays* at two locations as well as over the locations; Rajesh Singh, planned experiments and wrote the manuscript; Ram Prakash Srivastava, helped in molecular analyses; Pavan Devesh, Saket Kumar and Abhishek Singh, improved the first draft. All authors read and approved the final manuscript.

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