

Leaf Anatomical Studies of Maize (*Zea mays* L.) in Relation to Tolerance and Susceptibility to Turcicum Leaf Blight Disease

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Abstract

Screening of a set of 12 different composite and commercial hybrid varieties/land races against Turcicum leaf blight disease showed differences in disease reaction. These varieties differed significantly and Percent Disease index (PDI) ranged from 10 to 62 %. Among the different varieties, Allrounder, Prabal, DKC- 9081 showed significantly less (P=0.05) disease as compared to other varieties. Keeping this in mind, they were classified as Tolerant Type and Susceptible Type. The studies on leaf structural anatomy of the both the types revealed that difference in number of stomata per unit area, size of vascular bundle, lower and upper epidermis and number of trichomes per unit area. It may be concluded that the structural anatomy of maize leaf may play an important role in resistance and susceptibility to the Turcicum leaf blight disease.

Highlights

- Screening of maize varieties against Turcicum leaf blight disease showed PDI from 10 to 62 %.
- The studies showed difference in the leaf structural anatomy of tolerant and susceptible cultivars.

Keywords: Screening of maize varieties, Turcicum leaf blight, maize, leaf anatomy

Plants have developed a wide range of mechanisms to resist a variety of biotic and abiotic stresses (Wang Min *et al.*, 2013). Structural defenses like waxes on leaf, thick epidermal cell, broad and elevated guard cell and thick cuticle are present even before the pathogen comes in contact with the plant which act as a pre-existing structural defense against pathogens (Agrios, 2005). Maize is one of the most diverse grain crop found in nature (Reddy *et al.*, 2012), and the significance of these structures in plant

disease resistance has been reported by many workers. Rai *et al.*, (2000) studied the leaf anatomical structures of resistant and susceptible maize cultivars to Turcicum leaf blight disease caused by *Exserohilum turcicum* and they reported that the leaves of resistant cultivar showed wider epidermis, lesser number of stomata per sectioned piece, lesser number of germ tube penetrating the host surface, and very few vascular bundles penetrated by the infection threads, compared to the susceptible cultivar but the resistant



and susceptible cultivars had almost similar stomata dimension. This paper tries to explore the leaf anatomy of different local land races of Darjeeling Himalayas as well as commercial hybrids of maize in relation to resistance against Turcicum leaf blight disease

Materials and Methods

Screening of different maize cultivar against TLB

Seeds of different composite and commercial hybrid varieties which are generally grown by the farmers in Darjeeling Himalayas were collected and grown following Randomized Block Design in experimental plot at District Seed Farm, Bijanbari, Darjeeling. Plot size measured 3X2m with spacing of 75X20 cm. which were replicated thrice. The agronomic practices followed as per recommendation. In each plot the number of infected plant were diagnosed visually on the basis of their symptoms. The occurrence of TLB was recorded and evaluated at 45, 60, 75, 90 and 105 days after sowing (DAS) following 0-5 scale (Payak and Sharma,1983), and per cent disease index (PDI) was calculated using the following formula.

$$\text{Percent Disease Index} = \frac{\text{Sum of individual disease ratings}}{\text{Total no. of plants observed} \times \text{Maximum grade}} \times 100$$

Area Under Disease Progress Curve (AUDPC) was calculated by using following the formula.

$$\text{AUDPC} = \sum_{i=1}^k \frac{1}{2} (S_i + S_{i-1})d$$

AUDPC = Area under disease progress curve

Si = Disease severity at the end of time i

k = Number of successive evaluation of blight severity

d = Interval between two observations

Leaf anatomical observations

In field screening for the natural occurrence of the Turcicum leaf blight disease, different maize varieties showed difference in disease reaction. Some varieties showed susceptibility to the disease whereas the other type showed some degree of tolerance to the disease to a variable extent. Keeping this in mind, the fresh leaves of both types were collected from screening plots. The slides were made from thin transverse sections of these leaves and observed under microscope. Simultaneously, epidermal tissues were stripped from the adaxial and abaxial surfaces of collected

leaves (Driscoll *et. al.*, 2006). The epidermal peels were mounted in citrate phosphate buffer (0.1 M sodium citrate, 0.1 M sodium phosphate, pH 6.5). These slides were examined under light microscopy to study the structural anatomy *viz.*, number of stomata per unit area, lower and upper epidermis and number of trichome per unit area.

Results and Discussion

The evaluation of a set of 12 different composite and commercial hybrid varieties/land races showed differences in disease reaction. The data on Per cent Disease Incidence (PDI) are presented in table-1. High severity of TLB occurred in the experimental plot as reflected by PDI of 62 %. The varieties differed significantly and PDI ranges from 10 to 62 % is indicative of the fact that which indicates that this set of maize varieties included materials ranged from tolerant to highly susceptible ones to TLB. This variation in disease reaction may be due to high diversity of the crop, as maize is one of the most diverse grain crop found in nature (Reddy *et al.*, 2012). Among the different varieties, Allrounder, Prabal, DKC- 9081 showed significantly less (P=0.05) disease as compared to other varieties this findings corroborates with the findings of Mahajan *et al.*, 1991; Kamikoga *et al.*, 1991; Mallikarjuna *et al.*, 2010). High disease incidence was recorded in variety *Saathiya* followed by NLD and *Seti*.

Based on the results the tested varieties/land races may be divided into two distinct groups with high and low resistance to the disease. The resistant groups consisted of Allrounder, Prabal, DKC-9081, BN-101, *Paheli Tall*, among which all are commercial hybrids except *Paheli Tall* which is an open pollinated variety with considerable amount of resistance which as reflected by the AUDPC calculated before senescence. The result also indicated the absence of resistance in old traditionally grown open pollinated or composite varieties.

Leaf Anatomical structures of maize in relation to tolerance and susceptibility to TLB.

Screening of different varieties of maize against Turcicum Leaf Blight disease showed difference in disease reaction. Some varieties showed susceptibility to Turcicum Leaf Blight disease, whereas the other types showed some degree of tolerance to the disease in a considerable way. On the basis of these findings they were classified as: 1) Tolerant Type and, 2) Susceptible Type. In the category of ‘Tolerant Type’ the varieties Allrounder, Prabal, DKC 9081 and BN-101 were included, whereas in susceptible types *saathiya*,

**Table 1:** Field screening of maize varieties against Turcicum Leaf Blight

Varieties	PDI (Mean of 2011-12)				AUDPC
	45 DAS	60 DAS	75 DAS	90 DAS	
Prabal	10.2 (18.625)	16.6 (24.044)	17.7 (24.88)	20.3 (26.779)	896.85
Allrounder	12.5 (20.705)	15.5 (23.185)	16.8 (24.197)	19.22 (26.002)	863.25
A de Cuba	24.4 (29.601)	35.8 (36.75)	40.4 (39.465)	46.3 (42.878)	2025
NLD white	23.6 (29.065)	38.7 (38.469)	45.9 (42.648)	51.7 (45.974)	2217.75
Alipurduar local	22.5 (28.316)	36.8 (37.346)	45.5 (42.418)	-	1401.75
Sugar-75	20.1 (26.637)	33.2 (35.183)	42.5 (40.687)	-	1320
DKC-9081	15.7 (23.343)	20.4 (26.85)	22.1 (28.041)	23.6 (29.065)	1092.75
Seti	26.7 (31.113)	46.7 (43.108)	48.3 (44.026)	-	1569.75
Paheli	27.5 (31.628)	36.1 (36.93)	41.2 (39.932)	-	1395.75
Paheli tall	20.2 (26.708)	23.5 (28.997)	25.6 (30.395)	26.7 (31.113)	1287
BN -101	15.8 (23.421)	19.7 (26.35)	21.2 (27.415)	22.5 (28.316)	1053.6
Saadthya	28.8 (32.456)	62.38 (52.168)	-	-	683.85
CD (P=0.05)	3.0735	3.991	3.08	3.14	
SEm+	1.053	1.36	1.046	1.018	

(Figures inside the parenthesis are transformed values)

Table 2 : Leaf Anatomical studies of different maize varieties:

Type of Varieties	Upper epidermal cell (μ)		Lower epidermal cell (μ)		Guard Cell Size ($\mu \times \mu$)	Mean No. of trichomes / cm^2
	Mean Length	Mean Breadth	Mean Length	Mean Breadth		
Tolerant Type	19.68	13.23	15.34	12.16	53.6	280.7
Susceptible Type	15.21	10.64	12.63	9.36	48.7	242.3

Seti, *Paheli* were included. The structural anatomy of the both the types were investigated to make probable inference of the cause of resistance and susceptibility to the disease.

Maize has isobilateral leaves where the upper and lower sides of a leaf are generally symmetrical and does not have difference in cell arrangement or tissue structure. In the present investigation, the following observations were recorded (Table – 2). Epidermal cells were present in both upper and lower surface of the leaf. The epidermis composed of a single layer of cells which were more or

less oval in shape. The epidermal cells of both upper and lower regions were investigated and it is found that in resistant varieties the thickness of the upper (19.68 X 13.23 μm) and lower (15.34 X 12.16 μm) epidermal cells was found to be more than that of the susceptible type (15.21 X 10.64 and 12.63 X 9.36 μm). This findings corroborates with the findings of Rai *et al.*, (2000), however, it may be inferred that the differences are subjected to further investigations. Stomata were present in both the epidermis. So it is called amphistomatic type of leaf. The size of the



stomata was not so different between the tolerant and susceptible genotype although the sizes of the guard cells differed. In tolerant type, the mean size of the guard cells was $53.6 \mu\text{m}^2$, whereas, it was of $48.7 \mu\text{m}^2$ in susceptible type. Similar findings were reported by Rai *et al.*, (2000).

The sizes of the vascular bundles were not significantly in resistant and susceptible types. Xylem consisted of a few enlarged vessels, tracheids and xylem parenchyma, whereas phloem consisted of sieve tubes, companion cells and phloem parenchyma which are clearly visible in leaves of susceptible and tolerant types. The mean number of trichome per unit area was more ($280.7/\text{cm}^2$) than that of the susceptible type ($242.3/\text{cm}^2$).

Conclusion

Field screening of 12 different composite and commercial hybrid varieties / land races against TLB indicated that the varieties like Allrounder, Prabal and DKC-9081 had significantly less disease compared to local land races like *Saathiya*, *Seti Paheli* etc. The tolerance and susceptibility to the disease may be attributed to anatomical structures of studied genotypes, however it needs further investigations.

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