

Biochemical Studies in Bt and Non-Bt Cotton Genotypes Against Alternaria Blight Disease (*Alternaria macrospora* Zimm.)*

GOVINDAPPA N. HOSAGOUDAR, S. N. CHATTANAVAR AND SRIKANT KULKARNI

Department of Plant Pathology
University of Agricultural Sciences, Dharwad - 580 005, Karnataka, India
E mail: s_n_chattannavar@yahoo.com

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Abstract : Biochemical studies were carried out on Laxmi, Abhadita, DCH-32 and RCH-2 Bt, JKCH-1 Bt, JKCH-2 Bt, of non-Bt and Bt genotypes of cotton, respectively. All the test genotypes were found susceptible to the *Alternaria* blight disease. The results indicate that non-Bt genotypes recorded high amount of total protein (7.19 to 9.18%) as compared to Bt genotypes, but total phenol (-17.79 to -18.39 %), total sugar (-12.17 to -12.82 %) and reducing sugar (-4.51 to -19.05%) recorded lower amount compared to Bt genotypes. Further, the decrease in total protein (-18.43 to -19.85%), total phenol (-32.76 to -38.63), total sugar (-31.00 to -33.36%) and reducing sugar (-27.99 to -40.86) was more in infected plants of *Alternaria* blight disease.

Key words: Bt cotton, biochemical, *alternaria* blight.

Introduction

Cotton is the most important fiber crop. It is primarily used in textile industries. Cotton is cultivated in 70 countries of the world with total coverage of 34 million hectare and India now produces around 242.50 lakh bales of cotton ranging from short staple to extra long staple from an area of 88.17 lakh hectares with productivity of 465 kg per hectare (Anon., 2006). In Karnataka, the area under cotton cultivation is 3.81 lakh hectares with a production of 7.00 lakh bales and an average productivity of 312 kg per hectare (Anon., 2006). In recent years, it is becoming increasingly evident that several natural and induced defense mechanisms operate in host plants against different diseases. One such defense mechanism is the presence of certain compounds inhibitory to the pathogen. Sometimes, the host plant is induced to synthesize these compounds on infection. In view of present day of Bt cottons it was thought to analyze the biochemicals in Bt cottons and accordingly non-Bt and Bt cotton genotypes were selected for biochemical analysis. The experiment was carried out at two different growth stages to understand their role in resistance/susceptibility of *Alternaria* blight pathogen.

Material and Methods

In order to study the factors responsible for resistance to *Alternaria* blight disease of cotton caused by *Alternaria macrospora* Zimm., totally six genotypes (non-Bt and Bt) were selected during 2006-07. Among them, Laxmi, Abhadita and DCH-32 were the non-Bt genotypes, RCH-2Bt, JKCH-1 Bt and JKCH-2 Bt were the Bt genotypes. In the field one set was maintained healthy and one separate set was artificially inoculated twice with *A. macrospora*, at an interval of 30 and 50 days after sowing. Biochemical analysis was done at an interval of 90 and 120 days after sowing (DAS). For sampling, five plants were selected at random from each treatment. The top three healthy and diseased leaves were chosen for the purpose of biochemical analysis and scoring was done for those leaves. Estimation of

protein content in leaves was done as per the method given by Lowry et al. (1951), phenols were estimated by folin ciocalteau reagent (Bray and Thorpe, 1954). Sugars were estimated by Nelson's modification of Somogy's method (Nelson, 1944). The data was statistically analysed by following completely randomized design with two factor in MSTATC Programme

Results and Discussion

Total protein : Total protein recorded at different stages is presented in Table 1. It was evident that significant differences existed among the genotypes, treatments and their combination at both stages i.e., 90 and 120 DAS. There was an increase in protein content from 90 to 120 DAS. But, there was decrease in the protein content under infected condition in all Bt and non-Bt cotton genotypes. This result is in agreement with the findings of Patel and Vaishnav (1987). This may be the protein biosynthesis of the host is widely assumed to be significant feature of pathogenesis, particularly during incompatible reaction. Quantitatively the total protein synthesis is much enhanced in the tissues around the infected tissues. This additional protein is considered to be entirely of host origin (Dasgupta, 1988). At 90 DAS, the per cent increase in the mean protein content was more in the non-Bt cotton genotypes, i.e., Laxmi, Abhadita and DCH-32 (12.76%) than the Bt cotton genotypes, RCH-2 Bt, JKCH-1 Bt and JKCH-2 Bt (9.45%) both at healthy and infected condition, respectively. In addition, there was a decrease in the per cent mean protein content in the infected condition irrespective of whether it is non-Bt (-19.95%) or Bt cotton genotype (-16.91%). Similarly, at 120 DAS with an increase in the per cent mean protein content in the non-Bt genotypes both at healthy (5.61%) and in infected condition (4.94%), but the non-Bt genotypes which infected exhibited more per cent decrease in the mean protein content (-20.14%) when compared to Bt genotypes (-19.57%).

Total phenol : The total phenol content was recorded at different stages of crop growth in six non-Bt and Bt genotypes is presented in Table 2. Total phenol content increased from 90 to

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120 DAS both in healthy and in infected plants. Total phenol content differed significantly among genotypes, treatments and their interaction at both the stages of crop.

At 90 DAS, there was decrease in the phenol content in infected genotypes compared to healthy genotypes in both non-Bt and Bt genotypes. All the genotypes under healthy recorded maximum mean phenol content (2.797 mg/g fresh weight) and differed significantly superior over the infected genotypes (1.856 mg/g fresh weight). This result is in agreement with the findings of Borthakur and Addy (1988), Borkar and Verma (1991) and

Chakrabarthy *et al.*(2002). This may be the high concentration causing an instant lethal action by a general tanning effect while, low concentration causes gradual effect on the cellular constituent of the parasite. If the concentration does not occur in toxic level, the inhibition will be obviously slow. Besides, the pathogen readily detoxifies low concentrations of the toxicants rather than high concentrations (Dasgupta, 1988).

The per cent decrease in the mean phenol content was more in the non-Bt genotypes, Laxmi, Abhadita and DCH-32 in both healthy and infected conditions (-35.92% and -27.86%,

Table 1: Total protein content in different Bt and non-Bt cotton genotypes as influenced by *Alternaria macrospora*

Genotypes	Total protein (mg/g fresh weight)							
	90 DAS				120 DAS			
	Healthy	Infected	Mean	% decrease over healthy	Healthy	Infected	Mean	% decrease over healthy
Laxmi	5.859	3.683	4.771		8.075	5.464	6.769	
Abhadita	6.854	6.342	6.598		10.119	8.719	9.419	
DCH-32	6.746	5.552	6.149		9.915	8.264	9.089	
Mean	6.486	5.192		-19.95	9.369	7.482		-20.14
RCH-2 Bt	6.839	6.208	6.523		10.257	9.313	9.785	
JKCH-1 Bt	5.340	4.028	4.684		8.008	6.099	7.053	
JKCH-2 Bt	4.795	3.869	4.332		8.264	5.924	7.094	
Mean	5.658	4.701		-16.91	8.843	7.112		-19.57
Per cent increase or decrease over non-Bt	-12.76	-9.45			-5.61	-4.94		
Grand mean	6.072	4.946			9.106	7.297		
Source	SEm±		CD (0.01)		SEm±		CD (0.01)	
Genotypes (G)	0.104		0.413		0.163		0.645	
Infected (I)	0.060		0.238		0.094		0.372	
G × I	0.147		0.584		0.230		0.913	

DAS – Days after sowing

Table 2. Total phenol content in different Bt and non-Bt cotton genotypes as influenced by *Alternaria macrospora*

Genotypes	Total phenol (mg/g fresh weight)							
	90 DAS				120 DAS			
	Healthy	Infected	Mean	% decrease over healthy	Healthy	Infected	Mean	% decrease over healthy
Laxmi	2.586	1.870	2.228		5.452	2.410	3.931	
Abhadita	1.887	1.457	1.672		3.038	2.547	2.792	
DCH-32	2.083	1.342	1.712		3.077	1.887	2.482	
Mean	2.185	1.556		-28.78	3.855	2.281		-40.83
RCH-2 Bt	3.369	1.627	2.498		3.910	2.838	3.374	
JKCH-1 Bt	4.023	2.242	3.132		4.681	2.566	3.623	
JKCH-2 Bt	2.838	2.603	2.737		3.078	2.012	2.545	
Mean	3.410	2.157		-36.74	3.889	2.472		-36.43
Per cent increase or decrease over non-Bt	35.92	27.86			0.87	7.72		
Grand mean	2.797	1.856			3.872	2.376		
Source	SEm±		CD (0.01)		SEm±		CD (0.01)	
Genotypes (G)	0.043		0.173		0.042		0.169	
Infected (I)	0.025		0.100		0.024		0.098	
G × I	0.062		0.245		0.067		0.240	

DAS – Days after sowing

respectively) as compared to the Bt genotypes, RCH-2 Bt, JKCH-1 Bt and JKCH-2 Bt. Similarly at 120 DAS, infected plants of six genotypes recorded mean minimum phenol content (2.376 mg per g fresh weight) and differed significantly decrease over the mean phenol content of healthy plant (3.872 mg/g fresh weight).

Total sugar : The results from the Table 3 indicated that the total sugar recorded at different stages of non-Bt and Bt cotton under the influence of *A. macrospora*. The total sugar decreased from 90 to 120 DAS. The differences due to genotypes, treatments and their interaction were found significant at both the stages.

At 90 DAS, healthy genotypes recorded highest mean total sugar (8.670 mg/g fresh weight) when compared with infected genotypes (5.778 mg/g fresh weight). This result is in agreement with the findings of Ram Dayal and Joshi (1968). Differences in sugar level between resistant and susceptible genotypes was due to inherent characters of the genotypes. The per cent decrease in the mean total sugar content was more in the non-Bt genotypes, Laxmi, Abhadita and DCH-32 (2.80%) than the Bt genotypes, RCH-2 Bt, JKCH-1 Bt and JKCH-2 Bt (5.12%) both at healthy and infected condition, respectively. In addition, there was decrease in the per cent mean total sugar content in the infected condition irrespective

Table 3. Total sugar content in different Bt and non-Bt cotton genotypes as influenced by *Alternaria macrospora*

Genotypes	Total sugar (mg/g fresh weight)							
	90 DAS				120 DAS			
	Healthy	Infected	Mean	% decrease over healthy	Healthy	Infected	Mean	% decrease over healthy
Laxmi	8.975	7.067	8.021		5.122	3.533	4.327	
Abhadita	7.373	5.059	6.216		4.408	3.065	3.736	
DCH-32	9.295	4.753	7.024		3.065	2.147	2.606	
Mean	8.547	5.626		-34.17	4.198	2.915		-30.56
RCH-2 Bt	9.229	5.671	7.450		4.567	3.300	3.933	
JKCH-1 Bt	9.295	6.602	7.948		5.867	4.248	5.057	
JKCH-2 Bt	7.859	5.518	6.688		5.621	3.457	4.539	
Mean	8.794	5.930		-32.56	5.351	3.668		-31.45
Per cent increase or decrease over non-Bt	2.80	5.12			21.54		20.52	
Grand mean	8.670	5.778			4.774	3.291		
Source	SEm±		CD (0.01)		SEm±		CD (0.01)	
Genotypes (G)	0.126		0.498		0.078		0.308	
Infected (I)	0.072		0.287		0.045		0.178	
G × I	0.178		0.704		0.110		0.437	

DAS – Days after sowing

Table 4. Reducing sugar content in different Bt and non-Bt cotton genotypes as influenced by *Alternaria macrospora*

Genotypes	Reducing sugar (mg/g fresh weight)							
	90 DAS				120 DAS			
	Healthy	Infected	Mean	% decrease over healthy	Healthy	Infected	Mean	% decrease over healthy
Laxmi	4.755	3.228	3.991		3.845	2.492	3.168	
Abhadita	2.917	2.612	2.764		3.090	1.951	2.520	
DCH-32	3.227	2.814	3.020		1.755	1.009	1.382	
Mean	3.633	2.884		-20.61	2.896	1.817		-37.25
RCH-2 Bt	4.667	3.437	4.052		2.472	1.614	2.043	
JKCH-1 Bt	5.335	2.612	3.973		4.497	2.492	3.494	
JKCH-2 Bt	3.509	2.681	3.095		3.731	1.789	2.760	
Mean	4.503	2.910		-35.37	3.566	1.965		-44.48
Per cent increase or decrease over non-Bt	19.32	0.89			18.78	8.14		
Grand mean	4.068	2.897			3.231	1.891		
Source	SEm±		CD (0.01)		SEm±		CD (0.01)	
Genotypes (G)	0.068		0.272		0.049		0.193	
Infected (I)	0.039		0.157		0.028		0.111	
G × I	0.097		0.384		0.069		0.274	

DAS – Days after sowing

of whether the genotypes is not-Bt (-34.17%) or Bt cotton genotypes (-32.56%). Similarly at 120 DAS, the per cent mean total sugar content decreased in non-Bt genotypes in healthy (-21.54%) and in infected condition (-20.52%). Further, the mean per cent of total sugar content decreased at infected condition in both non-Bt (-30.56%) and Bt genotypes (-31.45%). Reducing sugar : It is clear from the data presented in Table 4 that the reducing sugar content decreased from 90 to 120 DAS, both in healthy and in infected plants. It was found that there was significant difference in the reducing sugar content in genotypes, treatments and their interaction. At 90 DAS, healthy genotypes recorded highest mean reducing sugar (4.068 mg/g fresh weight) when compared with infected genotypes (2.897 mg/g fresh weight). Nevertheless, the mean reducing sugar content of healthy genotypes differed significantly superior over the infected genotypes, irrespective of whether the genotypes may be susceptible or resistant. This result is in agreement with the findings of Ram Dayal and Joshi (1968). The per cent decrease in the mean reducing sugar content was more in the non-Bt

genotypes, Laxmi, Abhadita and DCH-32 (-19.32%) than the Bt genotypes, RCH-2 Bt, JKCH-1 Bt and JKCH-2 Bt in healthy, but also per cent decrease in the mean reducing sugar content in infected condition (-0.89%). In addition, there was decrease in the per cent mean reducing sugar content in the infected condition of whether the genotypes is non-Bt (20.61%) or Bt cotton genotypes (35.37%). Similarly at 120 DAS, the per cent mean reducing sugar content decreased in non-Bt genotypes in healthy (-18.78%) and in infected condition (-8.14%). Further, the mean per cent of reducing sugar content decreased at infected condition in both non-Bt(-37.25%) and Bt genotypes (-44.48%). In brief it was observed that the protein content was reduced from healthy to infected conditions and also the protein content was little higher in non Bt cotton genotype than the Bt cottons. The phenol content was high in Bt cotton than the non Bt cotton and there was reduction from healthy to infected conditions. In contrast to this it was observed that the sugar content was high in Bt cotton than the non Bt cotton and there was reduction from healthy to infected conditions. Similar trend was observed with respect to reducing sugars.

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