

# Biochemical factors imparting resistance to leaf blight of barley

Nagaveni, T. and I.K. Kalappanavar\*

Department of Plant Pathology, University of Agricultural Sciences, Dharwad-580 005 (Karnataka) India

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**Overall the resistant genotypes of barley recorded higher amount of total phenol, total protein, sugars, chlorophyll content as compared to genotypes susceptible to leaf blight disease. The amount of these biochemical components increased from 30 days after sowing (DAS) to 60 days after sowing (DAS). However, healthy counterparts of plant samples recorded higher biochemical content (total phenol, total protein, sugars, chlorophyll content) as compared to inoculated counterparts at both the stages. The study revealed that higher sugars, phenols, proteins, chlorophyll contents are some of the possible reasons for resistance to leaf blight disease in barley.**

**Key words :** Resistance, Leaf Blight Barley.

## INTRODUCTION

It is well known that the disease resistance mechanism is a complex phenomenon and in response to invasion by a disease causing organism, plant produces various kinds of reactions. 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 biochemical compounds inhibitory to the pathogen (Prabhu *et al.*, 1984; Singh and Chand, 1982). During these processes considerable changes takes place in biochemical and physiological aspects like changes in the concentrations of total phenol, total protein, reducing sugar, non reducing sugar, total sugar, chlorophyll-a, chlorophyll-b and total chlorophyll (Sharma and Sharma, 1994) in plant tissues and at the same time activities of various isozymes are also modified. Therefore, analysis of biochemical in selected resistant and susceptible genotypes to leaf blight disease was carried out at two different stages to understand their role in resistance / susceptibility to blight pathogen.

## MATERIAL AND METHODS

Four barley genotypes were selected for the study. Among the

4 genotypes, DWR 28 and PL 760 were moderately resistant to leaf blight pathogen and RD 2508, RD 2653 were found to be susceptible to leaf blight pathogen by considering the data resulted by field experiments conducted at the Main Agricultural Research Station, University of Agricultural Sciences, Dharwad during Rabi 2003- 04 and 2004-05. PL 760 and RD 2508 are six rowed barley genotypes where as DWR 28 and RD 2653 are two rowed barley genotypes. The genotypes were allotted in Randomized Block Design (RBD) with three replications of 1m x 1 m plots and four rows in each plot. In the field one set was maintained healthy and another set was artificially inoculated with leaf blight pathogen (*Helminthosporium sativum* Pam., king and Bakke). For different biochemical analysis, top two leaves were collected at 30 and 60 days after sowing (DAS) from random plants and composite leaf sample was made for estimation of total phenol, total protein, sugars, total chlorophyll in the ethanol extracts of fresh leaves of resistant and susceptible genotypes. Total phenol was estimated by Folin – ciocalteu reagent method (Bray and Thorpe, 1954). Total protein was estimated by following the procedure of Lowery *et al.* (1951) sugars were estimated by Nelson's modification of Somogyi's method (Nelson, 1944). Chlorophyll were estimated

**Table 1 :** Total Phenol content in different barley genotypes as influenced by *H. sativum*

Genotypes	Total phenol (mg / g fresh weight)							
	30 DAS				60 DAS			
	Healthy	Inoculated	Mean	% increase or decrease over healthy	Healthy	Inoculated	Mean	% increase or decrease over healthy
DWR 28	2.120	2.607	2.362		2.293	3.107	2.700	
PL 760	2.307	2.817	2.562		2.933	3.813	3.373	
Mean	2.214	2.712		18.36	2.613	3.460		24.47
RD 2653	1.607	1.703	1.655		1.160	2.200	1.680	
RD 2508	1.793	1.857	1.825		0.990	1.993	1.492	
Mean	1.700	1.780		4.49	1.075	2.096		48.71
% increase or decrease over resistant	- 23.21	- 34.3			- 59.01	- 39.42		
Grand mean	1.957	2.246			1.844	2.778		
Source	S. Em±	CD (0.01)			S. Em±	CD (0.01)		
Genotypes (G)	0.006	0.024			0.009	0.039		
Inoculation (I)	0.004	0.017			0.007	0.028		
G x I	0.008	0.034			0.013	0.055		

DAS - Days After Sowing

NS - Non Significant

\* Author for Correspondence

by following the method of Arnon (1949).

Phenol molecule is constructed with lipophilic and hydrophilic portions.

## RESULTS AND DISCUSSION

### Total phenols

The phenolic content under both the situations increased from 30 DAS to 60 DAS. There is an increase in the phenol content ranging from 12.86 to 33.62 per cent. Further, decrease in the phenol content was more in the susceptible genotypes, RD 2653 and RD 2508 at both the stages ranging from 29.35 to 47.78 per cent as compared to the resistant genotypes, DWR 28 and PL 760. The increased rate of phenolic contents from 30 DAS to 60 DAS under diseased condition is in confirmation with the findings of Kiraly and Farkas (1962), Tripathi and Chiranjeevi (1977). The variation in phenol content in different crop plants may be attributed to the unique characters of the phenolic compounds.

### Total protein

The rate of increase in the protein content in response to the disease infection was more in resistant genotypes compared to susceptible genotypes. The protein content under both healthy and inoculated situations increased from 30 DAS to 60 DAS. Also the mean total protein content was more in resistant genotypes than the susceptible genotypes. There is an increase in the protein content ranging from 29.26 per cent to 33.72 per cent when compared to the resistant genotypes with susceptible genotypes. This result is in agreement with the findings of Malhotra (1993) and Malli *et al.* (2000). The protein biosynthesis of the host is widely assumed to be significant feature of

**Table 2 :** Total protein content in different barley genotypes as influenced by *H. sativum*

Genotypes	Total phenol (mg / g fresh weight)							
	30 DAS				60 DAS			
	Healthy	Inoculated	Mean	% increase or decrease over healthy	Healthy	Inoculated	Mean	% increase or decrease over healthy
DWR28	6.143	6.723	6.433		7.033	9.047	8.040	
PL 760	6.237	6.890	6.563		7.810	9.150	8.480	
Mean	6.190	6.806		9.05	7.421	9.098		18.43
RD 2653	4.283	5.043	4.663		5.707	6.270	5.988	
RD 2508	4.123	4.940	4.532		5.073	4.847	4.960	
Mean	4.203	4.990		15.77	5.390	5.558		3.02
% increase or decrease over resistant	- 32.10	- 26.68			- 27.36	- 38.90		
Grand mean	5.197	5.899			6.406	7.328		
Source	S. Em $\pm$	CD (0.01)	S. Em $\pm$	CD (0.01)				
Genotypes (G)	0.026	0.110			0.017	0.070		
Inoculation (I)	0.019	0.078			0.012	0.049		
G x I	0.037	NS			0.023	0.099		

DAS - Days After Sowing                      NS -Non Significant

**Table 3 :** Total sugar content in different barley genotypes as influenced by *H. sativum*

Genotypes	Total phenol (mg / g fresh weight)							
	30 DAS				60 DAS			
	Healthy	Inoculated	Mean	% increase or decrease over healthy	Healthy	Inoculated	Mean	% increase or decrease over healthy
DWR28	5.853	5.040	5.447		3.163	3.037	3.100	
PL 760	5.550	4.993	5.242		2.997	2.933	2.965	
Mean	5.701	4.986		-12.54	3.080	2.985		- 3.08
RD 2653	4.880	3.077	3.978		2.387	2.157	2.272	
RD 2508	4.117	2.847	3.482		2.147	1.987	2.067	
Mean	4.498	2.962		- 34.14	2.267	2.072		- 8.60
% increase or decrease over resistant	- 21.10	- 40.59			- 26.39	- 30.58		
Grand mean	5.100	3.974			2.673	2.528		
Source	S. Em $\pm$	CD (0.01)			S. Em $\pm$	CD (0.01)		
Genotypes (G)	0.025	0.107			0.016	0.066		
Inoculation (I)	0.018	0.075			0.011	0.047		
G x I	0.036	0.151			0.022	Ns		

DAS - Days After Sowing                      NS -Non Significant

**Table 4** : Total chlorophyll content in different barley genotypes as influenced by *H. sativum*

Genotypes	Total phenol (mg / g fresh weight)								
	30 DAS				60 DAS				
	Healthy	Inoculated	Mean	% increase or decrease over healthy	Healthy	Inoculated	Mean	% increase or decrease over healthy	
DWR28	1.481	1.340	1.411		2.325	1.261	1.793		
PL 760	1.366	1.287	1.311		2.202	1.076	1.639		
Mean	1.408	1.313		- 6.74	2.263	1.168		- 48.38	
RD 2653	1.221	1.190	1.205		1.989	0.996	1.492		
RD 2508	1.162	1.110	1.131		1.836	0.922	1.379		
Mean	1.191	1.150		- 3.42	1.912	0.959		- 49.84	
% increase or decrease over resistant	- 15.41	- 12.41			- 15.51	- 17.89			
Grand mean	1.300	1.229			2.088	1.064			
Source	S. Em±	CD (0.01)			S. Em±	CD (0.01)			
Genotypes (G)	0.010	0.043			0.003	0.014			
Inoculation (I)	0.007	0.031			0.002	0.010			
G x I	0.015	NS			0.005	0.020			
DAS - Days After Sowing		NS - Non Significant							

pathogenesis, particularly during incompatible reaction. Quantitatively speaking, 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).

### Sugars

The resistant genotypes, DWR 28 and PL 760 exhibited more total sugar content when compared with the susceptible genotypes RD 2653 and RD 2508. Further, observations revealed that there was reduction in total sugar due to infection in both resistant and susceptible genotypes. The per cent reduction ranges from 22.07 (at 30 DAS) to 5.42 (at 60 DAS) that is almost about seven times reduction in the early stage and about two times reduction at later stage.

Sugars act as precursor for synthesis of phenolics, phytoalexins, lignin and cellulose which play an important role in defense mechanism of plants against invading pathogens. In the present investigation resistant genotypes recorded higher sugars and these results corroborate the findings of Tripathi and Chiranjeevi (1977).

### Chlorophyll content

In general, total chlorophyll content significantly reduced under inoculated condition in all the genotypes compared to their corresponding values in healthy condition. There was 5.46 per cent reduction in total chlorophyll content at 30 DAS and 49.04 per cent reduction in total chlorophyll at 60 DAS. This shows that the drastic reduction in chlorophyll content occurs as the disease advances. One of the reasons for this may be the breakdown of chlorophyll, pigments by the pathogens. These results are in agreement with the findings of Hardev Singh and Chand (1982) and Nandagopal (1995).

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