INTERNATIONAL JOURNAL OF PLANT PROTECTION / VOLUME 6 | ISSUE 1 | APRIL, 2013 | 42- 44

IJPP

RESEARCH ARTICLE

Biochemical changes in greengram leaves due to infection by anthracnose pathogen

■ SUNIL KULKARNI[1] AND V.I. BENAGI[2]

[1]Department of Plant Pathology, Krishi Vigyan Kendra (U.A.S.) BIDAR (KARNATAKA) INDIA
[2]Department of Plant Pathology, University of Agricultural Sciences, DHARWAD (KARNATAKA) INDIA

ARITCLE INFO

Received : 04.07.2012
Revised : 02.01.2013
Accepted : 15.01.2013

Key **W**ords :
Greengram, Anthracnose, Resistance, Biochemical changes

*Corresponding author:
kvksunil@gmail.com

ABSTRACT

Changes in chlorophyll, sugar and phenolic compounds in resistant and susceptible genotypes of greengram were studied. The chlorophyll and sugar content were found to decrease due to the infection of *Colletotrichum truncatum* and the rate of decrease was more in susceptible genotypes (Chinamung, Yellowmung and S-4) than resistant genotypes (TM-96-2 and TARM-18). However, phenol content was found increased due to infection and the rate of increase was higher in resistant genotypes. Comparatively lesser sugar and higher phenol content were observed in resistant and moderately resistant genotypes than in susceptible genotypes.

How to view point the article : Kulkarni, Sunil and Benagi, V.I. (2013). Biochemical changes in greengram leaves due to infection by anthracnose pathogen. *Internat. J. Plant Protec.*, **6**(1) : 42-44.

INTRODUCTION

Anthracnose caused by *Colletotrichum truncatum* (Schw.) Andrus and Moore, is one of the most important fungal diseases of greengram [*Vigna radiata* (L.) Wilczek]. Though, there are fungicides that can reduce disease development, they are not economical and cause environmental pollution. Development of resistant varieties is the most appropriate approach to control the disease. Generally, resistance in plants against various diseases has been correlated with phenolic compounds. Carbohydrates and mineral elements also play an important role in inducing disease resistance (Sindhan and Parashar, 1996). However, information on relationship between resistance to anthracnose and biochemical changes in greengram is not available. The present investigations deals with some biochemical alterations which occur due to anthracnose pathogen in resistant and susceptible genotypes of greengram.

MATERIALS AND METHODS

Thirty greengram genotypes obtained from AICRP (MuLLARP), Dharwad were screened against *Colletotrichum truncatum* under artificial condition in greenhouse during 2007. To carryout the screening work, 60 earthen pots were taken with sterilized soil and kept in greenhouse. Twenty seeds of each genotype were sown in each pot and two such pots were maintained for each genotype. Later, five healthy seedlings per pot were maintained for screening purpose. Conidial suspension of *C. truncatum* (10^5 conidia/ml) was prepared and spray inoculation was done to 30 days old seedlings. The inoculated seedlings were covered with polythene hood for 48 hours to create high humidity. Observations on anthracnose incidence were recorded on 20th day of inoculation by following 0 to 9 scale given by Mayee and Datar (1986).

Healthy and diseased leaves from genotypes TM-96-2, TARM-18 resistant, BGS-9, TM-97-55 moderately resistant and Sel-4, Chinamung, Yellowmung, susceptible to anthracnose were collected 20 days after inoculation and fresh leaves were extracted in 80 per cent ethanol separately. The extracts were analysed for total chlorophyll, chlorophyll 'a' and chlorophyll 'b' (Arnon, 1949), total phenols (Swain and Hills, 1959), ortho-dihydroxy phenols (Johnson and Schaal, 1952), total sugars (Dubois *et al.*, 1956) and reducing sugars

(Somogyii, 1952). Non-reducing sugars were calculated by subtracting the amount of reducing sugars from total sugars.

RESULTS AND DISCUSSION

Among the thirty genotypes screened, none of the genotypes was found immune (Table 1). However, two genotypes *viz.*, TM-96-2, TARM-18 showed resistant reaction, three genotypes *viz.*, BGS-9, TM-98-50 and TM-97-55 showed moderately resistant reaction, while remaining genotypes were found susceptible (2) to highly susceptible (23) (Table 1). Rathaiah and Sharma (2004) reported that, the greengram the

Table 1: Reaction of greengram genotypes to anthracnose under artificial inoculated condition

Sr. No.	Grade	Disease reaction	Name of the genotypes
1.	0	Immune	Nil
2.	1	Resistant	TM-96-2, TARM-18
3.	3	Moderately resistant	BGS-9, TM-98-50, TM-97-55
4.	5	Moderately susceptible	Nil
5.	7	Susceptible	Pusa baisaki, Sel-4
6.	9	Highly susceptible	China mung, Vaibhav, KG-06-1, Yellowmung, DGGV-04, DGS-052, BPMR-1, DLGG-22, DGGS-16, BPMR-145, HMV-6, ML-5, HVM-1, PD-51, OBGG-52, ML-131, KM-6-146, DGS-051,KM-5-133,DGGS-27,KM-5-141,KM-5-134,KM-6-210

Table 2: Chlorophyll content (mg/g of fresh weight) in varieties of greengram with different levels of resistance

Chlorophyll	Type of leaves	Resistant		Mean	Per cent decrease over healthy	Moderately resistant		Mean	Per cent decrease over healthy	Susceptible/ Highly susceptible			Mean	Per cent decrease over healthy
		TM-96-2	TARM-18			BGS-9	TM-97-55			Sel-4	China mung	Yellowmung		
Chlorophyll 'a'	Healthy	0.813	1.306	1.060	4.25	1.102	0.850	0.976	4.41	0.653	0.701	0.645	0.666	58.11
	Diseased	0.780	1.250	1.015		1.060	0.806	0.933		0.262	0.310	0.265	0.279	
Chlorophyll 'b'	Healthy	0.601	0.733	0.667	31.78	0.495	0.512	0.504	39.29	0.380	0.422	0.337	0.379	74.93
	Diseased	0.503	0.407	0.455		0.311	0.301	0.306		0.077	0.119	0.088	0.095	
Total chlorophyll	Healthy	1.414	2.039	1.727	14.88	1.597	1.362	1.480	16.28	1.033	1.123	0.982	1.046	64.24
	Diseased	1.283	1.657	1.470		1.371	1.107	1.239		0.339	0.429	0.353	0.374	

Table 3: Sugar content (mg/g of fresh weight) in varieties of greengram with different levels of resistance

Sugars	Type of leaves	Resistant		Mean	Per cent decrease over healthy	Moderately resistant		Mean	Per cent decrease over healthy	Susceptible/ highly susceptible			Mean	Per cent decrease over healthy
		TM-96-2	TARM-18			BGS-9	TM-97-55			Sel-4	China mung	Yellowmung		
Reducing sugar	Healthy	9.32	9.15	9.24	8.01	8.03	9.29	8.66	8.78	9.51	9.85	9.74	9.70	9.07
	Diseased	8.10	8.90	8.50		7.12	8.68	7.90		8.74	8.75	8.97	8.82	
Non-reducing sugar	Healthy	5.79	6.09	5.94	5.40	6.11	6.15	6.13	5.38	7.01	6.31	6.87	6.73	5.49
	Diseased	5.31	5.93	5.62		5.52	6.08	5.80		6.68	6.01	6.40	6.36	
Total sugar	Healthy	15.11	15.24	15.18	6.98	14.14	15.44	14.79	7.36	16.52	16.16	16.61	16.43	7.61
	Diseased	13.41	14.83	14.12		12.64	14.76	13.70		15.42	14.76	15.37	15.18	

Table 4: Phenol content (mg/g of fresh weight) in varieties of greengram with different levels of resistance

Phenols	Type of leaves	Resistant		Mean	Per cent increase over healthy	Moderately resistant		Mean	Per cent increase over healthy	Susceptible/ Highly susceptible			Mean	Per cent increase over healthy
		TM-96-2	TARM-18			BGS-9	TM-97-55			Sel-4	China mung	Yellowmung		
Total phenols	Healthy	5.44	5.24	5.34	25.47	5.27	5.35	5.31	20.90	4.12	4.16	4.10	4.13	19.61
	Diseased	6.41	6.99	6.70		6.16	6.68	6.42		5.15	5.21	4.46	4.94	
Orthodihydroxy phenols	Healthy	3.55	3.80	3.68	26.36	3.64	3.46	3.55	25.35	2.72	3.21	2.95	2.96	21.61
	Diseased	4.41	4.89	4.65		4.59	4.31	4.45		3.42	3.80	3.58	3.60	

cultivars *viz.*, MLTG-2 and TARM-18 were highly resistant to *C. truncatum*.

Analysis of biochemical constituents revealed that the resistant and moderately resistant genotypes recorded higher amounts of chlorophyll 'a', 'b' and total chlorophyll than susceptible genotypes in both healthy and diseased leaves (Table 2). All varieties showed decrease in chlorophyll 'a', chlorophyll 'b' and total chlorophyll content in diseased leaves. The least decrease in chlorophyll 'a' was 4.25 per cent in TM-96-2 and TARM-18 (both resistant) followed by 4.41 per cent in BGS-9 and TM-97-55 (both moderately resistant), whereas highest decrease of 58.11 pre cent in Sel-4, Chinamung and Yellowmung (all susceptible and highly susceptible). But, chlorophyll 'b' was found to be more sensitive which was also reduced at higher rates in all genotypes and maximum in susceptible genotypes (74.93 per cent).

Total chlorophyll was also in the same trend, as it was cumulative effect of chlorophyll 'a' and 'b'. Its reduction was more in susceptible varieties (64.24%) than resistant (14.88%) and moderately resistant (16.28%) varieties. Similar results were reported by Mesta (2006), while working on Alternaria blight of sunflower. Abnormalities in the form and destruction of chloroplasts are common features of diseased tissue in plants infected with pathogens, which usually exhibited reduced photosynthetic rate, phosphorylation, hill reaction and carbondioxide assimilation (Bawden, 1999).

The observations (Table 3) revealed that, total sugars, reducing sugar and non-reducing sugar contents were higher in healthy leaves of susceptible genotypes than of resistant ones. In diseased leaves their amount deceased in both resistant and susceptible genotypes. But, this decrease in sugar content was at higher rates in susceptible varieties compared to resistant and moderately resistant varieties. Sindhan *et al.* (1999) reported similar results with Cercospora leaf spot of greengram. The reduction in carbohydrate contents after infection may be due to rapid hydrolysis of sugars during pathogenesis through enzymes secreted by pathogens and subsequent utilization by pathogens for their development (Jayapal and Mahadevan, 1968).

In the present investigation, the healthy leaves of resistant and moderately resistant genotypes contained higher amount of total phenol and O.D. phenol than susceptible one. In diseased leaves their amount increased in all groups of genotypes, but this increase in phenol content was at higher rates in resistant and moderately resistant genotypes, while it was at lower rates in susceptible genotypes (Table 4). Similar results were reported by Sindhan *et al.* (1999), while working on Cercospora leaf spot of greengram. The higher amount of phenolic compounds in diseased leaves may be due to either enhancement of synthesis or translocation of phenolics to the site of infection or hydrolysis of phenolic glycosides by fungal glycosides to yield free phenols (Sharma *et al.*, 1983), which helped in arresting the spread of the pathogen. The results indicate that high amount of phenolic compounds and low amount of carbohydrates in resistant genotypes may be responsible for resistance of genotypes against anthracnose of greengram.

REFERENCES

Arnon, D.J. (1949). Copper enzymes in isolated chloroplasts polyphenoloxidase in *Beta vulgaris. Pl. Physiol.,* **24** : 1-15.

Bawden, F.C. (1999). *Plant diseases*. Green World Publishers, Lucknow (U.P.) INDIA p.206.

Dubois, M.K., Gills, A., Hamilton, J.K., Rebarg, P.R. and Smith, F. (1956). Colorimeteric method for determination of sugars and related substances. *Ann. Chem.*, **28** : 350-356.

Jaypal, R. and Mahadevan, A. (1968). Biochemical changes in banana leaves in response of leaf spot pathogens. *Indian Phytopath.,* **21**: 43-48.

Johnson, G. and Schaal, L. A. (1952). Relation of chlorogenic acid to scab resistance in potato. *Science*, **115** : 627-629.

Mayee, C.D. and Datar, V. V. (1986). "*Phytopathometry*". Technical Bulletin-I, Marathawad Agricultural University, Parbhani (M.S.) INDIA, 146 pp.

Mesta, R.K. (2006). Epidemiology and management of Alternaria blight of sunflower caused by *Alternaria helianthi* (Hansf.) Tubaki and Nishihara. Ph. D. Thesis, University Agricultural Sciences, Dharwad, KARNATAKA (INDIA).

Rathaiah, Y. and Sharma, S.K. (2004). A new leaf spot disease on mungbean caused by *Colletotrichum truncatum*, *J. Mycol.& Pl. Pathol.,* **34**: 176-178.

Sharma, S.G., Ram Narayan, Sangam Lal and Chaturvedi, C. (1983). Role of phenolic compounds in resistance of maize to leaf blight caused by Drechslera state of *Cochliobolus heterostrophus*. *Indian Phytopathol.,* **36**: 43-46.

Sindhan, G.S. and Parashar, R.D. (1996). Biochemical changes in groundnut leaves due to infection by early and late leaf spot pathogens. *Indian J. Mycol.& Pl.Pathol.*, **26**: 210-212.

Sindhan, G. S., Indra Hooda and Parashar, R.D. (1999). Sources of resistance of Cercospora leaf spot in mungbean and biochemical parameters for resistance. *J. Mycol.& Plant Pathol.*, **29**: 130-132.

Somogyii, M. (1952). Note on sugar determination. *J.Biol. & Chem.*, **195** : 19-23.

Swain, T. and Hills, W.E. (1959). Phenolic constituents of *Pronus domestica*. Quantitative analysis of phenolic constituents. *J. Sci. & Food. Agric.,* **10** : 63-68.

****