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BRIEF COMMUNICATION

Biochemical and physiological response to salicylic acid in relation to the systemic acquired resistance

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Abstract

In five genotypes of cowpea (*Vigna unguiculata*), the influence of salicylic acid (SA) on photosynthetic activity and biochemical constituents including peroxidase activity at the genotypic level was determined. After SA treatment the total free sugar content increased in IFC 8401 and IGFRI 450 genotypes, whereas the content of total leaf soluble proteins decreased significantly in IFC 902. The high chlorophyll (Chl) (*a* + *b*) content in IFC 902 showed a good correlation with the net photosynthetic rate (P_N), as in this genotype a significant increase in P_N was found after the SA treatment.

Additional key words: carotenoids; chlorophyll; cowpea; net photosynthetic rate; peroxidase; proteins, sugars, *Vigna unguiculata*.

The salicylic acid is one of numerous phenolic compounds found in plants. Phenolics in general may function as the plant growth regulators (Aberg 1981). Exogenously supplied SA affects a large variety of processes in plants, including stomatal closure, seed germination, fruit yield, and glycolysis (Cutt and Klessig 1992). The SA is also a signal molecule in disease resistance (Klessig and Malamy 1994). The involvement of SA in inducing the systemic acquired resistance in all plants generally has opened an extremely active area of investigation. The metabolisms of phenolic compounds and the photosynthetic apparatus functioning are involved in a different manner, and under diseased conditions these two systems may interact with one another (Prokharchik and Marshakova 1983). In addition, plants are the major source of reduced carbon compounds directly or indirectly used by all animals, and thus the functioning of photosystems requires a particular attention, especially when an exogenous chemical is involved (Bounias *et al.* 1988). Therefore the aim of the

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present work was to examine changes in the chlorophyll and carotenoid contents, and photosynthetic activity due to SA treatment, as well as further exploring a possible implication of saccharides and total soluble proteins in relation to SA on the efficiency of photosynthesis.

Five genotypes of cowpea (*Vigna unguiculata*), viz. IFC 8401, 8402, 902, Bundel 2-8503, and IGFRI 450 were sown in pots in three replications at a nursery farm of the Indian Grassland and Fodder Research Institute, Jhansi during August, 1995. All genotypes were sprayed with 0.02 % solution of salicylic acid (pH 6.5) on 30th and 60th d after the sowing. The control plants were sprayed with water accordingly. Photosynthetic activity and photosynthetic pigment contents were determined one week after the second spraying. At the same time, the green leaves were kept in an oven to obtain complete dryness which was later utilized for the total free sugar and starch estimation (Hedge and Hofreiter 1962). The soluble protein content was determined according to Lowry *et al.* (1951), using bovine serum albumin (*Sigma*, St. Louis, USA) as a standard. The peroxidase activity was determined according to Chance and Machly (1955). The oxidation of guaiacol used as a substrate was measured by an increase in absorbance at 470 nm and the activity was expressed in units per fresh mass, whereas one enzyme unit was defined as a change of 0.01 absorbance per min caused by the enzyme aliquot. The contents of carotenoids and chlorophylls were determined by following the spectrophotometric method of Arnon (1949). The P_N, transpiration rate (E), and internal cellular CO_2 concentration (C_i) were measured using a Portable Photosynthesis System (*LI-6200*, *Li-Cor*, Lincoln, USA) between 10:00 and 12:00 h in October, having an average bright sunshine of 9.9 h per day.

After the SA treatment, the Chl (a + b) content increased significantly in IFC 8401 and 902 genotypes while it decreased in IGFRI 450 (Table 1). Two of the genotypes, viz. IFC 8401 and Bundel 2-8503, showed a significant increase in the carotenoids content. According to Bounias *et al.* (1988), both viral and fungal infections share a common increase of most of the photosynthetic pigments in the leaves of a resistant cultivar of *Capsicum*. Similar findings in *Triticineae* (Soulié *et al.* 1984) show that this effect is not species-specific. The SA stimulated pigment formation and in turn the efficiency of photosynthetic apparatus in genotypes with a better potential for resistance. This is because in uninfected conditions the photosynthetic apparatus of resistant leaves can be more efficient than that of the susceptible ones, and a decrease in photophosphorylation rate usually occurring after an infection can be compensated by an increase in efficiency of the photosynthetic apparatus (Hutcheson and Buchanan 1983). The ratio of Chl a : b may be used as an index relating the antenna and reaction centre of photosystems (Platt *et al.* 1979). In our study, IFC 8401 and 8402 showed significant reductions in the Chl a : b ratio while other genotypes did not show any noticeable changes after the SA treatment (Table 1).

After the SA treatment, P_N was significantly decreased in IFC 8401, 8402 and in IGFRI 450, whereas it increased in IFC 902 (Table 1). The genotype showing a maximum increase in P_N, *i.e.* IFC 902, showed a general increase in peroxidase activity during all three days of observation. On the other hand, IFC 8401 showing

Table 1. Net photosynthetic rate (P_N), transpiration rate (E), internal CO_2 concentration (c_i), chlorophyll (Chl) and carotenoid (Car) contents in five genotypes of cowpea influenced by exogenous application of salicylic acid. *Significant at $p < 0.05$.

Genotype	P_N [μmol m^{-2} s^{-1}]		P_N/E [mmol mol^{-1}]		P_N/c_i		Chl ($a+b$) [g kg^{-1}(f.m.)]		Chl a/b		Car [g kg^{-1}(f.m.)]	
	control	treated	control	treated	control	treated	control	treated	control	treated	control	treated
IFC 8401	24.27	10.06*	1.37	1.12	0.038	0.034*	0.816	0.938	3.58	3.18*	0.368	0.406*
IFC 8402	23.33	15.47*	1.38	1.30	0.036	0.061*	0.725	0.697*	3.39	2.96*	0.329	0.315
IFC 902	15.70	23.07*	1.17	1.40*	0.059	0.086*	0.742	0.863*	3.05	3.18	0.342	0.338*
Bundel 2-8503	24.46	26.16	1.61	1.46	0.091	0.091	0.719	0.771	3.27	3.48	0.280	0.319*
IGFRI 450	22.46	13.74*	1.35	0.93	0.071	0.046*	0.959	0.696*	3.72	3.51	0.340	0.286*
LSD ($p < 0.05$)	5.52		0.19		0.020		0.097		0.24		0.037	

Table 2. Total free sugar, starch and total soluble protein contents [g kg^{-1}] and peroxidase activity [kunit kg^{-1}(f.m.)] after 1, 4, and 6 d of treatment in control and salicylic acid treated leaves of cowpea seedlings.

Genotype	Total free sugar		Starch		Total soluble protein		Peroxidase 1 d		4 d		6 d	
	control	treated	control	treated	control	treated	control	treated	control	treated	control	treated
IFC 8401	21	62*	52	34*	54.6	51.6	360	457	225	262	740	690
IFC 8402	18	32	46	43	57.0	57.6	352	397	127	667*	142	427
IFC 902	18	102*	40	57*	55.2	44.4*	307	510	495	600	517	577
Bundel 2-8503	24	32	43	31*	63.0	59.4	622	1005*	502	1342*	2212	2467
IGFRI 450	32	82*	38	46**	54.0	57.6	375	277	615	420	1012	937
LSD ($p < 0.05$)	29		8		5.0		214		339		762	

a maximum depression in P_N possessed initially more peroxidase activity after the SA treatment (Table 2). The total free sugar and and starch contents increased in IFC 902 and IGFRI 450 with the application of SA (Table 2). This indicated that IFC 902 had a better potential over other genotypes to show the acquired resistance, as evidenced by the significant increase in the Chl (a + b), sugar and starch contents, P_N, P_N/E (water use efficiency) and P_N/C_i ratios, and the carboxylation efficiency after the SA treatment (Table 1). Only the cv. Bundel 2-8503 showed a significant increase in peroxidase activity despite of an insignificant change of the P_N, Chl (a + b) content and Chl a : b ratio. The total soluble protein content in one day SA-treated plant was significantly decreased in IFC 902 while other genotypes did not show any changes. This significant drop in the protein content after the SA treatment may be due to its compartmentation in light-harvesting complex protein or in some other activities related to a hypersensitive response. The exact nature of this drop requires a further investigation.

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