J. Phytol. Res. 7 (1) : 41-44, 1994

EFFECT OF CERTAIN PHENOLICS ON SENESCENCE OF RADISH *(RAPHANUS SATIVUS L.)* LEAF DISCS

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Effect of p-hydroxybenzoic acid, protocatechuic acid, gentisic acid, m-coumaric acid, ferulic acid, t-cinnamic acid, scopoletin, juglone, quercetin and 3,4-dihydroxybenzaldehyde on senescene of radish leaf discs was studied. The senescene was evaluated in terms of chlorophyll, nucleic acid and protein contents. All the compounds stimulated senescene. Among these compounds, t-cinnamic acid, juglone, ferulic acid and p-hydroxybenzoic acid were found to be most effective in inducing senescence.

Keywords: Leaf discs; Phenolics; Radish; Senescence.

Introduction

The physiological process of senescence is under the influence of phytohormones.[1] Phenolic compounds are fast emerging as non-hormonal plant growth regulators.[2,3] In addition to regulating growth, phenolics also regulate various other processes like seed germination, rhizogenesis, flowering, stomatal movement and ion uptake.[3] In the present study, the effect of some phenolic compounds on senescence of radish (Raphanus *sativus L.)* leaf discs was investigated.

Material and Methods

Raphanus sativus L. var. Japanese White plants were raised in the field. 18 mm diameter leaf discs were taken from 30 day old plants using cork borer. The leaf discs were washed thoroughly with sterile distilled water. Eleven leaf discs were put in Petriplate (10 cm diameter) provided with Whatman No.1 filter paper. 5 ml of test solution was added to each Petriplate.Each compound was tried at 4 concentration levels viz., 10^{-6}, 10^{-5}, 10^{-4} and 10^{-3} M. The Petriplates were kept under continous fluorescent illumination. After 4 days, the senescence of the leaf discs was recorded in terms of chlorophyll, nucleic acid and protein contents. Chlorophylls were estimated by Arnon[4] method. DNA and RNA were separated from alcohol homogenate of leaf discs by the procedure described by Jayaraman[5]. RNA was estimated adopting the process of Schneider[6] and DNA by the method of Burton.[7] Total nitrogen was estimated by microkjeldal procedure8 and total protein was calculated by multiplying the total nitrogen value with 6.25.

Results and Discussion

All the compounds tested reduced the levels of chlorophylls (Table1). Among all the compounds, t-cinnamic acid, juglone and ferulic acid were proved to be most effective in decreasing the levels of chlorophylls. The phenolic compounds decreased the levels

Table 1. Effect of phenolic compounds on the chlorophyll content of *Rephanus sativus* L. leaf discs.

S. No.	Compound	Chlorophyll 'a' (ug g^{-1} f.w.)*				Chlorophyll 'b' (ug g^{-1} f.w.)*				Total Chlorophyll (ug g^{-1} f.w			
		10^{-6} m	10^{-5} m	10^{-4} m	10^{-3} m	10^{-6} m	10^{-5} m	10^{-4} m	10^{-3} m	10^{-6} m	10^{-5} m	10^{-4} m	10^{-3} m
1.	P-Hydroxybenzoic acid	391±15.6	344±20.4	313±15.0	309±13.3	184±08.0	168±09.2	125±07.4	104±06.3	575±16.3	512±13.9	438±16.3	413
2.	Protocatechuic acid	386±13.7	357±13.1	334±16.1	303±11.9	175±10.6	164±09.5	150±09.4	107±07.6	561±24.2	521±22.6	484±24.9	410
3.	Gentisic acid	380±12.3	333±13.7	299±12.9	265±11.9	177±10.5	161±10.1	141±08.7	121±07.8	557±22.8	494±23.0	440±19.0	386
4.	m-Coumaric acid	385±12.6	356±09.8	323±10.5	271±08.9	186±10.4	175±08.1	143±07.3	119±05.6	571±24.6	531±17.6	466±17.5	390
5.	Ferulic acid	375±14.8	346±09.3	316±06.7	281±09.7	168±06.9	157±06.2	102±07.6	082±05.8	543±21.3	503±15.5	418±14.1	363
6.	t-Cinnamic acid	324±11.6	279±09.9	208±07.8	169±09.2	132±09.2	105±06.5	087±05.5	058±05.7	456±21.2	384±16.4	295±12.8	227
7.	Scopoletin	378±09.6	356±10.2	326±12.4	299±08.9	189±09.1	175±08.3	164±05.8	141±06.3	567±16.8	531±15.5	490±17.1	440
8.	Juglone	323±11.5	275±10.3	265±07.7	221±08.5	143±08.5	139±07.7	121±09.0	082±05.3	466±19.7	414±17.4	386±15.6	303
9.	Quercetin	380±08.0	326±09.6	294±13.4	266±09.3	177±12.7	164±10.3	132±12.3	109±06.2	557±20.7	490±19.7	426±25.5	375
10.	3,4 Dihydroxy benzaldehyde	353±11.7	326±13.4	309±09.5	294±07.2	198±10.9	164±12.5	159±11.2	132±09.3	551±17.6	490±19.3	468±15.6	426
11.	Control	401±20.4				202±12.3				603			

* Mean ± S.E.

Table 2. Effect of phenolic Compounds on the nucleic acid contents of *Raphanus sativus* L. leaf discs.

S. No.	Compound	RNA (µg g^{-1} f.w.)*				DNA (µg g^{-1} f.m.)*			
		10^{-6} m	10^{-5} m	10^{-4} m	10^{-3} m	10^{-6} m	10^{-5} m	10^{-4} m	10^{-3} m
1.	P-Hydroxybenzoic acid	1687±15	1500±13	1387±13	1312±08	675±21	563±12	525±11	468±09
2.	Protocatechuic acid	1612±07	1163±08	975±10	785±06	625±11	468±14	375±11	300±08
3.	Gentisic acid	1615±05	1465±05	1355±06	1127±04	625±12	562±09	487±11	431±14
4.	m-Coumaric acid	1315±11	1205±09	1125±08	1013±05	475±06	450±04	387±07	362±12
5.	Ferulic acid	1210±03	1130±04	1015±06	902±06	437±13	400±16	375±10	337±14
6.	t-Cinnamic acid	938±06	825±04	750±05	675±03	356±09	300±10	250±08	212±08
7.	Scopoletin	1580±29	1365±25	1140±24	1026±18	595±23	488±15	430±27	400±19
8.	Juglone	1050±15	938±10	825±07	675±09	425±12	375±08	300±06	287±06
9.	Quercetin	1052±13	940±06	790±08	710±04	412±13	350±11	300±06	250±08
10.	3,4 Dihydroxy benzaldehyde	1657±13	1497±18	1320±14	1210±11	625±09	563±12	469±08	438±15
11.	Control	2025±16				750±13			

* Mean ± S.E.

TABLE 3. Effect of phenolic compounds on the total protein content of *Raphanus sativus L. leaf discs.*

S. No.	Compound	Total Protein (mg g^{-1} f. w.)*			
		10^{-6} M	10^{-5} M	10^{-4} M	10^{-3} M
1.	p-Hydroxybenzoic acid	25.18±1.51	19.56±1.53	16.75±1.52	11.18±1.04
2.	Protocatechuic acid	37.75±1.30	28.00±1.55	23.75±1.86	19.56±1.04
3.	Gentisic acid	39.18±1.03	33.56±1.16	25.18±1.35	21.00±1.05
4.	m-Coumaric acid	37.75±0.96	33.56±0.67	29.37±1.02	19.56±0.70
5.	Ferulic acid	36.37±2.32	28.00±1.00	23.75±1.29	16.75±083
6.	t-Cinnamic acid	30.75±0.96	26.56±1.00	22.37±1.14	16.75±1.25
7.	Scopoletin	39.37±1.23	37.75±1.35	34.06±1.41	26.56±0.93
8.	Juglone	22.37±1.53	19.56±0.92	18.18±1.30	12.56±1.20
9.	Quercetin	32.18±0.87	29.37±1.31	25.18±0.74	22.37±1.18
10.	3, 4 Dihydroxy-benzaldehyde	39.25±1.05	32.19±1.17	26.62±0.89	21.06±0.97
11.	Control		44.75±1.59		

* Mean ± S. E.

of RNA and DNA (Table 2). t-Cinnamic acid, protocatechuic acid, juglone and quercetin were most effective in decreasing the nucleic acids level. A depression in the levels of total proteins was observed (Table 3). p-Hydroxybenzoic acid, juglone and ferulic acid lowered the contents of proteins to a maximum extent.

In earlier studies, gentisic acid9, scopoletin[10], juglone[11], ferulic acid and coumaric acid[12] were found to be growth inhibiting. In the present study, all these compounds were found to be stimulating senescence. Thimann[1] suggested a relationship between senescence of leaves and the opening and closure of stomata and that several senescence inducing substances also act as inducers of stomatal closure. Some of the phenolics, scopoletin, chlorogenic acid were found to induce stomatal closure in tobacco and sun flower[13] Rao14 found ferulic acid, protocatechuic acid, p-hydroxybenzoic acid and gentisic acid inhibiting stomatal closure in Commel*ina benghalensis* epidermal peelings. This study presents a case for the inclusion of certain phenolic compounds to the list of growth regulators which stimulate the process of senescence.

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