Histological analysis may be another rapid indication of the susceptibility of new tomato cultivars to cracking. The shape of the cells and deposition of cutin appear to play an important role in the cracking of tomato skins. Tomato cultivars resistant to concentric cracking possessed flattened epidermal and hypodermal cells for the first few rows. For tomato cultivars resistant to radial cracking, the cutin penetrated into the third layer of cells. Less total cutin resulted in a higher τ, η, and E_f. Flat cells in the first few rows of the hypodermis were associated with a reduced τ.

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Relationships and Interactions between Phenylalanine Ammonia-lyase, Phenylalanine Ammonia-lyase Inactivating System, and Anthocyanin in Apples[1]

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Abstract: **Low temperature (6°C) in light stimulated the accumulation of phenylalanine ammonia-lyase (PAL) and anthocyanin and reduced the level of phenylalanine ammonia-lyase inactivating system (PALS-IS) in the skin of whole apples. Without light, anthocyanin synthesis did not take place in the skin of whole apples, and the level of PAL was found to be very low. In skin disks held in the dark, however, PAL activity increased but no anthocyanin was synthesized. In both the light and dark conditions, PAL-IS in the skin of whole apples was about 3.5 times higher than that in the apple skin disks after 44 hours of incubation at 18°. In the skin of whole apples, cycloheximide (10, 25, and 50 μM), chloramphenicol (50, 100, and 200 μM), and puromycin (10 and 25 μM) increased the accumulation of PAL and anthocyanin and reduced the level of PAL-IS. In the apple skin disks, cycloheximide (0.1, 1.0, 5.0, and 10.0 μM) and chloramphenicol (10.0 and 50.0 μM) inhibited the accumulation of PAL and anthocyanin to varying degrees. Puromycin had little effect on the accumulation of PAL but strongly inhibited the synthesis of anthocyanin. None of the antibiotics, at the concentrations tested, had any effect on the accumulation of PAL-IS in apple skin disks.**

The development of red color is one of the serious problems confronting the growers of red apples in New York State. Since Extra Fancy or Fancy grades are based on quantity of color, the red coloration is an important economic factor in the production and marketing of red apples.

The major red pigment of apple is a soluble anthocyanin called cyanidin-3-galactoside (24). Its synthesis is influenced by phenylalanine ammonia-lyase (PAL), first described by Koukol and Conn (20). The synthesis of anthocyanin in several plant tissues is associated with increased PAL activity (6, 7, 13, 14, 19). PAL activity is affected by light, temperature, growth regulators, inhibitors of RNA and protein synthesis, wounding (3, 11), and by mineral nutrition (21). The recent finding that a phenylalanine ammonia-lyase inactivating system (PAL-IS) capable of inactivating PAL *in vitro* has increased interest in the study of the possible role of this inactivating system as a regulator of PAL in the plant. PAL-IS has been demonstrated in the extracts of leaf disks of sunflower (8, 9, 10, 26), red cabbage seedlings (9), and sweet potato root (27). In addition, a macromolecule inhibitor of PAL has been reported in gherkin hypocotyls (1, 17). The reaction of PAL inhibitor is reversible (1) while the reaction of PAL-IS is not reversible (27). The main difference between the PAL-IS from sunflower leaf and sweet potato root tissue is in their pH optima. The assay pH optima of the sunflower PAL-IS is 9.5, and that of sweet potato is 6.0.

The objective of the present investigation was to study the relationships and the interactions between the activity of PAL and PAL-IS and the synthesis of anthocyanin in apples. This paper reports on the effect of light, low temperature, and antibiotics on the synthesis of anthocyanin and the accumulation of PAL and PAL-IS in the skin of whole apples and in apple skin disks under these varying conditions.

Materials and Methods

Experimental materials and conditions.

Green apples ('Red Spy') were harvested in mid-September from shaded positions of trees in the Cornell orchard, Ithaca, during the 1976 and 1977 growing season and stored in a cold room for about 1½ month (about 2°C) until used for the whole apple and apple skin disk experiments. When whole apples were

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used in the experiment, a small slice on one side of the apple was removed in order to stand the apple in a tray with distilled water. The apple skin discs used were 1.0 cm in diameter and about 1 mm thick. When whole apples with spur leaves attached were required, they were picked freshly from the orchard. Light (2.4 ± 0.1 mW/cm^2) was supplied by fluorescent lamps and temperature was controlled (± 1.0°) by refrigeration in growth chambers.

Temperature and light experiments.

Whole apples: Whole apples with spur leaves attached were preincubated in constant light at 18°C for 8 hr. After preincubation, they were divided into 7 different groups. Table 1 shows the daily temperature and light regimes used for each group. Discs were sampled from apple skin (to be referred as skin of whole apples) of each group immediately before the changes in light and temperature, frozen in liquid nitrogen and stored in a freezer until used for PAL and PAL-IS analysis. For anthocyanin determination, discs were put directly into 0.1% HCl in methanol and stored in a freezer.

Apple skin discs: The apple skin discs newly cut from stored apples were floated on 0.1M sucrose at 18°C. For both the constant light and constant darkness experiments, the samples were collected at 0, 20, 44, and 68 hr after incubation.

Experiments with antibiotics.

Whole apples: The antibiotics (cycloheximide, chloramphenicol, and puromycin) were each dissolved in 0.1M sucrose with 0.1% Tween 20. Whole apples were dipped for 2 min in the antibiotic solution and then exposed to fluorescent light for 72 hr at 20°C.

Apple skin discs: The antibiotics were dissolved in 0.1M sucrose, and the disks were floated on the antibiotic solution in light for 72 hr at 20°C.

Extraction and assay.

PAL: Apple skin discs (20/sample) were shattered with the grinder of a tissue homogenizer in a 50 ml polycarbonate tube in liquid nitrogen until a fine powder was obtained. About 100 mg polyclar, polyvinylpolypyrrolidone (GAF Corp. N. Y.), was then added to the tube which was then shaken to mix the polyclar with the powdered skin. Next 4 ml 0.1M sodium borate, pH 8.8, was added to the mixture. The homogenate was centrifuged in a J-21C Beckman centrifuge at 2,000 g for 2 min. The pellet was discarded and the supernatant was centrifuged again at 20,000 g for 30 min. The pellet was then discarded and the supernantant was used for assaying PAL and protein. The reaction mixture for PAL assay consisted of 0.3 ml of the supernatant, 1.0 ml of 0.06 M L-phenylalanine, and 1.7 ml of 0.1 M sodium borate buffer at pH 8.8. In the reference mixture, L-phenylalanine was replaced by water. The activity of PAL was measured by recording the production of cinnamate over time at 40°C. Cinnamate was detected at 280 nm by using a Beckman DK-2A spectrophotometer. A unit of PAL was defined as the production of 1 μmole of cinnamate per min at 30°.

Table 1. Daily temperature and light regimes used in experiments with whole apples.

Treatment group	Temperature (°C)		Light (L) or Dark (D)	
	1st 12hr period	2nd 12hr period	1st 12hr period	2nd 12hr period
1	18	18	L	L
2	6	18	L	L
3	18	18	D	L
4	6	18	D	L
5	18	18	D	D
6	12	18	L	L
7	12	18	D	L

The activity of PAL-IS was very low when the extraction was carried out at pH 8.8 Therefore it was not likely that PAL was inactivated by PAL-IS *in vitro* during the course of assay and/or isolation.

PAL-IS. The procedure used to extract PAL-IS was similar to that used to extract PAL, except that 0.1M 2(N-morpholone) ethane sulfonic acid (MES) buffer at pH 5.5 was used instead of 0.1M sodium borate at pH 8.8. The pH of the supernatant after centrifugation at 20,000 g for 30 min was brought up to 8.8 by adding sodium borate powder. The supernatant was centrifuged once again at 1,000 g for 10 min to remove the excess sodium borate. This final supernatant was immediately used for assay of PAL-IS (no PAL activity was ever detected in this supernatant). PAL-IS activity was assayed by adding a known amount of PAL from *Rhodotorula glutinis* (from P.L. Biochemicals, Milwaukee, Wisc.), about 200 mU/ml, to 50 μl of the supernatant, and an appropriate amount of sodium borate buffer was added to make a total volume of 65 μl. The mixture was then incubated at 30°C. Controls were prepared with PAL from *R. glutinis* and buffer only. Samples were drawn immediately before incubation and 10 min after, and PAL activity was assayed. Preliminary experiments showed that the inactivation of PAL by isolated PAL-IS was logarithmic with time. A milliunit (mU) of PAL-IS was defined as 1% loss in PAL activity per min.

Anthocyanin: Apple skin discs (usually 4) were extracted in 3 ml of 0.1% HCl in methanol at room temperature (about 22°C). The anthocyanin absorbance, A, was determined from the absorption spectrum by means of the formula,

$$A_{anthocyanin} = (A_{550} - A_{620}) - 0.2(A_{650} - A_{620}).$$

The anthocyanin content was determined by using the molar extinction coefficient of 4.62 × 10^4 (28) in 95% ethanol and 0.1 N HCl(85:15).

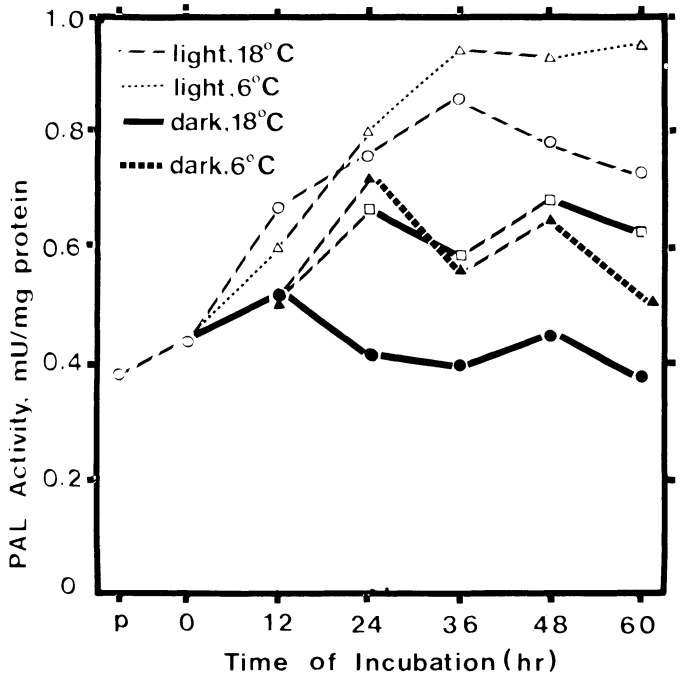

Fig. 1. Effects of light and temperature on the accumulation of PAL in the skin of whole apples. Each point represents the average of 3 replicates. The standard deviation value is between 5 and 10% of the average. (p = preincubation.)

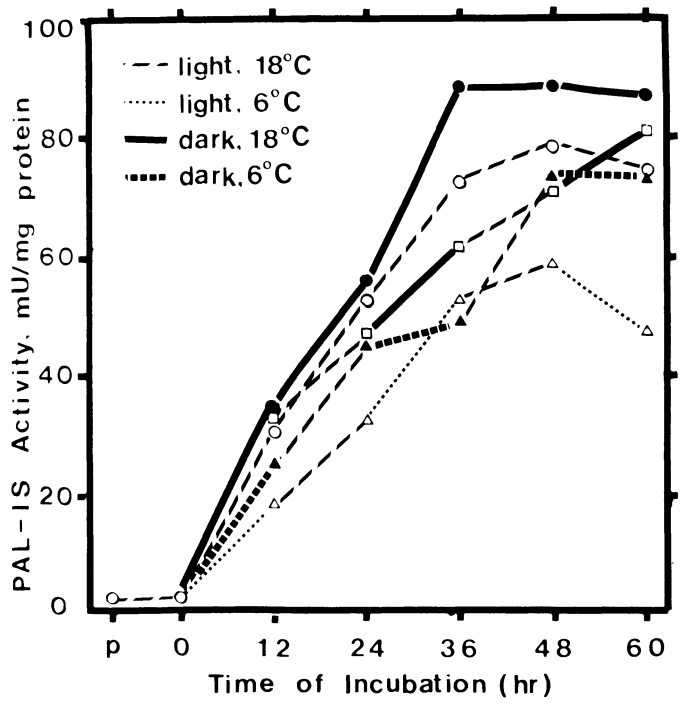

Fig. 2. Effects of light and temperature on the accumulation of PAL-IS in the skin of whole apples. Each point represents the average of 3 replicates. The standard deviation value is between 5 and 15% of the average. (p = preincubation.)

Protein: The samples were precipitated with 5% trichloroacetic acid and the content of protein was determined by using the biuret method (18) with bovine serum albumin as standard.

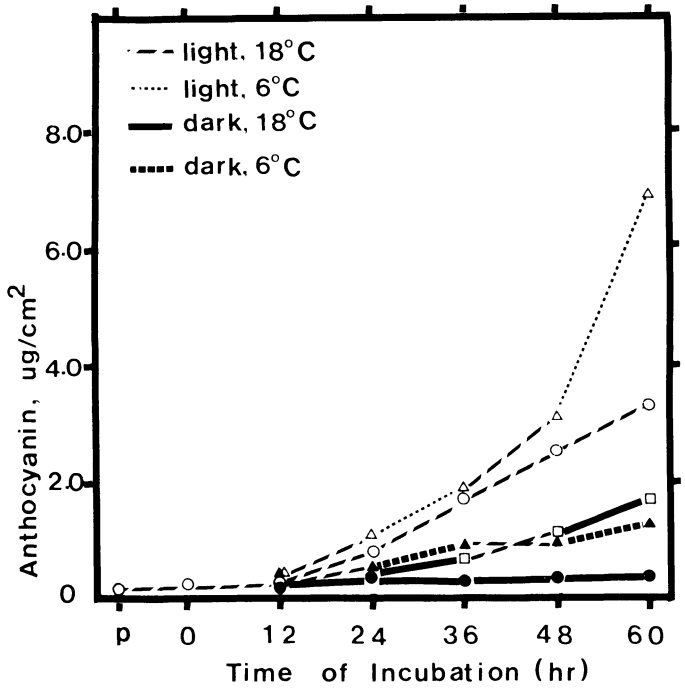

Fig. 3. Effects of light and temperature on the accumulation of anthocyanin in the skin of whole apples. Each point represents the average of 3 replicates. The standard deviation value is between 5 and 10% of the average. (p = preincubation.)

Table 2. Accumulation of PAL, PAL-IS, and anthocyanin in constant light at different temperature regimes.

Time of incubation (hr)	PAL (mU/mg protein)		PAL-IS (mU/mg protein)		Anthocyanin ($\mu g/cm^2$)	
	8-18°C[z]	12-18°C[y]	6-18°C[z]	12-18°C[y]	6-18°C[z]	12-18°C[y]
0	0.40	0.40	15.8	15.8	0	0
12	0.60**	0.39	18.2**	34.5	0.09**	0.04
24	0.80**	0.48	32.1**	54.6	0.87**	0.44
36	0.94**	0.67	52.9	60.7	1.64**	0.67
48	0.93**	0.82	58.8	65.8	2.95**	1.47
60	0.95**	0.80	46.2**	68.2	6.69**	2.43
LSD 5%	0.16	0.12	9.0	11.7	0.65	0.27

[z]Alternating 12 hr at 6°C and 12 hr 18°C, first 12 hr at lower temperature.

[y]Alternating 12 hr at 12°C and 12 hr 18°C, first 12 hr at lower temperature.

**Significantly different from that at 12-18°C at 5% level.

Results and Discussion

Effects of light and temperature on skin of whole apples. Low temperature (6°C) both in light and dark stimulated the accumulation of PAL (Fig. 1). The PAL level in apples that received alternating 6° and 18° treatment in light was 20% higher than that in apples at constant 18° in light after 48 hr of incubation, and 35% higher after an incubation of 60 hr. Low temperature reduced the level of PAL-IS (Fig. 2). In constant light, PAL-IS activity was relatively lower in the apples that received alternating 6° and 18° treatment when compared to that in apples receiving constant 18° after 60 hr of incubation. Low temperature also had a strong stimulatory effect on the synthesis of anthocyanin in the skin of whole apples (Fig. 3). The level of anthocyanin (6.92 $\mu g/cm^2$) in the apples that received alternating 6° and 18° treatment was more than 100% higher than that (3.27 $\mu g/cm^2$) found in apples that received 18° throughout the experiment.

The comparison between the effect of various low temperatures (6° and 12°C) on the accumulation of PAL, PAL-IS, and anthocyanin is shown in Table 2. Apples that received the alternating 6° and 18° treatment had higher levels of PAL and anthocyanin and lower levels of PAL-IS than apples that received alternating 12° and 18° treatment.

These results agree with the findings in earlier studies (7, 13, 14, 19) and also indicate that the rapid increase of anthocyanin accumulation following low-temperature treatments was preceded by increases in the level of PAL. No anthocyanin accumulation was observed when there was no increase in PAL (for example in the dark treatment).

In earlier studies, the activity of PAL followed the usual pattern of increase and decrease attributed to synthesis and inactivation (3, 8, 10, 17, 25, 26, 27, 29). In the present experiment, low temperature was found to reduce the level of PAL-IS, which allowed the accumulation of PAL and so promoted the synthesis of anthocyanin. It is well established that cool temperature is necessary to insure good coloration in apples (22). Explanation at the molecular level in here provided to support this finding.

The low-temperature effect on PAL has also been observed in tomato fruits (23) and gherkin seedlings (15). In tomato fruits, PAL activity increased at temperatures below 10°C and failed to increase at 15° or 20°. Engelsma (15) explained that at a temperature above 10°, PAL-IS compensated for PAL synthesis and the end product of the reaction catalyzed by PAL was involved in the induction and/or functioning of PAL-IS. He inferred that as a consequence of the low rate of synthesis of

Table 3. Accumulation of PAL, PAL-IS, and anthocyanin in whole apples and apple skin discs in constant light at 18°C.

Time of incubation (hr)	PAL (mU/mg protein)		PAL-IS (mU/mg protein)		Anthocyanin ($\mu g/cm^2$)	
	Whole apples	Skin discs	Whole apples	Skin discs	Whole apples	Skin discs
0	0.48	0.48	18.1	18.1	0	0
20	0.67**	1.27	30.6	28.3	0.06**	0.02
44	0.86**	2.94	72.7**	28.3	1.60**	0.51
68	0.73**	3.75	75.2**	30.2	3.10	3.50
LSD 5%	0.13	0.40	14.2	3.8	0.41	0.50

**Significantly different from that in skin discs at 5% level.

Table 4. Accumulation of PAL, PAL-IS and anthocyanin in whole apples and apple skin discs in darkness at 18°C.

Time of incubation (hr)	PAL (mU/mg protein)		PAL-IS (mU/mg protein)		Anthocyanin ($\mu g/cm^2$)	
	Whole apples	Skin discs	Whole apples	Skin discs	Whole apples	Skin discs
0	0.48	0.48	18.1	18.1	0	0
20	0.40**	0.87	32.3**	20.2	0.13	0.07
44	0.40**	1.44	88.2**	20.8	0.17	0.08
68	0.38**	1.90	86.3**	25.3	0.13	0.19
LSD 5%	0.06	0.15	12.0	4.2	0.09	0.13

**Significantly different from that in skin discs at 5% level.

these end products at low temperature, newly synthesized PAL was not inactivated and previously synthesized PAL was released from an enzyme inactivating system. Engelsma's data can also be explained if it is assumed that the cold treatment destroyed or suppressed the development of some of the inactivating system and then it was synthesized again when the tissues were returned to higher temperatures.

Skin of whole apples vs. apple skin discs. The accumulation of PAL, PAL-IS, and anthocyanin in the skin of 'Red Spy' whole apples and apple skin discs in constant light were compared (Table 3). In skin discs, PAL activity increased with time of incubation and reached its peak value of about 3.75 mU/mg protein after 68 hr of incubation whereas in the skin of whole apples it increased only to 0.73 mU/mg protein. The level of PAL-IS increased rapidly in the skin of the whole fruit but remained unchanged in skin discs. The anthocyanin level in apple skin discs (3.50 $\mu g/cm^2$) was only slightly higher than that in the skin of whole apples (3.10 $\mu g/cm^2$). These results further indicate that the physiology between whole apples and apple skin discs is different.

The anthocyanin level did not increase to the same extent as the PAL level in the apple skin discs. This difference might be because at these light (2.4 ± 0.1 mW/cm^2) and temperature (18°C) conditions, anthocyanin might reach a saturation level with respect to PAL at levels lower than 3.75 mU/mg protein. However, the amount of anthocyanin accumulated did depend on the initial increase in PAL activity, Higher PAL level in the apple skin discs is justified by the fact that besides accumulating anthocyanin, apple skin discs also produced a tremendous amount of other phenolic compounds (6).

When light was excluded, no anthocyanin was formed in either skin of whole apples or skin discs, although the PAL level increased from 0.59 to 1.90 mU/mg protein in skin discs (Table 4). Probably the synthesis of anthocyanin requires some other light dependent factors. In the skin of whole apples, the PAL level remained unchanged but the PAL-IS level increased substantially, from 15.6 to 86.3 mU/mg protein, during the incubation period. PAL-IS level remained unchanged in the apple skin disks.

These results indicate that PAL-IS regulates the activity of PAL in the skin of whole apples. The lower level of PAL-IS found in apple skin discs allowed the accumulation of PAL, whereas the higher levels of PAL-IS in the skin of whole apples inhibited the accumulation of PAL. Wounding (by cutting the whole apples into skin discs) seemed to destroy the ability of apple skin to synthesize PAL-IS. It is, therefore, concluded that in the skin of whole apples synthesis of anthocyanin depends on PAL activity as long as the light requirement is fulfilled.

Effects of antibiotics on the skin of whole apples and apple skin discs. Cycloheximide (10, 25, and 50 μM), chloramphenicol (50, 100, and 200 μM), and puromycin (10 and 25 μM) increased the accumulation of PAL and anthocyanin, and reduced the level of PAL-IS (Table 5). Under the same experimental conditions (20°C and constant light), apple skin discs responded differently (Table 6). Cycloheximide at concentrations of

Table 5. Accumulation of PAL, PAL-IS, and anthocyanin in the skin of whole apples with different treatments of antibiotics (± SE).

Treatment[z]	PAL (mU/mg protein)	PAL-IS (mU/mg protein)	Anthocyanin ($\mu g/cm^2$)
Sucrose	1.72 ± 0.12	26.2 ± 3.4	1.58 ± 0.08
Cycloheximide (μM)			
10	3.35 ± 0.30	6.6 ± 1.2	3.18 ± 0.24
25	3.35 ± 0.35	6.8 ± 1.3	3.40 ± 0.30
50	3.46 ± 0.28	9.8 ± 1.4	3.84 ± 0.38
Chloramphenicol (μM)			
50	3.23 ± 0.28	16.4 ± 2.1	4.99 ± 0.30
100	2.89 ± 0.18	16.4 ± 2.4	2.57 ± 0.15
200	2.71 ± 0.18	13.2 ± 1.4	2.31 ± 0.17
Puromycin (μM)			
10	2.65 ± 0.24	20.8 ± 3.4	3.16 ± 0.17
25	2.89 ± 0.22	20.0 ± 3.2	2.81 ± 0.22
50	1.79 ± 0.20	18.2 ± 3.0	1.70 ± 0.11

[z]At time 0, the activity of PAL was 0.81 mU/mg protein and PAL-IS was 9.8 mU/mg protein, and the level of anthocyanin was 0 $\mu g/cm^2$. Time of incubation was 72 hr.

Table 6. Accumulation of PAL, PAL-IS, and anthocyanin in apple skin discs with different treatment of antibiotic (± SE).

Treatment[z]	PAL (mU/mg protein)	PAL-IS (mU/mg protein)	Anthocyanin ($\mu g/cm^2$)
Sucrose	7.14 ± 0.60	10.0 ± 5.0	3.45 ± 0.22
Cycloheximide (μM)			
0.01	8.03 ± 0.55	5.4 ± 5.1	3.08 ± 0.12
0.05	8.07 ± 0.50	10.1 ± 5.4	3.67 ± 0.15
0.10	5.24 ± 0.49	8.8 ± 4.3	1.43 ± 0.10
1.00	2.43 ± 0.21	6.2 ± 4.9	0
5.00	1.64 ± 0.20	7.3 ± 4.4	0
10.00	1.86 ± 0.20	10.5 ± 4.2	0
Chloramphenicol (μM)			
0.10	7.83 ± 0.58	10.4 ± 3.9	2.97 ± 0.17
0.50	7.37 ± 0.68	12.9 ± 3.2	3.34 ± 0.20
1.00	7.57 ± 0.61	15.4 ± 2.3	3.24 ± 0.20
10.00	5.29 ± 0.50	20.0 ± 6.3	1.48 ± 0.15
50.00	5.29 ± 0.50	15.4 ± 5.4	0.90 ± 0.08
Puromycin (μM)			
0.10	6.43 ± 0.52	13.6 ± 5.4	2.49 ± 0.14
1.00	6.86 ± 0.55	15.4 ± 4.3	2.60 ± 0.20
5.00	7.00 ± 0.67	8.8 ± 4.8	2.17 ± 0.18
50.00	7.37 ± 0.48	13.0 ± 2.1	0.19 ± 0.01
100.00	5.18 ± 0.62	12.9 ± 3.4	0.11 ± 0

[z]At time 0, the activity of PAL was 0.97 mU/mg protein and PAL-IS was 13.6 mU/mg protein, and the level of anthocyanin was 0 $\mu g/cm^2$. Time of incubation was 72 hr.

0.01 and 0.05 μM and chloramphenicol at concentrations of 0.1, 0.5 and 1.0 μM had no effect on PAL and anthocyanin level. However, cycloheximide at concentrations of 0.1, 1.0, 5.0, and 10.0 μM and chloramphenicol at concentrations of 10.0 and 50.0 μM inhibited the accumulation of PAL and anthocyanin to varying degrees. At the concentrations tested, puromycin had little effect on the level of PAL but strongly inhibited the synthesis of anthocyanin in the apple skin discs. This may be due to the inhibition of the other enzymes in the pathway of anthocyanin biosynthesis (16) which are more sensitive to puromycin than PAL. None of the antibiotics tested had any effect on the level of PAL-IS in apple skin discs.

Low concentrations of antibiotics promoted the accumulation of PAL and anthocyanin in the skin of whole apples, presumably because they inhibited the synthesis of PAL-IS. The effect of cycloheximide on PAL has been studied in many other tissues (2, 6, 11, 12, 26, 29, 30), and some workers (12, 26) have stated that both PAL synthesis and inactivation were sensitive to cycloheximide, with inactivation being more sensitive than synthesis.

The level of PAL-IS remained unchanged throughout the course of the experiments with apple skin discs. This result provides further evidence to support the hypothesis that wounding destroys the ability of the tissue to synthesize PAL-IS. This might be one of the explanations why antibiotics did not stimulate the accumulation of PAL and anthocyanin in apple skin discs.

Where PAL activity was stimulated by antibiotics, an alternative mechanism is feasible. Cycloheximide has been shown to stimulate ethylene production which in turn increases the accumulation of PAL in 'Jonathan' whole apples (4, 5, 16). It was also suggested that cycloheximide reduced the accumulation of PAL and anthocyanin in apple skin discs because it inhibited ethylene biosynthesis.

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N Form and Concentration: Effects on N Absorption, Growth, and Total N Accumulation with Southernpeas[1]

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Abstract. **Nitrate and NH_4^+ absorption by southernpeas (*Vigna unguiculata* (L.) Walp.) during the growth cycle was determined in solution culture at 7-day intervals. Dry weight and total mg of N for each plant part were determined at bloom initiation, pod initiation, and seed maturity. When the N concentration was deficient virtually all of the NO_3^- was absorbed by southernpea plants from the 3rd week until harvest. At sufficient N concentrations a single peak NO_3^- uptake period occurred in the growth cycle prior to bloom initiation. Nitrate absorption predominated when NH_4^+ constituted 50% or more of the N form at both N concentrations. Ammonium had either 1, 2 or 3 absorption peaks depending upon its relative concentration with NO_3^-. The first NH_4^+ absorption peak was the largest and coincided with the growth cycle just prior to bloom initiation. Dry weight and N accumulation were greatest prior to bloom initiation. Subsequent accumulation occurred primarily in the seed. A reduction in N content and dry weight in the vegetative portions, particularly the leaves, coincided with the accumulation of N and dry weight in the seeds. Use of total mg of N in the vegetative tissue as a means of indicating the N status of the plant and subsequent yield was not accurate.**

Growth of southernpea seedlings is greater with combinations of NO_3^- and NH_4^+ ions as opposed to either ion used alone (11), but the influence of N form on N absorption throughout the growth cycle has not been investigated. Reports on the effectiveness of fertilizer N applied to southernpea have been conflicting in terms of growth and yield responses, though N form was generally not considered (2, 5, 8, 12, 13, 14, 15, 19).

The absorption of NH_4^+ lowers the medium pH, whereas NO_3^- absorption increases pH (16). These changes in pH may subsequently influence further absorption of NO_3^- and/or NH_4^+ as maximum NO_3^- uptake occurs at a solution pH of 4.5 to 5 and for NH_4^+, pH 6 to 6.5 (17). Since the pH change due to ion absorption occurs rapidly at the root surface (7), the pH at the root surface can differ from the medium pH by one pH unit or more (18). While most soils buffer against pH changes, this buffering action may have limited influence in the rhizosphere as rapid absorption of NO_3^- and/or NH_4^+ ions may result in a higher concentration of H^+ and/or OH^- ions at the sites of active ion uptake. Consequently, a fluctuating pH at the root surface likely occurs with outward diffusion of H^+ and OH^- ions from zones of higher concentrations to zones of lower concentrations and/or with periodic dilution by soil water from rain and irrigation. This interaction of N absorption and rhizosphere pH would be an important factor influencing N form utilization, especially if a high demand for a particular N form exists during the growth cycle.

For certain legumes N distribution within the plant varies depending on the N form supplied (1, 9, 10). In addition to the influence of N form on N content and distribution within the plant, the stage of plant development has been suggested as a factor in certain legumes (10).

The objectives of this study were to determine the effect of N concentration and form as combinations of NO_3^- and NH_4^+ at defieiency and sufficiency concentrations on N absorption and pH through the growth cycle, and dry weight and N content of southern pea plants at bloom initiation, pod initiation, and seed maturity.

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