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Relative Phosphorus Dependency of Citrus Rootstocks Is Reflected in Leaf Nitrogen Metabolism

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Abstract. **The relative P dependency obtained for 3 citrus rootstock cultivars was rough lemon [*Citrus limon* (L)] > Carrizo citrange [*Poncirus trifoliate* x *C. sinensis* (L.)] > trifoliate orange [*P. trifoliate* (L.)]. After only 12 weeks of P deprivation, the youngest, fully expanded leaves of rough lemon (about 21 days old) had amounts of total N, nitrate, and NH_3 that exceeded the levels in +P leaves of the same age by 8.4 mg total N, 2.6 mg nitrate, and 0.6 mg NH_3 per gram of dry weight. It took 7 months for similar levels of these compounds to accumulate in the youngest, fully expanded leaves of Carrizo citrange (a less P-dependent rootstock) when grown under low P conditions. After 12 weeks of P deficiency, the incorporation of $NaH^{14}CO_3$ into arginine plus urea was 13-, 7.4-, and 4.7-fold greater in P-deficient leaves than P-sufficient leaves for rough lemon, Carrizo citrange, and trifoliate orange, respectively. Concomitantly, arginine accumulated in −P leaves to a concentration 4.2-, 2.1-, and 1.4-fold greater than in +P leaves, respectively, for the same 3 rootstocks. The data clearly demonstrate that relative P dependency of citrus rootstocks influences their N metabolism and support the hypothesis that a more P-dependent rootstock will accumulate total N, nitrate, and NH_3 sooner or to a greater extent, exhibit a greater rate of *de novo* arginine biosynthesis, and accumulate a higher level of arginine than a less P-dependent rootstock. Thus, calculation of the N:P ratio during leaf nutrient analysis may be useful in evaluating the nutritional status of citrus trees. Whenever N:P ratios were >20, *de novo* arginine biosynthesis and arginine accumulation increased significantly, indicative of ammonia detoxification. The N:P ratio of P-sufficient plants was always <15 when arginine metabolism was normal.**

The capacity of about 20 plant species and cultivars, all commercially important citrus rootstocks, to take up essential nutrients has been reviewed extensively by Embleton et al. (2). Based on results reported in the literature, they ranked citrus rootstocks according to their tendency to maintain a specified concentration of each essential nutrient in the leaves of the same scion cultivars. The relative capacity of citrus rootstocks to maintain P in the leaves of their scions was of interest to us for 2 reasons. First, Menge et al. (9) determined the mycorrhizal dependency of 6 citrus rootstocks by growing them with and without vesicular–arbuscular mycorrhizae (VAM) at different levels of P nutrition. At the end of 6 months, a relative VAM dependency was calculated for each rootstock by expressing dry weight of the VAM plants as a percentage of the dry weight of the non-VAM plant for each P treatment. VAM dependency correlated with the P nutritional status of each rootstock, but with no other nutrient, and the ranking of Menge et al. (9) for VAM dependency correlated with the relative P dependency determined by Embleton et al. (2) for the same rootstocks. Second, we have recently investigated arginine metabolism in 4 citrus roostocks grown under 2 levels of P nutrition. For each roostock, the activity of the *de novo* arginine pathway and the amount of arginine in young, fully expanded leaves increased with increasing severity of P deficiency (11). When P was resupplied to the −P plants, the activity of the *de novo* arginine pathway was reduced to the level of the +P plants and arginine ceased to accumulate (11). We also have demonstrated that P deficiency results in a state of NH_3 intoxication in the leaves of members of the Cucurbitaceae and Rutaceae (12). This result suggests that accelerated arginine biosynthesis is a general mechanism for detoxifying leaf tissue of excess NH_3. The net increase in the rate of *de novo* arginine and urea synthesis during P deprivation (11) is positively correlated with the relative P dependency reported by Embleton et al. (2) for each rootstock and the greater VAM dependency of the rootstocks determined by Menge et al. (9).

It was of particular interest to us that, when rootstocks were listed by decreasing ability to maintain P in their scion leaves, the order was almost the direct opposite of what it was when the same rootstocks were rated by Embleton et al. (2) for their capacity to supply N to their scions. Thus, rootstocks that are more likely to cause P deficiency in their scions (i.e., those rootstocks that have greater P dependency) are also more likely to have increased levels of N in the leaves of their scions. This observation, taken together with the results of our previous research, suggested that the amount of N, nitrate, and NH_3 accumulating during P deficiency should be greatest in those rootstocks exhibiting the greatest VAM and P dependency. To test this hypothesis, we used both sand culture and hydroponic culture to confirm the relative P dependency of rough lemon, Carrizo citrange, and trifoliate orange and to demonstrate that this characteristic was related to the capacity of the rootstocks to maintain P in their leaves. These rootstocks represent the range of P and VAM dependencies observed for citrus rootstocks: a) high—rough lemon; b) moderate—Carrizo citrange; and c) low—trifoliate orange. We then quantified the influence of P nutrition on the levels of total N, nitrate, NH_3, arginine, and *de novo* arginine synthesis of young, fully expanded leaves from these 3 rootstocks.

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Materials and Methods

Chemicals. All radiolabeled chemicals were purchased from ICN Pharmaceuticals. Liquiscint (liquid scintillation cocktail) was purchased from National Diagnostics. Mineral salts for Hoagland's (4) and modified Shive's (1) nutrient solutions were of analytical reagent quality from Fisher. All other chemicals were purchased from Sigma.

Plant materials. For sand culture, seeds of rough lemon, Australian trifoliate orange, and Carrizo citrange were planted in sterile sand containing P applied as $Ca(H_2PO_4)_2$ to sand at the rate of 5 $\mu g \cdot g^{-1}$ (P deficient; −P) or 100 $\mu g \cdot g^{-1}$ (P sufficient; +P) and watered with half-strength Hoagland's solution as previously described (11). Analyses were carried out when plants were 6, 12, and 28 weeks old.

For hydroponic culture, seeds were imbibed in distilled H_2O for one day and germinated between paper towels for 7 days at 30°C. Seedlings of uniform appearance were selected and transferred to polyurethane breadboxes containing complete Shive's solution, aerated, and allowed to grow at 30° under continuous illumination (310 $\mu mol \cdot s^{-1} \cdot m^{-2}$). After 7 days, plants were transferred to fresh Shive's nutrient solution (+P) or to Shive's nutrient solution modified by replacing the 1 mM KH_2PO_4 with an additional 0.5 mM K_2SO_4 (−P). Nutrient solutions were decanted and replaced by fresh solutions every 7 days for a period of 9 weeks. All analyses employed the youngest, fully expanded leaves harvested and pooled from 5 to 20 plants for sand culture and from 50 plants for hydroponic culture. Experiments were initiated at seed germination, no two experiments were initiated in the same weeks, and several seed lots were used in the many experiments. Experiments consisted of single treatments. Each datum is the average value obtained for N experiments. All statistical analyses employed Duncan's multiple range test. In each table, the age at which one rootstock first becomes significantly different from the others is indicated in the vertical column row by a different letter.

Table 1. Phosphorus dependency of 4 citrus rootstocks.[z]

	Sand culture			Hydroponic culture
			Duration	
Rootstock	6 weeks	12 weeks	28 weeks	9 weeks
	Phosphorus dependency[y]			
Rough lemon	182	442	820	643
Carrizo citrange	114	211	677	137
Trifoliate orange	---	207	500	104
	Milligrams of P per gram dry weight of +P leaf tissue			
Rough lemon	2.3[x]	2.0 a[xw]	1.9	3.5
Carrizo citrange	3.0[x]	2.2 a[x]	2.2	2.5
Trifoliate orange	---	2.9 b[x]	3.1	3.2

[z]Calculated using the mean weight or height of 5 to 20 plants per experiment. Dry weight was used for plants 6, 9, and 12 weeks old; height for 28-week-old plants.
[y]P dependence = +P plant height or dry weight/−P plant height or dry weight.
[x]Data are the means of 2 to 4 different experiments; all other values were obtained in a single experiment.
[w]Data within a column separated by Duncan's multiple range test, $P = 5\%$.

Table 2. The amount of total N, NO_3^-, and NH_3-NH_4^+ in the leaves of −P plants in excess of the amounts observed for the leaves of +P plants.

	Sand culture			Hydroponic culture
			Duration	
Rootstock	6 weeks[z]	12 weeks[z]	28 weeks	9 weeks
	Milligrams of N per gram dry weight of leaf tissue			
Rough lemon	−3.5	8.4 b[x]	14.3	−0.02
Carrizo citrange	−1.4	0.6 a	8.1	−4.2
Trifoliate orange	---[y]	−3.1 a	5.2	−3.2
	Micrograms of NO_3^- per gram dry weight of leaf tissue			
Rough lemon	410	2572 b	2430	1720
Carrizo citrange	330	690 a	1780	−1390
Trifoliate orange	---	520 a	1383	790
	Micrograms of NH_3-NH_4 per gram dry weight of leaf tissue			
Rough lemon	154	615 b	---	455
Carrizo citrange	174	108 a	619	−380
Trifoliate orange	---	−230 a	---	−430

[z]Data are the means of 2 to 5 different experiments; all other values were obtained in a single experiment.
[y]Not enough tissue available to analyze all rootstocks.
[x]Values within a column separated by Duncan's multiple range test, $P = 5\%$.

Leaf tissue preparation. Excised leaves were immediately frozen in liquid N, lyophilized, and ground with a Wiley mill to a size fine enough to pass through a 40-mesh screen.

Leaf P and N content. Phosphorus content of leaves was determined for a 50-mg sample by a colorimetric assay employing molybdivanadophosphoric acid (6). Absorbance at 420 nm was linear for concentrations of P from 0 to 0.4 mg per 100 mg leaf tissue (dry weight).

Total N was determined for a 25-mg sample using the conventional micro-Kjeldahl method; nitrate for a 100-mg sample extracted in 100 ml of 16 mM $CaSO_4 \cdot 2H_2O$ using a Technicon Autoanalyzer (14); and NH_3-NH_4^+ for a 200-mg sample extracted in 50 ml of 1 N KCl using a Technicon Autoanalyzer (15). Amino acid content was determined using a Beckman 120 C Amino Acid Analyzer calibrated with commercial standards in the appropriate buffer at 570 nm (7).

Activity of the *de novo* arginine biosynthetic pathway was assessed by measuring the incorporation of $NaH^{14}CO_3$ [5 mM, 37.5 μCi (1 Ci = 37 Gbq)] into arginine plus urea in intact cells of the youngest, fully expanded leaves (500 mg fresh weight cut into 5 × 5 mm pieces, midveins removed) during a 3-hr incubation period as described previously (11). The amount [*guanidino*-^{14}C]arginine and [^{14}C]urea synthesized by the leaf tissue from $NaH^{14}CO_3$ during the 3-hr incubation period was determined using commercial arginase and urease as described by Lovatt and Cheng (8).

Results

Relative phosphorus dependency of citrus rootstocks. Phosphorus dependency is the degree to which the growth of a species or cultivar is reduced when P nutrition is less than optimal, but all other nutrients are maintained at optimal levels. P dependency = +P plant height or dry weight/−P plant height or dry weight. P dependency of the 4 citrus rootstocks calculated on a height or dry weight basis was rough lemon > Carrizo citrange > trifoliate orange (Table 1).

For plants grown in sand culture with P, the leaf concentration of P decreased sooner and to a greater degree in the more P-dependent than in less P-dependent rootstocks (Table 1). When the P concentration of the youngest, fully expanded leaves for all experiments employing P-sufficient conditions were averaged ($\bar{x}$ mg P per gram dry weight of leaf tissue ± SE), rough lemon had the lowest P concentration (2.26 ± 0.19; n = 10),

Table 3. Activity of the *de novo* arginine pathway in young, fully expanded +P and −P leaves of the 3 citrus rootstocks.

	Synthesis of arginine plus urea			
	Sand culture			
	6 weeks[z]		12 weeks[z]	
Rootstock	+P	−P	+P	−P
Rough lemon	15 ± 2(6) b[y]	149 ± 13(6) b	12 ± 2(5) b	156 ± 10(7) c
Carrizo citrange	3 ± 3(4) a	9 ± 4(4) a	5 ± 2(4) a	37 ± 12(4) a
Trifoliate orange	---	---	17 ± 4(3) ab	80 ± 8(3) b

[z]Data are the mean ± SE $NaH^{14}CO_3$ (nmol) incorporated into arginine plus urea per gram of fresh weight of leaf tissue during a 3-hr incubation with number of experiments given in parentheses.
[y]Values in a column separated by Duncan's multiple range test, $P = 5\%$.

Table 4. Effect of P deficiency on the total arginine content of the youngest fully expanded leaves of the citrus rootstocks.

	Total arginine content			
	Sand culture			
	6 weeks[z]		12 weeks[z]	
Rootstock	+P (μmol·g^{-1} dry wt)	−P (% of +P control)	+P (μmol·g^{-1} dry wt)	−P (% of +P control)
Rough lemon	64 a[y]	195 b	58 a	418 b
Carrizo citrange	73 a	84 a	55 a	213 a
Trifoliate orange	---	---	64 a	140 a

[z]Data are the average values of 2 to 4 separate experiments; differences between experiments did not exceed 30%.
[y]Values in a column separated by Duncan's multiple range test, $P = 5\%$.

followed by Carrizo citrange (2.50 ± 0.21; n = 6), and trifoliate orange (3.03 ± 0.09; n = 4). Rough lemon was significantly less than trifoliate orange at the 5% level.

Levels of total N, nitrate, and NH_3. The total N level in young, fully expanded leaves during P sufficiency (averaged for all experiments; $\bar{x}$ mg N per gram dry weight of leaf tissue ± SE) was not significantly different ($P < 0.05$) for rough lemon (29.4 ± 0.6; n = 10), Carrizo citrange (31.2 ± 1.2; n = 6), or trifoliate orange (32.4 ± 0.6; n = 4). However, during P deficiency, total N, NO_3^-, and NH_3-NH_4^- all increased (Table 2). By the end of 12 weeks of P deprivation, the youngest, fully expanded leaves of rough lemon had significantly more N ($P < 0.01$), nitrate ($P < 0.05$), and NH_3 ($P < 0.05$) than the other rootstocks. It took 28 weeks of P deficiency for Carrizo citrange exhibiting intermediate P dependency to accumulate similar levels of N, nitrate, and NH_3. At the end of 28 weeks of P deficiency, the least P-dependent rootstock, trifoliate orange, had not yet accumulated levels of these compounds approaching those observed for rough lemon after only 12 weeks of −P treatment. Consistent with our hypothesis, a more P-dependent citrus rootstock cultivar always accumulated N, nitrate, and NH_3 sooner or to a greater degree than a less P-dependent cultivar.

P dependency and the activity of the de novo *arginine biosynthetic pathway.* The activity of the pathway for *de novo* arginine synthesis in intact tissue of young, fully expanded leaves increased dramatically by the end of the first 6 weeks of P deprivation for the most P-dependent rootstock, rough lemon ($P < 0.01$) (Table 3). After an additional 6 weeks of P deprivation, the less P-dependent rootstocks also exhibited a significant increase in *de novo* arginine biosynthesis ($P < 0.01$). By the end of 12 weeks of P deficiency, the incorporation of $NaH^{14}CO_3$ into arginine plus urea per gram fresh weight of tissue during the 3-hr incubation was 13-, 7.4-, and 4.7-fold greater than that of the +P plants for rough lemon, Carrizo citrange, and trifoliate orange, respectively.

P dependency and the arginine content of leaves. The concentration of arginine increased markedly in young, fully expanded leaves of rough lemon, the most P-dependent rootstock, after 6 weeks of P deprivation (Table 4). Marked increases in leaf arginine levels were not observed for the less P-dependent rootstocks until they had been deprived of P for 12 weeks. By the end of 12 weeks of −P treatment, the amount of arginine per gram dry weight of tissue had accumulated to levels 4.2-, 2.1-, and 1.4-fold greater than that of the +P plants for rough lemon, Carrizo citrange, and trifoliate orange, respectively. This accumulation represented net increases in arginine of 184, 62, and 24 μmol·g^{-1} dry weight of leaf tissue for the 3 rootstocks, respectively. In both instances, rough lemon was significantly greater ($P < 0.05$) than the 2 less P-dependent rootstocks.

Discussion

In this study, experimentally determined plant height and weight values were used to calculate P dependency. Embleton et al. (2) based P dependency on the capacity of a rootstock to maintain a specific level of P in the leaves of the same scion cultivar. Their data were compiled from results obtained with field-grown

Table 5. Nitrogen : phosphorus ratio in the leaf tissue of 4 citrus rootstocks during P deficiency and P sufficiency.

	Sand culture							
	Duration						Hydroponic culture (9 weeks)	
	6 weeks		12 weeks		28 weeks			
Rootstock	+P	−P	+P	−P	+P	−P	+P	−P
Rough lemon	14	19	14	38	13	23	9	50
Carrizo citrange	13	12	14	26	12	34	13	32
Trifoliate orange	---	---	11	32	11	20	11	31

trees, and each rootstock was judged in combination with a number of different scion cultivars. The relative P dependency of the rootstocks obtained by the 2 independent methods was in good agreement, suggesting that P dependency is probably a direct function of the rootstock rather than the scion.

The results presented demonstrate: i) that NH_3 accumulation is an early response of plants to P deficiency, with the exception of trifoliate orange; ii) that for each of the rootstocks, even trifoliate orange, NH_3 levels increased in parallel to the increasing duration of P deprivation; and iii) that the rate of *de novo* arginine biosynthesis and the concentration of arginine increased during P deprivation in a manner consistent with the detoxification of ammonia via the *de novo* arginine pathway.

The increased activity of the *de novo* arginine pathway in trifoliate orange during P sufficiency might be the basis, either totally or in part, for the failure of NH_3 to accumulate and for the maintenance of the lowest level of NH_3-NH_4^+ during P deficiency of any of the rootstocks tested (Table 2).

Taken together, the data support our hypothesis that citrus rootstocks that are sensitive to P deficiency (i.e., more P-dependent), will accumulate N, nitrate, NH_3, and arginine sooner or to a greater degree than less P-dependent rootstocks.

The observation that the level of total N in leaves of P-sufficient plants was not significantly different for rough lemon, Carrizo citrange, and trifoliate orange suggests that differences in P dependency are not due to differences in N metabolism of the rootstocks, but are due to the capacity of the rootstocks to provide P. The more P-dependent rootstocks, even when grown under +P fertilization, had significantly lower leaf P ($P < 0.05$) than trifoliate orange (Table 1). Phosphorus becomes limiting to the growth of the more P-dependent rootstocks sooner and to a greater degree, causing a reduction in growth that results in the accumulation of N in the leaves. Failure of the plant to regulate N uptake is consistent with the fact that land plants evolved under conditions in which N was limiting. Thus, there was little selection pressure for mechanisms regulating N uptake or reduction (10). In light of this, it is clear that ranking rootstocks for their tendency to maintain a given level of N in scion leaves under field conditions (2) would be compromised by the fact that P deficiency, or probably any other stress that limits canopy growth (except N deficiency), would result in N accumulation in the leaves.

A number of considerations logically follow from the results of our research. First, the N:P ratio, rather than the actual levels of these nutrients, may be more useful in evaluating the severity of P deficiency. Whenever N:P ratios >20 were observed (Table 5), *de novo* arginine biosynthesis was increased significantly (Table 3), and significant amounts of excess arginine accumulated (Table 4). These changes are consistent with NH_3 detoxification. The N:P ratio for P-sufficient plants was always <15 (Table 5); arginine metabolism was normal. The importance of the N:P ratio is supported by the observation of Hepper (3) that, for all P levels, mycorrhizal infection increased as more nitrate was supplied to the host plants, while increasing amounts of applied phosphate generally depressed VAM infection. The extent to which this infection occurred depended on the N:P ratio.

Second, leaf nutrient analysis is used routinely to design fertilizer programs for citrus cultivars and most other crops. Our results suggest that leaf total N content, which is currently the basis for N fertilizer recommendations, may not be as valuable as a measurement of leaf NH_3-NH_4^+ and arginine concentrations. For example, with *Cucurbita pepo*, leaf total N content did not increase during P deficiency, but nitrate and NH_3 were significantly increased ($P < 0.05$) in both young and mature leaves. Symptoms characteristic of NH_3 toxicity were observed for mature leaves of *C. pepo* that contained 160 μg of NH_3-NH_4^+ per gram of dry weight in excess of the level found in P-sufficient control plants. No symptoms were observed with young leaves having an excess of only 52 μg of NH_3-NH_4^+ per gram of dry weight (12).

While NH_3 is known to be toxic to plants, we are not aware that an upper threshold value for leaf NH_3 concentration has been determined for any plant species. If this value were known, leaf NH_3 content might provide a good indicator of plant health. In addition, leaf arginine content could be used to monitor stress.

Finally, the results presented in this study suggest that the P dependency of citrus rootstocks used commercially and the P nutritional status of the scions should be taken into consideration when designing a fertilization program, especially with regard to the application of N when soil P levels are marginal.

The relative P dependency of the rootstocks determined in this study paralleled their dependency on vesicular–arbuscular mycorrhizae (VAM) as determined by Menge et al. (9). In addition, relative P dependency appears to be inversely related to the hydraulic conductivity of roots (13). Trifoliate orange and Carrizo citrange have higher conductivities (resulting in higher mass flow of water and minerals to the shoots) relative to sour orange, a rootstock that ranked very similar to rough lemon for both P dependency (2) and VAM dependency (9).

Differences in hydraulic conductivity may prove to be the physiological basis for differences among citrus rootstocks in providing P to the leaves of their scions. Rootstocks with low hydraulic conductivity would be expected to be sensitive to mineral nutrient deficiencies in general.

Finally, the relative P dependency appears to be indicative of the susceptibility of citrus rootstocks to citrus nematode [*Tylenchulus semipenetrans* (Cobb)]. Sour orange and rough lemon, which are highly P-dependent (2), were demonstrated to be highly susceptible to root infection by citrus nematode. Troyer citrange was moderately susceptible, and trifoliate orange was resistant to *T. semipenetrans* (5).

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^{14}C-Gibberellic Acid Uptake, Translocation, Persistence, and Metabolism in Grapefruit

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Abstract. **The uptake, translocation, and metabolism of ^{14}C-gibberellic acid (^{14}C-GA_3) was studied in 3-year-old container-grown 'Marsh' grapefruit trees (*Citrus paradisi* Macf.). A total of 1.65 × 10^5 disintegrations per min (dpm) in 200 µl of solution was applied evenly over the entire fruit surface, or, on both surfaces of 3 to 5 subtending leaves of a fruit. Absorption of ^{14}C-GA_3 by leaves and peel began within 1 hr of application and continued for 8 hr. Translocation of labeled material from leaves to peel and the reverse began 4 to 8 hr after application and continued for 4 weeks. No labeled material was recovered from juice or seeds. Labeled material persisted in albedo, flavedo, and leaves for 8 weeks with the highest accumulations in the peel. Separation of ^{14}C-GA_3 metabolites from the 95% EtOH extract by reversed-phase HPLC produced 2 ^{14}C-labeled peaks. Analysis of these 2 peaks by β-D-glucosidase hydrolysis, *n*-butanol partitioning, and cochromatography with ^{14}C-GA_3 standards suggested that the major component was ^{14}C-GA_3 and the other a polar metabolite.**

Gibberellic acid has been applied to grapefruit for many years to delay senescence and prolong the harvest season (1, 5, 7, 10, 15); yet little is known about GA_3 metabolism in grapefruit. Goldschmidt and Eilati (11) applied GA_3 to the flavedo of 'Shamouti' oranges [*C. sinensis* (L.) Osbeck] before and after harvest. Delay of peel color development indicated that GA_3 was absorbed by both attached and detached fruits, although this effect was more marked and persistent in the former. Delay of peel color development beyond the area of application suggested lateral movement of GA_3. Goldschmidt and Galili (12) applied ^{3}H-GA_3 to detached 'Shamouti' orange peels and recovered measurable amounts of labeled material from the flavedo after 5 days of storage. In a subsequent study (13) they applied ^{14}C-GA_3 to 'Valencia' oranges on the tree and recovered only 2% of applied material from the flavedo after 24 hr.

The work of Goldschmidt et al. (12, 13) indicates the absorbed ^{14}C-GA_3 is localized in the flavedo. However, the low levels of recovery after one (13) and 5 (12) days suggests translocation of the ^{14}C-GA_3.

Exogenously applied GA_3 delays loss of chlorophyll, RNA, and proteins (8, 9) in the flavedo and maintains a compact structure in the albedo (18), suggesting that it or its metabolites are localized in these areas. Moreover, field application of GA_3 has no effect on juice quality (1, 7, 10, 15).

Evidence for persistence of exogenously applied GA_3 in citrus peel is both indirect and direct. Sprays of GA_3 delay peel color development and loss of rind firmness for as long as 7 months (5). This effect could be an indication of GA_3's persistence in citrus peel or it could indicate that the processes GA_3 effects persist this long. Jordan et al. (15) reported 0.10 to 0.16 ppm GA_3 in lemon peel 7 days after application. However, their methods could not prove this was residue in excess of endogenous gibberellins. Goldschmidt and Galili (13) applied ^{14}C-GA_3 to 'Valencia' oranges on the tree and recovered 1% of the labeled material after 100 days.

Metabolism of GA_3 has been studied in sweet oranges but not in grapefruit. Goldschmidt and Galili (12, 13) found that diethyl ether or ethyl acetate fractions of orange peel extracts contained about one-half of the recovered label on all sampling dates. The other half remained in the aqueous fraction. The organic fractions cochromatographed with GA_3 standard on silica gel H-coated plates. Total recovery decreased over time, but the ratio of ethyl acetate to water-soluble activity remained relatively constant. Consequently, they suggest this ratio was the result of a gradual catabolism of ^{14}C-GA_3 to a polar metabolite and that the rate of catabolism may be controlled by gradual release of the ^{14}C-GA_3 from vacuoles or other compartments.

Our objectives were to study the uptake, in vivo translocation, persistence, and initial metabolism of ^{14}C-GA_3 applied to 'Marsh' grapefruit peel and leaves.

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