THE NITROGEN NUTRITION OF THE PEACH TREE

III.* METABOLISM AND TRANSLOCATION OF L-[*GUANIDO-14C*]ARGININE HYDROCHLORIDE AND L-[U-14C]ASPARAGINE IN YOUNG DORMANT TREES

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Summary

Solutions of L-[guanido-¹⁴C]arginine hydrochloride and L-[U-¹⁴C]asparagine were applied to dormant two-year-old peach trees using a cut-shoot technique. Radioactivity was recovered in soluble sugar plus acid and protein amino acid fractions indicating that both compounds were metabolized by the trees. The results also suggest that there was a turnover of arginine in the dormant tissues. Most of the applied ¹⁴C from both compounds remained in the treated shoot or neighbouring tissues, but small amounts were translocated both upwards and downwards from the point of application.

I. INTRODUCTION

Deciduous fruit trees accumulate nitrogen in their woody tissues in proportion to supply during late summer, autumn, and winter, and the evidence suggests that this nitrogen is utilized for new growth during the next growing season irrespective of the external nitrogen supply (Murneek 1930; Oland 1959; Taylor and May 1967). In dormant peach trees stored nitrogen consists largely of soluble organic nitrogen; free arginine is the principal constituent of this fraction while the amides (asparagine + glutamine) are a minor constituent (Taylor and May 1967). Since these compounds may accumulate in dormant peach trees it might be expected that they would be metabolically inert in tree tissues during winter.

This hypothesis was tested by applying ¹⁴C-labelled arginine hydrochloride and ¹⁴C-labelled asparagine to dormant peach trees and assaying for radioactivity in compounds other than those fed to the trees. Translocation of the applied ¹⁴C through tree tissues was also studied.

II. METHODS

(a) Source and Storage of Isotopes

L-[guanido-¹⁴C]Arginine monohydrochloride (specific activity 13.6 mc/m-mole) and L-[U-¹⁴C]asparagine (specific activity 22.5 mc/m-mole) were obtained from the Radiochemical Centre, Amersham, England. 0.1 mc of each compound was taken up in 5.0 ml distilled water and deep frozen until required.

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amounts of 14 C (μ c) in tree tissues and amongst fractions containing free amino acids, soluble sugars and acids, and protein amino acids following application of $^{14}\mathrm{C}$ labelled arginine hydrochloride and asparagine to tree shoots

Values in parenthesis are percentages of ¹⁴C recovered in tree; those marked with asterisk are percentage recoveries of ¹⁴C taken up by tree

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banoamoD	IIntaka of 140	Treatment		Treated Shoot	l Shoot		Stock	Stock + Stem + Side Shoots	+ Side Si	noots		Roots	ots		Whole Tree
Applied and Tree No.	per Tree (nc)	Time (hr)	Free Amino Acids	Soluble Sugars and Acids	Protein Amino Acids	Total	Free Amino Acids	Soluble Sugars and Acids	Protein Amino Acids	Total	Free Amino Acids	Soluble Sugars and Acids	Protein Amino Acids	Total	Total
[¹⁴ C]Arginine HCl Tree 1	19.87	9.25	10.75	2.36	$1 \cdot 30$	14.41	2.10	0.30	0.44	2.84	0.05	0	0.24	0.29	17.54
			$(61 \cdot 29)$	$(13 \cdot 46)$	$(7 \cdot 41)$	$(82 \cdot 16)$	(11.97)	(17.1)	$(2 \cdot 51)$	$(16 \cdot 19)$	(0.28)	(0)	$(1 \cdot 37)$	$(1 \cdot 65)$	(88·27)*
Tree 2	19.68	7.50	12.56	3.49	1.83	17.88	0.18	0.03	0.02	0.23	0.03	0	0.06	60.0	$18 \cdot 20$
			$(69 \cdot 01)$	(19.18)	(10.05)	$(98 \cdot 24)$	(66.0)	$(0 \cdot 16)$	(0.11)	$(1 \cdot 26)$	$(0 \cdot 16)$	0	(0.33)	$(0 \cdot 49)$	$(92 \cdot 48)^*$
Tree 3	19.66	$24 \cdot 50$	$4 \cdot 19$	2.68	1.69	8.56	0.63	0.19	0.21	1.03	0.02	0	0.02	0.04	9.63
			$(43 \cdot 51)$	$(27 \cdot 83)$	(17.55)	(88.89)	(6.54)	$(1 \cdot 97)$	$(2 \cdot 18)$	(10.69)	$(0 \cdot 21)$	(0)	(0.21)	(0.42)	(48·98)*
Tree 4	19.90	$25 \cdot 25$	2.62	1.18	1.56	5.36	1.62	0.93	1.07	3.62	0.06	0	0.01	0.07	$9 \cdot 05$
			$(28 \cdot 95)$	$(13 \cdot 04)$	$(17 \cdot 24)$	$(59 \cdot 23)$	$(17 \cdot 90)$	(10.28)	$(11 \cdot 82)$	$(40 \cdot 00)$	$(99 \cdot 0)$	0)	$(0 \cdot 11)$	(22.0)	$(45 \cdot 48)*$
[¹⁴ C]Asparagine												19 ⁻¹ 66 A			
Tree 5	19.74	10.00		4.73	0.45	16.46	0.19	0.04	0.03	0.26	0.02	0	0.02	0.04	16.76
				(28.22)	$(2 \cdot 69)$	$(98 \cdot 21)$	$(1 \cdot 13)$	(0.24)	$(0 \cdot 18)$	$(1 \cdot 55)$	$(0 \cdot 12)$	0	$(0 \cdot 12)$	$(0 \cdot 24)$	$(84 \cdot 90)*$
Tree 6	19.83	9.25		3.41	0.35	$15 \cdot 69$	2.73	0.31	0.18	3.22	0.03	0	0.04	0.07	18.98
			$(62 \cdot 86)$	(17.97)	$(1 \cdot 84)$	(82.67)	$(14 \cdot 38)$	$(1 \cdot 63)$	$(96 \cdot 0)$	(16.96)	$(0 \cdot 16)$	(0)	$(0 \cdot 21)$	(0.37)	(12·36)
Tree 7	19.96	$25 \cdot 00$		3.79	$1 \cdot 00$	6.68	0.30	0.22	0.11	0.63	0.03	0	0.02	0.05	7.36
				$(51 \cdot 49)$	$(13 \cdot 59)$	$(90 \cdot 76)$	$(4 \cdot 08)$	$(2 \cdot 99)$	$(1 \cdot 49)$	(8.56)	(0.41)	0)	(0.27)	(0.68)	(36·87) *
Tree 8	17.38	$25 \cdot 00$	0.85	1.89	0.47		3.55		2.55	2.90	0.04	0	0.01	0.05	$11 \cdot 16$
			(2.62)	(16.93)	(4·21)	(28.76)	$(31 \cdot 81)$	$(16 \cdot 13)$	(22.85)	(20.79)	(0.36)	(0)	(60.0)	(0.45)	$(64 \cdot 21)*$

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(b) Distribution and Translocation of ^{14}C

Data relating to the distribution and translocation of 14 C in tree tissues approximately 10 and 24 hr after treatment are set out in Table 1 and Figure 1. Results of only one treatment are shown in Figure 1 since this is representative of the results obtained in all treatments. In all trees except Nos. 4 and 8 most of the applied 14 C remained in the treated shoot irrespective of the compound applied or the treatment time (Table 1). The treated shoots on trees 4 and 8 were short in comparison with those of the other trees and this is probably the reason why considerable amounts of 14 C were found in the stock plus stem plus side shoots of these trees; i.e. a spill-over mechanism operated. The applied liquid presumably moved through tree tissues until the negative tension of the xylem was balanced by the resistance to flow through the fine xylem spaces (Roach 1938).

Although most of the applied 14 C remained in the treated shoot or nearby tissues, the data in Figure 1 show that small amounts of 14 C were translocated both upward and downward from the point of application. There was no evidence to suggest that further movement of 14 C took place when the treatment time was extended from approximately 10 to 24 hr. This implies that if movement took place it was not on a sufficient scale to be detected by visual inspection of radioautographs, or that uptake and movement of 14 C through tree tissues were concomitant and interdependent.

IV. DISCUSSION

The finding that ¹⁴C-labelled arginine hydrochloride and asparagine were metabolized in dormant peach tissues was unexpected since it was thought that, in order for these compounds to accumulate in concentration and amount in tree tissues during autumn and winter, they would be metabolically inert at that time. It is possible that the method used to apply the isotopes to the trees may have influenced the results, e.g. enzymes may have been released at the cut surface, but further metabolism occurred after uptake was complete and therefore this possibility could not completely account for the observed results. Alternatively, exposing the trees to an elevated temperature during the experiment could have given rise to the observed results even though the trees remained visibly dormant. It is worth noting, however, that dormant trees would be exposed to such temperatures for short periods on warm, sunny days in winter. In addition it is known that dormant peach buds rapidly take up and metabolize [¹⁴C]thiourea when exposed to normal winter temperatures (Blommaert 1963). Therefore the odserved results may well be a true indication of the metabolic activity of dormant peach trees.

Although ¹⁴C-labelled arginine hydrochloride was metabolized in the dormant trees, chromatographic analysis of extracts showed that free arginine remained the major constituent of the free amino acid fraction over the experimental period. It is suggested that concurrent accumulation and degradation of arginine in dormant peach trees could only occur if there is a turnover of arginine in the tissues.

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VI. References

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