WEEKLY ABSORPTION OF NITRATE BY YOUNG, BEARING ORANGE TREES GROWING OUT OF DOORS IN SOLUTION CULTURES[1]

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(WITH THREE FIGURES)

Introduction

As part of a nitrogen nutritional experiment with young, bearing Valencia orange trees growing out-of-doors in complete solution cultures, a continuous record of the weekly absorption of nitrate and water, together with a thermographic record of air and solution temperatures, has been made for a period of approximately three and one-third years. Although these data do not indicate, save by inference, what periods of the year are most vital as regards the nutrient supply of nitrogen, they do show clearly the seasons of maximum and minimum absorption and afford information of interest as bearing on the relation of nitrate absorption to temperature conditions, periods of root and top growth, and transpiration. Data concerning total leaf number and leaf area and observations on their seasonal fluctuations and relations are included in this report.

To the knowledge of the authors, no studies of like nature have been recorded for citrus, although with reference to the seasonal requirements of these trees, Cameron (**1**), Cameron and Appleman (**3, 4**), and Cameron, Appleman, and Bialoglowski (**6**), from analyses of leaves, flowers, and young fruit from field trees, have indicated the importance of the nitrogen supply within the tree prior to and during the period of blossoming, spring growth, and fruit setting. These investigators and, more recently, Martin (**10**), and Hilgeman, Smith, and Draper (**7**), working with grapefruit in Arizona, found a marked reduction in the nitrogen of old leaves at the time of the spring blossom and growth cycle. Cameron *et al.* have also shown that the nitrogen of both top and root bark undergoes a reduction at this time and that substantial quantities of nitrogen are removed from the tree with the abscissed blossoms and small fruit. Unpublished data and observations accumulated by the authors in connection with nutritional and field experiments are in harmony with these findings, and it is widely held that ample nitrogen should be available to the citrus tree in the spring.

Experimental procedure

The detailed record of weekly nitrate and water absorption by the young

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citrus trees and of air and solution temperatures was begun September 27, 1937 and continued until January 13, 1941.

Two budded Valencia orange trees (nos. 25, 26) were employed for this study. These trees had been transplanted from the field nursery to solution cultures in May, 1936 (one year after budding) and had been grown in the solutions for a preliminary period of one year and five months.

The containers used in the preliminary period and during the first year in which records were kept were 100-liter vitrified tile sealed at the lower end and about three-quarters buried in the ground. Subsequently, the trees outgrew these tile containers and were transferred (November 14, 1938) to 600-liter concrete conduit provided with concrete bottoms. The concrete containers were painted inside and out with a water-emulsion type of

FIG. 1. Appearance of tree no. 25: A, in February, 1937, eight months before absorption record was begun; B, in January, 1941, at termination of experiment. Note the size of the foot scale at the right of each tree.

asphaltum. These, like the tile, were about three-quarters buried in the soil. With both the tile and concrete installations, concrete lids made in semicircular sections with semicircular holes at their adjoining centers, to accommodate the tree trunk, were used. These lids were provided with convenient holes to accommodate the aerator tube and to facilitate sampling and water additions (fig. 1).

The trees were grown in continuously aerated culture solutions of the following composition expressed in milliequivalents per liter: Calcium, 5.7; magnesium, 2.8; potassium, 1.0; sodium, 1.3; nitrate, 1.0; phosphate, 0.2; sulphate, 9.0; and chloride, 0.6. Manganese, boron, and aluminum in amounts to provide 0.5 p.p.m. of each, respectively, were added each time the solutions were renewed and small amounts of ferrous sulphate were added daily.

Analyses for nitrate by the phenoldisulphonic-acid method were made on samples of solution taken every Monday morning, the solutions having been made up to a fixed volume by the addition of distilled water and thoroughly mixed by increasing the aeration rate before taking the sample. After the analysis, the nitrate concentration was adjusted by the addition of a mixed solution of calcium, magnesium, and potassium nitrate, in an amount to compensate for the week's absorption of nitrate. A second set of samples was then taken for analysis, to check on the resultant concentration of nitrate. No regular analyses were made for calcium, magnesium, or potassium, but the pH of the solutions was tested every 2 to 3 days and adjustments made with KOH or H_2SO_4 as required. Phosphate was determined weekly and adjustments made with KH_2PO_4.

A record of the amounts of distilled water added during the week to compensate for transpiration and the small loss from evaporation was also kept.

New solutions were provided every month. The slow growth of the trees, the large volume of solution employed, the weekly additions of nitrate to compensate for absorption, and the frequent pH and phosphate adjustments, together with complete renewal of the solutions every month, sufficed to maintain the concentration of the major ions reasonably constant.

At the time measurements of nitrate absorption were begun, the concentration of this constituent was reduced from 2.0 to 1.0 milliequivalent of NO_3 per liter. (Previous studies (**6**) with citrus seedlings in solution culture have shown that a nitrate concentration in the amount of 0.1 milliequivalent per liter is ample, provided the concentration is maintained and the solution stirred continuously.) On July 18, 1938, while the trees were still in the tile containers, it became necessary to increase the nitrate to 1.5 milliequivalents per liter in order to avoid the absorption of all of this constituent during the weekly period. Subsequently, on November 14, 1938, when the trees were transplanted to larger containers, the nitrate was reduced to 0.2 milliequivalent of NO_3 per liter; then, during the summer of 1939, the amount was gradually raised to 0.3 milliequivalent. On May 6, 1940, owing to the increased growth of the trees, it was necessary to raise the nitrate to 0.6 milliequivalent per liter. These adjustments were all made by decreasing or increasing the mixed nitrate solution additions to the culture solutions.

In four of the weekly periods during this experiment, namely, in the weeks ending July 18, August 1, and August 22, 1938, and May 15, 1939, nearly all of the nitrate present at the beginning of the week was absorbed by the end of the week. The remainder of the time, the amount of nitrate remaining in solution at the end of the week was above the minimum requisite for good citrus growth. Save for the changes in concentration during the four weeks mentioned, when the nitrate was practically exhausted, and for the minor adjustment in nitrate concentration which became necessary as the trees grew, the nutrient conditions from week to week were essentially constant. Hence, the weekly quantities of nitrate absorbed may be regarded as primarily a reflection of the physiological activity of the tree.

At no time in the course of this experiment did these trees show detectable signs of malnutrition. The leaves were always deep green and the cycles of top growth coincided, in the main, with those of nearby field trees.

As stated, a continuous thermographic record of air and solution temperatures was kept, the recording instrument being housed in a suitably ventilated container adjacent to the cultures. The accuracy of the thermograph was checked at the outset, and maximum- and minimum-registering thermometers were installed in the thermograph house to check the accuracy of the air maxima and minima. The solution temperatures were also occasionally taken and compared with the thermograph reading. In general, agreement to within 1° F. was obtained throughout. The thermograph charts were changed at 9:00 A.M. on Monday mornings, and the mean weekly air and solution temperatures were calculated from planimeter measurements of the areas under the curves. The continuous stirring of the solution incident to aeration served to minimize inequalities of temperature within the solution.

The solution temperatures agreed closely with soil temperatures in nearby citrus orchards. The diurnal fluctuations of temperature in the solutions in the tile containers were of the same order of magnitude as those at the 12-inch soil depth. Those in the larger concrete containers were more like the fluctuations at the 24-inch soil depth. A series of comparative values are given in table I. Reference to this table shows that during some weeks the average temperature of the solution was somewhat lower and at other times higher than that of the soil at a depth of 24 inches. In view of the fact that there is a temperature gradient with depth in soils, it is evident that the temperature conditions under which these experimenal trees were grown were approximately similar to those which obtain in the field.

At periodic intervals during the three-and-one-third-year period, a count of all the leaves of each of the two trees was made. The leaves were graded into several size groups based on length, and counts of the number of leaves in each group were made each time. Having determined the area of leaves within each size group, it became possible to estimate total leaf area quite satisfactorily. Owing to new cycle growth on the one hand, and leaf senescence and abscission on the other, these counts are valid during the growing season for periods of only a few weeks before and after the leaf

TABLE I

COMPARISON OF MEAN WEEKLY TEMPERATURES OF SOLUTION CULTURE AND OF SOIL IN NEARBY CITRUS ORCHARD

DATE	MEAN WEEKLY TEMPERATURE	
WEEK ENDING	SOLUTION CULTURE	SOIL, AT DEPTH OF 24 INCHES
	°F.	°F.
1939		
Feb. 20	51.6	51.7
" 27	56.8	53.1
Mar. 6	55.0	50.1
" 13	57.0	56.4
June 5	74.0	69.5
" 12	77.0	71.2
" 19	77.0	72.2
July 3	81.5	73.5
" 31	84.0	78.1
Oct. 2	72.0	74.8
" 9	69.0	71.7
" 16	73.0	68.0
" 23	75.0	69.8
" 30	70.0	67.5
Dec. 4	59.0	61.2
" 11	62.0	61.3
" 18	60.0	61.3
" 25	58.5	60.0
1940		
Jan. 1	52.0	57.0
" 8	53.5	59.1
" 15	52.0	58.2
" 22	57.0	55.6
" 29	57.0	55.0
Average	64.52	63.32

count. During the first year (1938–1939), the fruit which set in April was picked early in June. In the two years of 1939–1940 and 1940–1941, however, all the fruit retained by the trees was allowed to ripen.

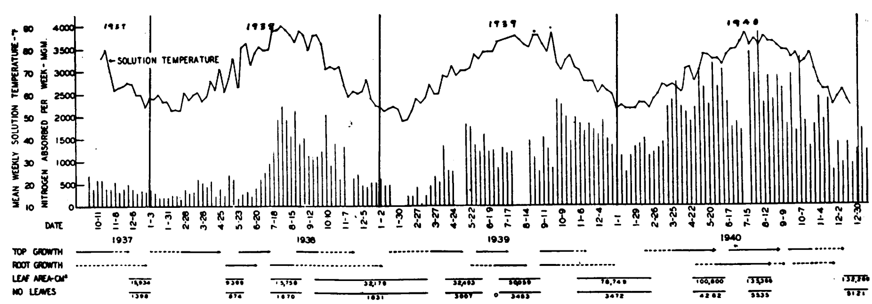

FIG. 2. The weekly absorption of nitrate nitrogen by a young Valencia orange tree (no. 25) growing out-of-doors in solution culture from September 27, 1937, to January 13, 1941.

From time to time it was necessary to spray the trees with an oil emulsion, with nicotine sulphate, and with tartar emetic to control red scale, aphids, and thrips, respectively.

The appearance of tree no. 25 in February, 1937 (eight months before the start of this experiment), and again in January, 1941, at the termination of the experiment, is shown in figure 1. The leaf count for this tree on the latter date was 5,121. This tree bore 36 pounds of fruit during the 1940–1941 season.

Results

In order that this paper may not be burdened with a large amount of tabular material, only the most pertinent data derived from this study are included.

The weekly absorption of nitrate by trees no. 25 and 26 from September 27, 1937 to January 13, 1941, together with the mean weekly temperatures of the culture solutions, are shown in figures 2 and 3 respectively. The mean air temperature, though averaging slightly lower than the mean solution temperatures, showed parallel weekly and seasonal changes. Since these data add nothing essential to the interpretation of the data given, they are omitted from the charts. The periods of top and root growth are shown by

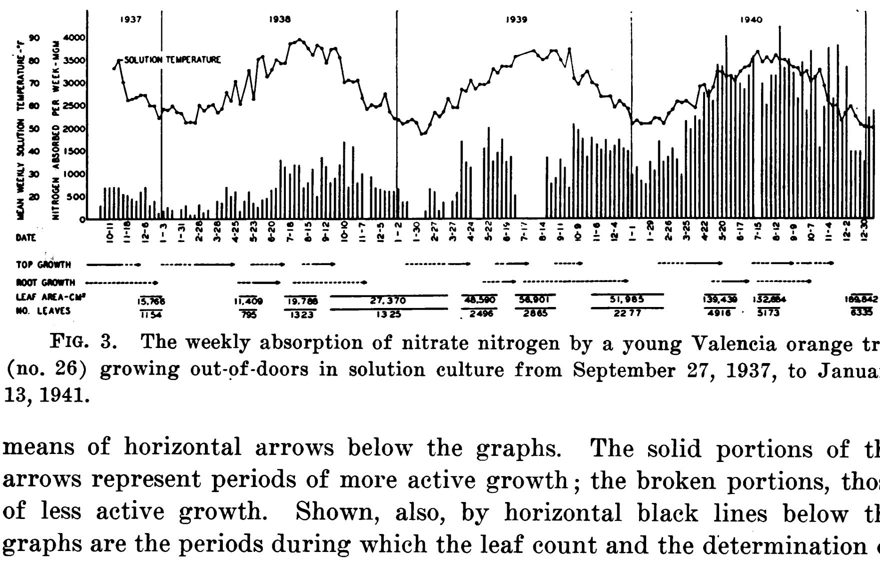

FIG. 3. The weekly absorption of nitrate nitrogen by a young Valencia orange tree (no. 26) growing out-of-doors in solution culture from September 27, 1937, to January 13, 1941.

means of horizontal arrows below the graphs. The solid portions of the arrows represent periods of more active growth; the broken portions, those of less active growth. Shown, also, by horizontal black lines below the graphs are the periods during which the leaf count and the determination of leaf area were reasonably accurate. The total leaf area, in square centimeters, and the leaf count for these periods are indicated immediately below the lines.

It is apparent that, under the climatic conditions prevailing in Riverside, California, nitrate absorption occurred throughout the year. As might be expected, however, it differed greatly in rate at various seasons. For the three years of this experiment, the period of least absorption was in January and February. This was also the period of minimum solution and air temperatures. The period of greatest absorption and also of maximum solution and air temperatures was in the summer and fall. Although a broad correlation was thus obvious between the quantity of nitrate absorbed and solution temperatures, there were weeks and periods when the correlation was not close. The lowest mean solution temperature for any week in which nitrate absorption data were available was that of 50.2° F. for the week ending January 13, 1941. Substantial absorption of nitrate took place during this week and other weeks when the temperature was almost as low.

It is of interest to note that substantial absorption of nitrate occurred in September, October, November, and even in December in every year. While there was usually some top and root growth during this period, it was ordinarily very much less than that occurring in the spring and summer. The favorable solution and air temperatures which prevailed throughout these months in the particular years during which this experiment was conducted, were no doubt the controlling factors in this absorption.

From January until May of each year there was no root growth. During this period the roots assumed the characteristic light-brown to brown color of mature, healthy feeder roots. While, as will be shown later in this paper, periods of more intense nitrate absorption were associated with periods of active root growth, it is clear that the older suberized and non-growing roots were capable of absorbing substantial quantities of nitrate.

Beginning in March of each year and coincident with the period of active bloom and spring-cycle growth, nitrate absorption increased, even though there was at that time no sign of root development. It was not until most of the blossoms had fallen and the new spring-cycle leaves had begun to harden that the first signs of root growth became manifest.

The tendency toward alternation of root and top growth, though striking at this time, became less marked as the growing season progressed and was least prominent in the fall. In the fall a certain amount of root and top growth proceeded simultaneously.

Root and top growth in relation to nitrate and water absorption

In an experiment of this type with no control over climatic factors, it is difficult to determine the differential influence of many conditions, both climatic and physiological, on nitrate absorption. An indication, however, of the relative importance of root versus top growth, as affecting nitrate and water absorption, respectively, was observed in the spring and summer of 1938.

After the spring-blossom growth cycle of 1938, the two trees of this experiment (nos. 25 and 26) got out of phase with one another in respect to root and top growth. Tree no. 25 began, on May 23, 1938, to send out a very profuse flush of new top growth accompanied by very little root growth; whereas new top growth on tree no. 26 was limited, but the root growth was abundant. The leaf count for these two trees on May 18, 1938, just before the onset of this differential behavior, was approximately the same, being 874 for tree no. 25, and 795 for tree no. 26. The amounts of water and nitrate absorbed during the several weeks preceding May 23 were also approximately the same for each tree. On May 30, however, as shown in the data of table II, differences in the relative absorption rate became

TABLE II

ABSORPTION OF NITRATE NITROGEN AND WATER IN RELATION TO ROOT AND TOP GROWTH OF TREES

Absorption period, 1938	Tree no. 25* (top growth profuse, root growth limited) †			Tree no. 26* (root growth profuse, top growth limited) †		
	Nitrogen absorbed	Water absorbed	Absorption ratio, nitrogen (mg.) / water (liter)	Nitrogen absorbed	Water absorbed	Absorption ratio, nitrogen (mg.) / water (liter)
	mg.	*liter*		*mg.*	*liter*	
May 9–16	600	6.47	92.7	600	6.80	88.2
May 16–23	198	.........‡	....	350	.........‡	....
May 23–30	280	8.92	31.4	250	10.95	22.8
May 30–June 6	320	7.52	42.5	420	9.14	45.9
June 6–13	200	.........‡	....	450	.........‡	....
June 13–20	400	10.64	37.6	630	7.91	79.6
June 20–27	600	14.80	40.5	680	9.97	68.2
June 27–July 4	750	15.24	49.2	1300	8.00	162.5
July 4–11	950	15.70	60.5	1150	10.00	115.0
May 23–July 11	3500	72.82	45.3	4880	55.97	79.1

* Leaf count May 18, 1938: tree no. 25, 874; tree no. 26, 795.
† Beginning week of May 23, 1938.
‡ Rain during this week.

manifest; tree no. 25 (with the heavy leaf growth) absorbed more water and less nitrate than tree no. 26, whereas the latter absorbed more nitrate and less water. These data provide a striking illustration of the differential influence of leaf growth versus root growth on water and nitrate absorption. In going over all of the water and nitrate absorption data for the two trees of this experiment, it became apparent that while in general large increases in water absorption were ordinarily accompanied by increases in nitrate intake there were, as illustrated in table II, many exceptions.

Another interesting, though by no means unexpected, observation was that in none of the weekly intervals during the course of this experiment were nitrate and water taken in by the plant in the same proportion as existed in solution. In all cases, nitrate was absorbed relatively more rapidly than the water. This is brought out by the data of table II, where the absorption ratio of milligrams nitrogen per liter of absorbed water varied in the experimental period covered from 22.8 to 162.5. During this period, nitrate nitrogen was adjusted at the beginning of each week to 14 p.p.m. It is evident that if nitrate and water were absorbed in the proportion existent in the solution, the absorption ratio should have been 14 or less (nitrate concentration gradually diminished during the week).

All of these observations merely constitute further evidence in a field which has been under investigation by HOAGLAND (**8**, **9**) and others over a long period: different, though not necessarily disassociated, mechanisms are concerned with the intake of water on the one hand, and nutrients on the other.

LEAF COUNT AND LEAF AREA

Of interest aside from their use as indices of top growth in relation to nitrate absorption, are the data with respect to leaf area and leaf count at different times of the year.

The increase in leaf number and leaf area was not smoothly progressive. For example, the leaf count in May, 1938, was less than that in December, 1937; and in December, 1940, the count was less than in August, 1940. These fluctuations are of course the result of seasonal leaf fall which may exceed in amount the concomitant or ensuing new-cycle growth. CAMERON (**2**) has shown that this seasonal fluctuation is marked in the case of mature, bearing citrus trees in the field.

Of interest, also, is the fact that there was no constant relation between leaf count and total leaf area. It is a common observation that certain cycles of growth may consist, predominantly, of small leaves and others of large leaves. When senescence and abscission of a cycle of small leaves coincide with a new cycle of large leaves, an increase in leaf area occurs without great change in total leaf number. Such a condition was noted in the fall of 1938. The leaf count of tree no. 25 in July was 1,670, and the leaf area was 15,758 sq. cm. (fig. 2); in November, after the fall-cycle growth, the leaf count of this tree was 1,831 (a relatively small increase) but the leaf area had doubled to 32,179 sq. cm. The converse took place in the spring of 1939 when, owing to the emergence of a large number of small leaves and the abscission of older and larger leaves, the leaf count increased from 1,831 to 3,007, whereas the leaf area increased only from 32,179 to 32,493 sq. cm. It is clear from these data that the relation of leaf number to leaf area is not constant and that any attempt to correlate physiological activity with leaf number must take account of the sizes as well as of the age of the leaves.

Discussion

With reference to the seasonal nitrogen requirements of citrus, the data of this paper are suggestive. General experience and the experimental work of others on citrus orchard trees (**1, 3, 4, 5, 7, 10**) indicate, as mentioned at the outset, the importance of nitrogen reserves within the tree prior to and during the spring growth and blossom cycle. In California, the first signs of spring growth on citrus occasionally appear in January and usually in February although the maximum development of flowers and leaves does not ordinarily take place until late March and early April. A heavy draft upon the reserve nitrogen and carbohydrate of the tree occurs during this period. Nitrate absorption, however, is at its minimum in the immediately preceding winter months. This suggests that the major accumulation of the nitrogen reserves necessary for vigorous spring growth, blossoming, and fruit setting may take place in the preceding fall and early winter when large amounts of nitrate are taken in by the tree, as shown by the data. It seems quite unlikely that the nitrate which begins to be absorbed in increasing amount at the time of the blossom peak is of consequence in-so-far as it affects the amount and character of the blossoms and leaves; but it doubtless becomes of great value to the young developing fruit.

Summary

A record of the weekly absorption of nitrate by two young, bearing Valencia orange trees growing out of doors in complete nutrient solutions for a continuous period of over three years has been made.

The period of least absorption in each of the three years was during January and February. The period of maximum absorption was that of late spring, summer, and early fall. These periods of varying absorption, as is to be expected, were related to solution and air temperatures. The week to week absorption varied widely at times, however, even when temperature conditions were essentially constant.

The relation between the quantities of nitrate and of water absorbed was quite variable from week to week. During one period when the two trees used in these experiments were out of phase with one another in respect to root and top growth, it became apparent that nitrate absorption was more closely related to periods of active root growth; on the other hand, water absorption was more closely related to cycles of active top growth or, more specifically, to the physiological activities associated with root and top growth. Under the conditions of low nitrate concentration prevailing throughout the experimental period, nitrate was absorbed relatively more rapidly than water, although the degree of selectivity varied quite widely

from time to time. While periods of rapid nitrate absorption were usually associated with periods of active root growth, substantial amounts of the constituent were taken in by roots which had become brownish colored and partially suberized.

From January to May in every year no new root growth occurred, the spring blossom and leaf cycle having completely emerged before the first signs of new root growth became manifest.

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