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# Absorption of Phosphorus by Foliage of Malus Robusta

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# THESIS

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ABSORPTION OF PHOSPHORUS BY
FOLIAGE OF MALUS ROBUSTA

by

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# BY FOLIAGE OF MALUS ROBUSTA

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## ABSTRACT

### ABSORPTION OF PHOSPHORUS BY FOLIAGE OF *MALUS ROBUSTA*

Five different phosphorus salts were applied to the foliage of *Malus robusta* trees growing in the greenhouse. The radioactive isotope of phosphorus ($P^{32}$) was used as a tracer in studying the absorption and translocation of the foliarly applied phosphorus.

Observations were made on the translocation of phosphorus by taking radioactive counts on fifteen-sixteenths inch leaf discs at weekly intervals. At the end of the experiment, samples from the old and new leaves and stems and from the roots were analyzed and counted to determine the total $P_2O_5$ in the sample. The percent $P_2O_5$ which came from foliar application was determined from the radioactive counts per minute obtained from the samples.

The results show conclusively that water soluble phosphorus applied as a foliar spray to foliage and small branches of apple trees can be absorbed and translocated to other parts of the trees. Under conditions of this experiment di-ammonium phosphate was absorbed and translocated in larger amounts than any of the other phosphorus salts used.

The phosphorus applied as a foliar spray was absorbed and translocated to the roots in more than double the quantity that was translocated to foliage in the unsprayed area on the same tree. Translocation of phosphorus was greater to young leaves in the unsprayed area than to older leaves in the same area.

# THE ABSORPTION OF PHOSPHORUS BY FOLIAGE OF *MALUS ROBUSTA*

## OBJECTIVES

The objectives of the research in this thesis are:

1. To determine if phosphorus salts can be absorbed by the foliage of apple trees when it is applied as a foliar spray.
2. To determine which of the five phosphorus salts used will be absorbed in the largest quantities by the foliage of apple trees.
3. To determine the rate of absorption of phosphorus when applied to the foliage and its distribution to various parts of the tree after absorption.

# INTRODUCTION

Many of the apple trees in New Hampshire are grown on soils of the Paxton series. It has been observed that some fruit trees failed to respond to phosphate fertilizers which were applied to soils low in available phosphorus.(16)(22) There have been no phosphorus deficiency symptoms reported on apple trees growing in Paxton soils, but soil analyses show that the amount of phosphorus available to the trees is very limited.(22) Other crops growing in the same soil series show phosphorus deficiency symptoms but have responded to phosphate fertilizer applications. Under these conditions the question arose as to whether the apple trees failed to respond to soil applications of phosphorus fertilizers because they did not need that element in larger quantities, or because they could not get it. The University of New Hampshire Agronomy Department (unpublished data) has shown that soils of the Paxton series have a fixing capacity equivalent to 10,000 pounds of 20 percent superphosphate per acre.

There is no record in the literature of any attempt to increase the phosphorus content of apple trees by foliar sprays. It has been found, however, that apple trees can absorb nitrogen, magnesium, zinc and iron through their foliage. The radioactive isotope of phosphorus ($P^{32}$) is a quantitative means of determining the rate of absorption of phosphorus fertilizers when applied either to the soil or to the foliage of plants. Radioactive phosphorus has been shown to react the same chemically as normal phosphorus except at very high concentrations.(6)(28)

## PREVIOUS WORK AND PRESENT OUTLOOK

Studies have been made of phosphorus deficiency symptoms by growing apple trees in nutrient cultures. Wallace (30) described the symptoms of phosphorus deficiency as being similar to a nitrogen deficiency, except that the leaves were darker green and more dull in color than with a nitrogen deficiency. In the more advanced stages the leaves deficient in phosphorus tended to exhibit a purple color with bronze tints over the entire surface. Later, brown spots appeared on the leaves and become dry. The fruit from such trees lacked firmness and had no desirable commercial qualities. Blake, Nightingale and Davidson (3) found that apple tree foliage became low in starch and proteins and high in sugars after growing in nutrient culture without phosphorus for four months. What little phosphorus was left in the plants tended to accumulate in the stem tips or the fruit and the cambium became relatively inactive. McMurtrey (18) in a study of phosphorus deficiency symptoms reported that distinctive symptoms do not always appear which will serve to accurately identify a shortage of the element.

Phosphorus plays an important role in plant metabolism. A deficiency of phosphorus will slow down normal cell division and elongation in the meristematic regions. It causes a reddening of leaves, petioles and branches due to an accumulation of carbohydrates which would normally be translocated to other areas of the plant and stored as starch. Phosphorus is found in nucleic acid, nuclein and nucleo-proteins, substances which are always present in the cell nucleus (10)(12)(19).

It plays a major role in the various enzyme processes of photosynthesis and respiration and is very important in the breakdown of starch (phospherolysis) to glucose 1 - phosphate. The most important function of phosphorus is in the metabolism associated with pyruvic acid in the manufacture of lipids, carbohydrates and proteins. Phosphorus also acts as an energy buffer in the release of energy during oxidation involving the enzyme adenosine. The energy is stored temporarily and released in the reduction of acetic acid to fatty acids (21).

In California a study was made of fruit trees and their response to phosphate fertilizers in comparison with field crops by Lilleland, Brown and Conrad (16). Eighteen different annual crops were planted between the rows of fruit trees. The field crops tested failed to make any growth unless phosphorus was applied. The trees, however, showed no response to phosphate fertilizer applications, although they were growing in soils deficient in the element. In Ohio, Gourley and Smock (11) made a survey of 27 orchards and found very little phosphorus available except in less acid soils but phosphorus did not directly benefit trees when it was applied to the soil. Potter and Fisher (22) in a study of apple orchards in New Hampshire found no increase in yield by supplying phosphorus in addition to nitrogen. They observed that a sod plot which had received 400 pounds of superphosphate per acre, each year, for 18 years had no more available phosphorus two inches beneath the surface than a plot which had received no phosphorus. The University of New Hampshire Agronomy Department has found (unpublished data) by the Heck Method (14) that Paxton soils have a fixing capacity of up to 10,000 pounds of twenty percent superphosphate per acre. In spite of these

conditions no phosphorus deficiency symptoms have been reported on apple trees growing in this soil series.

Heinicke (15) in a study of mature apple trees found, that on a tree producing twenty-five bushels of apples per year, approximately 0.12 pound of phosphorus was used by the fruit, 1.10 pounds by the leaves and 0.27 pound by the new tissues, each year. A study was made on the seasonal trend of phosphorus in apple trees by Butler, Smith and Curry (5) in New Hampshire, and a summary of the composition of apple leaves and other tissues at various times of the year was reported by Gardener, Bradford and Hooker (10).

In the soil, phosphorus is tied up in an insoluble form. Soils high in lime tie up the phosphorus as calcium and magnesium phosphate, forms which are not soluble in water but are available to trees. Soils which have a low pH tie the phosphorus up as iron and aluminum phosphates which are difficultly available to fruit trees (29)(33).

Thomas (29) concluded from experiments with apple trees that the omission of any one element (N. P. K.) from a complete fertilizer is followed by a decreased absorption of the remaining elements, due to a lack of balance of nutrients within the plant. He found that the ratio of nutrients for apple trees was approximately 6 parts nitrogen, 1 part phosphorus and 4 parts potassium.

Harley, Moon and Regeimbal (13) found that apple trees growing in distilled water exhibited growth which was directly related to that made the previous season in the nutrient cultures containing three levels of nitrogen, phosphorus, and potassium. This indicates the presence of a reserve supply of nutrients in the trees to withstand adverse conditions for the major part of one growing season.

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Bolaldes (15) in a study of mature apple trees found, that on a tree producing twenty-five bushels of apples per year, approximately 0.19 pound of phosphorus was used by the fruit, 1.50 pounds by the leaves and 0.27 pound by the new tissues, each year. A study was made on the seasonal trend of phosphorus in apple trees by Nation, Smith and Curry (5) in New Hampshire, and a summary of the composition of apple leaves and other tissues at various times of the year was reported by Gardner, Bradford and Hooker (10).

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There is no record in the literature of any attempt to increase the phosphorus content of apple trees by foliar sprays. There has, however, been a great deal of work done on the absorption of nitrogen, magnesium, zinc and iron by apple trees. In some places it is a common practice to spray trees with these elements.

Wolfenbarger (32) observed in experiments with phosphatic insecticides that increased yields resulted from application of these insecticides beyond results which could be explained by insect control. Silberstein and Wittwer (25) applied phosphorus to the foliage of vegetables by dipping the foliage into solutions containing phosphorus salts. They concluded that O-phosphoric acid was the most effective of all the compounds which they used as a source for plant utilization of foliarly applied phosphorus.

The radioactive isotope of phosphorus ($P^{32}$) offers an excellent tool for studying the absorption of elemental phosphorus ($P^{31}$) as well as for studying physiological processes within the plant (28). Radioactive phosphorus was found to react the same as normal phosphorus, up to concentrations as high as 20 microcuries of radioactive material per cubic centimeter of solution with an activity of $10^6$ disintegrations per cubic centimeter, per second (6).

Through the use of radioactive tracers it is possible to study not only the quantity of material present in any given tissue but also the rate of movement. Arnon, Stout and Sipos (1) observed radioactivity in leaves and stem tips of tomato plants six feet tall within 40 minutes after tagged phosphorus was added to the nutrient media. Biddulph (2) made a study of the migration of phosphorus within the plant during a

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24-hour period. He found the greatest downward migration was near 10 A. M. and the upward migration was at a maximum near noon but the amount was relatively small.

Rabideau, Whaley and Heimsch (23) made studies of absorption and distribution of radioactive phosphorus by tomato plants at various stages of development. Stout and Hoagland (27) studied the upward movement of radioactive salts in willow and geranium. By separating the bark from the woody tissue they observed that the nutrients were translocated upward in the xylem very rapidly, with lateral translocation to the phloem. The movement of nutrient solutions within the phloem was much slower than in the xylem. Colwell (6) found, that when a leaf was dipped in a solution containing radioactive phosphorus, the movement of phosphorus in the plant was predominently in the direction of "food" movement. Eggert, Yeager and others (8) found that radioactive phosphorus could be absorbed and translocated in excised red maple and apple trees comparatively fast.

# MATERIALS AND METHODS

This experiment was carried out in the greenhouse under controlled conditions, using one-year old Malus robusta No. 5 trees which were propagated asexually by mound layering. Thirty trees were selected for uniformity of size with tops approximately 36 inches high and roots as near uniform in size as it was possible to obtain. A small amount of top and root pruning was done at the time of planting to make the trees more uniform. They were planted on January 18, 1951, in No. 10 enameled tin cans. Each can had a single hole punched in the bottom of it for drainage. The soil used was Paxton sandy loam obtained from the University of New Hampshire Horticultural Experiment Station Farm near Durham, New Hampshire. The greenhouse in which the trees were grown had an average night temperature of 60° F. The trees were fertilized one week after planting with 2.5 grams of ammonium nitrate per cubic foot of soil.

On April 4, 1951, the trees were divided into six groups with five trees in each group. Spray applications of five different phosphate solutions were made on the leaves and branches, and each treatment was replicated five times. The five phosphorus salts used were: (a) Mono-ammonium phosphate, (b) Di-ammonium phosphate, (c) Di-sodium phosphate, (d) Tri-sodium phosphate, and (e) Mono-calcium phosphate. Treatment (f) was left unsprayed and used as a check.

The solutions used contained one gram of $P_2O_5$ per liter of distilled water with a specific radioactivity of approximately 0.00384 millicurie of $P^{32}$ per milliliter. Each group of five trees received a total of 125

milliliters of spray with approximately 0.48 of a millicurie of radioactivity. The upper half of the trees and the soil surface, which was to be left unsprayed, was covered with polyethylene bags to prevent contamination with the radioactive materials. The top half of the foliage on each tree was bagged, and the can and soil surface was also wrapped with a sheet of polyethylene. The collar of the cover on the can was sealed to the trunk of the tree with scotch tape to prevent the spray material from running down the trunk and into the soil: thus, avoiding contamination of the trunk, roots, soil, and can.

The trees were sprayed in a 50-gallon steel drum. The drum was placed on end and a small platform, capable of being rotated, was placed in the bottom of the drum. The trees were then placed in the steel drum one at a time and rotated slowly while the spray was being applied. The spray was applied by placing the nozzle of the sprayer through a 10-inch wide slit, cut the length of the drum. This permitted uniform coverage on all sides of the tree while giving maximum protection to the operator. The spray was applied with a 1-quart compressed air sprayer operated at 30 pounds pressure. The nozzle which was used gave a cone of very fine spray. The sprayer was modified from its original form by lengthening the intake pipe to the nozzle to insure removal of as much of the spray solution as possible from the supply tank. The spray nozzle and tank were carefully washed in distilled water after spraying with each solution to avoid mixing of materials. The operators used the proper protective clothing and monitoring devices. The person doing the spraying wore rubber gloves, boots, rain coat, mask, goggles and hat. Monitoring devices were used to check contamination with radioactive material in the area where the work was being done. The operators wore film badges while

they were working with the radioactive materials. The film badges were checked by the Tracerlab Company, Boston, Massachusetts, each week to be sure that the operators had not been exposed to an excessive amount of radiation.

Sampling Technique

After the spray on the leaves was thoroughly dried the bags were removed and the plants were placed on the greenhouse bench. Leaf samples were taken from the sprayed portions of the trees as soon as they had dried to determine the deposit of spray material on the leaves. Leaf samples were taken from the unsprayed area 24 hours after the spray was applied and at weekly intervals thereafter. Counts were made on fifteen-sixteenths inch leaf discs obtained by means of a cutting tool made from a 6-inch piece of steel pipe sharpened on one end to the desired diameter. The discs were taken from the center portion of the leaves on one side of the midrib. The discs were then placed in individual sample bottles and put into the refrigerator to prevent wilting until they were checked for radioactive counts.

Samples from the unsprayed area were taken from old leaves and from newly-developed full-sized leaves. Leaf samples were also taken from terminal growth in the sprayed area. These samples were taken from leaves which had developed since the spray was applied. This was accomplished by marking the growing tip with a ring of India ink shortly after the spray was applied. The ring was placed as near as possible to the growing tip. On May 3, 1951, the samples were taken from the last fully developed leaves on the terminal branches. They were taken at a point well beyond the India ink rings in every case to be sure they had not been contaminated by the spray.

The count recorded for each leaf disc was calculated by subtracting the average background for the day and calculating to the original activity of the material at the time of application. Any counts which were less than twice the background were discarded as not being high enough to indicate the presence of radioactive material. An analysis of the total phosphorus and a count on the radioactivity was taken at the completion of the project on the new leaves and stems, old leaves and stems, and on the roots. Ten-gram samples were taken from the unsprayed areas on the trees. One sample was obtained from the new leaves and twigs and another from the old leaves and twigs, on each tree. The root samples were obtained by carefully washing all the soil free from the roots and drying the entire root system except a few large roots which were on the tree at the time it was planted. Most of the root system was made up of numerous small feeder roots which had developed since planting. After the samples were dried, a three-gram composite sample was taken for analysis.

## Final Analysis

The samples were wet ashed in boiling nitric and perchloric acid and evaporated until approximately 10 milliliters of liquid remained. Hot water was added to the solution and it was filtered in a Buchner filter to remove the silica. The phosphorus was then precipitated, first as ammonium phosphomolybdate and then as magnesium ammonium phosphate as described by Winton (30). The final precipitate was filtered onto a fifteen-sixteenths inch filter paper with a stainless steel filter apparatus manufactured by the Tracerlab Company. The purpose of the special filtering procedure was to place the precipitate on the filter paper of

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Weighings were made of the samples of phosphorus by the following method: The planchet, with a clean filter paper disc in place, was brought to 50 percent relative humidity in a desiccator over sulphuric acid. The magnesium ammonium phosphate was then precipitated onto the filter paper and dried by pouring over it ten millileters of 95 percent ethyl alcohol followed by five millileters of ether. The sample, filter paper and planchet were then placed into the desiccator and returned to 50 percent relative humidity. They were then weighed on a chainomatic balance to an accuracy of .0001 of a gram and the results expressed as $P_2O_5$. The samples were then checked by placing them under the window of the tube on an automatic recording Geiger counter and counted for one minute. The counts thus obtained were divided by the counts per milligram of spray material per minute at the time it was applied to the foliage (288923 C. P. M.). This gave the milligrams of $P_2O_5$ in the analyzed sample which came from the foliarly applied phosphorus.

# PRESENTATION OF DATA

## SPRAY DEPOSIT

The first samples for counting were taken as soon as the spray on the foliage had dried. They were taken as a check of the spray deposit on the leaves. The amount of radioactive material present in the leaves (Table 1) was found to be quite variable, although every attempt was made to get as uniform coverage as possible. The counts per leaf disc ranged from 52 to 6,322 counts per minute. The amount of $P_2O_5$ on the leaves, calculated from the radioactive counts, was between .0146 and .0067 milligrams with an average of .0112 milligrams per fifteen-sixteenths inch leaf disc. The variation in coverage is a normal variation much the same as has been observed when the leaves of apple trees were sprayed to control insects and fungi.

The concentration of the spray solution was kept relatively low to avoid any possibility of injury to the foliage which might have affected the absorption of the phosphorus. There was, however, no observable injury to the foliage due to the phosphorus salts or the radioactive radiation at the concentrations used.

## LEAF DISC SAMPLES

Radioactive counts were taken at weekly intervals on leaf discs from the old and new leaves in the unsprayed areas. The phosphorus was absorbed by the sprayed foliage and translocated to the old and new leaves in the unsprayed areas in varying quantities. In Tables 2 and 3, each figure is an average of the counts obtained from five trees, with two samples

Table 1 Leaf samples from the sprayed area showing spray concentrations (a)

(Samples taken twenty-four hours after the spray was applied)

| Tree number | $NH_4H_2PO_4$ Foliar spray | $(NH_4)_2HPO_4$ Foliar spray | $Na_2HPO_4$ Foliar spray | $Na_3PO_4$ Foliar spray | $Ca(H_2PO_4)_2$ Foliar spray |
|---|---|---|---|---|---|
| 1 | 5443 | 4418 | 52 | 3877 | 2071 |
| 2 | 6082 | 5430 | 860 | 1743 | 2422 |
| 3 | 5311 | 3586 | 1055 | 4369 | 2453 |
| 4 | 3176 | 2426 | 1413 | 3673 | 4668 |
| 5 | 1120 | 1645 | 6322 | 3576 | 3939 |
| Average | 4226 | 3501 | 1940 | 3448 | 3111 |
| Mg. of $P_2O_5$ Per Leaf Disc | .0146 | .0121 | .0067 | .0119 | .0112 |

(a) All counts recorded as counts per minute and corrected to the counts per minute of the spray material at the time it was applied.

taken from each tree. The individual weekly counts were extremely variable with some samples showing relatively high counts and others showing none. In many of the samples which were considered to have no counts, they were not significantly higher than the average background for the day. These counts were assigned values of zero when they were averaged in order to be sure that no counts were included which should not have been. There was no relationship between the external appearance, or the area sprayed, and the amount of phosphorus absorbed and translocated to the unsprayed area. Neither was there any correlation between the amount of deposit from the spray and the amount of absorption and translocation.

As is clearly demonstrated in Tables 2 and 3, the phosphorus was translocated to the new leaves in larger quantities than to the old leaves. Each week the concentration of the radioactive phosphorus in each sample increased over the samples taken the previous week, on both the old leaves and the new leaves. The counts increased with each successive sampling even though the samples were obtained from positions progressively farther out on the limbs each week.

The course of absorption is shown in Figures 1 and 2. They demonstrate more clearly the increase in counts per leaf disc over each previous week's samples. The only exception to these was in the old leaves where the di-ammonium phosphate and the mono-calcium phosphate decreased slightly between April 19 and April 26 and on the new leaves where the di-sodium phosphate decreased slightly during the same sampling dates.

The concentration of radioactive phosphorus in the unsprayed area was significantly higher when di-ammonium phosphate was used compared to

**Table 2** Old leaves from the unsprayed area

| Treatment | 4/7/51 | 4/12/51 | 4/19/51 | 4/24/51 | 5/3/51 |
|---|---|---|---|---|---|
| $NH_4H_2PO_4$ | (a) | (e) | 170 (b) | 145 | 279 |
| $(NH_4)_2HPO_4$ | | | 193 | 277 | 450 |
| $Na_2HPO_4$ | | | 90 | 157 | 365 |
| $Na_3PO_4$ | | | 61 | 153 | 347 |
| $Ca(H_2PO_4)_2$ | | | 86 | 80 | 221 |

(a) Counts not significantly higher than background

(b) Each figure represents the average of 10 - 15/16 inch leaf discs counted for one minute each.

(c) No samples taken on 4/12/51 on old leaves.

**Table 3** New leaves from the unsprayed area

| Treatment | 4/7/51 | 4/12/51 | 4/19/51 | 4/24/51 | 5/3/51 |
|---|---|---|---|---|---|
| $NH_4H_2PO_4$ | (a) | 79 (b) | 211 | 330 | 563 |
| $(NH_4)_2HPO_4$ | | 139 | 438 | 838 | 1346 |
| $Na_2HPO_4$ | | 34 | 225 | 211 | 504 |
| $Na_3PO_4$ | | 18 | 117 | 183 | 536 |
| $Ca(H_2PO_4)_2$ | | 39 | 117 | 346 | 535 |

(a) Counts not significantly higher than background.

(b) Each figure represents the average of 10 - 15/16 inch leaf discs counted for one minute each.

Figure 1. Radioactive phosphorus (c.p.m.) in discs of old leaves from the unsprayed areas

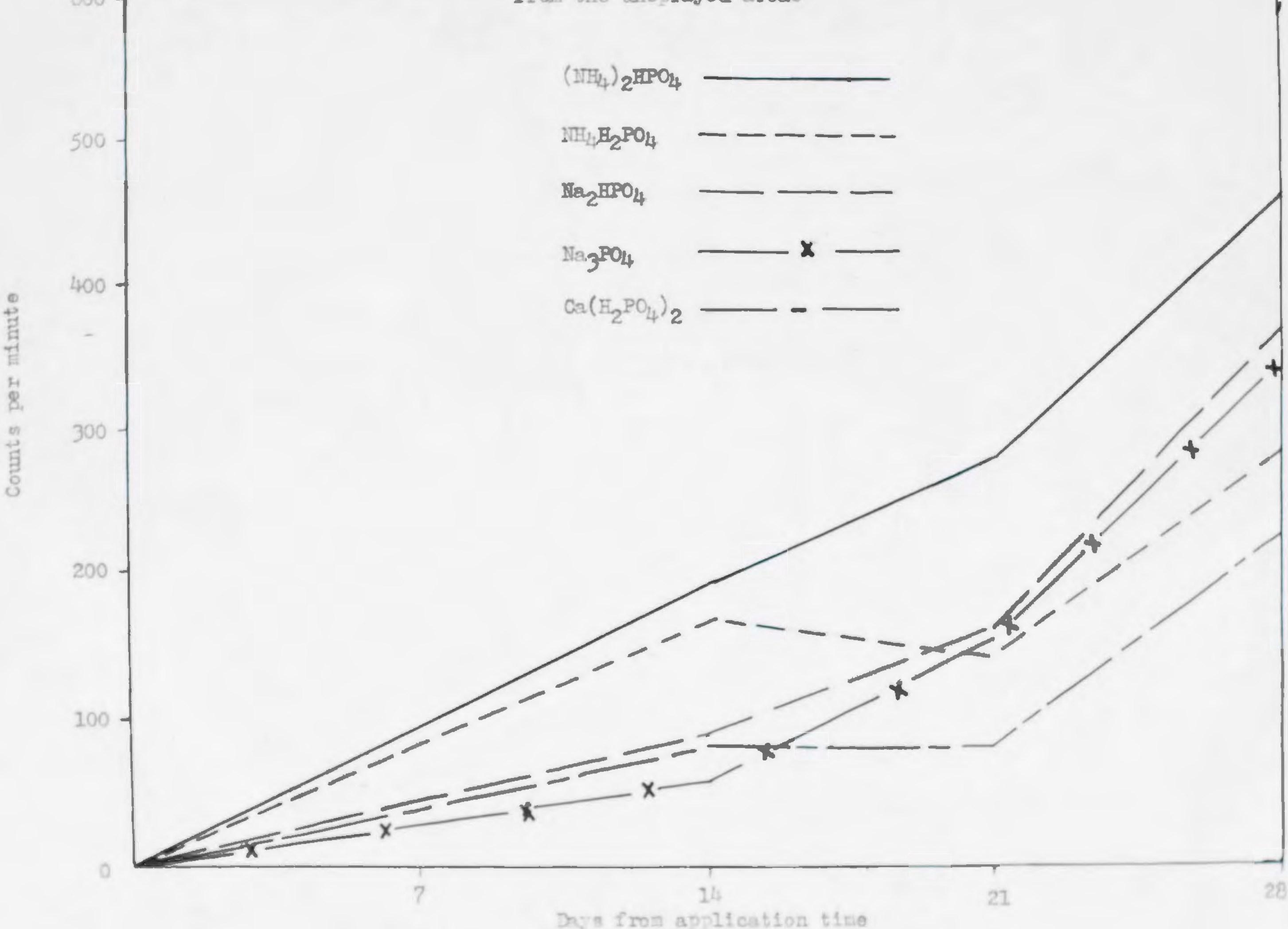
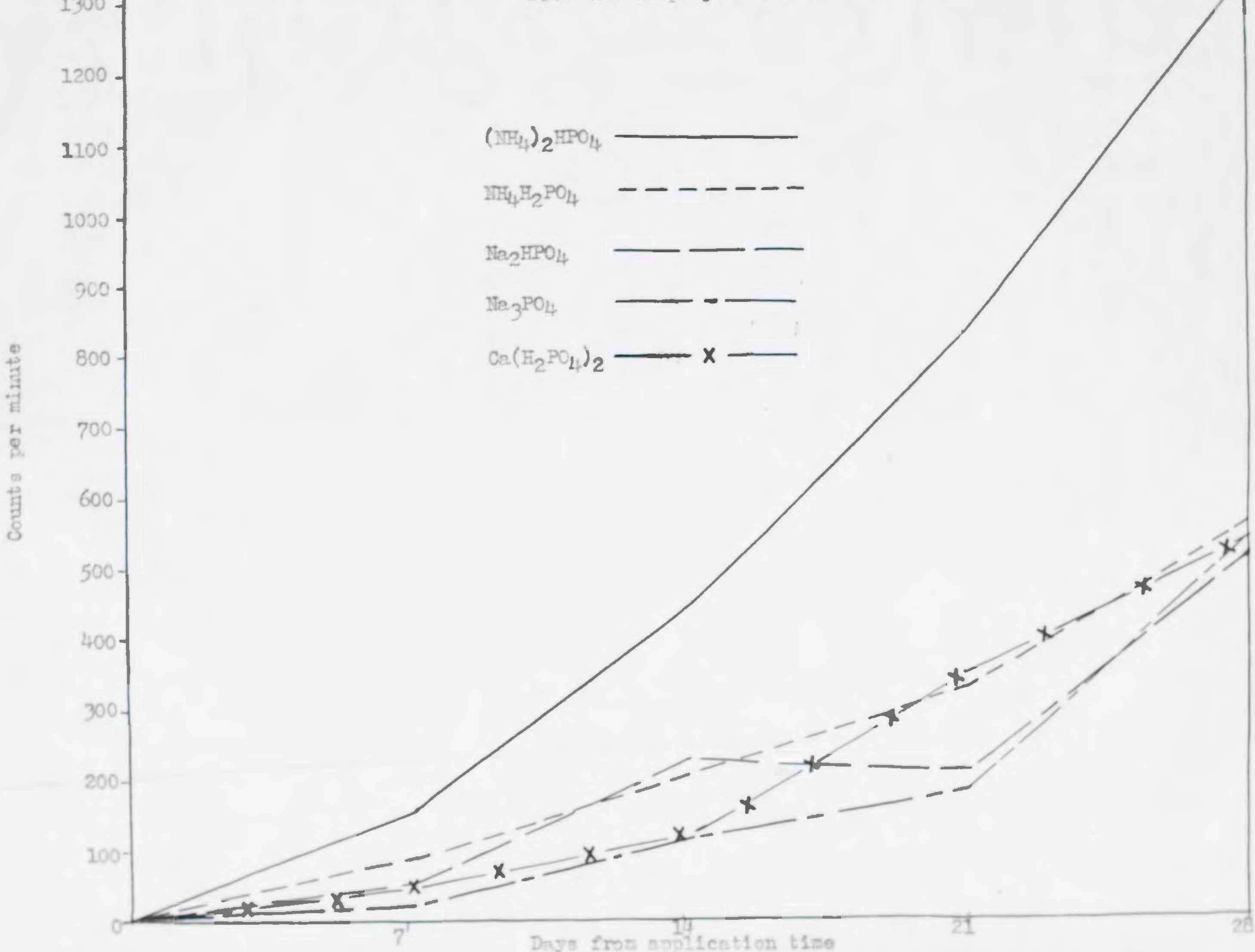


**Figure 2.** Radioactive phosphorus (c.p.m.) in discs of new leaves from the unsprayed areas



the other salts used. It was higher in both the old leaves and the new leaves with the difference being magnified in the new leaves. Twenty-eight days after the spray was applied there was an average of 1346 counts per minute for the di-ammonium phosphate spray compared with an average of 504 to 563 counts per minute for the other four salts used (Figure 2). In the old leaves the difference between the di-ammonium phosphate and the other salts was not quite so large but is significant under the conditions of this experiment (Figure 1).

TERMINALS FROM SPRAYED AREA

Leaf samples were taken from terminal growth in the sprayed area which had developed after the spray was applied (Table 4). The terminals on these limbs were marked with blue India ink bands shortly after the spray was applied. The samples were taken 28 days after the application of the spray (on May 3) and there was no possibility of any contamination with radioactive material. All the radioactive material present was absorbed through leaves in the sprayed area and translocated to the terminals which developed after the spray was applied. The activity was much higher in the newly developed leaves from the sprayed area than from samples in the unsprayed area. This was perhaps due to the fact that the material was translocated farther in the former than in the latter, and a given volume of phosphorus was spread over a larger area. The results shown in Table 4 clearly demonstrate that the phosphorus was absorbed and translocated to the terminal regions where high metabolic activity was taking place.

FINAL ANALYSIS

In the final analysis the percent of the $P_2O_5$ which came from the foliage application was found to be greatest in the roots (Table 5).

Table 4 Newly developed leaves from the sprayed area (a)

| Treatment | Average counts (b) |
|---|---|
| $NH_4H_2PO_4$ | 17,302 |
| $(NH_4)_2HPO_4$ | 17,933 |
| $Na_2HPO_4$ | 14,900 |
| $Na_3PO_4$ | 17,244 |
| $Ca(H_2PO_4)_2$ | 24,398 |
| Average | 18,356 |

(a) Samples taken May 3, 1951 - 28 days from the time the spray was applied

(b) Average of ten samples from five trees in each treatment

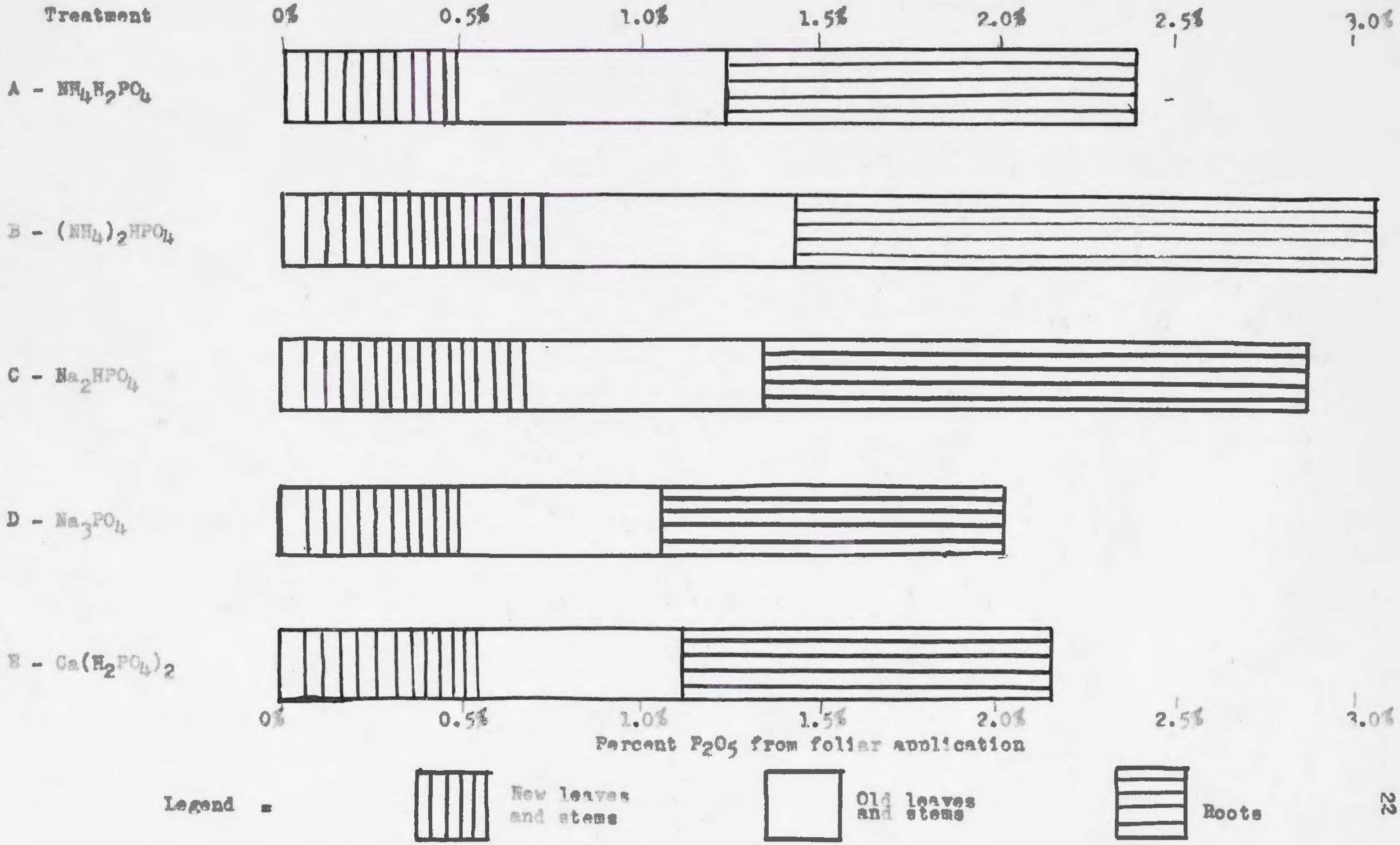
Although the total $P_2O_5$ in the roots was lowest of the three groups of tissue analyzed, the percent of $P_2O_5$ which was derived from the foliar spray was about twice as great. This would indicate perhaps that most of the phosphorus from the foliar spray was absorbed and translocated directly to the roots.

The proportion of the total $P_2O_5$ absorbed by the foliage was highest when di-ammonium phosphate was used as a spray. The difference was not as great in any given group of tissue, as in the leaf disc samples, but when the three groups of tissues are considered together, the di-ammonium phosphate showed a significant increase over any of the other treatments (Figure 3). A total of 3.081 percent of the $P_2O_5$ in the di-ammonium phosphate treatment came from the foliar spray. In the other treatments the percent of the total $P_2O_5$ which came from the foliar spray ranged between 2.005 and 2.860 percent.

Table 5. Analysis of samples from unsprayed section of the trees used in greenhouse studies, showing the percentage of $P_2O_5$ which came from the foliar spray

| Treatment (see fig. 3) | | C.p.m. per mgm. of $P_2O_5$ | Percent $P_2O_5$ in tissues | Percent of the $P_2O_5$ from foliar application |
|---|---|---|---|---|
| New leaves and stems | A | 1407 | .116 | .487 |
| | B | 2032 | .170 | .703 |
| | C | 1988 | .198 | .688 |
| | D | 1429 | .242 | .566 |
| | E | 1636 | .242 | .566 |
| | | av. | .225 | .585 |
| Old leaves and stems | A | 2131 | .085 | .738 |
| | B | 2091 | .135 | .725 |
| | C | 1893 | .197 | .655 |
| | D | 1425 | .256 | .493 |
| | E | 1651 | .209 | .571 |
| | | av. | .176 | .636 |
| Root samples | A | 3345 | .122 | 1.158 |
| | B | 4776 | .120 | 1.653 |
| | C | 4383 | .171 | 1.517 |
| | D | 2939 | .130 | 1.017 |
| | E | 2947 | .075 | 1.021 |
| | | av. | .123 | 1.273 |

Figure 3. Analysis of samples from unsprayed section of the trees, showing the percentage of $P_2O_5$ which came from the foliar spray



# DISCUSSION

There was considerable variation in the amount of spray solution which was retained by individual leaves when the spray was applied. This difference in concentration is an example of what might be expected, whether the spray is applied as a fertilizer or as an insecticide or fungicide. Some of the variation may have been due to a difference in the texture of leaves at different stages of maturity. The amount of solution which stayed on each leaf may have been due to the condition of the cuticle layer and the size of the leaf hairs present upon the leaf surface. There was an accumulation of spray material in droplets on the leaves. When the leaf samples were taken from this area, the counts were much higher than the average counts for the rest of the samples.

Under conditions of the experiment all phosphorus salts used in the foliar sprays were absorbed in varying amounts by the leaves and translocated to the unsprayed parts of the trees. The di-ammonium phosphate salt was absorbed by the leaves in the sprayed area and translocated into both the young and the old leaves in the unsprayed area in much larger quantities than any of the other salts used. The amount of material translocated to the old leaves was not as great as it was to the new leaves. Since phosphorus is used in cell differentiation, division and elongation to a large extent (19), it would be expected that there would be more phosphorus in the young leaves than in older leaves. Phosphorus is an essential constituent of the nucleus and cytoplasm of all

cells according to Blake (3) and, therefore, a limited amount would also be expected to remain in the mature leaves.

During the course of sampling several samples were obtained which were not sufficiently higher than background to consider that any radioactivity was present. Roach (24) working with dye injections found that when a dye was injected into the petiole of the leaf, it was translocated through definite channels to some other specific parts of the plant, the "injection pattern depending on the phyllotaxis and vascular anatomy of the stem". From these observations it might be concluded that unless any leaf which was sampled was connected with a translocation channel from a leaf which had been sprayed, no tagged phosphorus would be expected in the leaf. Such a situation is suggested as being responsible for certain leaf disc samples not showing any radioactivity.

The radioactive phosphorus which was found in the roots was more than double that found in the leaves and stems. The route of translocation is not known. The question did arise as to whether the large amount of phosphorus absorbed was translocated directly to the roots, then redistributed to the foliage, or whether it was translocated directly to the plant tissues where it was found.

It has been demonstrated that the movement within the phloem is mostly in a downward direction with only a limited amount of lateral translocation taking place. It is believed that phosphorus moves downward in the phloem in combination with carbohydrates which are synthesized in the leaves of plants (21). Phosphorus also has an important function in the plant in the storage and utilization of carbohydrates in plant metabolism. One of its more important functions in a plant is as an "energy buffer", when stored plant materials are converted to energy(9).

In the Xylem the movement is mostly upward with considerable lateral translocation. In beans it was found that the initial migration of phosphorus from the leaf is predominately downward, with only a small amount of upward migration of phosphorus at a time when transpiration was the greatest. The upward movement of phosphorus was due to the lateral translocation into the xylem from the phloem (2).

The counts increased on each successive sampling date and in every case the later samples were taken several inches farther out on the limb. There was a difference of 8 to 10 inches in the length of the terminals between the first and last date of sampling. From these observations it may be concluded that phosphorus was being translocated in increasing quantities toward the terminals on the tree. Blake (3), found that when phosphorus was low in apple trees, it tended to accumulate in the stem tips. This would indicate that it is needed most at the terminals and would, therefore, have a tendency to accumulate in this area where cell division and elongation is taking place.

The final analyses show that twice as much phosphorus was translocated to the roots as to any of the other areas outside of the sprayed area. Indications are that the phosphorus was transferred downward in the phloem. It was perhaps transferred downward in combination with the carbohydrates which were manufactured in the leaves and translocated to the roots. The large amount of small feeder roots with their rapidly developing and elongating root tips were perhaps responsible for the use of large amounts of the phosphorus which remained in the roots.

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The terminals from the sprayed area showed a very high concentration of phosphorus. This would indicate, perhaps, that the absorbed phosphorus which got into the conduction channels in the xylem was translocated directly to the stem tips on the same limb. Therefore, it had less area to be distributed in than the phosphorus which was translocated to other areas on the same tree and was not diluted as much.

In all the other samples the counts are relatively low: this can be accounted for by the small area of the tree sprayed and by the relatively low concentration of the spray applied. By the time the phosphorus was absorbed and translocated throughout the tree, with the high concentration found to be present in the roots, it was not surprising that the phosphorus was diluted to such a point that the counts from an individual leaf disc were relatively low. As was mentioned in the procedure the spray each group of five trees received was only 1/8 of a gram of $P_2O_5$ in 125 milliliters of water. The solution had only 0.48 millicurie of activity. The counts could not be very high under these conditions.

On the basis of the amount of phosphorus applied to the trees in the single spray used, recovery of the material was considered much higher than would be obtained from the soil applied phosphates.

To indicate the scope of this experiment a total of 570 samples were taken. Ninety of these were chemically analyzed for total phosphorus and 480 were counted by the leaf disc technique. The leaf disc technique made it possible to study the absorption and translocation of the phosphorus within the plant to a much greater extent than would have been possible with any other technique known. With this technique it was possible to take a large number of samples on small trees without affecting the growth of the trees.

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To indicate the scope of this experiment a total of 970 samples were taken. Ninety of these were chemically analyzed for total phosphorus and 880 were counted by the leaf disc technique. The leaf disc technique made it possible to study the absorption and translocation of the phosphorus within the plant to a much greater extent than would have been possible with any other technique known. With this technique it was possible to take a large number of samples on small trees without affecting the growth of the trees.

## SUMMARY & CONCLUSIONS

1. The results of this experiment show conclusively that water soluble phosphorus salts applied as sprays to foliage and small branches of apple trees can be absorbed and translocated to other parts of the trees.
2. Under conditions of this experiment di-ammonium phosphate was absorbed and translocated in larger amounts than any of the other phosphorus salts used.
3. Of the total phosphorus in the roots, the percent derived from the foliar spray was more than double that found in the leaves and stems from the unsprayed area.
4. Translocation of phosphorus was greater to young leaves in the unsprayed area than to older leaves in the same area.
5. Large amounts of phosphorus was found in leaves, which had developed on growth from the sprayed area, after the spray was applied.

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