

A QUANTITATIVE INVESTIGATION OF THE ABSORPTION OF  
CERTAIN CATIONS BY WHOLE PLANTS AND PLANT TISSUES.

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for the Degree of Master of Science

by

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## Summary

### Part 1

Greenhouse studies were conducted to investigate the absorption of sodium, potassium, calcium and magnesium by lemon seedlings. The plants were grown in controlled nutrient solutions and analysis of the plant material was made to determine the relationship between the four cations in the different plant organs. Results of the experiment may be summarized as follows:-

The occurrence of leaf burn appeared to be associated with a low calcium content, together with a high sodium, potassium or sodium + potassium content. The development of the seedlings was shown to be markedly influenced by the level of calcium supply and to a much lesser degree by the level of potassium supplied.

The concentration of sodium, potassium, calcium and magnesium in the stem and leaf varied with position of the tissues on the main axis.

The level of sodium was found to influence the distribution of sodium in the leaves, and the sodium content of the leaf, stem and root tissues gave a good reflection of the level of sodium supplied. The effect of the sodium treatment on the uptake of potassium appeared to be dependent on the calcium content of the tissues. Sodium treatment was found not to affect the leaf calcium content. In the stem and root tissues the calcium content was reduced when the calcium level in the nutrient medium was low, and increased the calcium content of these two tissues at the higher level of calcium supply. The sodium supply was shown to reduce the leaf magnesium only slightly, and its affect on the magnesium content of the stem and root tissues was shown to be dependent on the level of calcium supplied.

The potassium content of the leaves, stems and roots increased when the supply of potassium was raised. Increasing the potassium

level in the nutrient supply was found to cause a very slight reduction in the sodium content of the leaf, but did not affect the sodium content of the stem and root. Potassium did not affect the calcium content of the leaves and increased the stem calcium when sodium was not present in the nutrient media in high concentrations. In the roots potassium only increase the calcium content at the upper level of calcium supply. The magnesium content of the leaf and stem tissues was reduced as the potassium supply was increased. In the roots the general trend was for the magnesium content to increase when the level of potassium treatment was raised.

The calcium content of all the plant parts increased with a rise in the calcium supply, the stem and root tissues having a greater percentage increase than the leaves.

The sodium content of the plant tissue was reduced as the level of calcium supply was raised.

The leaf potassium content decreased with a higher calcium level of supply. In the stem and roots the potassium content was increased by doubling the calcium supply.

The calcium treatment had no influence on the magnesium content of the foliage, but in the stem and root tissues a decrease in magnesium content resulted from an increase in the calcium supply.

## Part II

Potato tuber tissue and carrot root tissue were used as experimental material in the investigation of sodium, potassium, and calcium uptake by storage tissue. It was shown that when the tissue disks are transferred from distilled water to a solution of salts there is a rapid initial uptake of cation which is neither particularly selective, nor related to metabolism, but dependent on the external concentration. On the other hand, the prolonged active accumulation of cation exhibits selectivity.

Potassium absorption by potato tuber tissue was shown to be stimulated by sodium, whereas the potassium and calcium absorption by carrot root tissue was shown to be reduced by sodium. Similarly the sodium absorption by the carrot root tissue was reduced by potassium, thus suggesting that the cations compete with one another for the same absorption mechanism.

## CHAPTER I

### INTRODUCTION.

It is well established that plant growth is dependent upon certain essential chemical elements. These elements are found to be present in the plant, together with elements non essential for growth, in varying concentrations, depending largely on the nature of the element, and the specific requirements of the plant. The elements may enter the plant from the external source by three independent pathways, namely; (1) Root uptake, (2) Plant Injection, (3) Leaf absorption. The first method is the more usual, and the one with which the present investigation is primarily concerned.

It has been recognised by numerous workers in this field, whose observations are reviewed in the following pages, that salt absorption by plant cells is not a simple process, which can be explained by osmosis. It has generally been assumed, from experimental evidence, that salts are taken up independently of one another and in varying amounts depending on the salt concentration, the particular plant in question, and environmental conditions. The passage of the nutrients into the cell is considered to be independent of the passage of water (Stiles, 1942), and generally the rate of ion uptake is the function of the concentration of the solutes outside and inside the cell. Ion absorption is further complicated by the selectivity of certain plants, some ions being absorbed in preference to others, regardless of their relative concentrations in the external supply. The entry of an ion into a plant cell may be retarded by the presence of another ion, a phenomenon to which the name "ion antagonism" was given by Stiles (1936). There are however instances where the entry of an ion is accelerated by the presence of another to which the term "syngery" has been suggested by Stiles.

The present investigation is an attempt to elucidate some of the problems associated with ion uptake by plants and storage tissues, and has been divided into parts, namely:-

#### Part 1.

A study of the effects of increasing the concentrations of sodium, potassium, and calcium in the nutrient supply, on the external

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appearance of lemon seedlings, and on the uptake and distribution of sodium, potassium, calcium and magnesium in the plants. A water culture technique was used for this experiment.

## Part 2.

A study of the effects of increasing the concentrations of sodium, and potassium, in the external media, on the absorption of sodium, potassium, and calcium by carrot tissues.

## A Review of the Literature

It was established during the first decade of the century that certain cations may have a mutual repressive effect on the absorption of one another. In 1906 Osterhout devised a number of methods for investigating this problem. In his earlier work, ion antagonism in plants was examined indirectly by observing the reduction of toxicity in mixed salt solutions, as compared with the toxicity of single salt solutions, containing the same substances in the same concentrations. Marine plant material was used for these preliminary experiments. Similar results were obtained in his later work when fresh water, and terrestrial species, including Spirogyra, Vaucheria, Lunularia gemmae, and Equisetum spores, as well as wheat seedlings were used.

A second approach to the problem was introduced in 1907 by Osterhout, who determined the rate of root growth, and used this as a measure of salt absorption. He found that root development of wheat seedlings proceeded more rapidly in solutions containing both salts of sodium and potassium, sodium and ammonium, sodium and calcium, and sodium and magnesium, than in single salt solutions of the same ionic concentration. Adopting the above method Hansteen (1910) using wheat, and McCool (1913) using peas, observed an antagonism between different ions. Hansteen found antagonisms between potassium and calcium, and to a lesser degree between potassium and magnesium, and potassium and sodium, while McCool found antagonisms to exist between calcium, and magnesium, potassium, sodium, ammonium, barium, and strontium, and between sodium and ammonium.

In 1907 Benecke produced direct evidence that the presence of one salt retards the entry of another into plant cells. Using Spirogyra

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he observed that the entrance of ferrous sulphate into the cells was much delayed by the addition of a calcium salt to the solution of ferrous sulphate. Szucs (1912) later confirmed the observations of Benecke and showed further that not only calcium salts but also those of other metals could bring about the same results. He concluded that the antagonistic action was dependent upon the valency of the ion.

More advanced methods have been used to demonstrate the phenomenon of differential ion absorption. Brenner (1920) used plasmolysis and deplasmolysis, while Osterhout (1912, 1914, 1915) measured the electrical conductivity of living tissue, both methods were effective for demonstrating ion antagonism. Plant analysis together with controlled nutrient supply has been used by the more recent workers in demonstrating the effects of different ions upon one another in ion uptake by plants.

Numerous research workers have shown that an antagonism exists between calcium, and sodium ions, in ion uptake by plants. Sodium was found to reduce calcium absorption by Thorne (1944) using tomato plants, Leonard, Anderson and Geiger (1948) using sweet potatoes, Gauch and Wadleigh (1945) using dwarf red kidney bean plants, garden beets and Rhodes and Dallis grasses. Similarly calcium was found to reduce the absorption of sodium by Haas (1957) using avacodo seedlings, Cooper, Paden and Phillipe (1953) using cotton plants, Smith and Reuther (1954) using various fruit crops and Cope, Bradfield and Peech (1953) using field crops.

The interactions between the two ions may not affect all parts of a plant in the same way, thus Lunt and Nelson (1950) using mature cotton plants found that sodium had no effect on the accumulation of calcium in the young cotton roots, but reduced the calcium content in the tops, seed and mature roots. Similarly Martin and Ervin (1957) showed that increasing the sodium supply depressed leaf calcium to a greater extent than root calcium in avacodo seedlings. The pH was also shown by Martin and Ervin to affect the degree to which the two ions interacted, sodium reducing the calcium absorption more in acid soils than in base saturated soils, or excess lime soils. Lundegardh (1944) found that cells in the interior of the root, or alternatively in organs distant from the roots,

displayed ionic relationships which deviated markedly from those in the nutrient solutions, due to the different migration speeds, as well as mutual interaction of ions.

Sodium and potassium ions have been found to be antagonistic to one another. A large number of workers using numerous plant species have found increases in the sodium external supply lead to a decrease in the plant potassium concentration; Reed and Haas (1923) and Haas and Brusca (1954) using citrus leaves, Hones, Pearson, Parker and Huberty (1952) using feeder roots of citrus, Thorne (1944) using tomato plants, Leonard, Anderson and Geiger (1948) using sweet potato plants, Bower and Wadleigh (1948) using dwarf kidney beans, garden beets, Rhodes and Dallis grasses, Cope, Bradfield and Peech (1953) using oats. However, Cooper, Paden and Phillippe (1953) using cotton plants, Larson and Pierre (1953) using table beets, flax, oats and corn, and Epstein and Hagen (1952) using excised barley roots found that living cells were able to discriminate between the absorption of potassium and sodium, accumulating potassium in preference to sodium even when the external environment was rich in sodium and poor in potassium.

Sodium has been found to interfere with magnesium uptake by the plant. Bower and Wadleigh (1948) using dwarf kidney beans, garden beet and Rhodes and Dallis grasses, Elgabaly (1955) using barley plants, Larson and Pierre (1953) using table beets, flax, oats and corn, Jones, Pearson, Parker and Huberty orange trees, and Jones, Martin and Bitters (1957) young lemon trees, all found that additions of sodium to the nutrient medium usually depressed the uptake of magnesium. Chapman and Brown (1943) found that the addition of sodium to trees deficient in potassium delayed the expression of visual symptoms of potassium deficiency, and suggest that this was due to the antagonistic action of sodium on calcium and magnesium.

Potassium and calcium also have been found to interact with one another, causing a nutritional unbalance in the plant (Lundegardh 1944). Antagonism between these two elements has been further demonstrated by Thorne (1944) using tomato plants, Sideris and Young (1945) using Ananas comosus, Lunt and Nelson (1950) using cotton plants, Leonard, Anderson and Geiger (1948) using sweet potatoes, Gauch and Wadleigh (1945)

using dwarf red kidney bean plants, Chapman and Brown (1943), and Haas (1949) using citrus. All these workers found that increasing the potassium concentration in the nutrient media adversely affected the absorption of calcium. Similarly calcium has been found to inhibit potassium absorption by several investigators including Leonard, Anderson and Geiger (1948) using sweet potatoes, and Fudge (1946) and Haas (1949) using grapefruit. Pierre and Bower (1943), and Richards (1944) found potassium to have a higher "competitive ability" than other cations, since divalent ions do not reduce potassium absorption to the same extent as potassium reduces that of the divalent ions. However Martin and Ervin (1957) using avocado, and Overstreet, Jacobson and Handley (1952) using barley roots found that potassium uptake may be increased by increasing the calcium supply, this effect being related to the potassium concentration in the external media.

Several independent investigators have shown that an interaction exists between potassium and magnesium. Increasing potassium in the external supply has been found to suppress the uptake of magnesium by citrus according to Chapman (1943), Chapman, Brown and Rayner (1947), Haas (1949), McColloch, Bingham and Aldrich (1957), and in the work of Leonard, Anderson and Geiger (1948) using sweet potatoes, Lunt and Nelson (1950) using cotton, Sideris and Young (1945) using Ananas comosus and Larson and Pierre (1953) using several plants. Correspondingly magnesium has been shown to have a slight depressing effect on potassium uptake by Fudge (1946) and Haas (1949) using citrus, and Smith and Reuther (1954) who used several fruit plants.

Increase in the calcium concentration has been shown to decrease the magnesium concentration in the plant by Reed and Haas (1923) and Smith and Reuther (1954) in citrus, and Leonard, Anderson and Geiger (1948) with sweet potatoes. However Smith and Reuther (1954) have found magnesium to have virtually no antagonistic action towards calcium.

The relationship between the ionic concentration in the external media and the concentration in the plant appears to vary with the plant species and also with the plant organ. The effects of increasing the sodium concentration in the external supply, upon sodium

absorption by plants was found to vary with the plant species, and also the position of the organ on the plant. Cooper, Paden and Phillipe (1953) using cotton plants, Cope, Bradfield and Peech (1953) using oats, and Elgabaly (1955) using barley, found that the sodium concentration in the plant rose markedly with increase in the sodium concentration in the external supply. Gauch and Wadleigh (1945) using dwarf red kidney bean plants however observed that the addition of sodium to the nutrient solution produced only a slight increase over the low sodium normally present in the leaves, but there was a moderate increase in the stem and a large increase in the roots. Haas (1952 a) (1952 b) and Haas and Brusca (1954) using a water culture technique for growing Lisbon Lemon seedlings, found that the sodium values were generally low and not uniform in the seedling leaves and bore no relation to the concentration of the sodium added. However the sodium content of the roots increased with the concentration of the sodium in the nutrient supply. Jones, Pearson, Parker and Huberty (1952) obtained similar results with orange trees. Richards (1944) considered that citrus was incapable of absorbing large quantities of sodium. Injury may result if large amounts of sodium accumulate in the plant, Haas (1950) and Martin and Bingham (1954) found large amounts of sodium accumulated in the leaves of avacodo seedlings, producing leaf burn on the margins of the leaves.

A relationship appears to exist between the potassium supply and the amount of potassium present in the foliage in most plants, as demonstrated by Sideris and Young (1945) using Ananas comosus, Jones, Martin and Bitters (1957) using citrus, Frear, Anthey, Haskins and Hewetson (1948) using peach trees. Haas (1949) found the potassium concentration decreased as the maturity of the leaves increased, and in the case of a deficiency of potassium the leaves produced visual symptoms of a disorder (Chapman and Brown 1947).

Citrus plants receiving very low levels of calcium supply were shown by Reed and Haas (1923) frequently to suffer from leaf abscission. Additions of calcium to the external supply produced increases in the concentration in the leaves, stem and roots; this increase was found not always to be proportional to the amount added.

Magnesium supplied to the external media was found by McCulloch (1957) to result in an increase in leaf magnesium. Smith and Reuther (1954) found severe magnesium deficiency to have a very deleterious effect on citrus trees.

In order to reduce the complicating effect on absorption of the transport of materials away from the absorbing regions, frequently attempts have been made to examine ion uptake using less complex systems than those which are presented by the intact angiosperm. Some investigators, (Hoagland, Davies and Hibbard 1928, Brooks 1937 and Jacques 1938) have used coenocytic algae such as Volvox, Halicystis and Nitella spp, where the problem of translocation is less involved. However difficulty has been encountered in culturing these algae in the laboratory. Many research workers (Lundegardh and Burstrom 1933, Hoagland and Broyer 1936 and Humphries 1950) have favoured the use of excised roots as experimental subjects, as they are easier to grow in culture under controlled conditions. Another important type of material which has been used in ion - absorption studies consists of tissue discs of various storage organs (Nathanson 1904, Stiles and Jorgensen 1915, Steward 1937, Robertson 1941 and Sutcliffe 1952). This method has the advantage over excised roots in offering greater uniformity of cells comprising the tissue. The work of Steward and his collaborators, using potato tuber tissue, have shown moreover that even this method is not without certain complicating features. A study of the absorption by disks of different thicknesses led to the conclusion that only the surface cells were involved in metabolic absorption. Using carrot disks Russel and Ayland (1955) demonstrated that potassium, rubidium and caesium compete with one another for ion uptake. Sutcliffe (1954) using washed beet disks, found that both sodium and potassium were absorbed at comparable rates from equimolar solutions of single salts. When the two ions are supplied together in the same medium unequal uptake may occur. The uptake of sodium was found to occur more rapidly than that of potassium at 15°C. and 25°C. but at 2°C. approximately equal absorption of the two ions were observed. Sutcliffe (1957) continuing his investigations on the selective uptake of alkali cations by red beet root tissue, found that

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when the disks were transferred from distilled water to a solution of salts, there is a rapid initial uptake of cations, which is neither particularly selective, nor directly related to metabolism. On the other hand, the prolonged active accumulation of cations exhibits strong selectivity, sodium being preferred to other ions.

It is widely recognised that the uptake of inorganic solutes by plants involves a complex of interrelated processes, any one of which may, in suitable circumstances, limit the overall absorptive capacity of the organism or tissue. Amongst these controlling factors are the rate of utilization and transportation of ions from one part of the plant to another. Both are intimately related to growth, and the influence which growth exerts on the course of mineral salt absorption may be attributed, at least in part, to these associated processes.

The present investigation was therefore planned with a view to studying some of the problems associated with cation absorption by whole plants and excised storage tissue.

PART I

## CHAPTER II

### Part I

#### Experimental Methods

##### Introduction

The value of the water culture technique for the study of problems in plant nutrition is generally recognised. This method was selected for the growth of the plants in the present experiment, as a stricter control in the external supply of nutrients to the plants could be maintained, than in most other methods used for plant culture work.

It is well established that the more exactly the chemical constitution of the culture solutions is controlled the more effective the method may be utilised. Therefore it is absolutely essential to use purified nutrients wherever permitting, and every effort made to ensure that the containers and any other vessels used are clean and chemically inactive.

##### Water Purification

All the water used throughout this work was tap water distilled in an electric aluminium still, and then repurified by the following two methods :-

- (1) Demineralisation
- (2) Glass Distillation

Both the above methods gave pure water of the quality required for the present investigation, i.e. containing no traces of sodium, potassium, calcium or magnesium.

##### (1) Demineralisation

The distilled water obtained from the aluminium still was siphoned from an aspirator (A), see Fig.I, the rate of flow of water being controlled by a screwclip (B). The water was passed through the ion-exchange column (C) containing the resin Amberlite M.B.3. The resin was packed into the column in three layers (D) separated one from another by "Pyrex" glass wool (E), degeneration of the resin was indicated by a

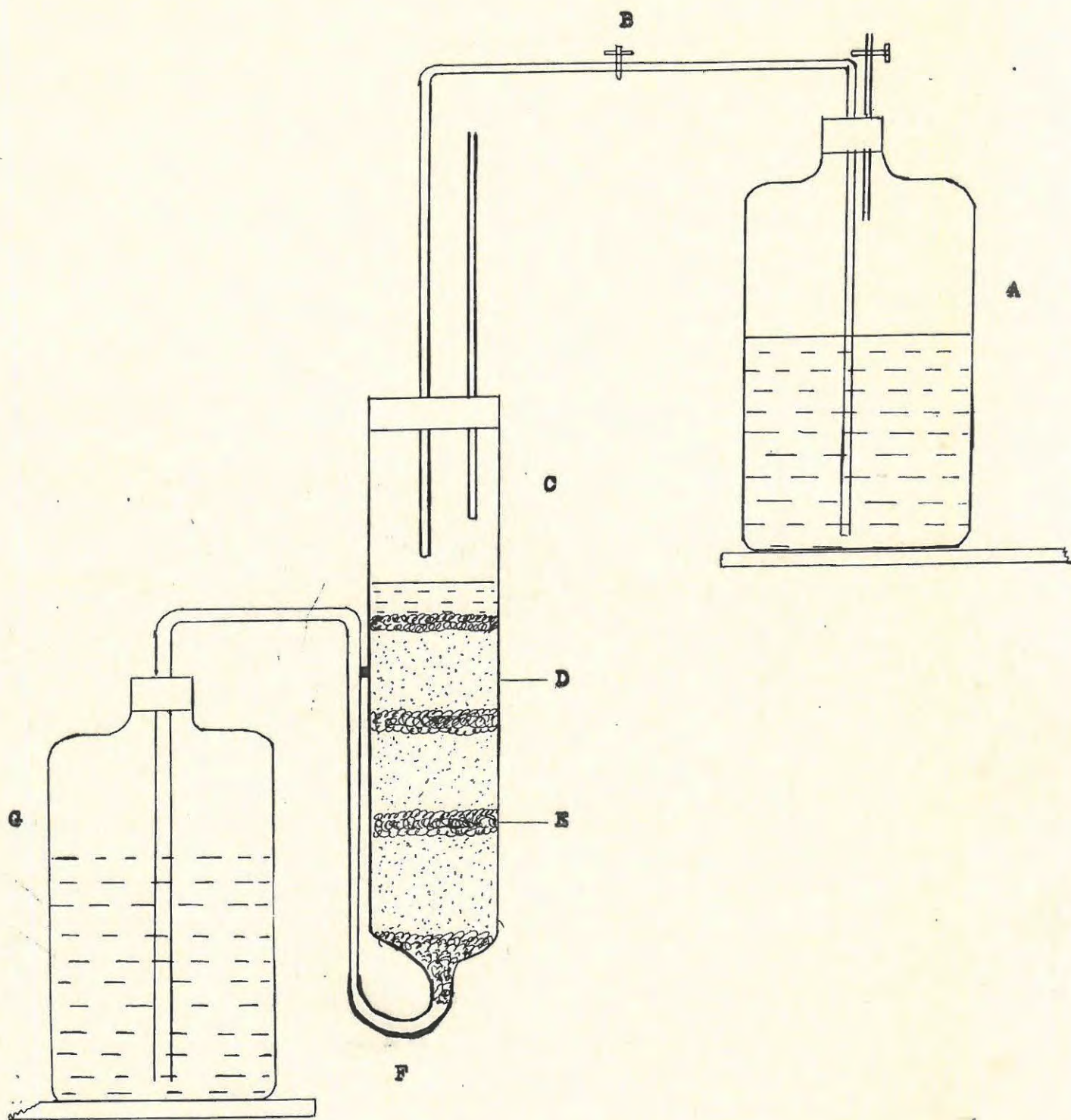


Fig.1 Apparatus used for Demineralisation of Water.

colour change in the resin from blue to green. The resin was renewed whenever the indicator in the second layer showed any signs of colour change.

A capillary tube (F), was attached to the base of the column through which the demineralised water flowed into a "Pyrex" glass aspirator (G). The capillary tube was arranged so as never to allow the level of the water in the column to fall below the uppermost layer of resin. Approximately 20 litres of demineralised water was produced per day using this method.

## (2) Glass Distillation

The water obtained from the aluminium still was redistilled in a constant level "Pyrex" glass still, as described by Wilson and Strickler (1939), and the redistilled water was collected in a "Pyrex" glass aspirator. This method produced approximately eight litres of pure water daily.

## Glassware

The glassware used throughout the present work was of "Pyrex" grade. All the nutrient stock solutions, and standard solutions used in calibrating the instruments, were stored in either polythene containers or "Pyrex" two litre flat bottomed flasks, as a precaution against sodium contamination from ordinary glassware.

## Purification of Glassware

All the containers used in the plant culture work were cleaned using the method described by Hewitt and Jones (1947). The containers were rinsed in 1% HCl water solution then thoroughly washed out with purified water, and finally autoclaved for thirty minutes at 20 lbs/sq.in.

## Chemical Reagents

The chemical reagents used throughout this work were British Drug Houses "Analar" grade reagents, unless otherwise stated.

### Containers

Wide mouth 1 litre conical flasks were used for the growing of the plants in the experimental solutions. The plants were supported in the vessels, with the aid of corks, which as well as supporting the plants prevented contamination of the culture solutions by dust. All the corks were impregnated with paraffin wax. This prevented impurities in the cork from being carried into the nutrient solution by condensing vapour. Each cork had a hole one inch in diameter in its centre for supporting the lemon seedlings, and two smaller holes towards the periphery, one for the aeration of glass tubing, and the other for a small right angled capillary glass tubing open at both ends, for maintaining constant pressure during aeration.

The lemon seedlings were supported by loosely wrapping non-absorbent cotton wool around the hypocotyl region of the main axis, thus forming a plug in the cork hole. The roots of the seedlings were allowed to dip into the nutrient solution. Light was excluded from the culture solution and roots by covering the glass flasks with black cartridge paper wrappers.

### Aeration

It has been shown by Haas (1932) that aeration is essential for the growth of citrus in water culture. The plants having an oxygen requirement of between 6 - 8 p.p.m. (Hewitt 1952). Aeration of the nutrient solutions was achieved by admitting compressed air to the nutrient solutions through the glass tubing in the corks. The tip of each glass tube was drawn out to form a capillary tube, in order to give a constant flow of small bubbles, which agitated the solution at the same time as aerating them. The flow of air was regulated by a screw clip on the rubber tubing connecting the main compressed air pipe to the glass tubing entering the nutrient solution.

### Lighting

All the side windows of the greenhouse were fitted with Venetian blinds and the glass roofing was coated with whitewash, in an effort to protect the seedlings from receiving direct sunlight. It was found that

direct sunlight was too intense, and also brought about high temperatures, inducing wilting of the plants.

Ordinary commercial blue fluorescent lighting was fitted over the growing plants, and was used to increase the light intensity on overcast days.

#### Temperature

The greenhouse was heated by means of electric coil heaters fitted into pipes beneath the benches, supporting the growing plants. Every attempt was made to maintain the temperature at 70°F, but during the winter months the heating supply was inadequate to maintain this temperature, and the temperature frequently dropped during the night to 50° F. There was no means of controlling the high temperatures during the summer months, and temperatures of 98°F were recorded during that part of the growing season. Scorching of the leaves occurred at these high temperatures, this matter will be dealt with in detail under "Visual Appearance".

#### Humidity

A humid atmosphere was maintained in the greenhouse by keeping the cement floors constantly wet. In addition asbestos troughs containing water were placed beneath all the benches in the greenhouse.

#### Plant Material

Rough lemon seedlings were used as plant material in the present investigation. The fruit containing the seed was picked on the 3rd February 1956, from the farm "Baddaford" in the Kat River Valley, Fort Beaufort District. The fruits were placed in the laboratory for six weeks, during which period they were allowed to dry out, prior to the collecting of the seed. As a precaution against fungal attack during this period, the fruit were swabbed once weekly with a mercuric chloride solution.

#### Germination

On the 15th March, six weeks after the harvesting of the fruit, the lemons were cut open, and the seeds extracted. In order to hasten germination the testa of the seeds were nipped with the aid of a scalpel.

Open topped circular dishes eight inches high, and varying diameters from eight to fourteen inches, of ordinary glass, were used for seed germination. The centre of each dish was packed with coarse filter paper (Postlip Mills No 633), and the dishes were lined with No. 1 Whatman filter paper. The seeds were sown between the outer layer of filter paper and the sides of the dishes. The filter paper was kept continually moistened with a modified Knop's four salt solution (Knop 1861), prepared as follows:

$\text{KH}_2\text{PO}_4$ .....	0.17 g/L
$\text{Ca}(\text{NO}_3)_2$ .....	0.94 g/L
$\text{KNO}_3$ .....	0.60 g/L
$\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ .....	0.49 g/L

There was always slight excess of solution present at the bottom of the germinating dishes, ensuring that the filter paper was continually saturated.

The glass dishes containing the seeds were covered with glass lids, and placed in the greenhouse, where the temperature was thermostatically maintained at approximately 70°F, in a humid atmosphere. It was found necessary to transfer the germinating seeds into dishes containing new filter paper and fresh nutrient solution, on the 9th April, because of fungal contamination.

By the 24th April, five and a half weeks after sowing the seeds, a large proportion of the seedlings had an overall length of 8 cms. At this stage the seedlings were removed from the germinating dishes, and placed singly into 1 litre culture flasks, containing nutrient solution similar to that used for germination. This solution had a pH value of 5.9.

The level of the nutrient solution in the flasks was maintained by daily making up to the mark with water, compensating for the loss due to transpiration and evaporation. The nutrient solutions were aerated for one hour per day during this period.

On the 9th May the nutrient solutions were renewed. The seedlings received their first dose of micro-nutrients on the 1st June, the iron solution was administered separately from the remainder of the

micro-nutrients. The micro-nutrient solutions were made up according to Arnon (1938), with slight modifications, as follows:-

Iron solution : 0.5 g.  $\text{FeSO}_4$  made to 100 ml with 0.4% Tartaric Acid solution.

Micro-nutrient Solutions

$\text{H}_3\text{BO}_3$ .....	2,8347 g
$\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$ .....	1,8008 g
$\text{ZnCl}_2 \cdot 7\text{H}_2\text{O}$ .....	0.1263 g
$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ .....	0.0738 g

made to 1 litre with water.

1 ml of the iron solution, and 1 ml of the micro-nutrient solution were added to each nutrient solution. Thereafter the iron solution was added to the nutrient solution in 1 ml doses every fourth day, whereas 1 ml of the micro-nutrient solution was added weekly when the complete nutrient solution was changed.

On the 3rd October it was observed that an interveinal "spotting" had developed on the lower leaves on some of the seedlings, see Fig.2. Closer inspection showed these "spots" to be areas in which epidermal collapse had occurred. The cause of this anatomical disorder is unknown although, it is thought to be of a physiological nature. This disorder did not re-occur, and had no detrimental effect on the future growth and health of the seedlings. Plants affected were noted, and mention will be made of these plants used in the experiment.

On the 23rd October (thirty two weeks after germination), when the seedlings had reached approximately 30 cms. in height, from the hypocotyl region to the stem apex, it was decided to place the plants in their respective experimental solutions.

Selection of Experimental Material

It was found that the experimental seedlings could be grouped into two categories with respect to height, the smaller seedlings having an average height of 29.4 cms, the larger seedlings with an average height of 31.5 cms. For the purpose of the experiment the seedlings were grouped in two, (1) small seedlings, (2) large seedlings. There was insufficient material of either group to supply the needs for two



Fig. 2. Showing "Epidermal Collapse" of the leaves

replications of all the treatments, therefore the large seedlings were used for one set of treatments in Block A, and the smaller seedlings for the replicate in Block B.

All the treatments were randomized and the position of each treatment in the greenhouse is illustrated in Fig. 3.

#### Experimental Nutrient Solutions

The experiment was designed to study the effect of varying the supply of sodium, potassium, and calcium, on the concentration of sodium, potassium, calcium and magnesium present in the root, stem, and leaf tissues of lemon seedlings.

Four levels of sodium 0.00 p.p.m., 50.0 p.p.m., 195.00 p.p.m. and 505.0 p.p.m., four levels of potassium, 0.00 p.p.m., 117.00 p.p.m., 253.00 p.p.m., and 507.00 p.p.m. and two levels of calcium 180.00 p.p.m., and 360.00 p.p.m., were supplied in the nutrient solutions.

In the design of this experiment it was felt that the concentrations of each of the anions and the cations in the nutrient solutions should be kept constant, except for the ion being varied in each series, even though it was necessary to vary the total ionic concentration. The above was achieved by supplying the nitrogen in both the nitrate, and the ammonia state. It has been shown by Smith (1951) that either the nitrate or ammonia form of nitrogen is equally suitable for the growth of citrus seedlings. In the present experiment, however, in no nutrient solution was the nitrogen supplied solely in the one form or the other, both forms were always present.

Sodium was added either as the chloride or the nitrate salts, or in both forms, while the potassium and calcium was supplied as the nitrate salts. The chloride level was kept constant at 117.00 p.p.m., the nutrient solutions receiving no sodium were supplied with chloride as the ammonium salt. The magnesium concentration was kept constant at 97.00 p.p.m., and the sulphur concentration at 128.00 p.p.m., by adding magnesium sulphate. The phosphate concentration was kept at 153.00 p.p.m., and was added as phosphoric acid, and the nitrogen concentration was kept at 1202.00 p.p.m. by adding the nitrates of calcium, potassium, and ammonium, and in some cases part was added as



BLOCK A

T7. T4. T29. T26. T1. T8. T6. T19. T10. T28. T32. T21. T3. T12. T24. T2.

T11. T18. T23. T16. T21. T25. T14. T20. T27. T13. T15. T17. T30. T5. T31. T22.

door end

window end

T23. T16. T27. T17. T2. T19. T26. T1. T5. T24. T15. T11. T13. T14. T30. T28.

T31. T29. T7. T8. T20. T6. T25. T22. T21. T10. T3. T9. T4. T18. T36. T12.

BLOCK B

Fig. 3. Diagram of position of treatments in greenhouse.

TABLE I

pH Values of Nutrient Solutions

Treatment	23/10/56		30/10/56		30/10/56		6/11/56		6/11/56		13/11/56		13/11/56		20/11/56		20/11/56		27/11/56		27/11/56		4/12/56		4/12/56		11/12/56		11/12/56		18/12/56		18/12/56		1/1/57		1/1/57		15/1/57		15/1/57		22/1/57	
	New	Old	New	Old	New	Old	New	Old	New	Old	New	Old	New	Old	New	Old	New	Old	New	Old	New	Old	New	Old	New	Old	New	Old	New	Old	New	Old	New	Old	New	Old	New	Old	New	Old				
1 A	5.78	2.69	6.70	3.30	6.60	3.32	-	-	-	3.12	6.30	3.08	6.70	3.60	5.96	3.20	5.74	7.93	6.28	8.45	7.30	7.52																						
1 B	5.78	2.60	6.70	3.05	6.60	3.30	-	-	-	3.00	6.30	3.64	6.70	3.20	5.96	3.38	5.74	7.30	6.28	7.40	7.30	7.68																						
2 A	5.39	2.90	6.18	3.04	6.51	2.90	-	-	-	2.87	7.69	3.32	6.52	3.62	6.51	3.45	5.80	4.20	6.31	8.17	7.18	7.71																						
2 B	5.39	3.20	6.18	3.15	6.51	2.90	-	-	-	2.89	7.69	3.76	6.52	3.13	6.51	3.26	5.80	6.41	6.31	-	7.18	8.00																						
3 A	5.60	2.70	5.58	3.00	6.50	2.90	-	-	-	2.70	6.43	3.12	6.70	3.52	6.55	3.08	5.89	4.40	6.13	7.50	7.30	7.32																						
3 B	5.60	2.89	5.58	3.18	6.50	2.99	-	-	-	2.89	6.43	3.40	6.70	3.40	6.55	3.23	5.89	6.73	6.13	7.25	7.30	7.30																						
4 A	6.50	2.69	4.82	2.94	6.50	2.82	-	3.70	-	2.72	6.61	3.30	6.63	3.70	6.76	3.10	5.77	3.94	6.22	7.00	6.19	7.60																						
4 B	6.50	2.73	4.82	3.08	6.50	2.90	-	3.73	-	2.62	6.61	3.15	6.63	3.45	6.76	3.23	5.77	7.12	6.22	7.87	7.19	7.88																						
5 A	5.81	2.88	6.40	3.11	6.19	3.40	-	3.40	-	3.14	6.57	3.20	6.32	3.20	6.62	3.32	5.97	7.02	6.32	7.74	7.41	7.63																						
5 B	5.81	3.10	6.40	3.10	6.19	3.34	-	3.40	-	3.34	6.57	3.30	6.32	3.56	6.62	3.33	5.97	6.88	6.32	7.38	7.41	8.18																						
6 A	5.51	3.40	5.62	3.28	7.43	3.40	-	-	-	3.08	6.40	3.21	6.30	4.46	6.32	3.75	5.79	7.42	6.19	8.19	6.78	8.25																						
6 B	5.51	3.20	5.62	3.26	7.43	3.35	-	-	-	3.04	6.40	3.34	6.30	3.13	6.32	3.72	5.79	7.13	6.19	7.38	6.78	8.14																						
7 A	5.78	3.39	5.52	3.30	5.98	2.95	-	-	-	3.00	6.84	3.48	6.28	4.12	6.23	3.60	5.91	7.40	6.42	7.80	7.58	7.90																						
7 B	5.78	3.23	5.52	3.22	5.98	2.92	-	-	-	2.77	6.84	3.73	6.28	3.70	6.23	3.39	5.91	7.58	6.42	7.70	7.58	8.04																						
8 A	6.42	3.20	5.61	3.00	6.00	2.88	-	3.19	-	2.80	6.34	3.07	6.42	3.56	6.22	3.13	5.83	6.20	6.10	7.67	7.01	7.62																						
8 B	6.42	3.20	5.61	3.28	6.00	3.12	-	3.70	-	3.25	6.34	3.60	6.42	3.34	6.22	3.50	5.83	6.74	6.10	7.58	7.01	8.00																						
9 A	5.57	2.77	5.58	3.08	7.18	3.12	-	3.30	-	2.99	6.33	3.05	6.30	3.22	6.41	3.50	5.99	4.10	6.20	7.50	7.21	7.94																						
9 B	5.57	2.90	5.58	3.39	7.18	2.92	-	3.30	-	3.00	6.33	3.28	6.30	3.20	6.41	3.50	5.99	4.10	6.20	7.50	7.21	7.94																						
10 A	5.81	2.92	6.30	3.12	6.90	3.10	-	3.24	-	2.90	6.22	3.22	6.50	3.21	6.70	3.48	6.00	6.69	6.18	7.58	7.30	7.88																						
10 B	5.81	2.91	6.30	3.18	6.90	2.90	-	3.30	-	2.99	6.22	3.41	6.50	3.48	6.70	3.48	6.00	6.69	6.18	7.58	7.30	7.88																						
11 A	6.01	4.00	6.70	3.40	7.20	2.90	-	3.35	-	3.12	6.88	3.42	6.31	3.50	6.62	3.59	5.90	7.30	6.23	7.50	7.20	7.92																						
11 B	6.01	4.00	6.70	3.40	7.20	3.00	-	3.19	-	2.84	6.88	3.16	6.31	3.29	6.62	3.14	5.90	4.00	6.23	7.42	7.20	7.92																						
12 A	6.32	3.00	6.12	3.18	6.13	2.89	-	3.20	-	3.52	6.72	3.28	6.30	4.48	7.00	3.69	6.01	-	6.48	8.42	7.08	8.02																						
12 B	6.32	3.24	6.12	3.34	6.13	2.96	-	3.40	-	4.21	6.72	3.37	6.30	3.31	7.00	3.40	6.01	7.08	6.48	7.62	7.08	7.84																						
13 A	6.30	2.89	5.58	3.15	6.60	3.20	-	3.29	-	2.93	6.60	3.27	6.45	3.50	6.41	3.21	5.90	6.92	6.32	7.63	7.30	7.78																						
13 B	6.30	3.09	5.58	3.35	6.60	3.10	-	3.48	-	-	6.60	3.63	6.45	3.28	6.41	3.22	5.90	7.92	6.32	7.90	7.30	7.89																						
14 A	6.32	2.88	5.74	3.12	6.20	3.10	-	3.23	-	2.82	6.50	3.20	6.35	3.21	6.58	3.13	5.98	5.71	6.45	7.46	7.32	7.90																						
14 B	6.32	2.83	5.74	3.22	6.20	2.80	-	3.30	-	2.95	6.50	3.46	6.35	3.76	6.58	3.57	5.98	7.15	6.45	8.12	7.32	7.75																						
15 A	5.80	2.92	5.82	3.10	5.76	2.70	-	3.23	-	2.76	6.36	3.09	6.51	3.49	6.71	3.37	5.96	6.27	6.02	8.15	7.42	8.01																						
15 B	5.80	3.17	5.82	3.40	5.76	3.00	-	3.39	-	3.00	6.36	3.22	6.51	3.57	6.71	3.49	5.96	6.56	6.02	7.28	7.42	8.10																						
16 A	6.50	3.20	6.53	3.40	6.02	3.02	6.50	3.32	-	2.88	6.40	3.22	6.78	3.42	6.58	3.58	6.10	7.58	6.02	7.58	7.32	7.95																						
16 B	6.50	3.12	6.53	3.34	6.02	2.98	6.50	3.30	-	2.82	6.40	3.10	6.78	3.16	6.58	3.10	6.10	4.55	6.02	7.32	7.32	7.63																						
17 A	6.40	3.05	5.98	3.10	6.43	2.85	6.00	3.48	-	3.08	5.88	3.22	6.60	3.78	6.47	3.48	5.95	6.08	6.09	8.23	7.11	7.78																						
17 B	6.40	3.08	5.98	3.08	6.43	2.90	6.00	3.23	-	2.87	5.88	3.19	6.90	3.30	6.47	3.42	5.95	7.02	6.09	7.64	7.11	7.92																						
18 A	6.50	2.88	6.30	2.78	6.50	2.80	6.72	3.42	-	2.80	6.50	3.21	6.90	3.46	6.89	3.39	5.92	6.72	6.13	7.40	7.10	7.44																						
18 B	6.50	3.20	6.30	3.49	6.50	3.24	6.72	3.20	-	-	6.50	3.30	6.90	3.43	6.89	3.41	5.92	6.40	6.13	7.42	7.10	7.48																						
19 A	5.97	2.78	6.34	3.00	6.65	2.90	6.10	3.40	-	2.86	6.22	3.12	6.62	3.50	6.89	3.54	5.93	6.72	6.07	8.19	7.12	7.61																						
19 B	5.97	2.80	6.34	3.15	6.65	3.08	6.10	3.17	-	3.10	6.22	3.35	6.62	3.65	6.89	3.59	5.93	7.12	6.07	7.20	7.12	7.96																						
20 A	5.88	2.92	5.52	3.13	6.18	2.92	5.89	3.21	-	2.82	6.56	3.20	6.51	3.52	6.70	3.52	5.99	6.84	6.20	7.08	6.58	7.91																						
20 B	5.88	3.01	5.52	3.23	6.18	3.18	5.89	3.40	-	3.15	6.56	3.19	6.51	3.68	6.70	3.65	5.99	6.78	6.20	7.79	6.58	8.01																						
21 A	6.13	2.91	5.88	2.88	6.50	3.07	6.62	3.43	-	2.83	6.48	3.59	6.53	-	6.41	3.28	6.08	6.66	6.07	7.40	6.73	7.74																						
21 B	6.13	3.09	5.88	3.10	6.50	3.09	6.62	3.40	-	-	6.48	3.32	6.53	3.40	6.41	3.40	6.08	6.52	6.06	7.12	6.73	7.75																						
22 A	5.39	3.07	5.52	2.88	6.15	2.83	6.00	3.25	-	2.99	6.41	3.55	6.84	3.80	6.28	3.40	5.68	6.50	6.21	7.27	7.40	7.61																						
22 B	6.39	3.07	5.52	2.99	6.15	2.92	6.00	3.20	-	2.84	6.41	3.12	6.84	3.60	6.28	3.53	5.88	6.88	6.21	7.09	7.10	7.78																						
23 A	5.70	2.99	5.41	2.50	6.18	2.53	6.23	3.10	-	2.87	6.33	3.21	6.43	3.50	7.01	3.82	5.96	7.02	6.01	7.51	6.81	8.03																						
23 B	5.70	3.02	5.41	-	6.18	2.92	6.23	3.40	-	3.05	6.33	3.31	6.43	3.73	7.01	3.63	5.96	7.24	6.01	7.80	6.61	7.98																						
24 A	5.80	3.27	6.28	2.30	6.08	2.65	5.90	3.12	-	2.79	5.80	3.23	5.70	3.21	6.71	3.48	6.01	7.13	6.20	7.36	7.38	7.80																						
24 B	5.80	3.44	6.28	2.66	6.08	2.83	5.90	3.12	-	2.90	5.80	3.28	5.70	3.41	6.71	3.64	6.01	7.36	6.20	-	7.38	8.00																						
25 A	5.49	3.40	5.60	2.23	6.30	2.90	6.50	2.98	-	2.71	5.60	2.92	6.79	6.00	7.10	3.19	5.97	6.00	6.02	7.47	7.16	7.83																						
25 B	5.49	3.43	5.60	2.73	6.30	3.29	6.50	3.65	-	3.17	5.60	3.44	6.79	6.10	7.10	3.54	5.97	6.60	6.02	7.33	7.16	8.02																						
26 A	5.70	3.20	5.41	2.47	6.27	3.10	6.45	3.50	-	3.12	6.72	3.45	6.14	3.78	6.23	3.86	6.01	8.16	6.14	7.97	6.91	7.90																						
26 B	5.70	3.56	5.41	2.55	6.27	3.70	6.45	3.82	-	3.51	6.72	3.15	6.14	3.73	6.23	3.55	6.01	7.28	6.14	6.92	6.91	7.86																						
27 A	6.40	2.96	6.23	2.58	6.27	3.03	6.40	3.22	-	2.88	6.50	3.69	6.08	3.33	7.10	3.59	5.90	7.00	6.18	7.32	6.22	7.30																						
27 B	6.40	3.43	6.23	3.08	6.27	3.20	6.40	3.47	-	3.00	6.50	3.52	6.08	3.50	7.10	3.74	5.90	7.69	6.18	7.78	6.22	7.90																						
28 A	6.50	2.70	6.45	2.88	6.44	3.10	6.13	3.40	-	3.20	6.54	3.49	6.30	3.32	7.00	3.62	6.08	7.04	6.12	7.68	6.83	6.88																						
28 B	6.50	2.82	6.45	2.91	6.44	3.07	6.13	3.38	-	2.83	6.54	3.65	6.30	3.60	7.00	3.80	6.08	7.58	6.12	6.18	7.83	7.95																						
29 A	5.38	2.99	5.50	2.90	6.71	3.28	6.20	3.20	-	3.13	5.50	3.08	6.61	3.42	7.12	3.46	5.70	7.60	6.12	7.41	7.19	7.68																						
29 B	5.38	2																																										

ammonium chloride. See Appendix I for constituents of each solution.

Molar stock solutions were prepared for all the above salts, and the culture solutions were made up by adding the required amount of each salt to approximately 1 litre of water, in addition 2.00ml iron solution, and 2.0 ml micro-nutrient solution were added, and the solutions were made up to two litres with purified water, and the pH value of the solution was determined.

In each of the replicates the old solution was drained out of the flasks and replaced with 1 litre of the fresh culture solution, 1 ml of the iron solution was added every fourth day as previously mentioned.

#### pH Value

The pH values of the fresh nutrient solution, prior to being supplied to the plants, ranged from 5.32 - 6.50, the values at the time of renewal, after seven days, were found to have dropped considerably to 2.6 - 3.56. This lowering of the pH values was found to be consistent, as can be seen in Table I. Root damage was not observed at these low pH values, as would have been expected, as it has been reported (Hewitt 1952) that at low pH values, between 3.0 - 4.0 root injury generally occurs.

The addition of 3.0 ml Molar Ammonium acetate solution to two litres of nutrient solution, was found to stabilize the pH of the nutrient solutions. Over a period of a week the pH values of the nutrient solutions receiving ammonium acetate were found to have varied only slightly, with a tendency to become more alkaline. The values at the commencement ranged from 5.74 - 6.10, and at the time of renewal were from 4.0 - 7.0, see Table I.

#### Infection by *Tetranychus telarius* L.

During the course of the experiment the plants were infected with greenhouse red mite - *Tetranychus telarius*. The use of acaricides were inadmissible due to the possible chemical contamination of the nutrient solutions and the plants. Therefore the plants were washed with distilled water weekly, each leaf being swabbed down with cotton wool saturated with purified water, this method although tedious was

found to be most effective, in ridding the plants of the mite. Some of the plants were slightly damaged as a result of the attack by the mite, these will be mentioned under "Visual Symptoms".

#### Preparation of Plant Material

On the 19th January 1957, the plants were photographed, and their general appearance and condition recorded, the plants were then prepared for chemical analysis.

#### Preparation of Plant Material

Each plant was removed from its respective nutrient solution, any excess solution which may have adhered to the roots was removed by drying on sheets of No. 1 Whatman filter paper. The entire plant was then weighed on a rough balance for its fresh weight determination.

Each seedling was divided up into seven sections namely:  
(1) Upper Stem, (2) Middle Stem, (3) Lower Stem, (4) Upper Leaves, (5) Middle Leaves, (6) Lower Leaves, and (7) Roots, for dry weight determination and chemical analysis. The lengths of the section of the main axis were arbitrarily determined by ensuring each section from the same plant bore the same number of leaves. In the case of plants with branches, the laterals were assessed individually on the same basis, irrespective of their position on the main axis, the leaf and stem tissues from each plant were bulked into the upper, middle and lower leaf and stem groups. The base of the lower stem sections was fixed as being immediately above the hypocotyl and the root sections all tissue below this point.

The leaves from each group were washed in redistilled water to remove any dust, or any other surface contamination that might have been present on the leaves surface and were dried on sheets of No. 1 Whatman filter paper. The petiole and mid rib from each leaf were removed, using a pair of stainless steel scissors and the lamina was wrapped separately from the petiole and mid rib in No. 1 Whatman filter paper. Similarly the roots were washed in purified water, removing as far as possible any nutrient solution still adhering to the root surface. The root and stem sections were wrapped separately in sheets of No. 1 Whatman filter paper. The various groups were

appropriately labelled and placed in an "Analis" forced-draught oven kept at 105°C.

#### Dry Weights

The plant material was placed in an oven at 105°C for at least twenty four hours prior to weighing. The material to be weighed was removed from the oven and placed in a glass dessicator containing calcium chloride and allowed to cool for exactly thirty minutes, before weighing. The weighing operation was carried out as rapidly as possible, in an attempt to prevent moisture being taken up by the plant material from the atmosphere during weighing. The seven groups of material from each plant were weighed and recorded separately.

#### Grinding of Plant Material

The dried plant material was prepared for chemical analysis after its dry weight had been determined. The leaf material was ground to a fine powder using an agate ball and mill, a porcelain ball and mill was used for grinding the root material. All the stem tissue was ground by hand with a porcelain mortar and pestle. In each case when the desired degree of grinding had been reached the samples were transferred to glass specimen tubes for storage.

#### Mixing of Plant Material

In some instances e.g. upper stem, there was found to be insufficient plant material for chemical analysis. In order to overcome this difficulty the samples from corresponding treatments in Block A and Block B were bulked and thoroughly mixed in a mortar and pestle.

#### Chemical Analysis of Plant Material

##### Preparation of Plant Material for Analysis

The ground plant material was placed in petri dishes and dried for at least twenty four hours at 105°C prior to weighing, removing any moisture which may have been absorbed during storage. After removal from the oven, the sample was placed in a glass dessicator containing calcium chloride, and allowed to cool for exactly thirty minutes, prior to weighing. A sample (0.5 g approx.) of the powdered plant material was weighted out accurately and placed into a

silica beaker.

The silica beakers containing the samples were placed in a muffle furnace at 500°C, and kept at this temperature for at least fifteen hours, at the end of this period the plant ash was completely white. The beakers and contents were allowed to cool to room temperature. The ash was moistened with a few drops of purified water and was then dissolved in 20 ml 1:1 HCl solution. The silica beakers were covered with watch glasses and warmed on an electric hot plate for twenty five minutes. To each beaker, 1 ml of concentrated HNO<sub>3</sub> was added, ensuring the complete oxidation of any remaining organic matter. The solutions were then evaporated to dryness, the residue was dissolved in 10 ml 1:1 HCl and further diluted to about 20 ml total volume with redistilled water. This solution was heated for about ten minutes, and then filtered through No. 1 Whatman filter paper, which had been previously treated with hot dilute HCl solution to remove any calcium that may have been present in the paper. The filtrate was collected in clean 100 ml "Pyrex" beakers, and the residue was washed three times with hot water, and the washings were added to the filtrate, which was made up to approximately 50 ml.

It has been previously shown that the presence of either phosphate or sulphate ions interfere with the quantitative determination of sodium, potassium and calcium, using flame photometric methods. It was therefore necessary to remove these two anions from the plant solution. In the present work these ions were removed on an anion exchange resin De-Acedite E. The plant solution was poured into the resin column and allowed to leach through at the rate of one drop per second into 100 ml volumetric flask. The resin was washed with small portions of purified water, and these washings were added to the original solution, and made up to the volume with purified water. The solution was shaken well and the sodium, potassium, calcium and magnesium concentrations present in the solutions were determined as described below.

### Sodium, Potassium and Calcium Determination

An "EL" flame photometer was used for the determination of sodium, potassium, and calcium in the plant solutions. A standard solution containing 50.00 p.p.m. Na, 100.00 p.p.m. K, 350.00 p.p.m. Ca, and 100.00 p.p.m. Mg, and was made up using the following procedure:-

- (1) Weigh out 0.1271 g NaCl  
0.1906 g KCl  
0.8740 g CaCO<sub>3</sub>  
0.5732 g MgSO<sub>4</sub>·7H<sub>2</sub>O

(2) Place the above salts into 1 litre volumetric flask and dissolve in redistilled water.

(3) Add 3 ml 1:1 HCl

(4) Make up to 1 litre with redistilled water

All the chemicals used to make the above standard solution were of "Specpure" grade. The above solution was 10 X Standard Solution and referred to as Solution A, and was stored in a polythene container, as a precaution against any contamination from glassware. Solution A was diluted by half, giving Solution B, this solution was used for preparing the diluted standards used in calibrating the instrument.

The diluted standards were made up as follows:-

- (1) 20.0 ml Solution B made up to 100 ml with redistilled water
- (2) 15.0 ml Solution B made up to 100 ml with redistilled water
- (3) 10.0 ml Solution B made up to 100 ml with redistilled water
- (4) 5.0 ml Solution B made up to 100 ml with redistilled water
- (5) Redistilled water

The values obtained using the above standard solutions in calibrating the instrument can be found in Tables 2 - 4 below.

Table 2

Calibration of "EEL" flame photometer for estimating Sodium

Sodium concentration in p.p.m.

	0.00	1.26	2.53	3.79	5.06
Replicate	0.00	26.00	52.00	76.00	100.00
Instrument	0.00	26.00	52.00	76.00	100.00
Reading	0.00	26.00	52.00	76.00	100.00
Average	0.00	26.00	52.00	76.00	100.00

Table 3

Calibration of "EEL" flame photometer for estimating Potassium

concentration in p.p.m.

	0.0	2.43	4.87	7.31	9.75
Replicate	0.00	26.00	52.00	75.75	100.00
Instrument	0.00	26.00	51.75	75.25	100.00
Reading	0.00	26.00	51.50	74.25	100.00
Average	0.00	26.00	51.75	75.08	100.00

Table 4

Calibration of "EEL" flame photometer for estimating Calcium

Calcium concentration in p.p.m.

	0.00	8.75	17.50	26.25	35.00
Replicate	0.00	23.00	48.25	73.50	100.00
Instrument	0.00	23.00	48.00	73.50	100.00
Reading	0.00	23.00	48.50	73.50	100.00
Average	0.00	23.00	48.25	73.50	100.00

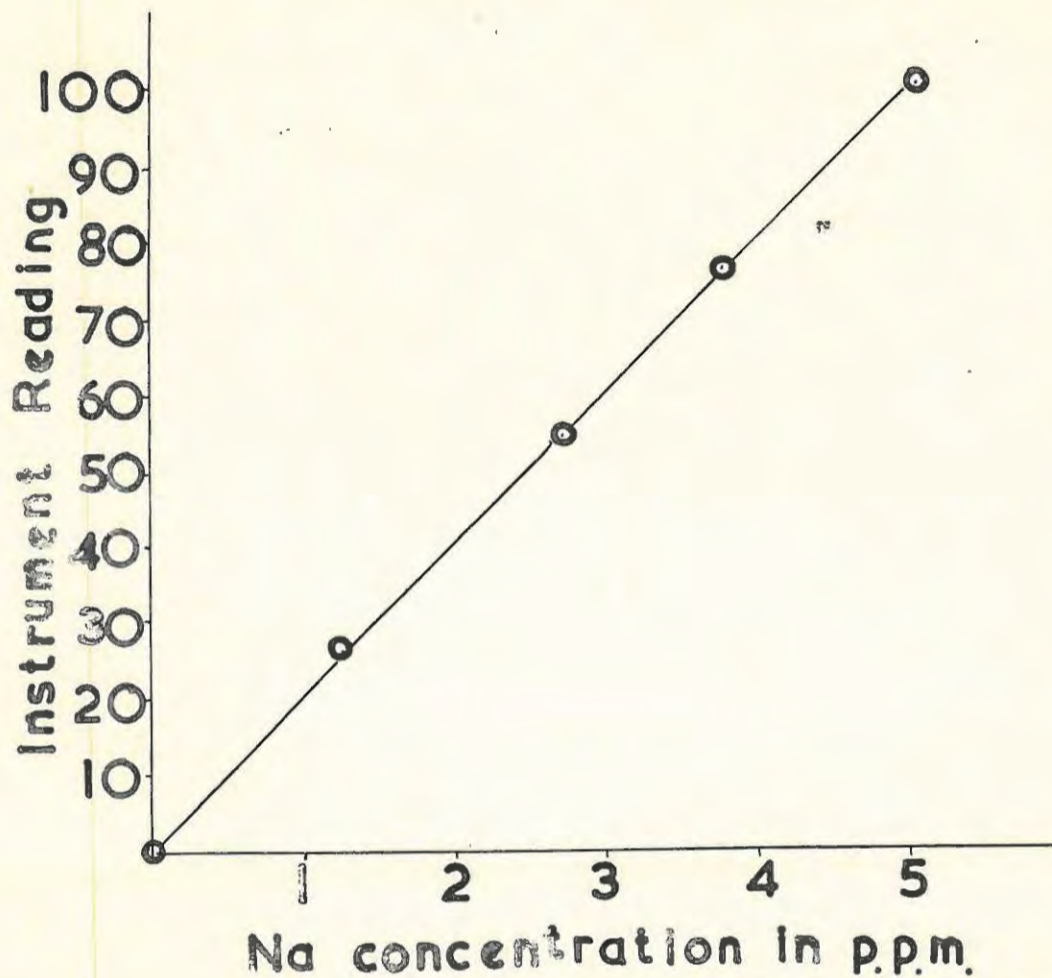


Fig. 4 Sodium calibration curve for E.E.L. flame photometer

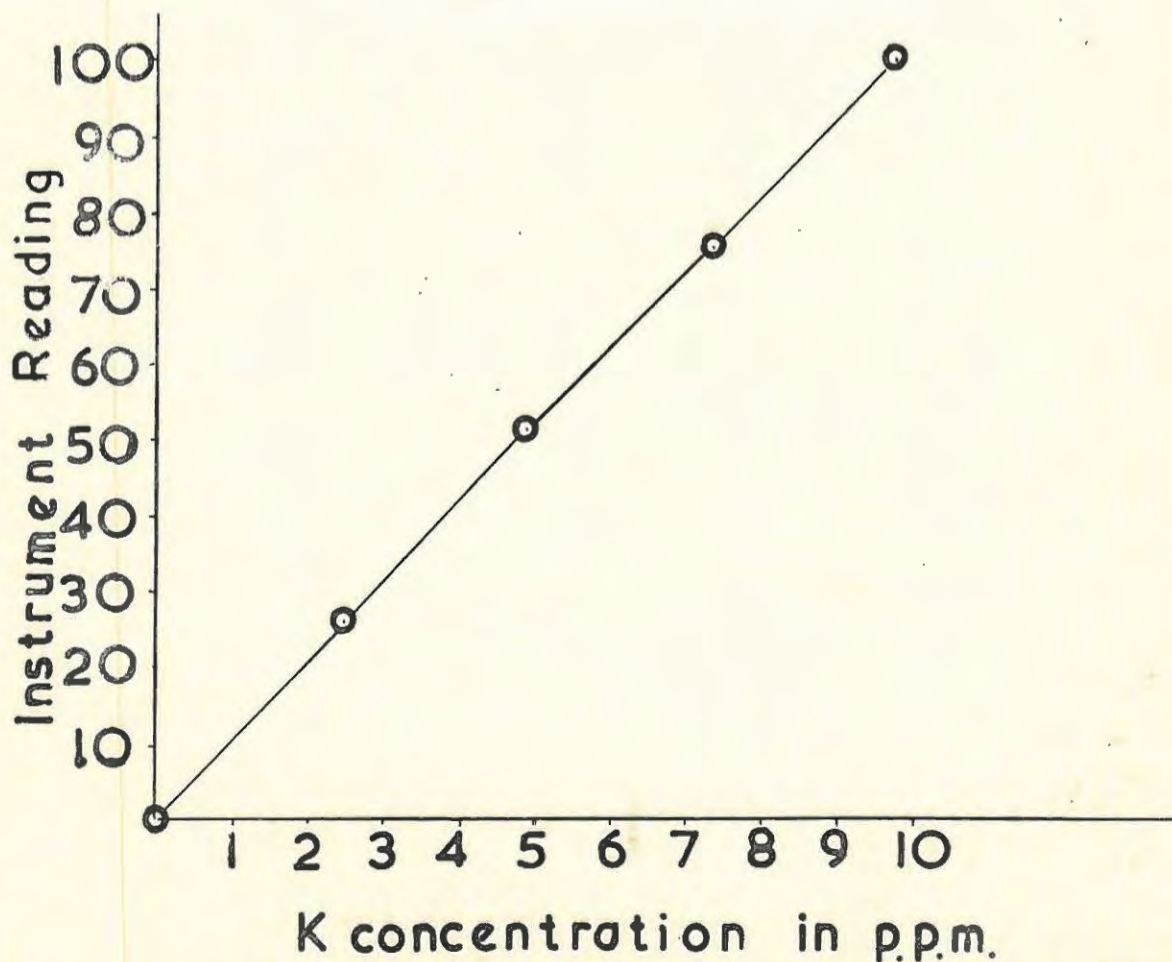


Fig. 5 Potassium calibration curve for E.E.L. flame photometer

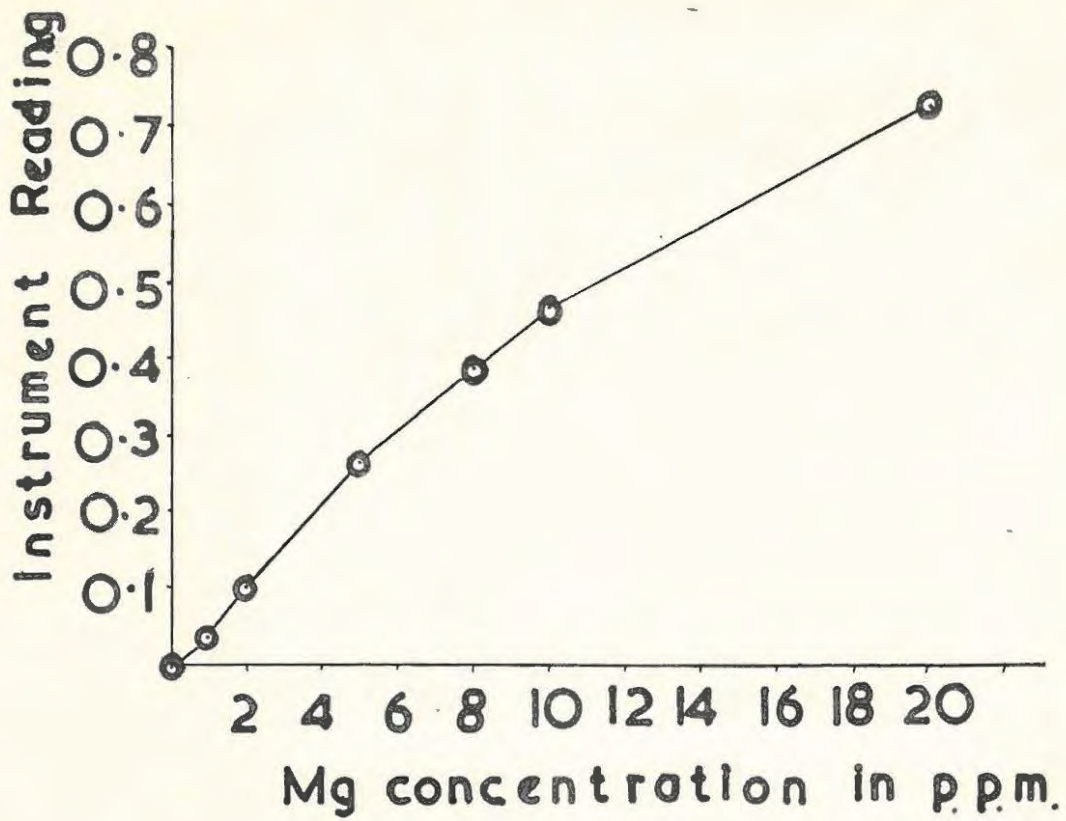


Fig. 6. Magnesium calibration curve for Spekker absorptiometer.

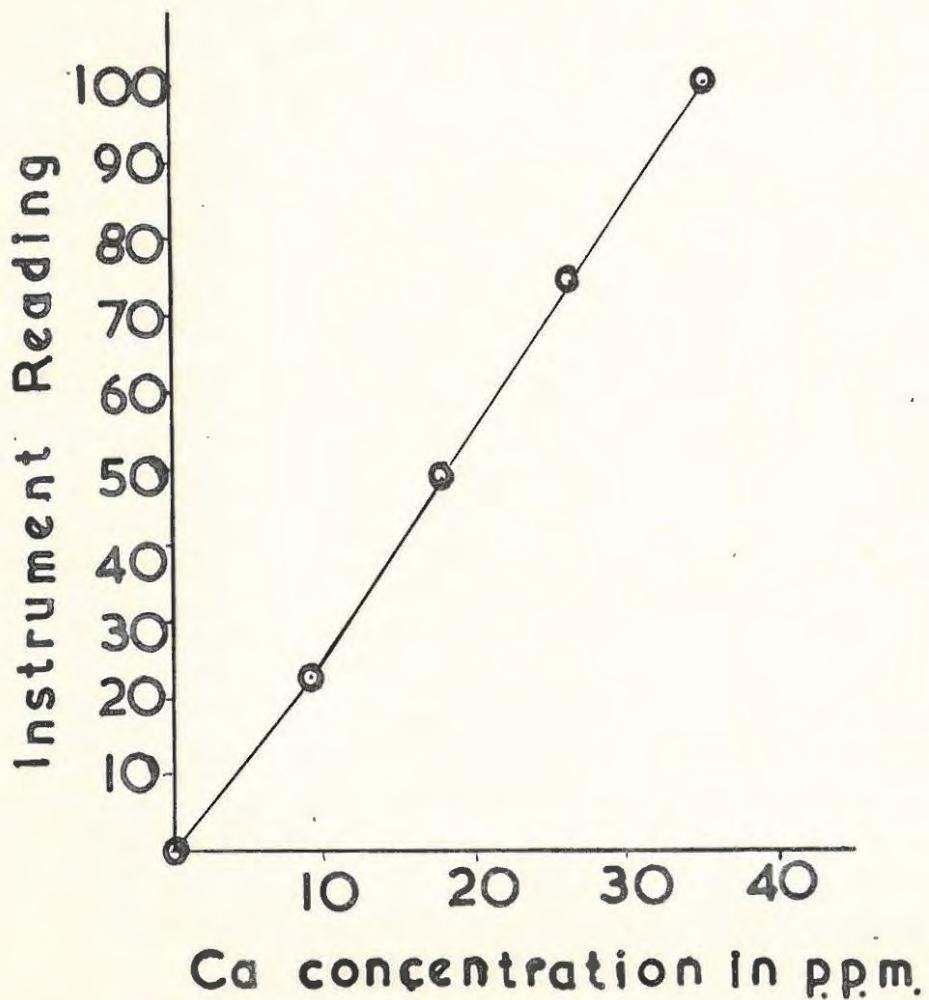


Fig. 7. Calcium calibration curve for E.E.L. flame photometer.

The relationship between the instrument readings and the concentrations of the above three elements was found to be almost linear, see Figs. 4,5 and 7.

In the case of the plant solution the sodium, potassium and calcium concentrations could be determined directly, however in some cases the solutions had to be diluted 5 X, 20 X, with purified water.

#### Magnesium Determination

For the magnesium determination in the plant solution the Titan Yellow Method was followed, using a Hilger "Spekker" absorptionmeter to measure the intensity of the colour produced by the reagent. The following reagents were used and prepared as follows:-

(1) Titan Yellow Solution

- (a) Weight out 0.30 g Titan Yellow Indicator (B.D.H.)
- (b) Dissolve in 150.0 ml Ethyl Alcohol.
- (c) Make up to 200.0 ml with redistilled water.

(2) Calcium Sulphate Solution

2.40 g  $\text{CaSO}_4$  were made up to 1 litre with purified water.

(3) 4% Hydroxylamine Hydrochloride Solution

4.0 g Hydroxylamine Hydrochloride was made up to 100 ml with redistilled water.

(4) Sodium Hydroxide Solution

100.0 g of Sodium Hydroxide solution was made up to 250 ml with purified water.

#### Calibration of "Spekker" absorption

A standard solution of magnesium solution was prepared as follows:-

- (1) Weight out 0.5067 g  $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ .
- (2) Dissolve in redistilled water.
- (3) Add 1.5 ml 1:1 HCl
- (4) Make to 500 ml with redistilled water.

The above solution contained 100.00 p.p.m.Mg. 1.0 ml, 2.0 ml, 5.0 ml, 8.0 ml, 10.0 ml and 20.0 ml aliquots of the standard solution were

taken and made up to 100 ml with redistilled water, giving dilute standards containing 1.0, 2.0, 5.0, 8.0, 10.0 and 20.0 p.p.m. magnesium.

The absorptiometer was calibrated using the following procedures:-

- (1) Pipette 5.0 ml of the diluted standard into a 200 ml conical flask.
- (2) Add 5.0 ml  $\text{CaSO}_4$ .
- (3) Add 20.0 ml redistilled water.
- (4) Place flasks in a water bath at  $25^\circ\text{C}$  for approximately fifteen minutes.
- (5) Remove the flasks from the water bath and add 1.0 ml 4% hydroxylamine hydrochloride, 0.7 ml Titan Yellow Solution, and 2.0 ml sodium hydroxide solution.
- (6) Shake the flasks.
- (7) Place the solution in a 4 cm. cell.
- (8) Use a Kodak No. 5 Yellow-Green filter.

A reagent blank solution was prepared similarly to the standard solutions with the exception that purified water was added instead of the  $\text{MgSO}_4$  solution. The readings obtained for the calibration curve are found in Table 5 below.

Table 5

Calibration of "Spekker" absorptiometer for estimating Magnesium

Mg concentration in p.p.m.	Drum Reading						Average
1.0	0.035	0.033	0.024	0.041	0.030	0.046	0.035
2.0	0.110	0.083	0.104	0.091	0.093	0.118	0.100
5.0	0.272	0.268	0.269	0.260	0.260	0.265	0.266
8.0	0.380	0.388	0.392	0.382	0.387	0.383	0.386
10.0	0.410	0.428	0.500	0.488	0.476	0.485	0.465
20.0	0.724	0.736	0.720	0.715	0.701	0.741	0.723

---

A curve was obtained when the drum readings were plotted against the concentration of magnesium, see Fig. 6.

The above method was found to be satisfactory for the determination of magnesium in plant material. The versenate (EDTA) method, which has been found satisfactory for the determination of magnesium in biological material was in this case found to be unsatisfactory due to the indicator being oxidized by the high iron content of the root.

## CHAPTER III

### Results

The effects of the experimental treatment on the visual appearance and condition of the lemon seedlings at the time of sampling are summarised in Table 6.

#### Visual Appearance

At the conclusion of the experiment it was observed that the leaves of some of the seedlings had produced visual symptoms suggesting the presence of a physiological disorder. The appearance of these symptoms was not confined to any single leaf position on the central axis. Generally the symptoms were more pronounced in the apical regions of the seedlings receiving the lower calcium level in the nutrient solution, and in the lower leaves of the seedlings receiving the higher calcium level of supply. These symptoms are described in greater detail below.

#### Leaf Symptoms

##### (1) Leaf Burn

For several days during the early part of January 1957, the greenhouse temperature rose considerably higher than normal (approximately 98°F), due to warm spell. It was during this period that leaf burn was first observed amongst the apical leaves of the seedlings.

Leaf burn was found to occur in seedlings receiving a low level of calcium together with high levels of (1) potassium, (2) sodium, or (3) sodium and potassium in the nutrient solution. At the time of sampling leaf burn was found not to have occurred on the foliage of seedlings receiving potassium treatments of 117.0 p.p.m. or less, or sodium treatments of 50.0 p.p.m. or less. Lemon seedlings receiving treatments with potassium levels higher than 117.0 p.p.m., together with levels of sodium of 50.0 p.p.m., were found not to produce leaf burn. Similarly the foliage of plants receiving levels of sodium of 195.0 p.p.m., together with 117.0 p.p.m. potassium, did not suffer from leaf burn. However, the seedlings receiving



Fig. 8. Showing Stage II of Leaf Chlorosis.



Fig. 9. Showing Stage III of Leaf Chlorosis



Fig. 10. Showing the various stages of leaf chlorosis on a single tree, from the Lower Leaf ( No 1) to the Apical Leaf ( No 31 ).



Fig. 11. Shows characteristic V shaped chlorotic area prior to leaf burn.



Fig. 12. Shows scorching of the apical leaves.



Fig. 13. Shows scorching of the apical leaves of a seedling receiving a high potassium and a low sodium treatment.



Fig. 14. A comparison between the appearance of seedlings receiving a high calcium (left), and a low calcium (right) treatment.



Fig. 15. A comparison between the appearance of seedlings receiving a high calcium (left), and a low calcium (right) treatment.



Fig. 16. Showing damage resulting from eruption of the bark

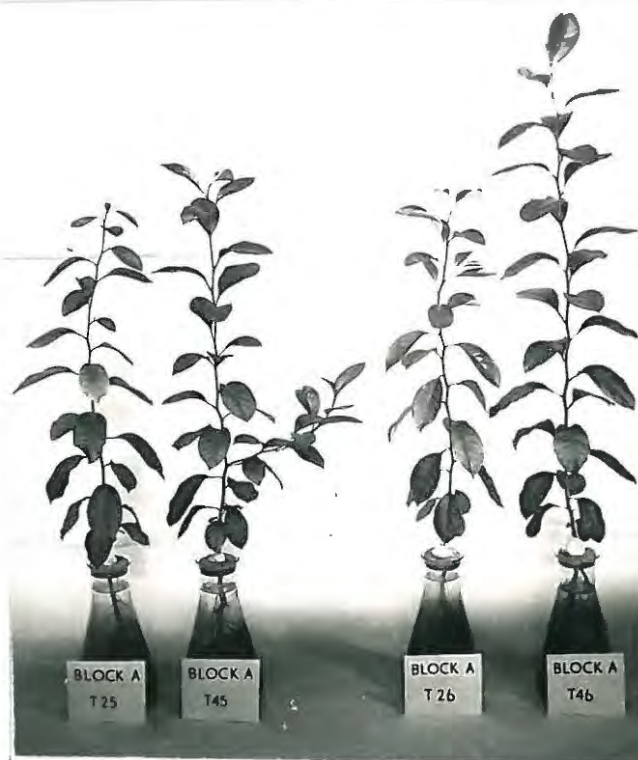


Fig. 17. A comparison of size between seedlings grown in high calcium treatment ( T 45, T 46 ), and low calcium treatment ( T 25, T 26 ).



Fig. 18. A comparison of size between seedlings grown in high calcium treatment (T47, T48) and low calcium treatment (T27, T28).

195.0 p.p.m. sodium, with potassium levels higher or lower than 117.0 p.p.m., suffered from leaf burn. Seedlings supplied with sodium concentrations higher than 195.0 p.p.m. produced leaf burn irrespective of the level of potassium. When the concentration of the external supply of calcium was increased, leaf burn was found not to occur, even in the presence of high levels of sodium and potassium.

The first visual sign suggesting that leaf burn is likely to occur is the appearance of light green areas in between the primary lateral veins, close to the leaf margins. These areas in the early stages are not always easily visible, and may only be observed by holding the leaf to a source of illumination.

The next stage is the progressive formation between the primary, lateral veins, on the lamina margin, of light green chlorotic areas. The leaf tissue between the veinlets becomes yellow-green in colour, while the primary lateral veins and veinlets maintain their original deep green colour, see Fig. 8. The chlorotic areas increase in size and eventually coalesce around the margin of the lamina to form a continuous band along the affected region. At the same time the chlorotic areas develop inwards towards the mid rib producing parallel chlorotic bands between the primary lateral veins, the latter remaining green, see Figs. 9 and 10. In the final stage the central tissue remains green, while the affected areas fuse to give a V-shaped chlorotic region which extends along the margin from the leaf tip to a position just beyond mid leaf. This stage is normally accompanied by the curling of the leaf margins towards the ventral leaf surface. Prior to leaf burn taking place, the yellow chlorotic regions develop a tangerine-orange colour (B.C.C.) see Figs. 11 and 12. In some instances leaf burn occurred prior to the merging of the chlorotic areas from both leaf margins. It was observed that there were instances of seedlings receiving high potassium levels and no sodium, where leaf burn occurred on the leaf margins and tips, without any of the above pre-burn symptoms occurring, see Fig. 13.

Frequently abscission of the leaves occurred after leaf burn, leaving a portion of the petiole still attached to the central axis. There were however, isolated cases of abscission of the leaves taking

place in the absence of any leaf burn, but in these particular instances the leaves were in an advanced stage of chlorosis.

## (II) Leaf Chlorosis

As mentioned above there were seedlings receiving the experimental treatments which, although not suffering from leaf burn, still showed signs of the presence of a leaf disorder. These visual symptoms resemble the stages occurring in pre-leaf burn described above, and these stages will be referred to in the present text as follows:-

### (i) Stage I

Light green areas formed in between the lateral veins, near the leaf margin. These areas were not always easily observed, and in some cases were only visible by holding the leaf to a light source.

### (ii) Stage II

Light yellow-green chlorotic areas formed in between the primary lateral veins, in a zone extending from the leaf tip to approximately mid leaf. These chlorotic regions were always close to the leaf margin. The primary lateral veins and veinlets maintained their original deep green colour.

### (iii) Stage III

The chlorotic areas coalesced, forming a continuous yellow-green region of tissue parallel to the leaf margin, and extending from the leaf tip to approximately mid-leaf.

### (iv) Stage IV

A V-shaped chlorotic area, with the two arms joined at the leaf tip, and each arm lying along the leaf margin. The chlorotic region had a characteristic light tangerine-orange colour (B.C.C.), while the central leaf tissue remained green.

Visual signs of a physiological disorder in the leaves were induced as a result of the experimental treatments. The plants receiving the lower level of calcium supply were found to suffer severely from leaf burn, and the leaves showed marked visual symptoms of an internal disorder. At the higher supply of calcium, no leaf burn was found to occur, and when symptoms of leaf disorder did occur, they were only very mild. A comparison between the visual

TABLE 6

Visual Appearance of Lemon Seedlings.

		BLOCK A	
Treat- ment.	Ca <sub>1</sub>	Treat- ment.	Ca <sub>2</sub>
Na <sub>0</sub>	1  K <sub>0</sub>	No leaf burn. All leaves show signs of a physiological disorder.  <u>Upper leaves</u> Stage I <u>Middle leaves</u> Stage II <u>Lower leaves</u> Stage III	17  No leaf burn. All leaves show signs of a slight physiological disorder.  <u>Upper leaves</u> Stage I <u>Middle leaves</u> Stage II <u>Lower leaves</u> Stage II
	2  K <sub>1</sub>	No leaf burn. No leaf symptoms of a physiological disorder.  <u>Upper leaves</u> (lateral) Healthy <u>Middle leaves</u> (lateral) Healthy <u>Lower leaves</u> (lateral) Healthy  <u>Remarks.</u> Bark eruption on central axis. Central axis dead above eighth basal leaf.	18  No leaf burn. Some leaves show symptoms of a physiological disorder.  <u>Upper leaves</u> Healthy <u>Middle leaves</u> Healthy <u>Lower leaves</u> Stage II
	3  K <sub>2</sub>	Leaf burn of five upper leaves. Abcission of third apical leaf. All leaves show signs of a physiological disorder.  <u>Upper leaves</u> Scorched and Stage III <u>Middle leaves</u> Stage II <u>Lower leaves</u> Stage II	19  No leaf burn. Some leaves show symptoms of a physiological disorder.  <u>Upper leaves</u> Healthy <u>Middle leaves</u> Healthy <u>Lower leaves</u> Stage II
	4  K <sub>3</sub>	Leaf burn of four upper leaves.  <u>Upper leaves</u> Scorched (marginal) Stage II <u>Middle leaves</u> Stage III, and Stage IV <u>Lower leaves</u> Stage II	20  No leaf burn. Lower leaves show symptoms of a physiological disorder.  <u>Upper leaves</u> Healthy <u>Middle leaves</u> Healthy <u>Lower leaves</u> Stage II

Table 6 contd.

BLOCK A

Treat- ment.	Ca <sub>1</sub>	Treat- ment.	Ca <sub>2</sub>
5  K <sub>0</sub>	No leaf burn. All leaves show signs of a physiological disorder.  <u>Upper leaves</u> Stage II <u>Middle leaves</u> Stages II and III <u>Lower leaves</u> Stage II  <u>Remarks</u> . Two lower leaves show signs of "Epidermal Collapse".	21	No leaf burn. Some leaves show signs of a physiological disorder.  <u>Upper leaves</u> Healthy. <u>Middle leaves</u> Stage II <u>Lower leaves</u> Stage II
6  K <sub>1</sub>	No leaf burn. Lower leaves show signs of a physiological disorder.  <u>Upper leaves</u> Healthy <u>Middle leaves</u> Healthy <u>Lower leaves</u> Stage II	22	No leaf burn. Some leaves show signs of a physiological disorder.  <u>Upper leaves</u> Healthy <u>Middle leaves</u> Healthy <u>Lower leaves</u> Stage II
7  K <sub>2</sub>	No leaf burn. All leaves show signs of a physiological disorder.  <u>Upper leaves</u> Stage IV <u>Middle leaves</u> Stage III <u>Lower leaves</u> Stage II	23	No leaf burn. Some leaves show signs of a physiological disorder.  <u>Upper leaves</u> Healthy <u>Middle leaves</u> Stage I <u>Lower leaves</u> Stage II
8  K <sub>3</sub>	No leaf burn. Some leaves show signs of a physiological disorder.  <u>Upper leaves</u> Healthy <u>Middle leaves</u> Stage III <u>Lower leaves</u> Stage II  <u>Remarks</u> . Curling of lower and middle leaves.	24	No leaf burn. Some leaves show signs of a physiological disorder.  <u>Upper leaves</u> Healthy <u>Middle leaves</u> Healthy <u>Lower leaves</u> Stage II  <u>Remarks</u> . Two lower leaves show signs of "Epidermal Collapse".

Table 6 Contd.

BLOCK A

		BLOCK A	
Treat- ment.	Ca <sub>1</sub>	Treat- ment.	Ca <sub>2</sub>
Na <sub>2</sub>	9  K <sub>3</sub>	Leaf burn. Abcission of two leaves. All leaves show signs of a physiological disorder.  <u>Upper leaves</u> Stage IV <u>Middle leaves</u> Stage III <u>Lower leaves</u> Stage II.	25  No leaf burn. Some leaves show signs of a physiological disorder.  <u>Upper leaves</u> Healthy <u>Middle leaves</u> Stage II <u>Lower leaves</u> Stage II
	10  K <sub>1</sub>	No leaf burn. All leaves show signs of a physiological disorder.  <u>Upper leaves</u> Stage I <u>Middle leaves</u> Stage II <u>Lower leaves</u> Stage I	26  No leaf burn. Some leaves show signs of a physiological disorder.  <u>Upper leaves</u> Healthy <u>Middle leaves</u> Stage I <u>Lower leaves</u> Stage I
	11  K <sub>2</sub>	Leaf burn. All leaves show signs of a physiological disorder.  <u>Upper leaves</u> Stage II <u>Middle leaves</u> Stage III <u>Lower leaves</u> Stage II	27  No leaf burn. Some leaves show signs of a physiological disorder.  <u>Upper leaves</u> Healthy <u>Middle leaves</u> Healthy <u>Lower leaves</u> Stage I  <u>Remarks.</u> "Epidermal Collapse" on three lower leaves.
	12  K <sub>3</sub>	Leaf burn. All leaves show signs of a physiological disorder.  <u>Upper leaves</u> Stage III <u>Middle leaves</u> Stage II <u>Lower leaves</u> Stage II	28  No leaf burn. No signs of a physiological disorder.  <u>Upper leaves</u> Healthy <u>Middle leaves</u> Healthy <u>Lower leaves</u> Healthy

Table 6 Contd.

## BLOCK A

Treat- ment.	Ca <sub>1</sub>	Treat- ment.	Ca <sub>2</sub>
Na <sub>3</sub>	13  K <sub>0</sub>	Leaf burn. Abcission of four apical leaves. All leaves show signs of a physiological disorder.  <u>Upper leaves</u> Stage IV <u>Middle leaves</u> Stage II <u>Lower leaves</u> Stage II  <u>Remarks.</u> Burn and die back of apical stem.	29  No leaf burn. Some leaves show signs of a physiological disorder.  <u>Upper leaves</u> Healthy <u>Middle leaves</u> Healthy <u>Lower leaves</u> Stage II
	14  K <sub>1</sub>	Leaf burn. Abcission of four apical leaves. All leaves show signs of a physiological disorder.  <u>Upper leaves</u> Stage IV <u>Middle leaves</u> Stage III <u>Lower leaves</u> Stage II	30  No leaf burn. Some leaves show signs of a physiological disorder.  <u>Upper leaves</u> Healthy <u>Middle leaves</u> Healthy <u>Lower leaves</u> Stage I  <u>Remarks.</u> Two leaves show signs of "Epidermal Collapse".
	15  K <sub>2</sub>	Leaf burn. All leaves show signs of a physiological disorder.  <u>Upper leaves</u> Stage IV. <u>Middle leaves</u> Stage II <u>Lower leaves</u> Stage II	31  No leaf burn. No signs of a physiological disorder.  <u>Upper leaves</u> Healthy <u>Middle leaves</u> Healthy <u>Lower leaves</u> Healthy  <u>Remarks.</u> Bark eruptions on lower stem.
	16  K <sub>3</sub>	Leaf burn. Abcission of ten apical leaves. All leaves show signs of a physiological disorder.  <u>Upper leaves</u> Burnt <u>Middle leaves</u> Stage IV <u>Lower leaves</u> Stage II	32  No leaf burn. Some leaves show signs of a physiological disorder.  <u>Upper leaves</u> Healthy <u>Middle leaves</u> Healthy <u>Lower leaves</u> Stage I  <u>Remarks.</u> Tree shows effects of infection by <u>Tetranychus telarius</u> .

Treat- ment.	Ca <sub>1</sub>	Treat- ment.	Ca <sub>2</sub>	
Na <sub>0</sub>	1	No leaf burn. All leaves show signs of a physiological disorder.  <u>Upper leaves</u> Stage II <u>Middle leaves</u> Stage II <u>Lower leaves</u> Stage II	17	No leaf burn. All leaves show signs of a physiological disorder.  <u>Upper leaves</u> Stage II <u>Middle leaves</u> Stage II <u>Lower leaves</u> Stage I  <u>Remarks.</u> Tree shows effects of infection by <u>Tetranychus telarius.</u>
	2	No leaf burn. All leaves show signs of a physiological disorder.  <u>Upper leaves</u> Stage I <u>Middle leaves</u> Stage II <u>Lower leaves</u> Stage II	18	No leaf burn. Some leaves show signs of a physiological disorder.  <u>Upper leaves</u> Healthy <u>Middle leaves</u> Stage I <u>Lower leaves</u> Stage I  <u>Remarks.</u> Tree shows effects of infection by <u>Tetranychus telarius.</u>
	3	Leaf burn. Abcission of one apical leaf. All leaves show signs of a physiological disorder.  <u>Upper leaves</u> Scorch, and Stage III <u>Middle leaves</u> Stage II <u>Lower leaves</u> Stage II	19	No leaf burn. Lower leaves show signs of a physiological disorder.  <u>Upper leaves</u> Healthy <u>Middle leaves</u> Healthy <u>Lower leaves</u> Stage I  <u>Remarks.</u> Tree shows effects of infection by <u>Tetranychus telarius.</u>
	4	No leaf burn. Abcission of one apical leaf. All leaves show signs of a physiological disorder.  <u>Upper leaves</u> Stage IV <u>Middle leaves</u> Stage III <u>Lower leaves</u> Stage II  <u>Remarks.</u> Tree shows effect of infection by <u>Tetranychus telarius.</u>	20	No leaf burn. Lower leaves show signs of a physiological disorder.  <u>Upper leaves</u> Healthy <u>Middle leaves</u> Healthy <u>Lower leaves</u> Stage I

Table 6 Contd.

BLOCK B

Treat- ment.	Ca <sub>1</sub>	Treat- ment.	Ca <sub>2</sub>
Na <sub>1</sub>	5  K <sub>0</sub>	No leaf burn. All leaves show signs of a physiological disorder.  <u>Upper leaves</u> Stage I <u>Middle leaves</u> Stage II <u>Lower leaves</u> Stage II	21  No leaf burn. All leaves show signs of a physiological disorder.  <u>Upper leaves</u> Stage I <u>Middle leaves</u> Stage II <u>Lower leaves</u> Stage II
	6  K <sub>1</sub>	No leaf burn. All leaves show signs of a physiological disorder.  <u>Upper leaves</u> Stage II <u>Middle leaves</u> Stage III <u>Lower leaves</u> Stage III  <u>Remarks.</u> Wilting of upper leaves.	22  No leaf burn. Some leaves show signs of a physiological disorder.  <u>Upper leaves</u> Healthy <u>Middle leaves</u> Stage I <u>Lower leaves</u> Stage II
	7  K <sub>2</sub>	No leaf burn. Some leaves show signs of a physiological disorder.  <u>Upper leaves</u> Stage I <u>Middle leaves</u> Healthy <u>Lower leaves</u> Healthy	23  No leaf burn. No signs of a physiological disorder.  <u>Upper leaves</u> Healthy <u>Middle leaves</u> Healthy <u>Lower leaves</u> Healthy  <u>Remarks.</u> Wilting of apical leaves.
	8  K <sub>3</sub>	No leaf burn. Some leaves show signs of a physiological disorder.  <u>Upper leaves</u> Stage II <u>Middle leaves</u> Stage I <u>Lower leaves</u> Healthy	24  No leaf burn. All leaves show signs of a physiological disorder.  <u>Upper leaves</u> Stage I <u>Middle leaves</u> Stage II <u>Lower leaves</u> Stage II  <u>Remarks.</u> Tree shows effects of infection by <u>Tetranychus telarius</u> .

Table 6 Contd.

BLOCK B

	Treat- ment	Ca <sub>1</sub>	Treat- ment.	Ca <sub>2</sub>
Na <sub>2</sub>	9	No leaf burn. Some leaves show signs of a physiological disorder.  <u>Upper leaves</u> Healthy <u>Middle leaves</u> Stage II <u>Lower leaves</u> Stage II  <u>Remarks.</u> Wilting of upper leaves	25	No leaf burn. No signs of a physiological disorder.  <u>Upper leaves</u> Healthy <u>Middle leaves</u> Healthy <u>Lower leaves</u> Healthy  <u>Remarks.</u> Wilting of upper leaves
	10	No leaf burn. All leaves show signs of a physiological disorder.  <u>Upper leaves</u> Stage III <u>Middle leaves</u> Stage II <u>Lower leaves</u> Stage II	26	No leaf burn. Some leaves show signs of a physiological disorder.  <u>Upper leaves</u> Healthy <u>Middle leaves</u> Healthy <u>Lower leaves</u> Stage I
	11	Leaf burn. All leaves show signs of a physiological disorder.  <u>Upper leaves</u> Stage III <u>Middle leaves</u> Stage II <u>Lower leaves</u> Stage II	27	No leaf burn. No signs of a physiological disorder.  <u>Upper leaves</u> Healthy <u>Middle leaves</u> Healthy <u>Lower leaves</u> Healthy  <u>Remarks.</u> Wilting of upper leaves
	12	Leaf burn. Abcission of five apical leaves. All leaves show symptoms of a physiological disorder.  <u>Upper leaves</u> Stage III <u>Middle leaves</u> Stage II <u>Lower leaves</u> Stage I	28	No leaf burn. Some leaves show signs of a physiological disorder.  <u>Upper leaves</u> Healthy <u>Middle leaves</u> Stage I <u>Lower leaves</u> Stage I

Table 6 Contd.

## BLOCK B

Treat-ment.	Ca <sub>1</sub>	Treat-ment.	Ca <sub>2</sub>
Na <sub>3</sub>	13  K <sub>0</sub>	Leaf burn. Abcission of nine leaves. All leaves show symptoms of a physiological disorder.  <u>Upper leaves</u> Burnt <u>Middle leaves</u> Stage III <u>Lower leaves</u> Stage II	29  No leaf burn. Some leaves show signs of a physiological disorder.  <u>Upper leaves</u> Healthy <u>Middle leaves</u> Healthy <u>Lower leaves</u> Stage I
	14  K <sub>1</sub>	Leaf burn. All leaves show signs of a physiological disorder.  <u>Upper leaves</u> Stage IV <u>Middle leaves</u> Stage III <u>Lower leaves</u> Stage II	30  No leaf burn. Some leaves show signs of a physiological disorder.  <u>Upper leaves</u> Healthy <u>Middle leaves</u> Stage I    2 leaves showing "Epidermal collapse". <u>Lower leaves</u> Stage I  <u>Remarks.</u> Bark eruption on main axis.
	15  K <sub>2</sub>	Leaf burn. Some leaves show signs of a physiological disorder.  <u>Upper leaves</u> Stage III <u>Middle leaves</u> Stage II <u>Lower leaves</u> Stage I	31  No leaf burn. Some leaves show signs of a physiological disorder.  <u>Upper leaves</u> Healthy <u>Middle leaves</u> Healthy <u>Lower leaves</u> Stage I
	16  K <sub>3</sub>	Leaf burn. All leaves show signs of a physiological disorder.  <u>Upper leaves</u> Stage IV <u>Middle leaves</u> Stage III <u>Lower leaves</u> Stage I	32  No leaf burn. No signs of any physiological disorder.  <u>Upper leaves</u> (lateral) Healthy <u>Middle leaves</u> (lateral) Healthy <u>Lower leaves</u> (main axis and laterals) Healthy  <u>Remarks.</u> Bark eruptions on lower main axis. Main axis died above basal leaf.

appearance of the seedlings receiving high and low levels of calcium can be seen in Figs. 14 and 15.

(III) "Epidermal Collapse"

During the early stages of growth the seedlings produced leaf spots, described previously (Chapter I). This visual leaf symptom will be referred to in the present work as "Epidermal Collapse", see Fig. 2. The spots produced on the upper and lower epidermal surfaces were the result of an unknown factor and appeared to be areas where the sub-epidermal tissue had broken down, resulting in epidermal collapse. The plants showing this phenomenon used in the present investigation are recorded in Table 6.

(IV) Effect of Tetranychus telarius L

The trees affected by Tetranychus telarius during the course of the present investigation were left with visual signs of their attack. The leaves lost their usual waxy gloss and both the upper and lower surfaces became rough in texture and took on a dull light green colour. In Table 6 a record has been made of experimental plants attacked by the mite.

Stem Symptoms

(I) Stem Burn

There were isolated occurrences amongst the experimental seedlings receiving the lower level of calcium in the nutrient solutions, of the apical region of the stem being burnt, and die back resulting, see Fig 11. These instances were only observed when there had been severe leaf burn, followed by leaf abscission.

(II) Bark eruption

In some of the experimental trees eruptions of the bark occurred in the lower stem regions exposing the woody tissue beneath. These erupted areas were approximately 1.5 x 0.2 cm. in extent, the greatest length of these areas lay in the same direction as the central axis. Usually there was more than one area on the infected stems. In three instances the damage brought about as a result of this bark eruption was so extensive that all the plant tissue above the affected area died, see Fig. 16. In each of these three cases, a

TABLE 7

Fresh Weight and Height of Lemon Seedlings on 9.1.57

Treatment			Weight in gms.		Height in cms.	
			Block A	Block B	Block A	Block B
T 1	K <sub>0</sub>	Ca <sub>1</sub>	22.3	18.2	51.1	50.1
T 17		Ca <sub>2</sub>	33.5	24.5	62.2	48.6
T 2	K <sub>1</sub>	Ca <sub>1</sub>	25.4	39.7	85.5	69.1
T 18		Ca <sub>2</sub>	47.2	31.5	77.5	57.4
T 3	K <sub>2</sub>	Ca <sub>1</sub>	36.6	27.4	68.2	58.2
T 19		Ca <sub>2</sub>	39.7	25.5	71.7	57.7
T 4	K <sub>3</sub>	Ca <sub>1</sub>	52.1	27.2	86.5	52.5
T 20		Ca <sub>2</sub>	46.3	77.0	77.0	66.9
T 5	K <sub>0</sub>	Ca <sub>1</sub>	23.2	23.7	54.5	47.4
T 21		Ca <sub>2</sub>	35.0	27.8	78.7	54.4
T 6	K <sub>1</sub>	Ca <sub>1</sub>	28.0	19.8	53.7	45.3
T 22		Ca <sub>2</sub>	44.9	33.2	74.6	62.6
T 7	K <sub>2</sub>	Ca <sub>1</sub>	33.3	30.7	69.0	59.5
T 23		Ca <sub>2</sub>	49.0	18.6	81.7	46.2
T 8	K <sub>3</sub>	Ca <sub>1</sub>	33.7	25.3	65.4	46.8
T 24		Ca <sub>2</sub>	51.0	32.2	75.5	57.2
T 9	K <sub>0</sub>	Ca <sub>1</sub>	29.1	22.6	60.1	52.1
T 25		Ca <sub>2</sub>	34.9	32.4	64.1	57.3
T 10	K <sub>1</sub>	Ca <sub>1</sub>	43.3	30.4	70.3	66.0
T 26		Ca <sub>2</sub>	49.9	34.5	75.9	53.0
T 11	K <sub>2</sub>	Ca <sub>1</sub>	30.0	26.3	61.0	58.0
T 27		Ca <sub>2</sub>	44.0	25.8	76.3	49.1
T 12	K <sub>3</sub>	Ca <sub>1</sub>	45.0	31.9	77.8	55.2
T 28		Ca <sub>2</sub>	42.8	41.6	104.6	88.0
T 13	K <sub>0</sub>	Ca <sub>1</sub>	28.0	20.5	59.7	52.6
T 29		Ca <sub>2</sub>	36.2	27.2	67.5	53.0
T 14	K <sub>1</sub>	Ca <sub>1</sub>	41.0	25.4	75.4	60.2
T 30		Ca <sub>2</sub>	50.6	35.9	84.3	65.3
T 15	K <sub>2</sub>	Ca <sub>1</sub>	37.9	27.2	71.1	48.6
T 31		Ca <sub>2</sub>	35.1	43.5	131.9	63.0
T 16	K <sub>3</sub>	Ca <sub>1</sub>	23.0	35.8	63.5	68.5
T 32		Ca <sub>2</sub>	45.6	27.7	95.3	82.5

Dry Weight expressed in grams. Block A

Treatment			Dry Wt. of Lower Leaves	Dry Wt. of Mid Leaves	Dry Wt. of Upper Leaves	Dry Wt. of Lower Stem	Dry Wt. of Mid Stem	Dry Wt. of Upper Stem	Dry Wt. of Roots	Total Dry Weight	
T 1	Na <sub>0</sub>	K <sub>0</sub> { Ca <sub>1</sub> <sup>a</sup> A	1.3238	1.3594	0.7131	1.5424	0.5987	0.2286	1.8645	7.6305	
T 17		K <sub>0</sub> { Ca <sub>2</sub> A	1.3191	2.1020	1.4268	1.9667	1.1096	0.4491	2.9096	11.2829	
T 2		K <sub>1</sub> {	Ca <sub>1</sub> A	0.6133	1.5122	2.0354	0.7301	0.8803	0.6172	2.2761	8.6646
T 18			Ca <sub>2</sub> A	2.5570	2.7393	1.6602	3.0646	1.6445	0.5601	3.9276	16.1533
T 3		K <sub>2</sub> {	Ca <sub>1</sub> A	1.5387	2.0660	1.3700	2.0742	1.3415	0.4481	2.6459	11.4844
T 19			Ca <sub>2</sub> A	1.4192	2.2867	1.6154	2.1122	1.3551	0.4947	2.7934	12.0767
T 4	K <sub>3</sub> {	Ca <sub>1</sub> A	2.2737	2.8251	1.9631	3.0602	1.7142	0.6525	4.1662	16.6550	
T 20		Ca <sub>2</sub> A	2.0846	1.9372	1.6208	2.6092	1.2563	0.5376	2.6553	12.7010	
T 5	Na <sub>1</sub>	K <sub>0</sub> { Ca <sub>1</sub> A	0.8566	1.4136	0.7003	1.1914	0.9545	0.2706	0.6086	5.9956	
T 21		K <sub>0</sub> { Ca <sub>2</sub> A	1.3920	2.1092	1.2557	1.6053	1.2689	0.4730	3.2113	11.3154	
T 6		K <sub>1</sub> {	Ca <sub>1</sub> A	0.9615	1.4819	0.6711	1.4210	0.8997	0.2481	2.2436	7.9269
T 22			Ca <sub>2</sub> A	1.8777	2.4530	1.6428	2.6389	1.6425	0.5197	3.7119	14.4865
T 7		K <sub>2</sub> {	Ca <sub>1</sub> A	1.5035	1.6266	0.9847	2.0021	1.0017	0.3410	2.3976	9.8572
T 23			Ca <sub>2</sub> A	2.2884	2.3712	1.6723	3.2670	1.5577	0.5299	3.8198	15.5063
T 8	K <sub>3</sub> {	Ca <sub>1</sub> A	1.4889	1.3842	0.9442	1.8822	1.0210	0.3080	2.5109	9.5394	
T 24		Ca <sub>2</sub> A	1.6540	2.7163	1.5964	2.8334	1.5968	0.4897	4.0128	14.8994	
T 9	Na <sub>2</sub>	K <sub>0</sub> { Ca <sub>1</sub> A	1.7340	1.8661	0.7732	1.5237	1.1763	0.2894	2.2645	9.6272	
T 25		K <sub>0</sub> { Ca <sub>2</sub> A	1.7728	1.9006	1.0296	2.0139	1.2972	0.3883	2.8474	11.2498	
T 10		K <sub>1</sub> {	Ca <sub>1</sub> A	1.6593	1.8804	1.3083	2.1250	1.1204	0.4269	3.6997	12.4200
T 26			Ca <sub>2</sub> A	1.6547	2.4125	1.6049	2.3969	1.5510	0.5428	4.0127	14.1755
T 11		K <sub>2</sub> {	Ca <sub>1</sub> A	1.2210	1.3717	1.0532	1.6540	1.0048	0.3592	2.2427	8.9066
T 27			Ca <sub>2</sub> A	1.8866	2.5868	1.7203	2.6363	1.0279	0.6042	3.5139	14.5760
T 12	K <sub>3</sub> {	Ca <sub>1</sub> A	1.7411	2.0416	1.5443	2.3205	1.2359	0.5213	2.6652	12.0699	
T 28		Ca <sub>2</sub> A	1.8066	2.0096	1.5333	2.3230	1.0752	0.4054	2.8721	12.0252	
T 13	Na <sub>3</sub>	K <sub>0</sub> { Ca <sub>1</sub> A	1.1684	1.5286	1.0722	1.6373	0.9742	0.4000	2.4705	9.2512	
T 29		K <sub>0</sub> { Ca <sub>2</sub> A	1.5064	1.7616	1.1545	1.8914	1.2290	0.4722	2.7748	10.7899	
T 14		K <sub>1</sub> {	Ca <sub>1</sub> A	1.6928	2.6004	1.7523	1.8539	1.6025	0.5033	3.1345	13.1397
T 30			Ca <sub>2</sub> A	2.0929	2.7855	1.7729	2.9442	1.8196	0.6726	4.2698	16.3575
T 15		K <sub>2</sub> {	Ca <sub>1</sub> A	1.2973	1.8241	1.3868	1.6715	1.1370	0.4217	3.2464	10.9848
T 31			Ca <sub>2</sub> A	2.5623	3.0147	1.8137	2.0262	1.6808	0.6358	3.1841	14.9176
T 16	K <sub>3</sub> {	Ca <sub>1</sub> A	1.3137	1.5814	1.0560	1.3860	0.6575	0.2890	1.4833	7.7669	
T 32		Ca <sub>2</sub> A	1.9330	2.8076	1.6333	2.4432	1.5631	0.4536	3.5494	14.3832	

TABLE 8 Contd.

Dry Weight expressed in grams. Block B

Treatment			Dry Wt. of Lower Leaves	Dry Wt. of Mid Leaves	Dry Wt. of Upper Leaves	Dry Wt. of Lower Stem	Dry Wt. of Mid Stem	Dry Wt. of Upper Stem	Dry Wt. of roots	Total Dry Wt.	
T 1	Na <sub>0</sub>	K <sub>0</sub> { Ca <sub>1</sub> B	0.5116	0.9825	0.7271	0.5690	0.5183	0.2240	1.8133	5.3458	
T 17		K <sub>0</sub> { Ca <sub>2</sub> B	0.8735	1.2223	0.7735	1.3172	0.7701	0.2820	2.4653	7.7039	
T 2		K <sub>1</sub> {	Ca <sub>1</sub> B	1.0614	1.9295	1.1683	2.0714	1.4253	0.4439	2.9249	11.0247
T 18			Ca <sub>2</sub> B	1.3149	1.3166	1.0662	1.6913	0.8571	0.3286	2.4812	9.0559
T 3		K <sub>2</sub> {	Ca <sub>1</sub> B	0.8051	1.1044	0.8783	1.5574	0.8963	0.3582	2.6847	8.2844
T 19			Ca <sub>2</sub> B	0.8315	1.1646	0.9948	1.0774	0.7632	0.4098	2.8062	8.0475
T 4	K <sub>3</sub> {	Ca <sub>1</sub> B	1.0008	1.6498	0.4977	1.2652	0.8880	0.2018	2.3335	7.8368	
T 20		Ca <sub>2</sub> B	1.2267	1.9813	1.5016	1.6793	1.4404	0.5301	3.4642	11.8236	
T 5	Na <sub>1</sub>	K <sub>0</sub> { Ca <sub>1</sub> B	0.7969	1.0279	0.7043	1.0353	0.6402	0.2459	1.8908	6.3413	
T 21		K <sub>0</sub> { Ca <sub>2</sub> B	0.8590	1.4070	0.9042	1.1168	0.8775	0.3012	2.3094	7.7751	
T 6		K <sub>1</sub> {	Ca <sub>1</sub> B	0.9001	0.9328	0.5799	1.0754	0.5059	0.2002	1.8561	6.0504
T 22			Ca <sub>2</sub> B	1.1793	1.4475	1.1370	1.5141	0.9733	0.4094	2.7505	9.4111
T 7		K <sub>2</sub> {	Ca <sub>1</sub> B	1.2025	1.2197	0.7726	1.4397	0.7261	0.2401	3.1065	8.7072
T 23			Ca <sub>2</sub> B	0.9152	1.2790	0.9241	1.1408	0.6166	0.2569	1.9700	7.1027
T 8	K <sub>3</sub> {	Ca <sub>1</sub> B	1.1834	0.9801	0.8463	0.9928	0.4735	0.2547	1.9220	6.6528	
T 24		Ca <sub>2</sub> B	1.4688	1.4229	0.7781	1.8062	0.8178	0.2616	2.2393	8.7947	
T 9	Na <sub>2</sub>	K <sub>0</sub> { Ca <sub>1</sub> B	0.9743	1.3088	0.6392	1.3032	0.7821	0.2135	2.1433	7.3644	
T 25		K <sub>0</sub> { Ca <sub>2</sub> B	0.9903	1.3672	0.9447	1.2611	0.9456	0.3591	3.4783	9.3463	
T 10		K <sub>1</sub> {	Ca <sub>1</sub> B	1.2009	1.4028	0.9624	1.7077	0.9772	0.3173	2.7862	9.
T 26			Ca <sub>2</sub> B	0.7333	1.5827	1.1204	1.0071	1.0430	0.3879	2.8685	8.7429
T 11		K <sub>2</sub> {	Ca <sub>1</sub> B	1.2174	1.2269	1.0015	1.5886	0.7317	0.2875	2.3097	8.3633
T 27			Ca <sub>2</sub> B	1.0593	1.0215	0.8150	1.0047	0.6224	0.2544	2.0500	6.8273
T 12	K <sub>3</sub> {	Ca <sub>1</sub> B	0.9705	1.3548	0.9619	1.3165	0.9014	0.3645	2.7731	8.6427	
T 28		Ca <sub>2</sub> B	1.4643	1.9989	1.6568	2.0828	1.1777	0.5010	2.9822	11.8637	
T 13	Na <sub>3</sub>	K <sub>0</sub> { Ca <sub>1</sub> B	0.8842	0.5930	0.6505	0.7348	0.5718	0.1464	2.0184	5.5991	
T 29		K <sub>0</sub> { Ca <sub>2</sub> B	0.8041	1.4661	0.8613	1.2978	0.9498	0.3793	2.3461	8.1045	
T 14		K <sub>1</sub> {	Ca <sub>1</sub> B	1.1421	1.1787	1.6258	1.5791	0.7404	0.1895	2.3743	7.8299
T 30			Ca <sub>2</sub> B	1.4028	1.4370	1.0993	1.8797	1.1361	0.4130	3.2192	10.5871
T 15		K <sub>2</sub> {	Ca <sub>1</sub> B	1.0595	1.0068	0.8070	1.1828	0.5903	0.2368	2.1085	6.9917
T 31			Ca <sub>2</sub> B	1.0104	1.8695	1.2030	1.6072	1.4867	0.4743	3.4830	11.1341
T 16	K <sub>3</sub> {	Ca <sub>1</sub> B	1.2723	1.7643	1.4310	1.8623	1.0529	0.4072	2.4809	10.2709	
T 32		Ca <sub>2</sub> B	1.0950	1.7307	1.5544	1.0908	0.7576	0.4770	2.0943	8.7998	

TABLE 9  
Log. Total Dry Weights

Treatment		Block A	Block B	
T 1	Na <sub>0</sub>	Ca <sub>1</sub>	0.8826	0.7280
T 17		Ca <sub>2</sub>	1.0522	0.8867
T 2		Ca <sub>1</sub>	0.9378	1.0422
T 18		Ca <sub>2</sub>	1.2081	0.9568
T 3		Ca <sub>1</sub>	1.0599	0.9182
T 19		Ca <sub>2</sub>	1.0816	0.9057
T 4	Na <sub>1</sub>	Ca <sub>1</sub>	1.2214	0.8941
T 20		Ca <sub>2</sub>	1.038	1.0727
T 5		Ca <sub>1</sub>	0.8022	1.5800
T 21		Ca <sub>2</sub>	0.8907	1.9442
T 6		Ca <sub>1</sub>	0.7818	1.6809
T 22		Ca <sub>2</sub>	0.9736	2.1347
T 7	Na <sub>2</sub>	Ca <sub>1</sub>	0.9398	1.9335
T 23		Ca <sub>2</sub>	0.8515	2.0421
T 8		Ca <sub>1</sub>	0.8230	1.8025
T 24		Ca <sub>2</sub>	0.9442	2.1174
T 9		Ca <sub>1</sub>	0.8671	1.8506
T 25		Ca <sub>2</sub>	0.9706	2.0217
T 10	Na <sub>3</sub>	Ca <sub>1</sub>	0.9710	2.0651
T 26		Ca <sub>2</sub>	0.9416	2.0932
T 11		Ca <sub>1</sub>	0.9224	1.8721
T 27		Ca <sub>2</sub>	0.8342	1.9980
T 12		Ca <sub>1</sub>	0.9366	2.0182
T 28		Ca <sub>2</sub>	1.0742	2.1544
T 13	Na <sub>3</sub>	Ca <sub>1</sub>	0.7481	1.7142
T 29		Ca <sub>2</sub>	0.9088	1.9419
T 14		Ca <sub>1</sub>	0.8938	2.0124
T 30		Ca <sub>2</sub>	1.0249	2.2387
T 15		Ca <sub>1</sub>	0.8446	1.8853
T 31		Ca <sub>2</sub>	1.0464	2.2202
T 16	Na <sub>3</sub>	Ca <sub>1</sub>	1.0115	1.9018
T 32		Ca <sub>2</sub>	0.9445	2.1022

Leaf Composition of Lemon Seedlings grown in Experimental Nutrient Solutions, expressed as a percentage of total Dry Weight

Treatment			Sodium			Potassium			Calcium			Magnesium		
			Lower	Middle	Upper	Lower	Middle	Upper	Lower	Middle	Upper	Lower	Middle	Upper
T1	K <sub>0</sub>	Ca <sub>1</sub>	0.13	0.03	0.03	1.06	0.45	3.03	1.97	0.43	0.11	0.16	0.21	0.22
T 17		Ca <sub>2</sub>	0.23	0.04	0.03	1.16	0.52	0.75	2.26	0.88	0.55	0.13	0.21	0.18
T 2		K <sub>1</sub>	Ca <sub>1</sub>	0.10	0.02	0.03	1.37	0.96	1.93	2.33	0.62	0.13	0.20	0.21
T 18	Ca <sub>2</sub>		0.19	0.04	0.02	1.20	0.76	1.31	2.27	1.26	0.40	0.16	0.13	0.16
T 3	K <sub>2</sub>	Ca <sub>1</sub>	0.17	0.06	0.05	1.49	1.24	2.56	2.05	0.26	0.06	0.10	0.15	0.11
T 19		Ca <sub>2</sub>	0.85	0.37	0.04	1.95	1.86	1.91	2.16	0.87	0.42	0.14	0.23	0.18
T 4		K <sub>3</sub>	Ca <sub>1</sub>	0.14	0.04	0.04	1.87	1.46	3.17	1.82	0.10	0.05	0.07	0.12
T 20	Ca <sub>2</sub>		0.23	0.29	0.07	1.91	2.52	2.26	2.19	1.18	0.44	0.11	0.17	0.17
T 5	K <sub>0</sub>	Ca <sub>1</sub>	0.15	0.14	0.31	0.68	0.85	0.99	2.46	0.45	0.08	0.17	0.18	0.15
T 21		Ca <sub>2</sub>	0.19	0.26	0.12	0.83	0.95	0.96	2.49	1.18	0.42	0.16	0.24	0.16
T 6		K <sub>1</sub>	Ca <sub>1</sub>	0.15	0.15	0.23	1.50	1.33	1.54	2.12	0.56	0.10	0.17	0.21
T 22	Ca <sub>2</sub>		0.23	0.23	0.09	0.94	1.14	1.52	2.17	1.02	0.45	0.12	0.19	0.17
T 7	K <sub>2</sub>	Ca <sub>1</sub>	0.16	0.13	0.14	1.77	1.77	2.28	1.99	0.32	0.05	0.11	0.19	0.14
T 23		Ca <sub>2</sub>	0.29	0.21	0.15	2.32	1.83	1.86	2.12	1.70	0.47	0.13	0.22	0.18
T 8		K <sub>3</sub>	Ca <sub>1</sub>	0.08	0.16	0.19	2.37	1.26	2.98	1.38	1.64	0.07	0.14	0.15
T 24	Ca <sub>2</sub>		0.17	0.12	0.22	2.56	2.09	2.41	2.10	1.59	0.28	0.09	0.14	0.16
T 9	K <sub>0</sub>	Ca <sub>1</sub>	0.24	0.30	0.35	0.69	2.14	0.87	1.05	0.28	0.05	0.14	0.16	0.22
T 25		Ca <sub>2</sub>	0.38	0.20	0.27	1.06	0.19	1.04	2.37	0.93	0.43	0.14	0.20	0.18
T 10		K <sub>1</sub>	Ca <sub>1</sub>	0.26	0.31	0.37	0.74	1.02	1.78	1.84	0.18	0.06	0.13	0.15
T 26	Ca <sub>2</sub>		0.47	0.30	0.25	1.63	0.39	1.46	2.71	1.19	0.49	0.11	0.20	0.19
T 11	K <sub>2</sub>	Ca <sub>1</sub>	0.18	0.31	0.38	0.98	1.46	2.20	1.67	0.21	0.07	0.11	0.14	0.15
T 27		Ca <sub>2</sub>	0.34	0.30	0.28	1.59	0.84	1.00	2.10	1.31	0.27	0.09	0.14	0.13
T 12		K <sub>3</sub>	Ca <sub>1</sub>	0.22	0.32	0.31	1.45	2.06	2.19	1.90	0.41	0.06	0.04	0.08
T 28	Ca <sub>2</sub>		0.31	0.36	0.21	2.32	1.73	2.00	1.96	1.62	0.42	0.08	0.13	0.15
T 13	K <sub>0</sub>	Ca <sub>1</sub>	0.52	0.62	0.82	0.67	0.61	1.08	1.74	0.29	0.06	0.11	0.13	0.11
T 29		Ca <sub>2</sub>	0.32	0.68	0.58	0.38	0.36	0.66	1.27	1.15	0.18	0.09	0.13	0.11
T 14		K <sub>1</sub>	Ca <sub>1</sub>	0.32	0.61	0.56	0.86	0.98	1.71	1.99	0.14	0.05	0.09	0.10
T 30	Ca <sub>2</sub>		0.37	0.68	0.44	0.43	0.60	1.35	1.95	1.24	0.20	0.06	0.08	0.15
T 15	K <sub>2</sub>	Ca <sub>1</sub>	0.60	0.81	0.66	1.28	1.32	1.85	1.80	0.25	0.05	0.08	0.11	0.14
T 31		Ca <sub>2</sub>	0.41	0.74	0.46	0.90	1.34	1.97	2.14	1.20	0.38	0.06	0.15	0.14
T 16		K <sub>3</sub>	Ca <sub>1</sub>	0.45	0.64	0.72	1.16	1.46	2.65	1.96	0.08	0.07	0.04	0.10
T 32	Ca <sub>2</sub>		0.40	0.58	0.40	1.32	1.76	1.81	1.94	1.50	0.26	0.07	0.11	0.13

Stem Composition of Lemon Seedlings grown in Experimental Nutrient Solutions, expressed as a percentage of Total Dry Weight

Treatment		Sodium			Potassium			Calcium			Magnesium			
		Lower	Middle	Upper	Lower	Middle	Upper	Lower	Middle	Upper	Lower	Middle	Upper	
7	K <sub>0</sub>	Ca <sub>1</sub>	0.01	0.02	0.03	0.13	0.13	0.14	0.07	0.05	0.06	0.05	0.06	-
		Ca <sub>2</sub>	0.02	0.23	0.03	0.14	0.13	0.17	0.18	0.16	0.16	0.04	0.03	-
	K <sub>1</sub>	Ca <sub>1</sub>	0.02	0.01	0.02	0.11	0.15	0.26	0.13	0.04	0.05	0.03	0.03	0.05
		Ca <sub>2</sub>	0.02	0.27	0.04	0.18	0.32	0.49	0.16	0.18	0.17	0.01	0.02	-
	K <sub>2</sub>	Ca <sub>1</sub>	0.01	0.03	0.02	0.15	0.15	0.20	0.07	0.03	0.03	0.03	0.04	0.05
		Ca <sub>2</sub>	0.02	0.25	0.10	0.38	0.34	0.39	0.18	0.16	0.20	0.03	0.02	0.03
K <sub>3</sub>	Ca <sub>1</sub>	0.01	0.21	0.02	0.38	0.34	0.25	0.05	0.04	0.04	0.02	0.05	0.08	
	Ca <sub>2</sub>	0.05	0.34	0.14	0.45	0.54	1.47	0.24	0.19	0.23	0.03	0.03	0.05	
1	K <sub>0</sub>	Ca <sub>1</sub>	0.03	0.04	0.07	0.16	0.13	0.10	0.04	0.05	0.04	0.07	0.05	-
		Ca <sub>2</sub>	0.03	0.30	0.15	0.14	0.11	0.39	0.19	0.13	0.13	0.03	0.03	-
	K <sub>1</sub>	Ca <sub>1</sub>	0.04	0.06	0.11	0.20	0.34	0.20	0.10	0.05	0.06	0.06	0.05	-
		Ca <sub>2</sub>	0.04	0.26	0.13	0.35	0.27	0.77	0.21	0.15	0.18	0.03	0.02	-
	K <sub>2</sub>	Ca <sub>1</sub>	0.05	0.08	0.15	0.16	0.11	0.26	0.06	0.04	0.05	0.03	0.05	-
		Ca <sub>2</sub>	0.03	0.14	0.08	0.38	0.25	0.14	0.27	0.19	0.32	0.03	0.03	0.07
K <sub>3</sub>	Ca <sub>1</sub>	0.06	0.05	0.10	0.24	0.32	0.48	0.06	0.04	0.05	0.04	0.05	-	
	Ca <sub>2</sub>	0.05	0.06	0.12	0.52	0.38	1.11	0.26	0.14	0.20	0.03	0.06	-	
5	K <sub>0</sub>	Ca <sub>1</sub>	0.07	0.14	0.27	0.14	0.17	0.31	0.05	0.03	0.06	0.05	0.03	-
		Ca <sub>2</sub>	0.06	0.03	0.08	0.14	0.12	0.10	0.15	0.12	0.09	0.04	0.03	-
	K <sub>1</sub>	Ca <sub>1</sub>	0.07	0.17	0.15	0.16	0.40	0.29	0.05	0.05	0.04	0.04	0.04	0.06
		Ca <sub>2</sub>	0.09	0.03	0.09	0.19	0.30	0.18	0.23	0.18	0.15	0.02	0.03	0.02
	K <sub>2</sub>	Ca <sub>1</sub>	0.07	0.08	0.18	0.34	0.39	0.36	0.10	0.05	0.05	0.06	0.04	0.05
		Ca <sub>2</sub>	0.07	0.05	0.10	0.50	1.17	0.23	0.24	0.16	0.07	0.03	0.03	-
K <sub>3</sub>	Ca <sub>1</sub>	0.15	0.15	0.16	0.57	0.65	0.67	0.09	0.04	0.04	0.05	0.04	0.07	
	Ca <sub>2</sub>	0.08	0.03	0.14	0.64	1.74	0.58	0.29	0.26	0.16	0.04	0.03	0.02	
13	K <sub>0</sub>	Ca <sub>1</sub>	0.13	0.23	0.24	0.12	0.16	0.13	0.08	0.03	0.05	1.07	0.05	-
		Ca <sub>2</sub>	0.16	0.06	0.25	0.17	0.90	0.11	0.28	0.25	0.21	0.04	0.04	0.07
	K <sub>1</sub>	Ca <sub>1</sub>	0.20	0.22	0.35	0.18	0.30	0.27	0.11	0.04	0.06	0.06	0.04	-
		Ca <sub>2</sub>	0.15	0.03	0.07	0.28	1.15	0.15	0.27	0.17	0.09	0.04	0.03	0.16
	K <sub>2</sub>	Ca <sub>1</sub>	0.23	0.28	0.25	0.37	0.29	0.23	0.10	0.05	0.05	0.06	0.06	-
		Ca <sub>2</sub>	0.20	0.03	0.07	0.35	0.86	0.13	0.29	0.19	0.10	0.04	0.03	0.06
K <sub>3</sub>	Ca <sub>1</sub>	0.14	0.21	0.32	0.42	0.30	0.62	0.08	0.04	0.04	0.05	0.05	-	
	Ca <sub>2</sub>	0.17	0.03	0.31	0.41	1.12	0.12	0.24	0.14	0.03	0.05	0.03	0.07	

large number of lateral shoots were produced in the axils of the lower leaves, compensating for the tissue that had died. No relationship between the occurrence of this disorder and the experimental treatments supplied could be established and its cause remained unknown.

#### Roots

The health and the condition of the roots of the experimental lemon seedlings appeared sound irrespective of the treatments they received.

#### Height

The lemon seedlings grown in nutrient solutions with the higher levels of calcium were found to be larger plants than those receiving the lower level of calcium supply, see Figs. 17 and 18. Increasing the concentrations of sodium and potassium in the nutrient supply appeared to have little or no effect on the height of the seedlings, except at the higher concentrations of both these ions, when a slight increase in size was produced, see Table 7.

#### Dry Weights

The dry weights were determined as described in Chapter II and the results are found in Table 8.

It appeared from the figures obtained for the dry weights of the experimental material, that the increase in weight brought about as a result of experimental treatments, was not a straightforward addition to the pre-treatment weight, but rather of a proportional type of increase. Further upon statistically examining the dry weights it was found that the ratio, Standard Error (natural variation of plants) : Mean, in the two blocks appeared to be consistent, i.e. Standard Error had not been independent of treatment. A log scale was therefore used, and found to be appropriate for estimating the effects of the different treatments, see Table 9.

Increasing the sodium level in the nutrient supply, for the experimental period, produced no apparent effect upon the weight of the plant. However, raising the potassium level in the external medium, produced an increase in the total weight of the plants,

TABLE 12

Composition of Lemon Seedling Roots grown in Experimental Nutrient Solutions, expressed as a percentage of the total dry weight.

Treatment		Sodium	Potassium	Calcium	Magnesium	
T 1	K <sub>0</sub> {	Ca <sub>1</sub>	0.03	0.16	0.11	0.04
T 17		Ca <sub>2</sub>	0.02	0.14	0.55	0.03
T 2	K <sub>1</sub> {	Ca <sub>1</sub>	0.03	0.25	0.12	0.04
T 18		Ca <sub>2</sub>	0.02	0.41	0.16	0.06
T 3	K <sub>2</sub> {	Ca <sub>1</sub>	0.03	0.31	0.07	0.03
T 19		Ca <sub>2</sub>	0.02	0.29	0.36	0.04
T 4	K <sub>3</sub> {	Ca <sub>1</sub>	0.05	0.38	0.07	0.04
T 20		Ca <sub>2</sub>	0.02	0.30	0.37	0.05
T 5	K <sub>0</sub> {	Ca <sub>1</sub>	0.04	0.10	0.15	0.04
T 21		Ca <sub>2</sub>	0.02	0.12	0.30	0.03
T 6	K <sub>1</sub> {	Ca <sub>1</sub>	0.04	0.21	0.07	0.12
T 22		Ca <sub>2</sub>	0.02	0.24	0.17	0.05
T 7	K <sub>2</sub> {	Ca <sub>1</sub>	0.05	0.35	0.13	0.08
T 23		Ca <sub>2</sub>	0.03	0.20	0.44	0.04
T 8	K <sub>3</sub> {	Ca <sub>1</sub>	0.03	0.49	0.13	0.06
T 24		Ca <sub>2</sub>	0.03	0.45	0.96	0.07
T 9	K <sub>0</sub> {	Ca <sub>1</sub>	0.06	0.22	0.12	0.04
T 25		Ca <sub>2</sub>	0.05	0.16	0.47	0.01
T 10	K <sub>1</sub> {	Ca <sub>1</sub>	0.05	0.23	0.07	0.05
T 26		Ca <sub>2</sub>	0.06	0.34	0.91	0.06
T 11	K <sub>2</sub> {	Ca <sub>1</sub>	0.05	0.31	0.10	0.04
T 27		Ca <sub>2</sub>	0.04	0.47	0.38	0.02
T 12	K <sub>3</sub> {	Ca <sub>1</sub>	0.00	0.37	0.11	0.05
T 28		Ca <sub>2</sub>	0.03	0.54	0.78	0.04
T 13	K <sub>0</sub> {	Ca <sub>1</sub>	0.07	0.13	0.07	0.05
T 29		Ca <sub>2</sub>	0.06	0.16	0.22	0.01
T 14	K <sub>1</sub> {	Ca <sub>1</sub>	0.06	0.19	0.07	0.05
T 30		Ca <sub>2</sub>	0.06	0.47	0.18	0.04
T 15	K <sub>2</sub> {	Ca <sub>1</sub>	0.08	0.32	0.11	0.06
T 31		Ca <sub>2</sub>	0.07	0.39	0.26	0.03
T 16	K <sub>3</sub> {	Ca <sub>1</sub>	0.06	0.38	0.72	0.04
T 32		Ca <sub>2</sub>	0.07	0.53	0.19	0.02

significant at the 0.1% level, see Table 13.

Table 13  
Statistical Analysis of Log Dry Weight

	D.F	Sum of Squares	Mean Sum of Squares	F Value	
Blocks	1	0.32691	0.32691	54.035	***
Na Treatment	3	0.02888	0.00963	1.592	n.s
K Treatment	3	0.13239	0.04413	7.294	***
Ca Treatment	1	0.16764	0.16764	27.709	***
Na x K Treatments	9	0.04514	0.00502	0.830	n.s
K x Ca Treatments	3	0.00929	0.00310	0.512	n.s
Na x Ca Treatments	3	0.02444	0.00815	1.347	n.s
Na x K x Ca Treatments	9	0.02742	0.00305	0.504	n.s
Residue	31	0.18761	0.00605		
Total	63	0.94972			

\*\*\* significant at 0.1% level, n.s not significant

It can be seen from Fig. 19 that the plant weight was increased by raising the external supply of potassium up to 117.0 p.p.m. but beyond this level no further increase in plant weight was brought about.

Increasing the external supply of calcium from 180.0 p.p.m. to 360.0 p.p.m. produced an increase in plant dry weight, significant at the 0.1% level.

The difference in weight between plants in Block A and plants in Block B, significant at 0.1% level, was due to the selection of experimental material for each Block, on a basis of size.

#### Chemical Composition of the Plant Material

In the analysis of the plant material, the concentration of each element was found to vary considerably between the different plant organs. In the cases of stems and leaves, variation in the concentration of the elements was found to exist with distance from the root.

Sodium was found to have its highest concentration in the roots, and lower concentrations in the stems and leaves. Potassium

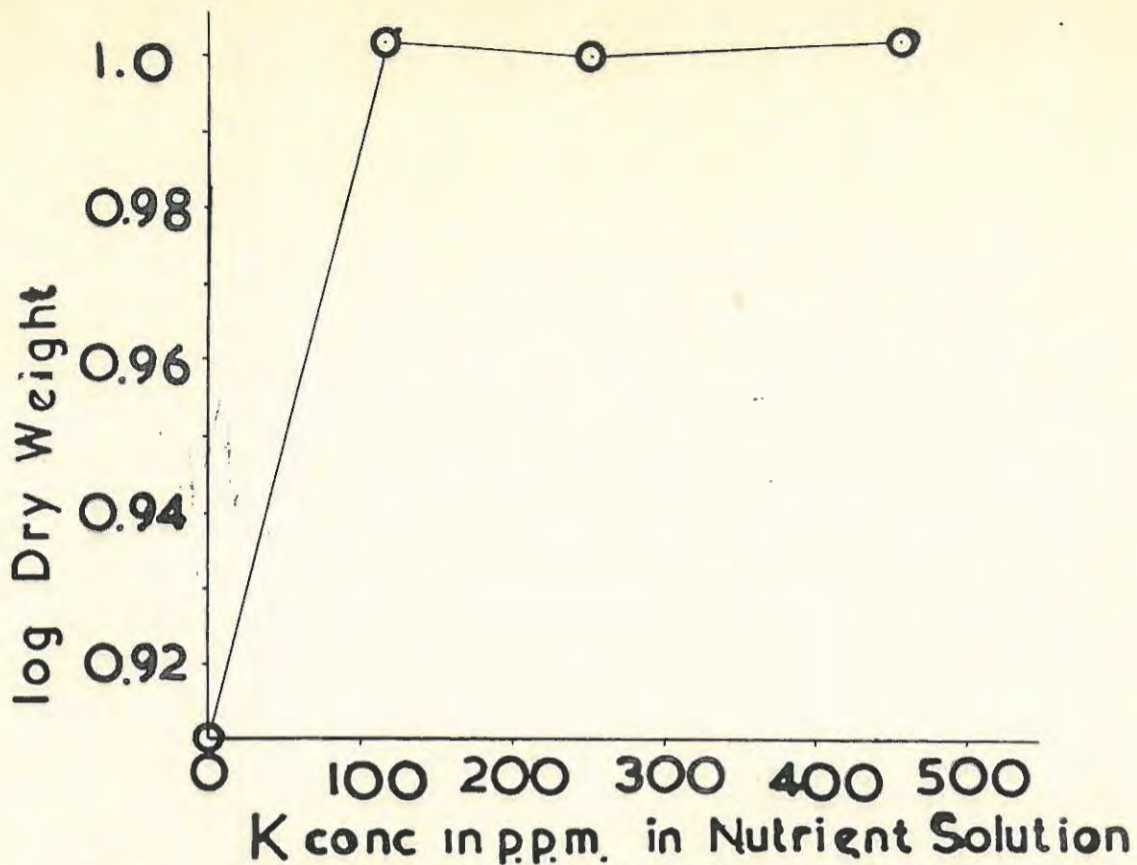


Fig. 19 The effect of Potassium concentration on log dry weight.

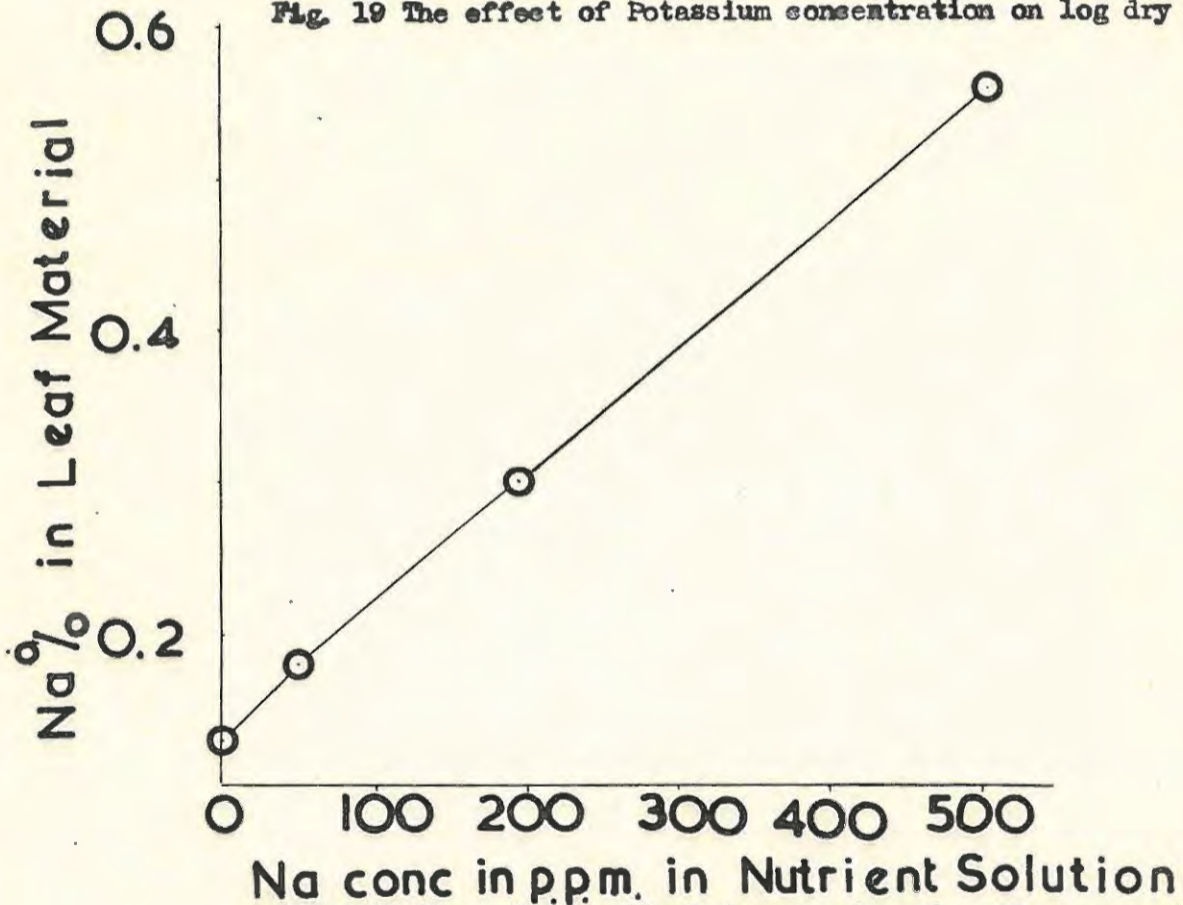


Fig. 20. The relationship between external sodium concentration and leaf sodium concentration.

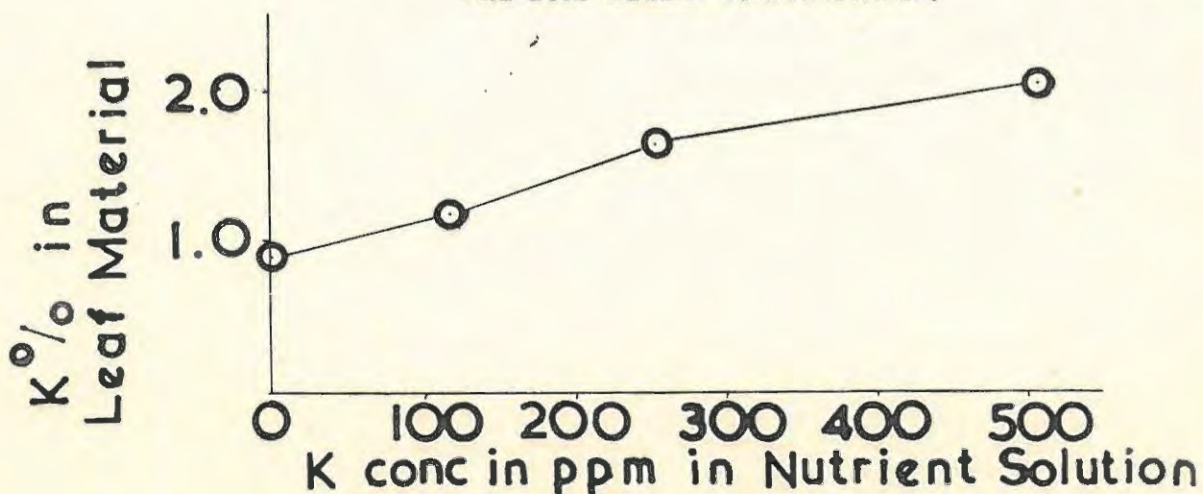


Fig. 21. The effect of external potassium concentration on the leaf potassium concentration.

had its greatest concentration in the leaves, the stem and roots containing about a quarter of that found in the leaves. Similarly the leaves contained higher concentrations of calcium and magnesium than either stems or roots.

The analysis of the roots showed the presence of a higher concentration of sodium, than of calcium, potassium or magnesium. However in the leaves the concentration of potassium was five times that of sodium, and ten times that of magnesium. Similarly in the stem, the concentration of potassium was the highest, being three times as great as that of sodium or calcium, and nine times as great as that of magnesium.

#### Leaf Chemical Composition

The chemical composition of the leaves of seedlings receiving various treatments can be found in Table 10.

#### (a) Sodium

The sodium leaf content ranged between 0.02 and 0.96%. Increasing the sodium level in the nutrient media, resulted in an increase in the percentage sodium concentration of the leaf, see Table 14.

Table 14

#### Statistical Analysis of Treatment Effect on Sodium Leaf Content

	D.F	Sum of Squares	Mean Sum of Squares	F Value	
Position	2	0.025253	0.012626	1.945	n.s
Na Treatment	3	2.597568	0.865856	133.065	***
K Treatment	3	0.068962	0.022987	3.533	*
Ca Treatment	1	0.005386	0.005386	0.828	n.s
Position x Na Treatment	6	0.424652	0.070775	10.874	***
Position x K Treatment	6	0.060617	0.010103	1.553	n.s
Position x Ca Treatment	2	0.161879	0.080940	12.439	***
Na x K Treatments	9	0.130264	0.014474	2.224	n.s
Na x Ca Treatments	3	0.167255	0.055752	8.568	***
K x Ca Treatments	3	0.026435	0.008812	1.354	n.s
Position x Na x K Treatments	18	0.119189	0.006622	1.018	n.s
Position x Na x Ca Treatments	6	0.040242	0.006707	1.031	n.s
Position x K x Ca Treatments	6	0.012821	0.002137	0.328	n.s
Na x K x Ca Treatments	9	0.087531	0.009726	1.495	n.s
Residue	18	0.117124	0.006507		
Total	95	4.045178			

\*\*\* significant at 0.1% level, \*\* significant at 1.0% level, \* significant at 5.0% level, n.s. not significant.

From the results obtained in the present investigation, an almost linear relationship was found between the leaf sodium content and the sodium concentration in the nutrient solution, see Fig. 20. Further, the leaf position on the central axis influenced the effect of sodium treatment on the leaf sodium concentration. The lemon seedlings receiving no sodium in the treatment were found to accumulate higher sodium concentrations in the lower leaves than in the upper leaves, whereas plants receiving 505.0 p.p.m. sodium accumulated high sodium concentrations in the apical leaves, which decreased towards the basal leaves.

The effect of the level of calcium supply in the external media on the sodium leaf concentration was related to the position of the leaf on the central axis. The lemon seedlings receiving 180 p.p.m. calcium were found to have their greatest sodium concentration in the apical leaves, with the sodium concentration decreasing progressively in the lower leaves. The plants receiving the higher level of calcium supply, 360 p.p.m., were found to have their highest concentration of sodium in the lower leaves, with a slight decrease in concentration in the middle leaves, and a proportionally larger decrease in the sodium concentration of the uppermost leaves. The sodium concentration in the lower leaves of plants receiving the higher calcium level was greater than that found in corresponding leaves of lemon seedlings receiving the lower calcium supply.

Increasing the potassium concentration in the external nutrient supply had only a very slight effect on the leaf sodium concentration, significant at the 5% level. The general tendency was for the leaf sodium concentration to decrease slightly with an increase in the potassium supply to the nutrient solution.

(b) Potassium

The leaf potassium concentration varied between 0.22 - 3.35%. The concentration of potassium present in the leaf varied with the position of the leaf on the central axis, the apical leaves having a higher concentration than the basal leaves. Increase in potassium concentration in the leaves from the basal to the apical position occurred irrespective of the calcium level of supply. However, the range in potassium concentration between the two extreme leaf positions

on the main axis was not as great in plants receiving 360.0 p.p.m. calcium, as in plants receiving 180.0 p.p.m. calcium, see Fig. 23.

Increasing the sodium level in the nutrient solutions decreased the leaf potassium content, significant at the 0.1% level, see Table 15.

Table 15

Statistical Analysis of Treatment Effect on Potassium Leaf Content

	D.F.	Sum of squares	Mean sum of squares	F Value	
Position	2	5.66824	2.83412	26.273	***
Na Treatment	3	3.09272	1.03091	9.557	***
K Treatment	3	17.6125	5.87083	54.425	***
Ca Treatment	1	0.57528	0.57528	5.333	*
Na x K Treatments	9	1.10052	0.12228	1.134	n.s
K x Ca Treatments	3	0.59377	0.19792	1.835	n.s
Na x Ca Treatments	3	0.21546	0.07182	0.666	n.s
Position x Na Treatment	6	1.41643	0.23607	2.188	n.s
Position x K Treatment	6	0.20957	0.03493	0.324	n.s
Position x Ca Treatment	2	1.97338	0.98669	9.147	**
Position x Na x K Treatments	18	1.99298	0.11072	1.026	n.s
Position x K x Ca Treatments	6	0.86592	0.14432	1.338	n.s
Position x Na x Ca Treatments	6	3.47935	0.57989	5.376	**
Na x K x Ca Treatments	9	0.72002	0.08000	0.742	n.s
Residue	18	1.94174	0.10787		
Total	95	41.45788			

\*\*\* significant at 0.1% level, \*\* significant at 1.0% level, \* significant at 5.0% level and n.s not significant.

Concentrations of sodium in the nutrient solution up to 50.0 p.p.m. had very little effect upon the potassium leaf concentrations, but concentrations above this level brought about a decrease in the concentration of the leaf potassium.

Increasing the potassium supply in the nutrient media, resulted in a higher potassium leaf level, significant at the 0.1% level, see Table 15.

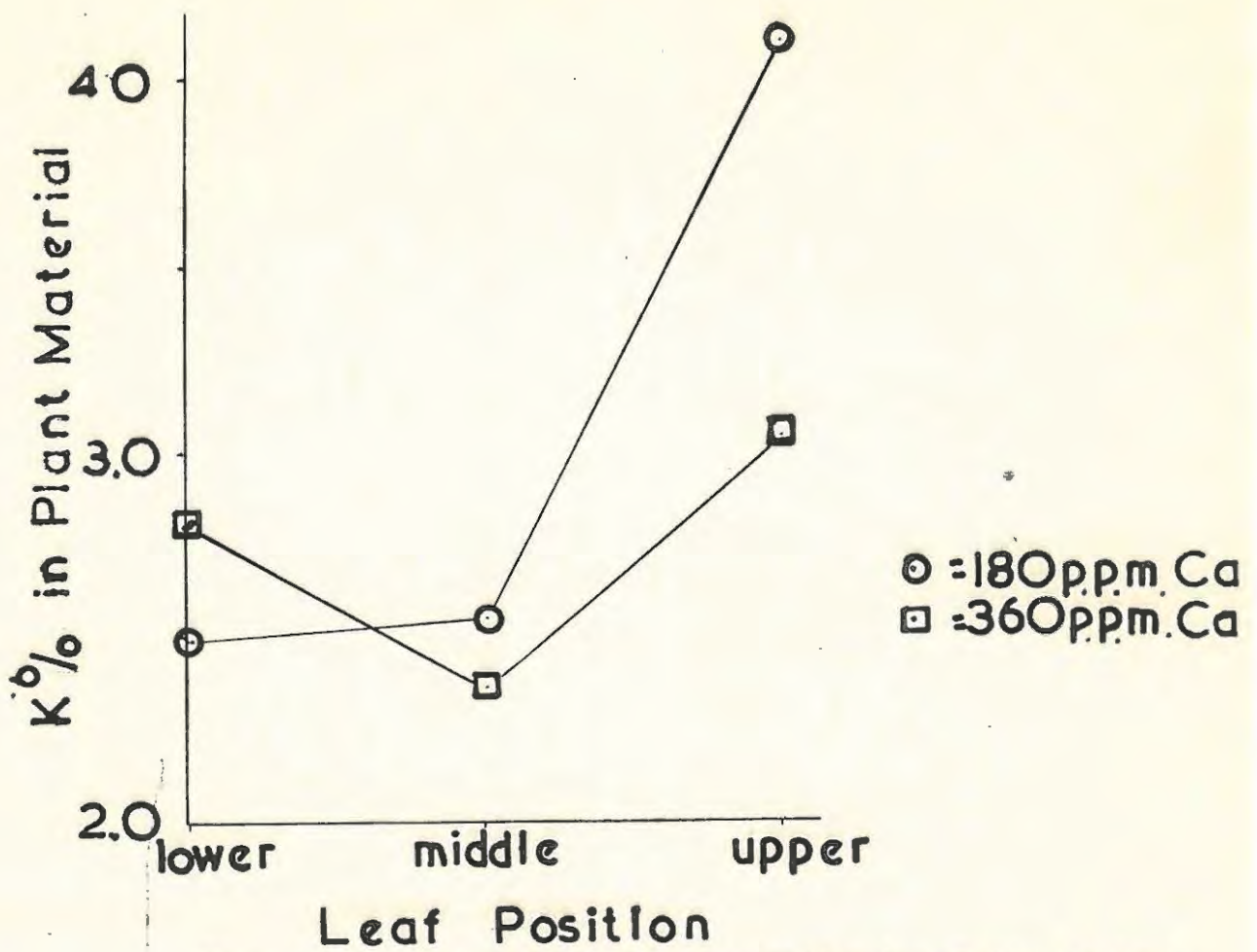


Fig. 23. The potassium leaf concentration in relation to leaf position and level of calcium supply.

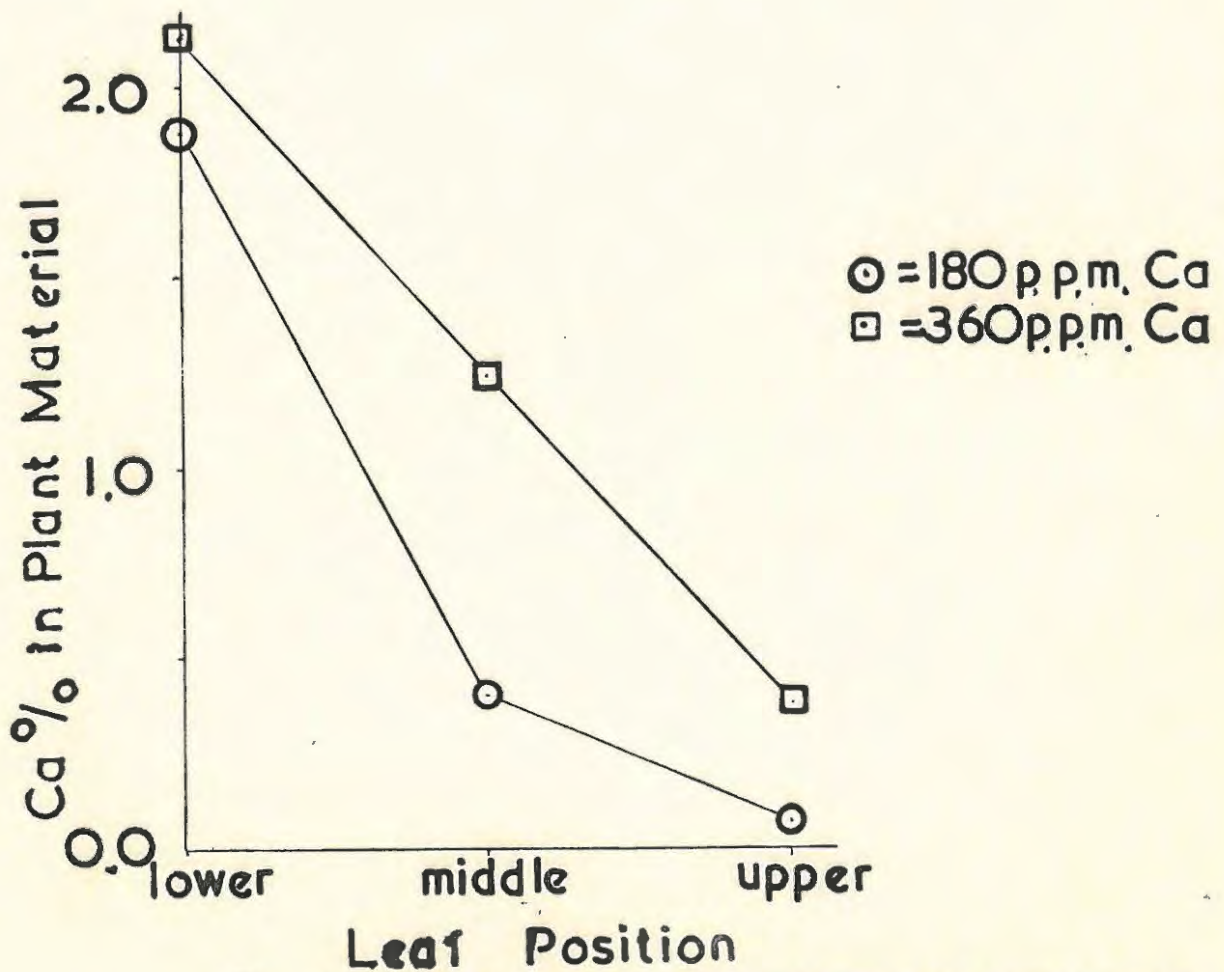


Fig. 24. The calcium leaf concentration in relation to leaf position and level of calcium supply.

An almost linear relationship was found to exist between the increase in the leaf potassium concentration and the increase in the external potassium concentration, see Fig. 21.

Increasing the calcium level in the nutrient supply from 180 p.p.m. to 360 p.p.m. produced a slight decrease in the potassium concentration in the leaf, significant at the 5.0% level.

(c) Calcium

The calcium concentration in the leaves of the lemon seedlings varied between 0.05 - 2.36%. The calcium concentration varied with position on the main axis, see Table 16, the lower leaves having the highest concentration and the apical leaves having the lowest. The variation in concentration from the lower to the apical leaves, was as pronounced for the seedlings receiving 180.0 p.p.m. calcium as for those receiving 360.0 p.p.m. calcium in the nutrient media, even though their respective internal calcium concentration differed, see Fig. 24.

Table 16  
Statistical Analysis of Treatments on Calcium Leaf Content

	D.F.	Sum of squares	Mean sum of squares	F Value	
Position	2	52.88051	26.44026	245.819	***
Na Treatment	3	0.62375	0.20792	1.933	n.s
K Treatment	3	0.14351	0.04784	0.445	n.s
Ca Treatment	1	5.36671	5.36671	48.895	***
Na x K Treatments	9	0.40747	0.04527	0.421	n.s
K x Ca Treatments	3	0.04259	0.01420	0.132	n.s
Ca x Na Treatments	3	0.31263	0.10421	0.969	n.s
Position x Na Treatment	6	0.50748	0.08458	0.786	n.s
Position x K Treatment	6	0.64691	0.10782	1.002	n.s
Position x Ca Treatment	2	1.73052	0.86526	8.044	**
Position x Na x K Treatments	18	0.60412	0.03356	0.312	n.s
Position x K x Ca Treatments	6	0.09702	0.01617	0.150	n.s
Position x Na x Ca Treatments	6	0.55747	0.09291	0.864	n.s
Na x K x C Treatments	9	0.37876	0.04208	0.391	n.s
Residue	18	1.93609	0.10756		
Total	95	66.23554			

\*\*\* significant at 0.1% level, \*\* at 1.0% level, \* significant at 5.0% and n.s. not significant.

Increasing the calcium supply in the external nutrient solution brought about a corresponding increase in the calcium leaf concentration significant at the 0.1% level. The increase in the calcium leaf concentration was almost of the same order as the increase in calcium concentration in the nutrient media.

(d) Magnesium

The leaf magnesium concentration varied between 0.04 - 0.25%. The leaf position on the central axis influenced the magnesium leaf composition, see Table 17. The highest concentration of magnesium being found in the basal leaves and decreasing progressively towards the apical leaves. The concentration in the lower leaves was twice as great as that in the upper leaves.

Table 17

Statistical Analysis of Treatments on Magnesium Leaf Content

	D.F.	Sum of squares	Mean sum of squares		
Position	2	0.037244	0.018622	68.970	***
Na Treatment	3	0.063200	0.021067	78.026	***
K Treatment	3	0.025585	0.008528	31.585	***
Ca Treatment	1	0.000595	0.000595	2.204	n.s
Na x K Treatments	9	0.007504	0.000834	3.089	*
K x Ca Treatments	3	0.005466	0.001822	6.748	**
Na x Ca Treatments	3	0.001428	0.000476	1.763	n.s
Position x Na Treatment	6	0.001543	0.000257	0.952	n.s
Position x K Treatment	6	0.007759	0.001294	4.793	**
Position x Ca Treatment	2	0.002634	0.001317	4.878	**
Position x Na x K Treatments	18	0.009466	0.000526	1.948	n.s
Position x K x Ca Treatments	6	0.001517	0.000253	0.937	n.s
Position x Na x Ca Treatments	6	0.001146	0.000191	0.517	n.s
Na x K x Ca Treatments	9	0.013807	0.001534	5.681	***
Residue	18	0.004853	0.000270		
Total	95	0.183749			

\*\*\* significant at 0.1% level, \*\* significant at 1.0% level, \* significant at 5.0% level and n.s not significant.

The leaf magnesium concentration decreased as the sodium concentration in the external media was raised. Concentrations of sodium up to 50.0 p.p.m. in the nutrient solution had little or no effect on the magnesium leaf concentration, but at higher sodium levels a decrease in leaf magnesium was found, see Fig. 25. Increasing the potassium concentration in the nutrient supply reduced the leaf magnesium concentration but had less influence on the leaf magnesium concentration than corresponding sodium levels of supply, see Fig. 26.

#### Stem Chemical Composition

A summary of the chemical composition of the stems from seedlings receiving experimental treatment can be found in Table 11.

##### (a) Sodium

The sodium concentration in the stem varied between 0.01 - 0.35%. The position of the stem on the main axis influenced the sodium concentration, higher concentrations being found in the apical and middle portions of the stem than in the lower portions, see Table 18.

Increasing the concentration of sodium in the external supply raised the sodium concentration in the stem, significant at 0.1% level, see Table 18. The level of calcium present in the external supply was found to affect the degree to which the sodium in the stem tissue increased, with raising the concentration of sodium in the external supply. Increasing the external supply of sodium to lemon seedlings receiving 180.0 p.p.m. calcium produced proportionally large increases in the internal stem sodium content, whereas plants receiving 360.0 p.p.m. calcium showed only very slight changes in internal stem sodium content, see Fig. 27.

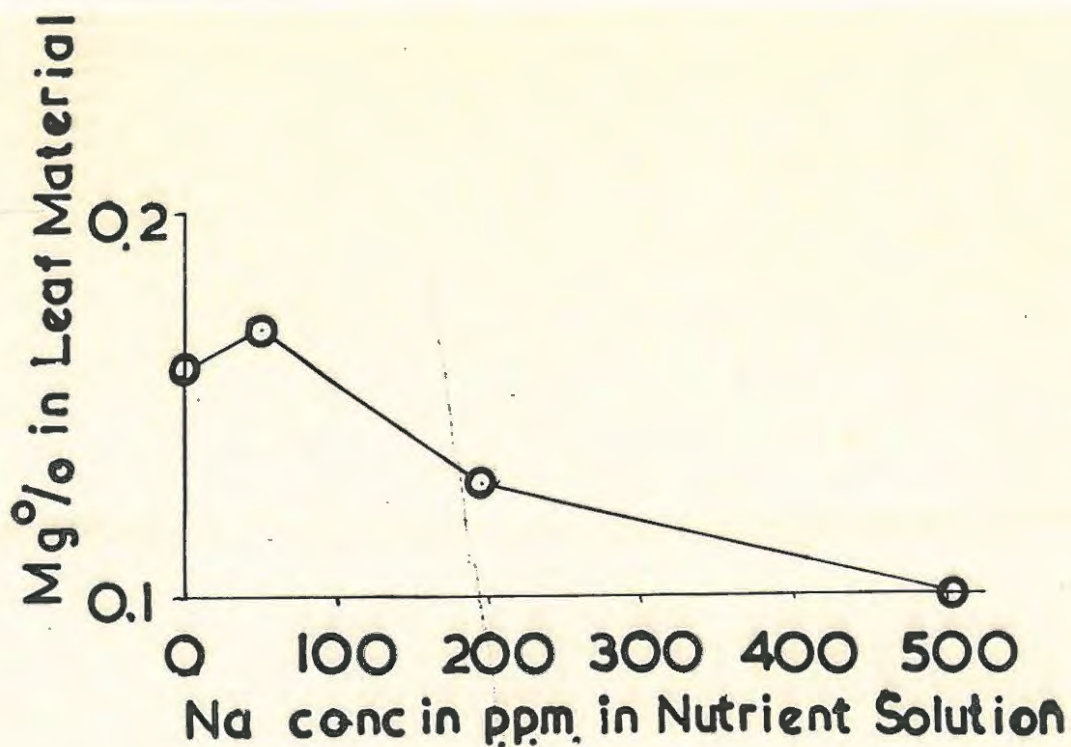


Fig. 25. The effect of the external sodium concentration on the leaf magnesium concentration.

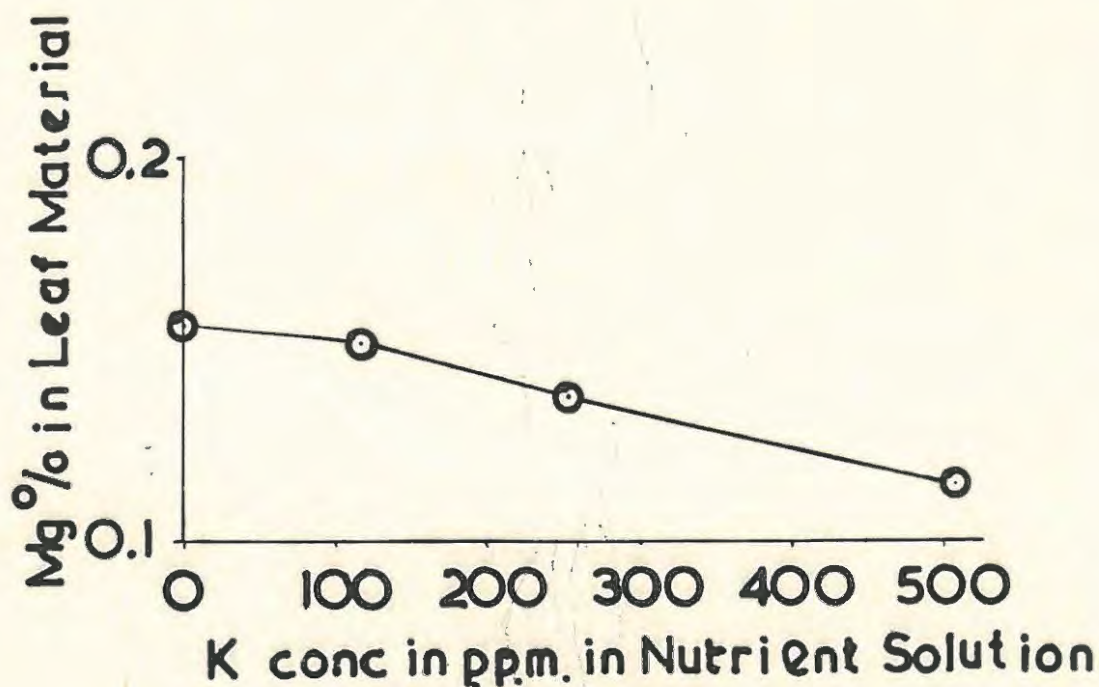


Fig. 26. The effect of the external potassium concentration on the leaf magnesium concentration.

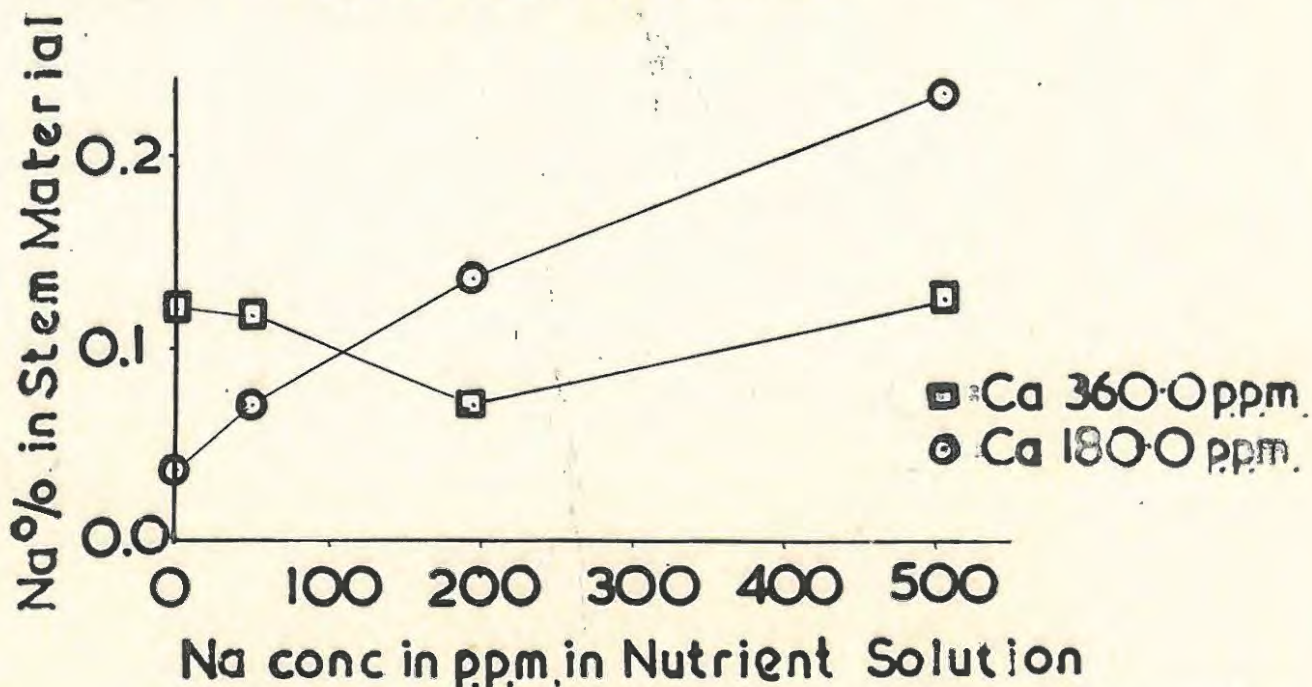


Fig. 27. The effect of the external calcium and sodium concentrations on the stem sodium concentration.

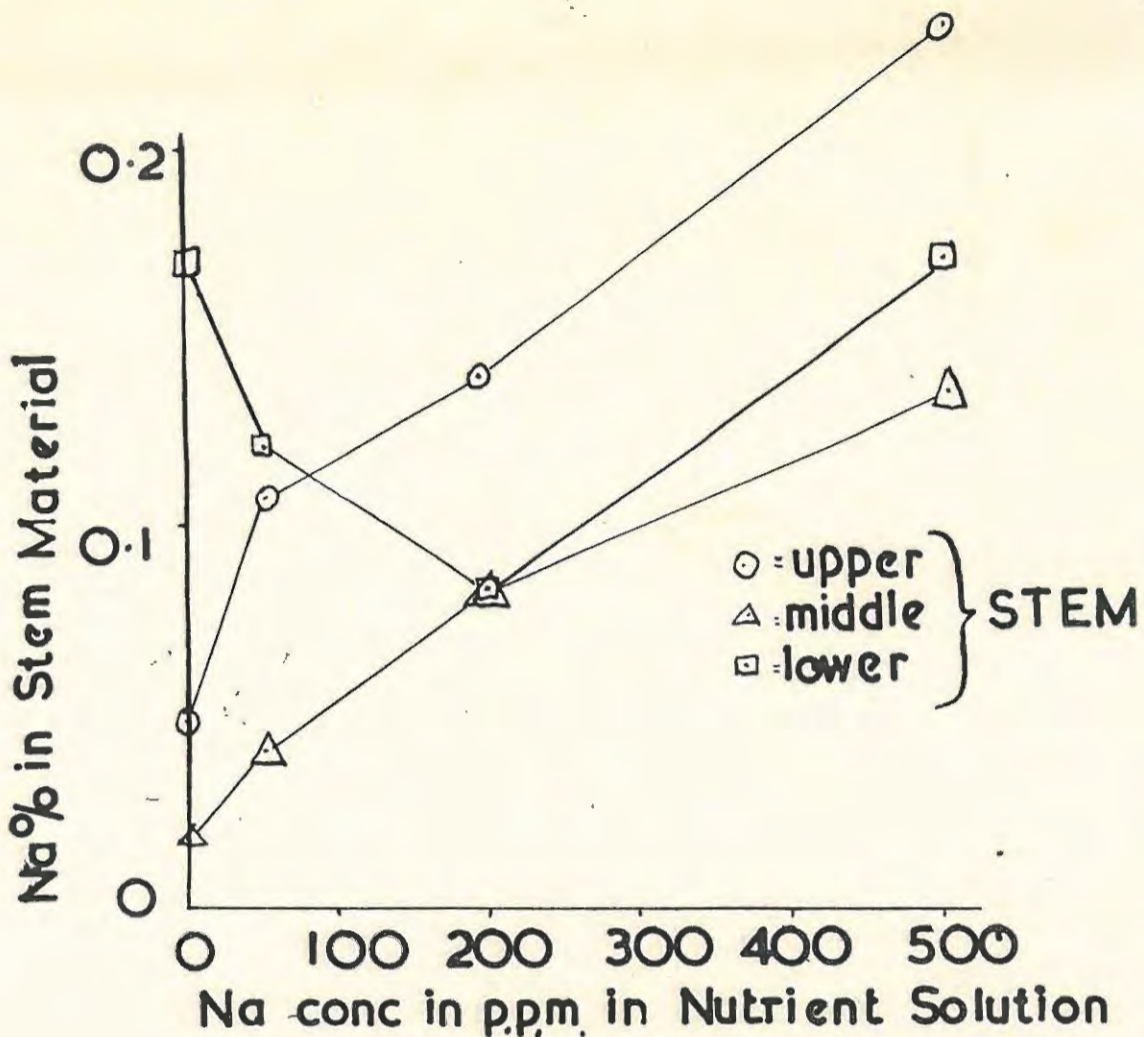


Fig. 28. The effect of the external sodium concentration, and stem position on the stem sodium concentration.

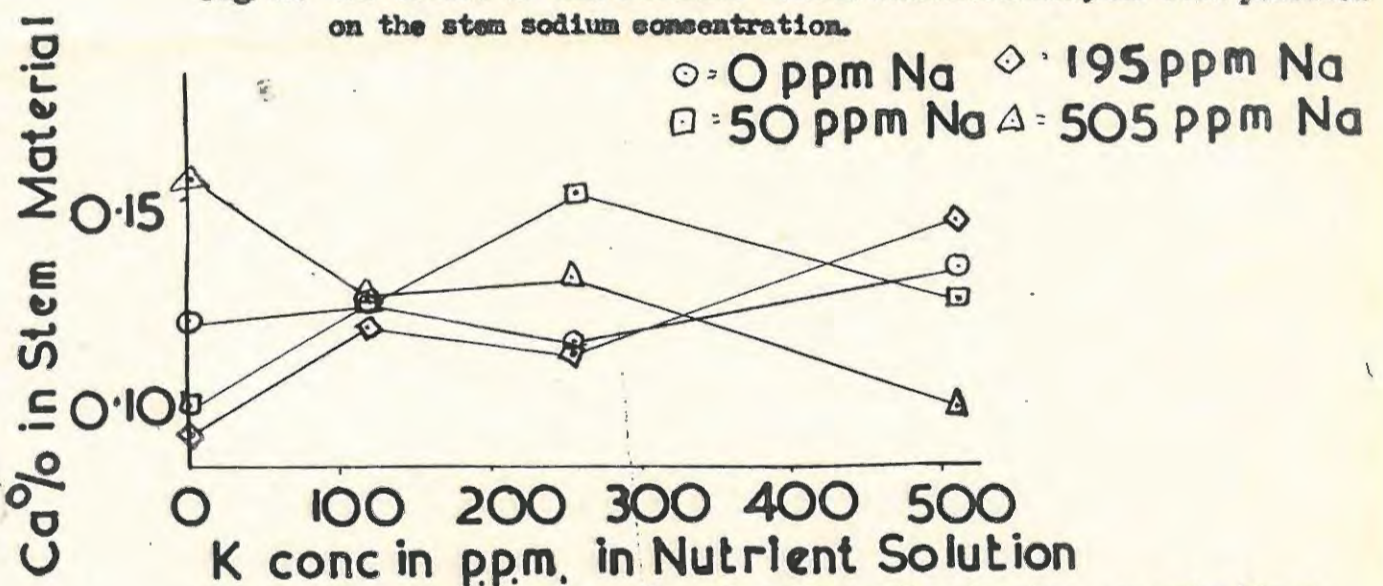


Fig. 29. The effect of the external sodium and potassium concentrations on the stem calcium concentration.

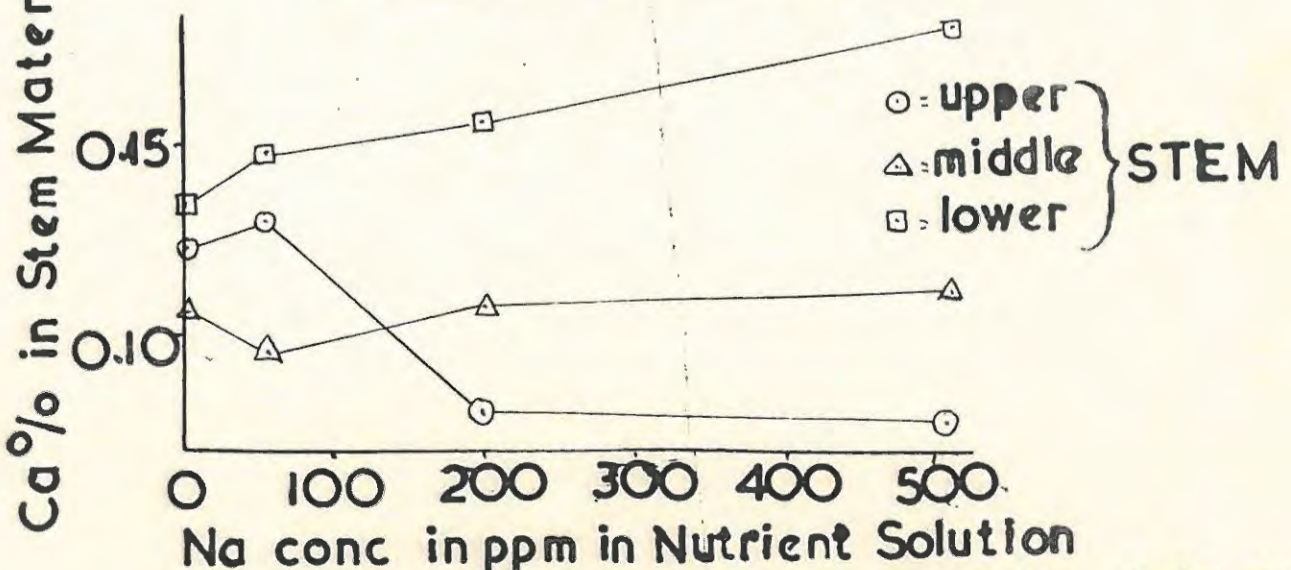


Fig. 30. The effect of the external sodium concentration and stem position on the stem calcium concentration.

Table 3)

## Statistical Analysis of Treatments on Sodium Stem Content

	D.F.	Sum of squares	Mean Sum of squares	F Value	
Position	2	0.0584381	0.0292191	21.024	***
Na Treatment	3	0.1456647	0.0485549	34.937	***
K Treatment	3	0.0074270	0.0024757	1.781	n.s.
Ca Treatment	1	0.0021822	0.0021822	1.570	n.s.
Na x K Treatments	9	0.0215818	0.0023980	1.725	n.s.
K x Ca Treatments	3	0.0036193	0.0012061	0.868	n.s.
Na x Ca Treatments	3	0.1551698	0.0517233	37.216	***
Position x Na Treatment	6	0.1292696	0.0215449	15.502	***
Position x K Treatment	6	0.0076858	0.0012810	0.922	n.s.
Position x Ca Treatment	2	0.0085150	0.0042575	3.063	n.s.
Position x Na x K Treatments	18	0.0547820	0.0030434	2.190	n.s.
Position x K x Ca Treatments	6	0.0173002	0.0028834	2.075	n.s.
Position x Na x Ca Treatments	6	0.1001129	0.0166855	12.006	***
Na x K x Ca Treatments	9	0.0350458	0.0038940	2.802	*
Residue	18	0.0250170	0.0013898		
Total	95	0.7718110			

\*\*\* significant at 0.1% level, \*\* significant at 1.0% level  
\* significant at 5.0% level, n.s. not significant.

The arbitrary three stem positions differed in their responses to an increase in the external sodium supply. The upper and lower stem portions followed similar trends, in having increased internal sodium concentrations with increased external supply, but the middle portion's sodium content slightly decreased with increases of sodium level in the nutrient supply, see Fig. 28.

(b) Potassium

The potassium concentrations in the stem varied between 0.10 - 1.74%. The stem potassium content was found to vary with the position on the central axis, the upper stem portions having greater potassium concentrations than the lower portions.

Increases in the concentration of potassium in the external medium raised the stem potassium content, significant at the 0.1% level, see Table 1).

Table 1)  
Statistical Analysis of Treatments on Potassium Stem Content

	D.F.	Sum of Square	Mean Sum of Square	F. Value	
Position	2	0.383961	0.191981	5.269	*
K Treatment	3	2.156657	0.718886	19.730	***
Na Treatment	3	0.249955	0.083318	2.287	n.s
Ca Treatment	1	0.765855	0.765855	21.019	***
Na x K Treatment	9	0.550517	0.061169	1.679	n.s
K x Ca Treatment	3	0.198532	0.066178	1.816	n.s
Na x Ca Treatment	3	0.033324	0.011108	0.305	n.s
Position x Na Treatment	6	1.148799	0.191467	5.255	**
Position x K Treatment	6	0.213254	0.035542	0.975	n.s
Position x Ca Treatment	2	0.305250	0.152625	4.189	*
Position x Na x K Treatments	18	0.458541	0.025476	0.699	n.s
Position x K x Ca Treatments	6	0.149258	0.024876	0.683	n.s
Position x Na x Ca Treatments	6	1.191866	0.198644	5.452	**
Na x K x Ca Treatments	9	0.307202	0.034134	0.937	n.s
Residue	18	0.655870	0.036437		
Total	95	8.768871			

\*\*\* significant at 0.1% level, \*\* significant at 1.0% level, \* significant at 5.0% level, n.s. not significant

The level of calcium in the external solution influenced the stem potassium concentration. The lemon seedlings receiving the higher level of calcium supply had larger stem potassium concentrations than the plants receiving the lower levels of calcium supply.

The effect of increasing the sodium level in the nutrient solution upon the stem potassium concentration varied with sampling position on the stem. The potassium concentration in the upper stem portions decreased with increases in the sodium supply, the potassium in the lower stem portions however increased only slightly, while

the middle portions of the stem showed considerable increase in potassium concentration.

(c) Calcium

The stem calcium values varied between 0.03 - 0.29%. Considerable difference was found in the stem calcium concentration with sampling position on the stem, the greatest concentration being in the lower portions, with a decrease towards the apex, significant at 0.1% level, see Table 20.

A fourfold increase in the internal calcium concentration resulted from raising the calcium supply in the external media from 180 p.p.m. to 360.0 p.p.m. Raising the calcium concentration in the external supply increased the calcium concentration of the lower stem portions to a slightly larger degree, than the middle and upper stem portions, see Table 20.

Table 20

Statistical Analysis of Effect of Treatments on Calcium Stem Content

	D.F	Sum of squares	Mean Sum of squares	F Value	
Position	2	0.0535631	0.0267816	61.638	***
K Treatment	3	0.0021065	0.0007022	1.616	n.s
Na Treatment	3	0.0014264	0.0004755	1.094	n.s
Ca Treatment	1	0.4150798	0.4150798	955.304	***
Na x K Treatments	9	0.0274821	0.0030536	7.028	***
K x Ca Treatments	3	0.0053354	0.0017785	4.093	*
Na x Ca Treatments	3	0.0013085	0.0004362	1.004	n.s
Position x Na Treatment	6	0.0270440	0.0045073	10.374	***
Position x K Treatment	6	0.0022103	0.0003684	0.848	n.s
Position x Ca Treatment	2	0.0086508	0.0043254	9.955	**
Position x Na x K Treatments	18	0.0099424	0.0005524	1.271	n.s
Position x K x Ca Treatments	6	0.0025430	0.0004238	0.975	n.s
Position x Na x Ca Treatments	6	0.0219297	0.0036550	8.412	***
Na x K x Ca Treatments	9	0.0297611	0.0033068	7.611	***
Residue	18	0.0078206	0.0004345		
Total	95	0.6162037			

\*\*\* significant at 0.1% level, \*\* significant at 1.0% level,  
\* significant at 5.0% level, n.s not significant.

Increasing the potassium in the external supply increased the stem calcium concentration, with the exception of the lemon seedlings receiving 505.0 p.p.m. sodium. At the highest level of sodium there was a decrease in the stem calcium content upon the addition of potassium to the external medium, see Fig. 29.

The effect of increasing the sodium concentration in the external media on the stem calcium concentration varied with the position on the main axis. The calcium concentrations in the upper and middle portions of the stem were found to decrease with an increase in external sodium. In the lower stem portion however the calcium concentration rose with increased sodium supply, see Fig. 30.

(d) Magnesium

The magnesium concentration of some of the upper stem portions were not determined due to an insufficiency of plant material for chemical analysis. The magnesium concentration in the stem portion was found to vary between 0.01 - 0.06%

Increasing the sodium concentration in the nutrient media increased the stem magnesium concentration, see Fig. 31. A decrease in stem magnesium concentration was produced when the calcium concentration in the external media was raised from 180.0 p.p.m. to 360.0 p.p.m., significant at 0.1% level, see Table 21.

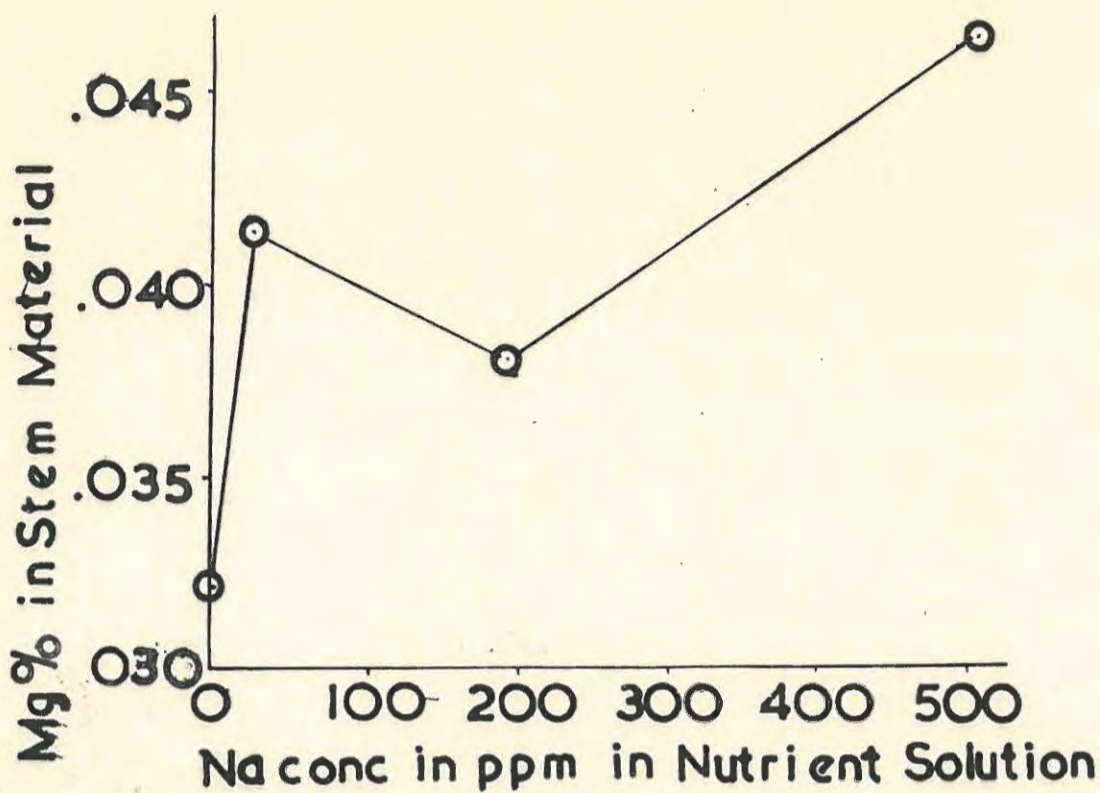


Fig. 31. The effect of the external sodium concentration on the stem magnesium content.

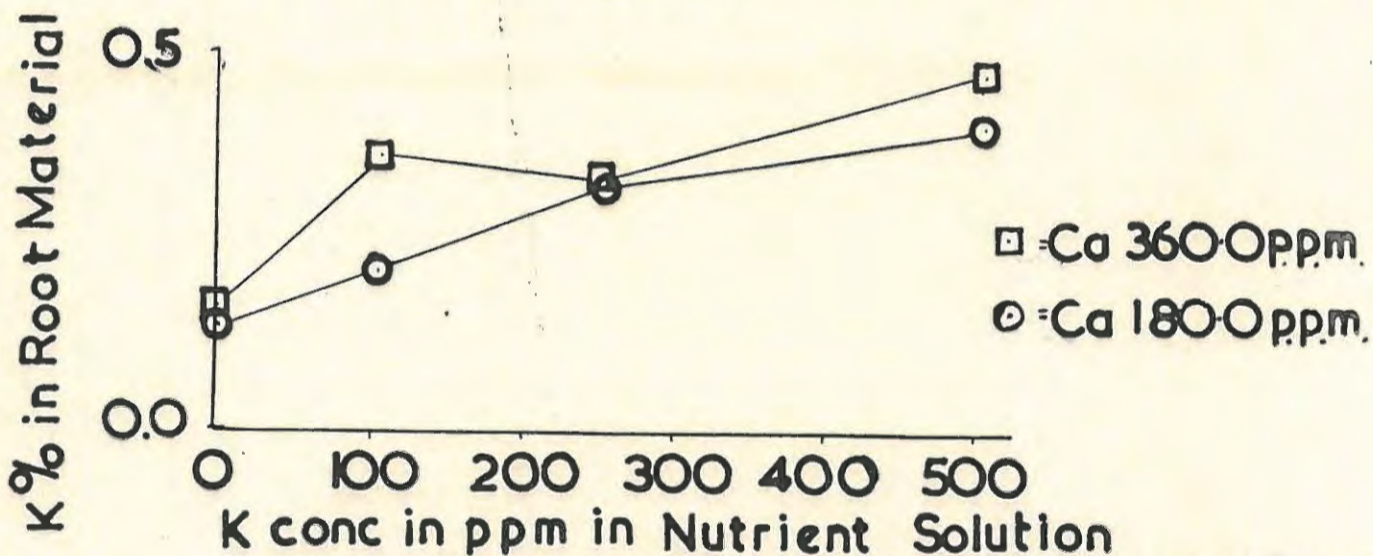


Fig. 32. The effect of the external potassium and calcium concentrations on the root potassium content.

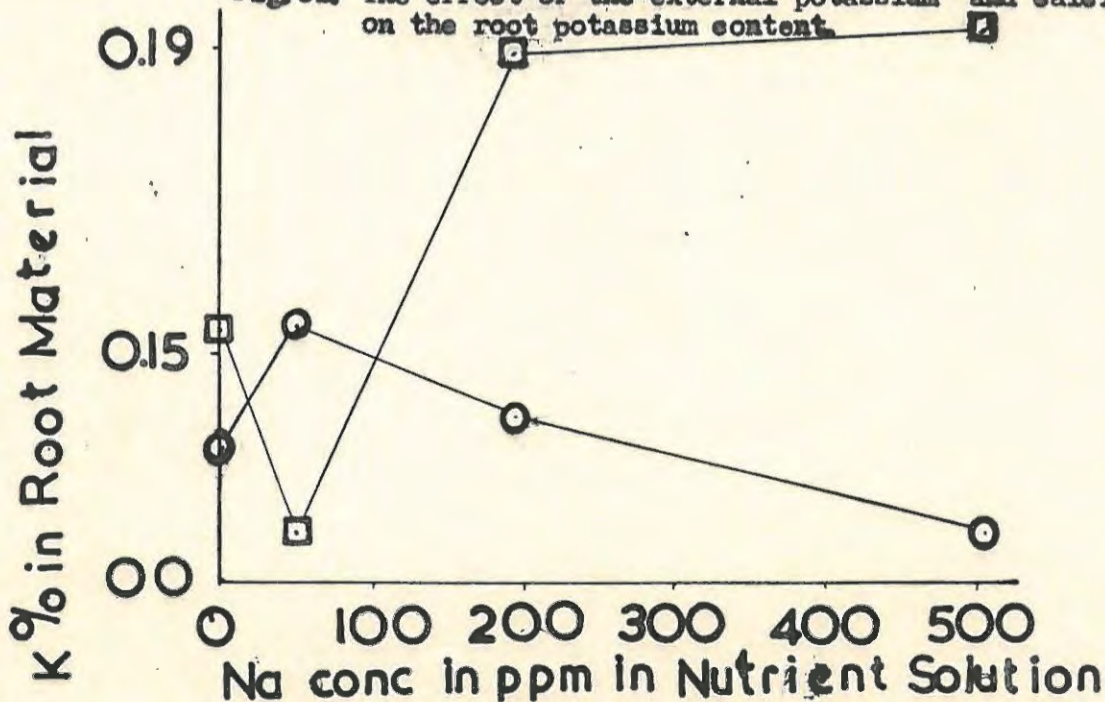


Fig. 33. The effect of the external sodium and calcium concentrations on the root potassium content.

Table 21

Statistical Analysis of the Effect of Treatments on  
Magnesium Stem Content

	D.F	Sum of squares	Mean Sum of squares	F Value	
Position	1	0.00011396	0.00011396	3.213	n.s
Na Treatment	3	0.00175901	0.00058634	16.531	***
K Treatment	3	0.00094687	0.00031562	8.898	**
Ca Treatment	1	0.00356409	0.00356409	100.482	***
Na x K Treatments	9	0.00080316	0.00008924	2.516	n.s
K x Ca Treatments	3	0.00045882	0.00015294	4.312	*
Na x Ca Treatments	3	0.00013943	0.00004648	1.310	n.s
Na x K x Ca Treatments	9	0.00091034	0.00010115	2.852	n.s
Position x Na Treatment	3	0.00066117	0.00022039	6.213	*
Position x K Treatment	3	0.00048080	0.00016027	4.518	*
Position x Ca Treatment	1	0.00004726	0.00004726	1.322	n.s
Position x Na x K Treatments	9	0.00036747	0.00004053	1.143	n.s
Position x Na x Ca Treatments	3	0.00034124	0.00011375	3.207	n.s
Position x K x Ca Treatments	3	0.00009911	0.00003304	0.093	n.s
Residue	9	0.00031919	0.00003547		
Total	63	0.01101192			

\*\*\* significant at 0.1% level, \*\* significant at 1.0. level,  
\* significant at 5.0% level, n.s not significant.

Root Chemical Composition

The chemical composition of the roots receiving the various treatments can be found in Table 12.

(a) Sodium

The sodium concentration in the root varied between 0.12 - 1.08%. Increases in the sodium concentration in the external media raised the sodium concentration in the roots, see Table 22.

Table 22

Statistical Analysis of Treatment Effect on SodiumRoot Content

	D.F	Sum of squares	Mean Sum of squares	F Value	
Blocks	1	0.0153	0.0153	0.680	n.s
Na Treatment	3	2.0046	0.6682	29.680	***
K Treatment	3	0.0565	0.0188	0.836	n.s
Ca Treatment	1	0.3271	0.3271	14.538	***
Na x K Treatment	9	0.2742	0.0305	1.356	n.s
K x Ca Treatment	3	0.1179	0.0393	1.747	n.s
Na x Ca Treatment	3	0.0033	0.0011	0.049	n.s
Na x K x Ca Treatment	9	0.1407	0.0156	0.693	n.s
Residue	31	0.6973	0.0225		
Total	63	3.6369			

\*\*\* significant at 0.1% level, n.s. not significant

The sodium concentration in the roots of the lemon seedlings was lowered when the calcium concentration in the external media was increased from 180.0 p.p.m. to 360.0 p.p.m., significant at 0.1% level.

(b) Potassium

The potassium concentration of the roots varied between 0.12 - 0.54%. Increasing the concentration of potassium in the external nutrient supply, resulted in higher concentrations of potassium in the lemon seedling roots, see Table 23.

The experimental plants receiving 360.0 p.p.m. calcium in the external nutrient solution had a slightly higher response to increases in the potassium external supply, than the seedlings receiving 180.0 p.p.m. calcium, see Fig. 32.

Table 23

Statistical Analysis of Treatment Effect on PotassiumRoot Content

	D.F	Sum of squares	Mean Sum of squares	F Value	
Blocks	1	0.023378	0.023378	8.157	**
Na Treatment	3	0.026812	0.008937	3.116	*
K Treatment	3	0.641794	0.213931	74.592	***
Ca Treatment	1	0.041769	0.041769	14.5364	***
Na x K Treatments <sup>2</sup>	3	0.067982	0.022661	7.901	***
Na x Ca Treatments	3	0.080671	0.026890	9.376	***
Na x K x Ca Treatments	9	0.039725	0.004414	1.539	*
Residue	31	0.088901	0.002868		
Total	63	1.070719			

\*\*\* significant at 0.1% level, \*\* significant at 1.0% level.  
\* significant at 5.0% level.

The effect of raising the sodium concentration in the external media on the potassium root concentration varied with the level of calcium in the external solution. The potassium concentration of the lemon seedlings receiving 180.0 p.p.m. calcium decreased slightly with increasing sodium supply, whereas the seedlings receiving 360.0 p.p.m. calcium increased their potassium concentration with increased sodium supply, see Fig. 33.

(c) Calcium

The calcium concentration of the roots of the experimental seedlings varied between 0.14 - 0.54%.

The effect of increasing the sodium concentration in the external supply on the root calcium content varied with the level of calcium supply, see Table 23. The calcium concentration of the seedlings receiving 180.0 p.p.m. calcium, decreased slightly with an increase in sodium supply. The lemon seedlings receiving 360.0 p.p.m. calcium in the nutrient media, increased their root calcium content when the sodium supply was raised to 195.0 p.p.m. At a still higher level of sodium supply, the calcium content decreased considerably, see Fig. 34.

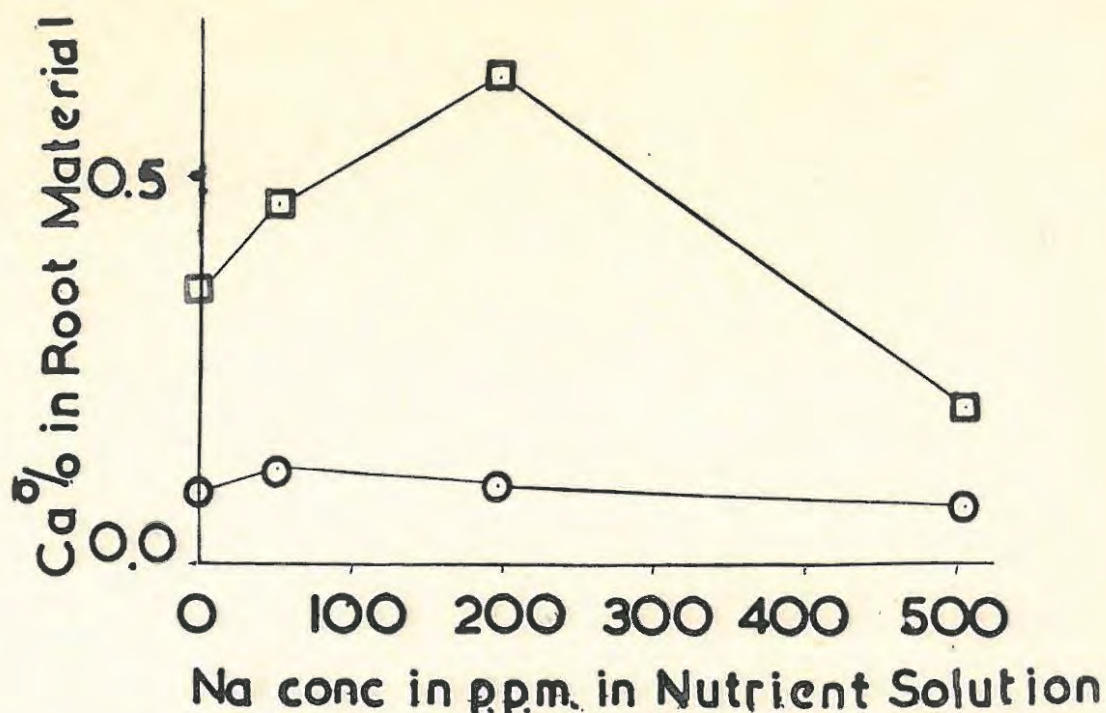


Fig. 34. The effect of the external sodium and calcium concentrations on the root calcium content.

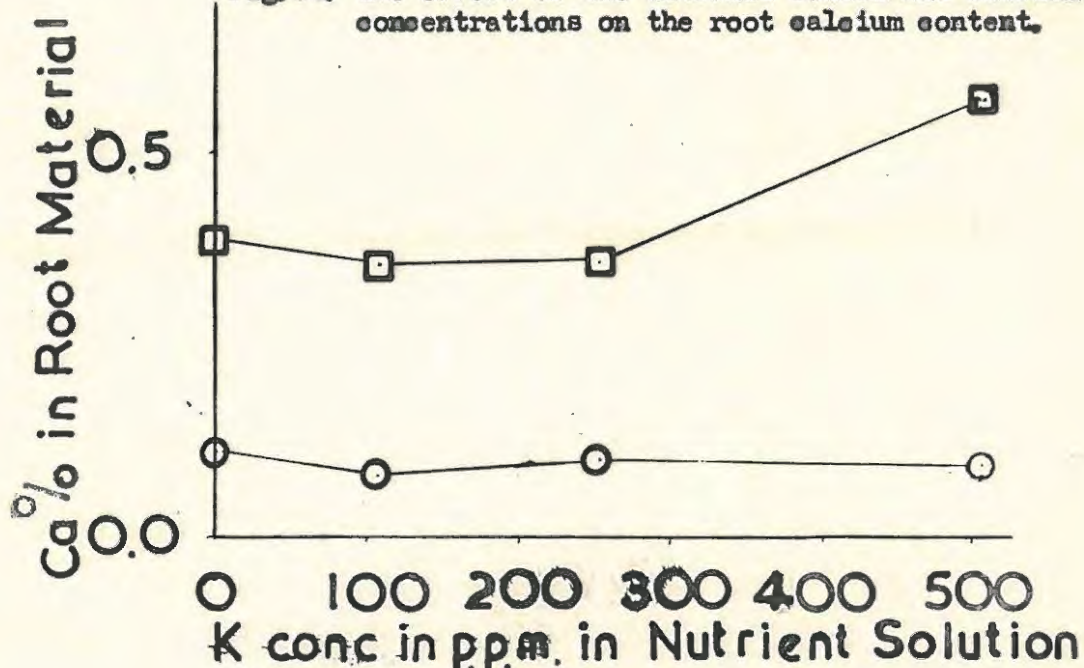


Fig. 35. The effect of the external potassium and calcium concentrations on the root calcium content.

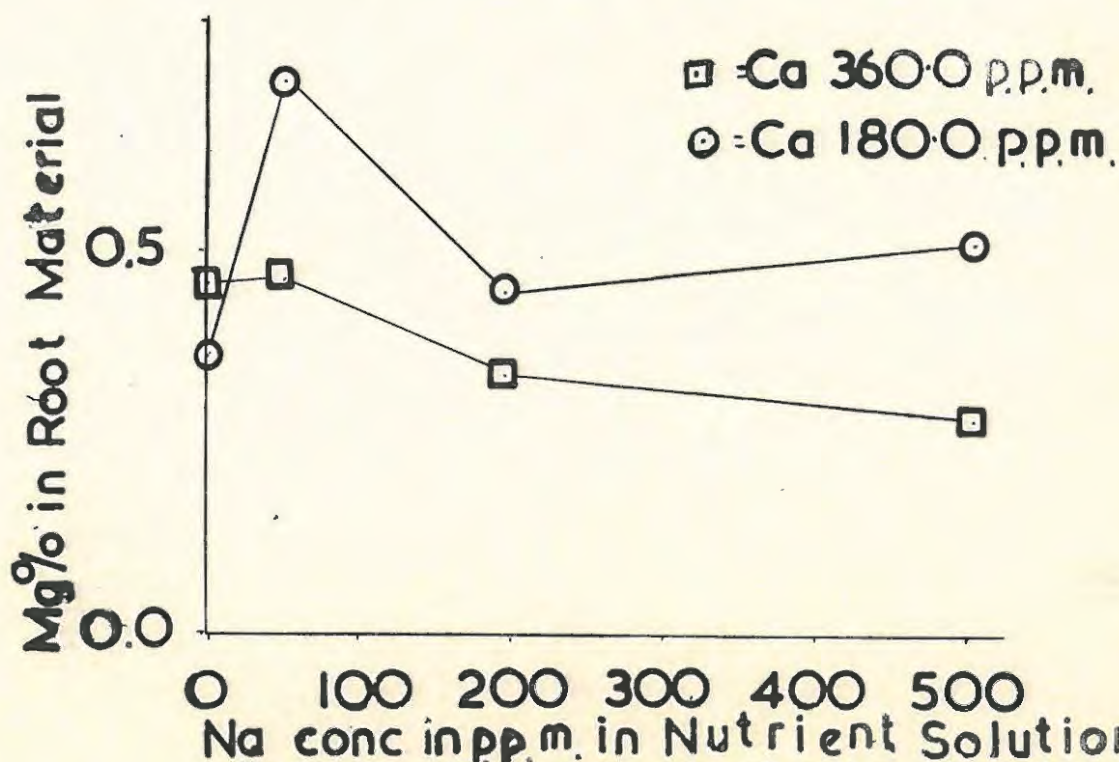


Fig. 36. The effect of the external sodium and calcium concentrations on root magnesium content.

Similarly the effect of increasing the potassium concentration in the external media, upon the internal root calcium content, varied with the level of calcium supply. Increasing the potassium concentration, at the lower level of calcium supply in the external media had very little effect upon the internal calcium concentration of the root. At the higher level of calcium supply, there was a trend towards an increased calcium concentration, with increase in potassium supply, see Fig. 35.

Table 24  
Statistical Analysis of Treatment Effect on Calcium  
Root Content

	D.F	Sum of squares	Mean Sum of squares	F Value	
Blocks	1	0.000050	0.000050	0.052	n.s
Na Treatment	3	0.431688	0.143896	150.834	***
K Treatment	3	0.133731	0.044577	46.227	***
Ca Treatment	1	1.635713	1.635713	1,714.584	***
Na x K Treatments	9	0.516553	0.057395	60.162	***
K x Ca Treatments	3	0.130820	0.043607	45.710	***
Na x Ca Treatments	3	0.340536	0.113512	118.983	***
Na x K x Ca Treatments	9	0.488850	0.054317	56.36	***
Residue	31	0.029560	0.000954		
Total	63	3.707501			

\*\*\* significant at 0.1% level, \*\* significant at 1.0% level,  
\* significant at 5.0% level, n.s. not significant.

Increasing the level of calcium in the external nutrient solution had a marked effect on the internal calcium concentration. The calcium root content was increased by more than four times its original concentration by doubling the calcium external supply from 180.0 p.p.m. to 360.0 p.p.m.

(d) Magnesium

The magnesium concentration of the roots varied between 0.02 - 0.12%. The effect of increasing the sodium concentration in the

external media on the root magnesium concentration varied with the level of calcium supply. The root magnesium concentration of the lemon seedlings receiving 360.0 p.p.m. calcium in the external media decreased with increase in the sodium treatment. However there was an increase in the root magnesium concentration in the lemon seedlings receiving 180.0 p.p.m. upon increasing the sodium concentration in the external media, see Fig. 36.

Table 25  
Statistical Analysis of Treatment Effect on Root Magnesium

	D.F	Content		F Value	
		Sum of squares	Mean Sum of squares		
Blocks	1	0.000648	0.000648	6.331	*
Na Treatment	3	0.004574	0.001525	14.891	***
K Treatment	3	0.006441	0.002147	20.966	***
Ca Treatment	1	0.002543	0.002543	24.841	***
Na x K Treatment	9	0.003119	0.000346	3.384	**
K x Ca Treatment	3	0.000705	0.000235	2.298	n.s
Na x Ca Treatment	3	0.003051	0.001017	9.934	***
Na x K x Ca Treatment	9	0.004491	0.000499	4.874	***
Residue	31	0.003174	0.000102		
Total	63	0.028751			

\*\*\* significant at 0.1%, \*\* significant at 1.0%,  
\* significant at 5.0% level, n.s. not significant

Generally increasing the potassium external supply resulted in an increase in the root magnesium concentration, see Table 25. This effect was found to vary with the level of sodium supply, the largest increase being obtained when the sodium concentration in the nutrient supply was 50.0 p.p.m.

Increasing the calcium level in the nutrient solution resulted in a decrease in the root magnesium concentration.

## CHAPTER IV

### Discussion and Conclusions

It has been well established by numerous workers that the level of nutritional supply markedly influences the visual appearance of plants. Under the conditions of the present experiment, similar observations were made.

The leaves of plants receiving the lower calcium treatment showed leaf patterns which corresponded with the calcium deficiency symptoms described by Camp, Chapman and Parker (1949), "a fading of the chlorophyll along the margins of the leaf and between the main veins, and in the case of lemon leaves the faded areas developed into larger burned areas".

A possible explanation for the presence of symptoms of calcium deficiency in plants grown in a nutrient media containing at least 180.0 p.p.m. calcium, is that the ammonia nitrogen present in the nutrient solution depressed the uptake of calcium. It has been shown by Jacobson and Swanback (1933), Nightingale (1937), McEvery (1946) and Hewitt (1947) that in the presence of ammonium nitrogen the uptake of calcium and magnesium was decreased.

The restriction of the leaf burn to the leaves of the apical region may be related to the calcium, sodium and potassium leaf content as suggested from the leaf analysis data. This leaf injury did not occur in leaves with a calcium content greater than 0.08 per cent. However in leaves with a calcium level below 0.08 per cent leaf burn did not occur if both the sodium and potassium leaf contents were low. Increasing the sodium, potassium or sodium + potassium leaf content above 1.80 per cent produced leaf burn in the apical leaves. The above observations suggest that the leaf injury may be attributed to a deficiency of calcium, together with high levels of sodium or potassium, or both ions together. The high sodium and potassium leaf contents were the result of high levels of supply of these ions to the nutrient solutions.

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It was observed that if intermediate levels of sodium (50.0 p.p.m.) were supplied together with high potassium levels in the nutrient supply, leaf burn did not occur, suggesting that intermediate levels of sodium may assist in preventing injurious effects produced by high concentrations of potassium. The leaf analysis data show that this may well be the case, as an increase in the level of supply of sodium was found to produce a decrease in the leaf potassium content. Likewise there was no occurrence of leaf burn in plants treated with 195.0 p.p.m. sodium, together with an intermediate level (117.0 p.p.m.) of potassium. The effects of potassium on high sodium treatments was not as marked as that of sodium on plants receiving high potassium treatments as leaf burn was produced on plants receiving sodium concentrations higher than 195.0 p.p.m. irrespective of the level of potassium supplied.

The restriction of leaf burn to the apical region alone, further suggested that the leaf injury was associated with low calcium content, together with high levels of sodium or potassium. It has been well established by Hoffer (1938), Skinner and Purvis (1949), Johem (1955) and numerous other workers that the basal leaves have a higher calcium content than the leaves of the apical region, whereas the sodium and potassium concentrations are higher in the apical leaves than in the lower leaves. The above phenomenon is ascribed to the varying mobilities of these elements within the plant.

Further it is probable that the occurrence of leaf burn was accentuated by the high temperatures reached in the greenhouse during the week prior to sampling as Ahi and Power (1938) and Magisted, Ayers, Wadleigh and Gauch (1943) have conclusively shown that high temperatures and rapid transpiration increase scorching and salt toxicity in plants.

Leaves of plants receiving no potassium treatment showed no definite or specific leaf patterns, which may characterize a deficiency of this element, this being possibly due to the plants still having sufficient potassium from the pre-experimental treatment. It is probable that if the experiment had continued under the same conditions for a longer period the trees would have shown signs of a lack of

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potassium, as potassium deficiency symptoms have been reported by Haas (1948 a, 1948 b) and Camp, Chapman and Parker (1949). Likewise lack of sodium was found not to have any effect on the appearance of the leaves.

The level of calcium treatment was shown to markedly influence the development of the seedlings. The seedlings receiving the higher calcium supply were better developed than the plants receiving corresponding treatments with the lower calcium supply. This is well illustrated by the height and weight results. The level of potassium supply only slightly influenced the development of the lemon seedlings, a small increase in dry weight resulting from an increase in the level of potassium supply. There did not appear to be any relationship between the level of potassium supplied and the increase in dry weight. Increase in the sodium supply appeared to have no influence on the growth of the seedlings, this being in agreement with the findings of Martin, Hardy and Murphy (1953).

Each of the elements studied in this investigation was found to vary in concentration between the different plant organs and with the position of the leaf stem samples on the main axis. Concentration variations in plants, similar to the above, have been shown by Pierre and Bower (1943) and numerous other workers, to be dependent upon (a) the mobility of the ion and (b) the presence of other ions which may interfere with its uptake and its mobility within the plant and (c) the plant species.

The lemon seedlings in this experiment had their highest sodium concentration in the roots, with lower concentrations in the stem and leaves. Potassium was shown to have its highest concentration in the leaves, while the stem and roots contained about a quarter of that found in the leaves. Similarly the leaves contained higher concentrations of calcium and magnesium than either the stem or roots. Considerable variation in concentration was found between the different elements within a plant organ. It was shown in the leaf analysis results that the concentration of potassium was ten times that of magnesium, five times that of sodium, and a third that of calcium. Similarly in the stem tissue the potassium

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concentration was the highest, being nine times as great as magnesium and three times as high as calcium or sodium. In the root tissue sodium was present in much higher concentrations than calcium, potassium or magnesium.

It was shown that the effect of the sodium treatment on the leaf sodium content varied with the position of the leaf on the central axis. Lemon seedlings receiving no sodium treatment were found to have larger sodium concentrations in the lower leaves than in the upper leaves, whereas plants receiving 505.0 p.p.m. sodium accumulated most of their sodium in the upper leaves and progressively less in the lower leaves. The above results suggest that the mobility of sodium in the leaves is dependent upon the external concentration of the element. The concentration of sodium in the stem tissue was found to vary with position on the central axis, the higher concentrations being found in the apical and middle portions and the lower concentrations in the basal portions. However, the distribution of sodium in the stem was found not to vary with the external concentration as was found to be the case in the leaf tissue.

The leaf analysis data showed the potassium concentration varied with the position of the leaf on the main axis. From the distribution of the potassium concentration levels in the leaves it would appear that potassium is mobile within the plant as a higher concentration of potassium was present in the apical leaves than in the lower leaves. Calcium on the other hand, appeared to be a comparatively non-mobile element, as the highest concentration of the element was present in the basal leaves and only a low concentration in the apical leaves. The variation in concentration from the apical leaves to the lower leaves was as pronounced for the seedlings receiving 180.0 p.p.m. calcium as for those receiving 360.0 p.p.m. calcium in the nutrient supply, even though their respective internal calcium concentrations differed.

Similarly the concentration of magnesium in the leaves was shown to vary with the position of the leaf on the central axis. Magnesium, like calcium, had its greatest concentration in the basal

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leaves, and was found in progressively smaller quantities in the apical leaves, the concentration in the lower leaves being twice as great as that in the upper leaves.

The distribution of sodium suggests this element to be mobile in the lemon stem tissue, higher concentrations being found in the apical and middle portions of the stem, than in the lower portions. The level of sodium supplied did not appear to alter the sodium distribution in the stem as it had done in the leaf tissue. The potassium and calcium contents of the stem tissue had a similar distribution pattern to that shown by the foliage.

As mentioned earlier, it has been well established that the amount of a given element taken up by the plant from the external source is influenced by (1) the concentration of the element in the external medium and (2) the concentration of other elements present in the nutrient medium. In the present investigation the chemical content of the seedlings was found to be influenced by the level of sodium in the nutrient supply. The sodium content of all the plant organs increased with a rise in the sodium level in the experimental treatments. A greater response was shown by the roots than either the stem or leaves to an increase in the sodium level in the solution. This trend is similar to the observations of James, Pearson Parker and Huberty (1952) who used citrus.

In the present investigation leaf, stem and root analyses were all found to be satisfactory indices of the sodium supply. Analysis of the leaves showed an almost linear relationship between sodium leaf content and supply of sodium. Haas (1952) and James, Pearson, Parker and Huberty (1952) found leaf analysis unsatisfactory as an index of sodium toxicity. From the results obtained in the present investigation root analysis appeared to give no further information than that obtained from the analysis of the leaves.

The effect of increasing the sodium supply on the potassium content of the plant was shown to vary with the plant organ. The leaf potassium content decreased as the sodium level in the nutrient media was raised. These observations are not in agreement with the

findings of Chapman and Brown (1942), James, Pearson, Parker and Huberty (1952), and Martin, Hardy and Murphy (1953), who showed that an effect of fertilising mature orange with sodium nitrate was to increase the leaf potassium content.

In the roots the effect of increasing the sodium supply on the potassium content was found to vary with the level of calcium supplied. Plants receiving 180.0 p.p.m. calcium had their root potassium content slightly decreased by raising the sodium level, whereas seedlings receiving 360.0 p.p.m. calcium had their potassium content increased upon being supplied with higher levels of sodium.

The potassium content of the upper stem portions was found to decrease as the sodium treatment was increased. The lowering of the potassium content could be associated with the low calcium content of the upper stem portions, as both the middle and lower stem portions with a higher calcium content, had their potassium content increased by supplying additional amounts of sodium.

The results of the stem and root analysis suggest that the level of calcium present in the tissue may determine whether additional supplies of sodium increase or decrease the potassium content of the plant. The difference between the observations of Chapman and Brown (1942), James, Pearson, Parker and Huberty (1952) and Martin, Hardy and Murphy (1953), and those in the present investigation may be due to the level of calcium in the leaves, as the foliage of the orange trees investigated by James, Pearson, Parker and Huberty (1952) contained at least twice the percentage of calcium present in the foliage of the lemon seedlings in the present study.

The sodium treatment was shown not to affect the leaf calcium content, irrespective of the level of supply. However with the stem tissue, the effect of increasing the sodium supply on the calcium content was found to vary with the position on the main axis. The calcium content of the upper and middle stem portions decreased with an increase in the level of sodium supply, whereas in the lower stem portions the calcium content increased with increased sodium supply. In the root tissue the effect of the sodium treatment on the calcium

content varied with the level of calcium supplied. The seedlings receiving 180.0 p.p.m. calcium were found to have their calcium root content decreased when the level of sodium treatment was raised, whereas the plants receiving 360.0 p.p.m. calcium in the nutrient media increased their root calcium content when the external level was raised to 195.0 p.p.m. At a still higher level of sodium supply, the calcium content decreased considerably. The above observations are in agreement with the findings of Cooper, Paden and Phillippe (1953) and Johem (1955).

The results of both the stem and root analysis suggest that a depression of calcium by sodium occurs when the calcium : sodium ratio in the tissue is low, but an increase in calcium is brought about by sodium when the above ratio is high. It is of interest to note that a different ion relationship appears to exist in the leaves to that found in the stem and roots, and this suggests that the base reciprocation in the leaves may be of an entirely different character both quantitatively and qualitatively from that of other parts of the plant. Studies of ion relationships in plants by Smith, Reuther, Specht and Hrncair (1954) emphasize that leaf analysis alone may give the wrong impression of reciprocation in other plant parts, and they suggest that this type of situation makes it desirable to consider the distribution of elements in various parts of the plant before conclusions are drawn in regard to nutrient availability, absorption behaviour, and the location of antagonistic reactions.

The sodium treatment was found to influence the magnesium content of the roots, stems and leaves differently. Low levels of sodium supply were shown not to influence the magnesium content of the leaves, but levels of sodium above 50.0 p.p.m. slightly decreased the magnesium content of the foliage. These results are in agreement with the findings of Cooper, Paden and Phillippe (1953) and Larson and Pierre (1953). The magnesium content of the stem portions showed an increase when the level of sodium supply was raised. The influence of the sodium treatment on the root magnesium concentration was found to vary with the level of calcium supplied. The magnesium content of roots receiving the higher calcium treatment, decreased with increased sodium supply, whereas in the plants receiving the

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lower calcium supply, there was an increase in the root magnesium concentration when the sodium concentration in the nutrient solution was raised.

Increasing the potassium concentration in the nutrient supply raised the potassium content of the leaves, stem and roots of the lemon seedlings. A similar response to fertilizing with potassium has been obtained by numerous workers on fruit nutrition, Chapman, Brown and Rayner (1947), Cain (1948) and McColloch, Bingham and Aldrich (1957). It was observed that the increase in the potassium content of the stem and leaf tissues was approximately five times as great as that of the root tissue.

The level of calcium was shown to influence the degree to which an increase in the potassium supply raised the potassium root content. The roots of plants receiving the higher calcium treatment showed slightly greater responses to increases in potassium supply than plants receiving the lower calcium treatment. The above effect may be due to the higher calcium level reducing the sodium uptake and thus allowing a larger uptake of potassium, since, as has been reported earlier in this chapter, sodium was found to reduce the potassium root content.

The leaf sodium content was only slightly altered by raising the external potassium supply. The tendency was for the leaf sodium to decrease slightly with increase in the level of potassium supplied. Potassium appears not to interact with sodium in the stem and root tissues, as in both these tissues the sodium content was found not to be influenced by increases in the external potassium supply.

The influence of the potassium treatments on the tissue calcium content was shown to vary with the plant organ. The calcium content of the foliage was not affected by increases in the level of potassium supplied. The stem portions were shown to increase their calcium concentrations when the supply level of potassium was raised except in the case of the lemon seedlings receiving 505.0 p.p.m. sodium where the increased potassium level brought about a decrease in the stem calcium content.

In the root tissue the effect on the calcium content of increasing the level of potassium supply was similar to the effect obtained by raising the sodium supply level - the effect varying with the calcium treatment received. Increasing the potassium supply at the low level of calcium treatment had little or no influence on the internal root calcium concentration. However at the higher calcium level there was a trend towards an increased internal calcium concentration when the potassium supply was increased.

The magnesium contents of the leaf, stem and root tissues were shown to be affected by the level of potassium supplied. Increasing the external potassium concentration was found to induce a decrease in the magnesium content of the leaves. Similar observations have been found in citrus by Camp, Chapman and Parker (1949), Chapman and Brown (1953) and McColloch, Bingham and Aldrich (1957). The effect of potassium treatments on the magnesium content of the foliage was shown to be less severe than for the corresponding levels of sodium treatment.

The magnesium content of the stem tissue also decreased with increase in the potassium supply, but to a lesser degree than found in the foliage. In the root tissue the reciprocation between sodium and magnesium appeared to be of a different nature to that found in the leaf and stem tissue. The general trend was for the root magnesium content to increase when the potassium level was raised.

The results from the plant analysis data show that the level of the external calcium influenced the calcium content of the plant tissues. The degree to which the calcium content of the tissues increased when the calcium supply was raised was shown to vary with the plant organ. In the root and stem tissues approximately a four-fold increase in the calcium content was found when the external supply of calcium was doubled from 180.0 p.p.m. to 360.0 p.p.m., whereas in the leaf tissue the increase was only of the same order as the increase in the concentration of the nutrient media. Similar observations were made by Chapman (1952) using orange trees. However Haas (1952) growing Lisbon lemon trees in water culture found a none too striking tendency for calcium to increase in the leaves as the

observation was reported for root bark tissue by Haas (1952 b)

using Lishon Lemon seedlings grown in pots with a soil (1952)

concentration of calcium was increased.

It was observed by James, Pearson, Parker and Huberty (1952) that citrus trees receiving sodium in the form of sodium nitrate showed a marked reduction in sodium concentration in the tissues after calcium had been supplied in the form of gypsum. A similar observation was made on the root tissue in the present work, where the sodium content was shown to decrease when the calcium supply level was raised from 180.0 p.p.m. to 360.0 p.p.m. It was shown that when the calcium was supplied at the higher level, it mitigated the influence previously mentioned, of the sodium level in the external supply on the stem sodium concentration.

The distribution of leaf sodium was observed to be markedly influenced by the level of calcium supply. Seedlings receiving the 180.0 p.p.m. calcium treatment were shown upon chemical analysis to have their maximum leaf sodium content in the apical leaves, and the concentration decreased progressively towards the basal leaves. The plants receiving the 360.0 p.p.m. calcium treatment were found to have their greatest concentration of sodium in the basal leaves, with a slightly lower concentration in the middle leaves, and an even larger decrease in the sodium concentration in the apical leaves. The above results further indicate the complexity of ion accumulation in plant organs.

The calcium level in the external supply has been shown by numerous workers on the nutrition of citrus, to influence the potassium content of the different plant organs. Chapman (1952) showed low potassium in the leaf analysis of plants grown with excess calcium and Camp, Chapman and Parker (1949) found an unusually large intake of potassium by citrus trees grown on low calcium, acid soils in Florida. It was shown in the present investigation that a slight decrease in the leaf potassium resulted from raising the external level of calcium, which is in agreement with the findings of the above workers.

The stem and root tissues responded in a different manner to the leaf tissue. The potassium content of the former two tissues increased when the external calcium supply was raised. A similar

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By observing the sodium-potassium interaction in the roots, it can be seen how results of interactions may be misleading if no

observation was reported for root bark tissue by Haas (1952 b) using Lisbon Lemon seedlings grown in water culture, and Viets (1944) showed an increased migration rate of potassium into roots when sodium was supplied in addition to potassium in the external medium.

The calcium treatment had no apparent influence on the magnesium content of the foliage. However in the stem and root tissues a decrease in the magnesium content resulted when the calcium supply was increased.

In the past it has been the general tendency to draw conclusions concerning the ion interrelationships of the whole plant from chemical analysis of one type of tissue only, usually the leaves. The unsoundness of this procedure has been well exposed in the current investigation. Results obtained in the present studies show that in order to obtain the overall picture of ion interrelationships within the plant, analysis of stem and root tissue, in addition, are required, since these relationships from the leaf, stem and root of the same plant vary considerably. It is suggested that the ion interrelationships between these three tissues in particular require further detailed comparison.

In the present study it has been shown that consideration of leaf analysis only may not give the true impression of ion reciprocation for all the plant parts. This is well illustrated by the calcium-potassium interaction in the plants. In the leaves, increased calcium supply was shown to reduce the potassium leaf content, whereas, in the stem and root tissues, when the calcium supply was increased the potassium content rose.

Another aspect requiring further investigation is the effect of the external concentration of sodium on the distribution of sodium in the foliage. As reported earlier, sodium appears to differ from the other elements studied, in having a different distribution in the leaves for a high and low external supply. This would suggest that the distribution of this element in the plant is not dependent only on its mobility in the tissues.

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By observing the sodium-potassium interaction in the roots, it can be seen how results of interactions may be misleading if no consideration is taken of the levels of elements other than the two being specially studied. In the example cited, it was shown how the potassium content of the tissue was depressed by sodium when the supply of calcium was low, and the potassium uptake was accelerated by sodium when there was a high level of calcium present. Thus great caution should be exercised in interpreting interactions between two ions from chemical analysis data without due consideration of the action complementary ions may be having.

PART II

## Part II

### CHAPTER I

#### Experimental Methods

Disks of storage tissue have been found to be convenient material for studies of salt accumulation, and were used by Nathanson (1904), Stiles and Jorgensen (1915), Steward (1937), Robertson (1941), and Sutcliffe (1957) for their investigations on ion absorption. When suitably prepared they are capable of absorbing ions rapidly in a reproducible manner over a considerable period of time against an activity gradient. Their amenability to experimental manipulation and their relatively simple morphology are other attractive features. An additional advantage of using storage tissue in preference to whole plants, is the effect that the transport of materials away from the absorbing region is reduced, further, there is a greater uniformity of cells in the tissue.

In this work, as in the previous section, every precaution was taken to ensure that the containers were clean and that there was minimum external contamination.

#### Water Purification

All the water used in the preparation of solutions was purified either by redistillation, or by using an ion exchange column as described in Part I.

#### Glassware

"Pyrex" and "Hysil" glassware were used throughout the present work as a precaution against contamination likely to occur from 'softer' grades of glass. The glassware was cleaned and purified as previously described in Part I.

#### Containers

The containers used for the present study were 500 ml narrow mouth conical flasks. The flasks were stoppered with corks which had been previously impregnated with paraffin wax. Each cork had

a hole  $\frac{1}{4}$ " in diameter in its centre to allow the passage of the aeration glass tube.

Polythene bottles of 50 ml capacity were used for storing the experimental solutions prior to chemical analysis.

#### Chemical Analysis

The chemical reagents used throughout this investigation were British Drug Houses "Analar" grade reagents.

#### Experimental Methods

Both potato tuber and carrot root tissue were used in the present investigation. Carrot tissue was found to have a number of advantages over potato tuber tissue as experimental material, the chief being that carrot tissue would remain healthy for longer periods in aerated solutions.

A core of tissue 1.25 cms. in diameter was bored using a cork borer, and then disks of tissue 0.18 cms. in thickness were cut by means of a hand microtome, the average weight of each disk being approximately 0.24 g. The disks were then washed in running tap water for twenty four hours to remove any damaged cell debris. At the end of this period batches of sixty disks were threaded on to fuse wire, each disk being separated from its neighbours by means of a porcelain bead, to enable the complete disk to be bathed in the experimental solution, see Fig. 36.

The strings of tissue disks were then bathed in aerated purified water for a further ninety six hours, the purified water being renewed every three hours during the first twelve hours, and thereafter every six hours. The washing of the disks was performed primarily to starve the tissue, and also to remove any absorption inhibitors that were present, as it has been conclusively shown by Rees and Skelding (1950), Skelding and Rees (1952) and Dale and Sutcliffe (1956) that tissue extracts inhibit the absorption of salts by tissues. The disks during the washing period were aerated continuously by a rapid air stream bubbling through the liquid, and under these conditions they usually showed no signs of bacterial contamination, such as loss of turgidity. After the disks had

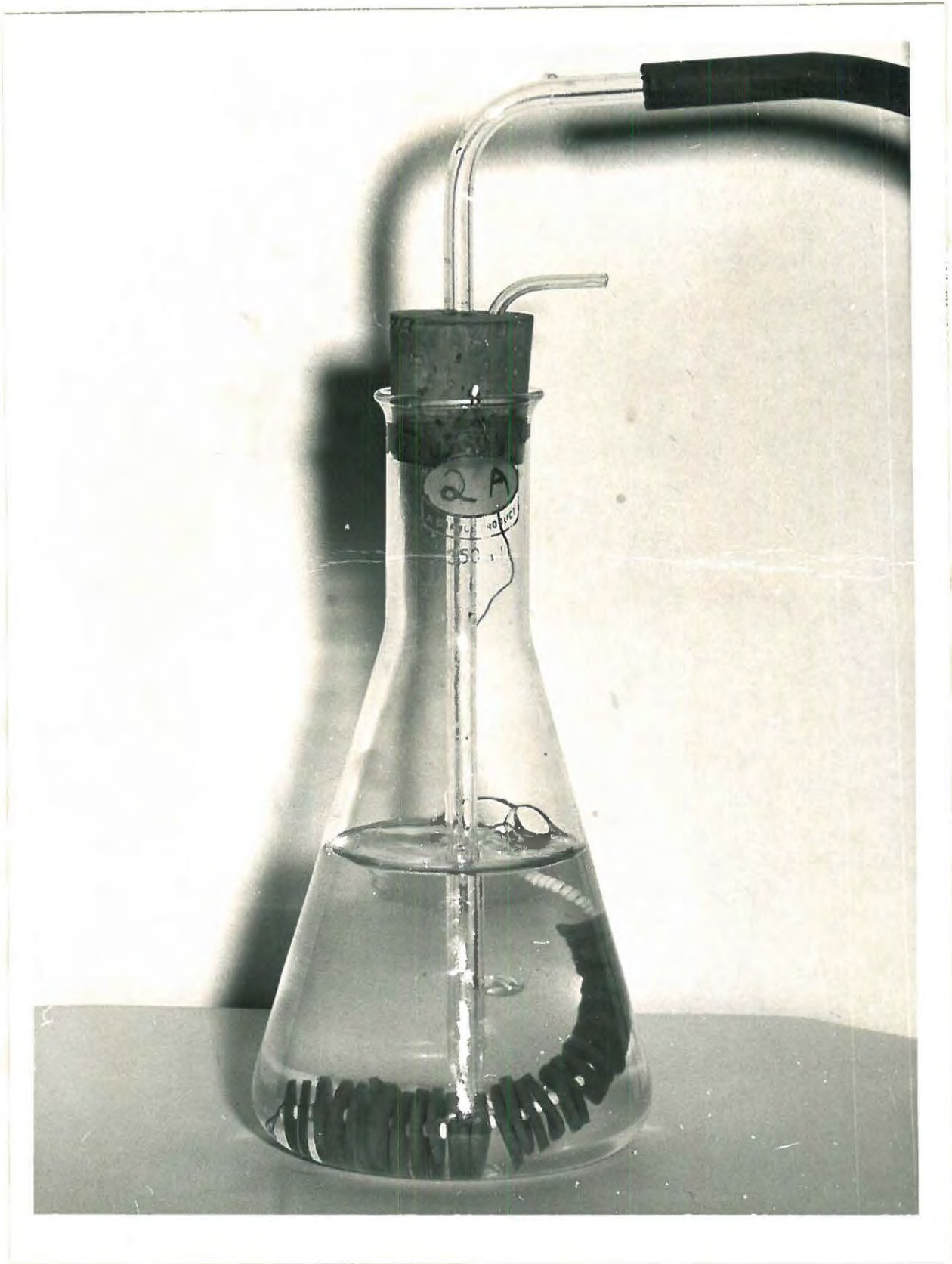
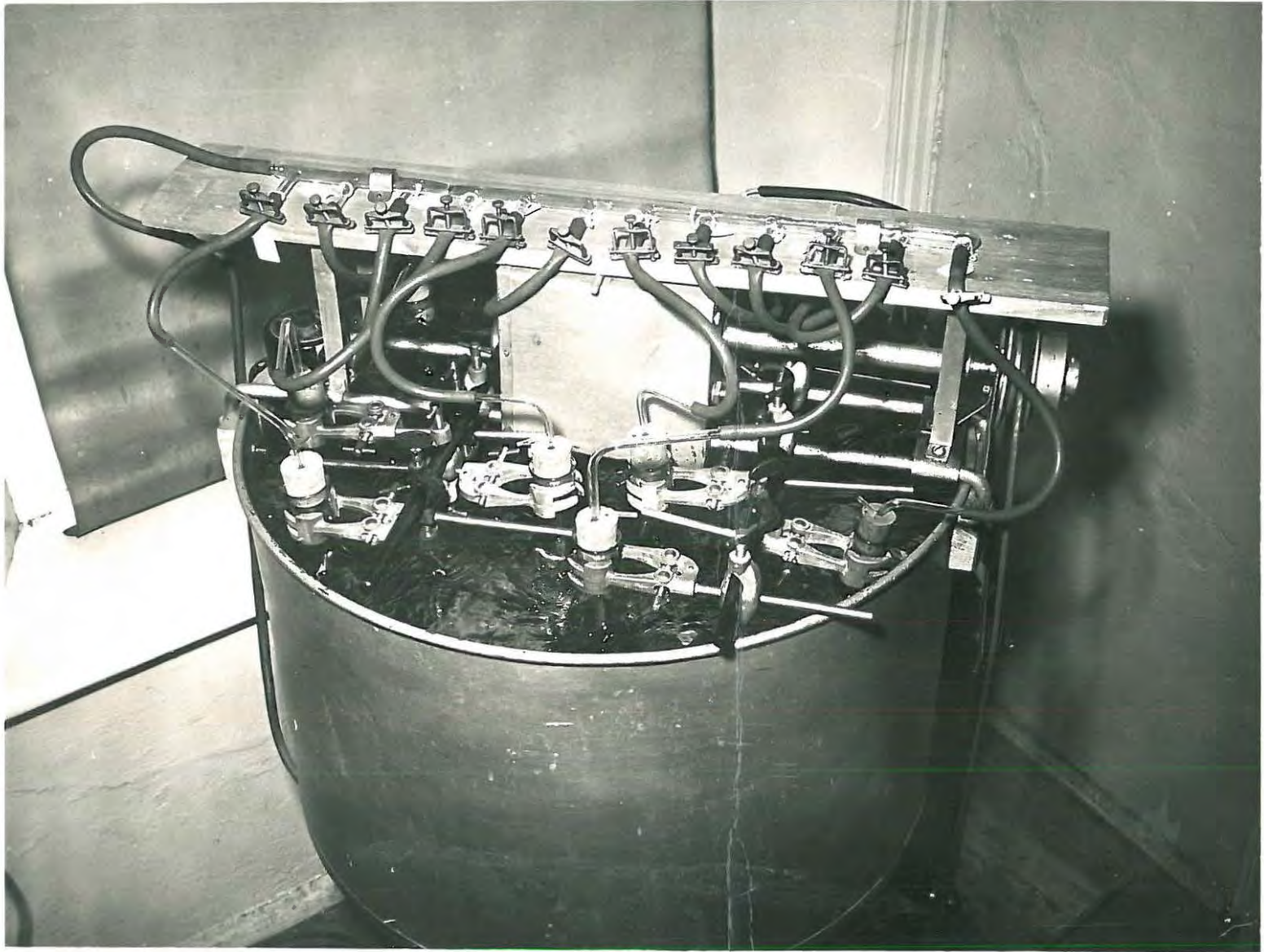


Fig. 36. String of carrot root disks in experimental solution.

Fig. 36. Showing flasks in water bath .



been washed in the above manner, the strings of sixty disks were taken and placed in 300 ml of their respective solutions, in 500 ml conical flasks. Each flask was then closed with a wax cork. The solutions and disks were continuously aerated by an air stream bubbling through the solutions. The disks were kept at 25°C throughout the experiment by partial immersion of the flasks in a temperature controlled water bath, see Fig. 37.

In order to follow the uptake of the salts by the tissue, the external solution was chemically analysed at intervals during the course of the experiment. 20 ml samples were taken of the experimental solution at the commencement, and then after 1, 2, 8, 24, 48, 72 and 96 hours. To ensure that the same volume ratio of tissue/medium was maintained after each sample was removed, five disks of tissue were removed from the string at each sampling. Each experiment was replicated on separate samples of material on one or more occasions and the results were usually found to be in agreement with one another.

#### Experimental Solutions

Sodium, potassium, calcium and magnesium were the only four cations present in the experimental solutions. The potassium was added as the dipotassium hydrogen phosphate, and the nitrate salts. Similarly calcium was added as the nitrate salt. Sodium was added as the chloride salt, and magnesium as the sulphate salt. Molar stock solutions of all the above salts were made, and used in the preparation of the experimental solutions.

#### pH Values

During the course of the experiment the pH values of the solutions were found to increase slightly. Initially, the solutions had a pH 6.5, which during the course of the ninety-six hour experimental period rose to pH 7.8. The pH of the experimental solutions was controlled largely by the addition of dipotassium hydrogen phosphate.

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### Chemical Analysis

The salt uptake measurements were made by means of chemical analysis of the external medium. In the analysis of these solutions, 5 ml aliquots of solution were diluted with purified water to 100 ml, and in instances where further dilution was required each 5 ml aliquot was diluted to 200 ml. Sodium, potassium and calcium were determined by flame photometry, following the same procedure as outlined in the previous section.

## Chapter II

### Results.

Sutcliffe (1957) reported that beet storage tissue would absorb either sodium or potassium at a comparable rate from equimolar solutions of single salts. However when the two ions were supplied together in the same medium unequal uptake was shown to occur. In the present investigation, the latter phenomenon was further investigated in six experiments, each experiment being designed to determine the extent of competition between sodium and potassium for their uptake by storage tissue.

#### Experiment 1

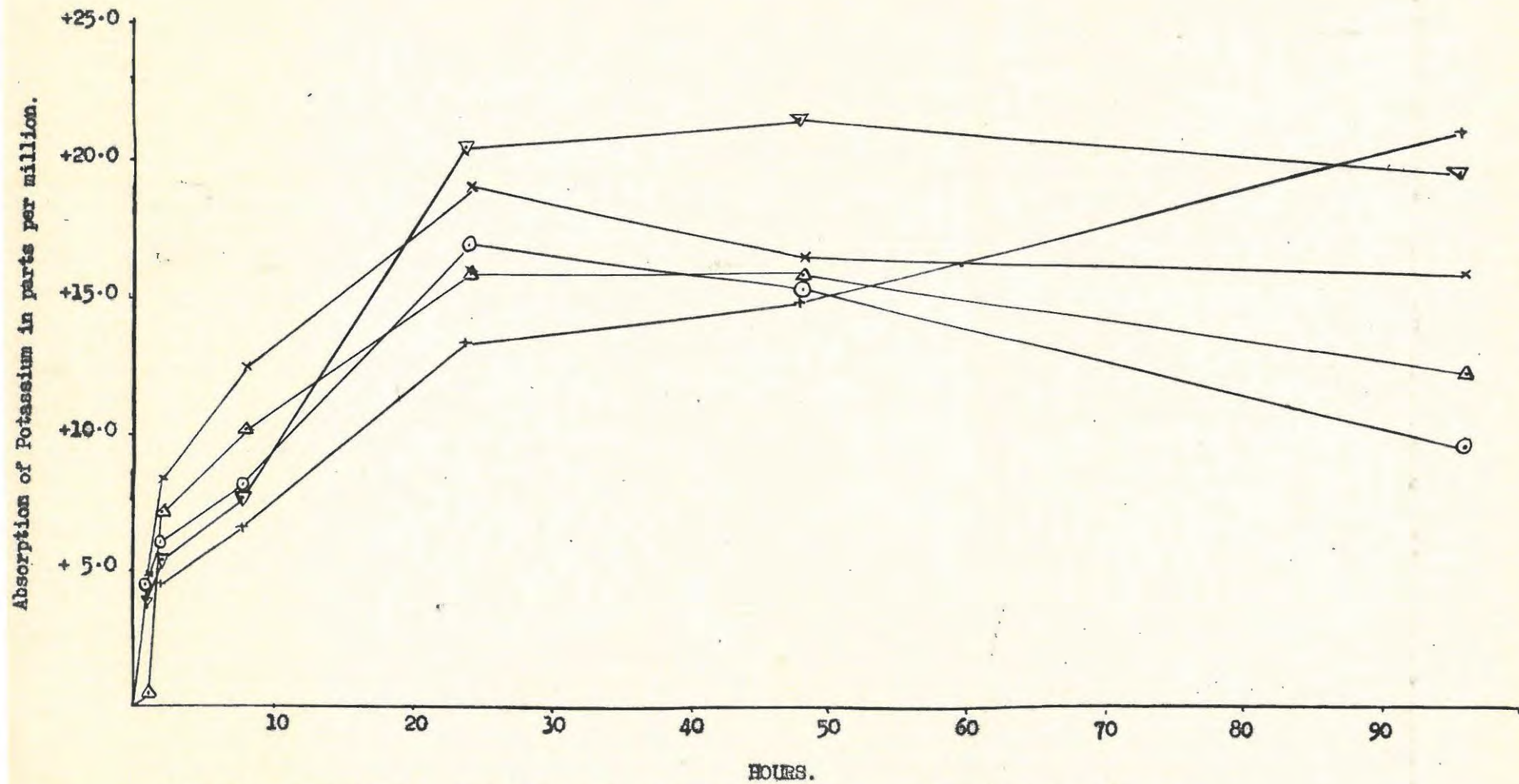
Potato tuber disks were used as the experimental material in the first experiment and carrot root tissue for all the remaining experiments. The tissue disks were pre-treated by washing for five days as previously described and then placed into their respective experimental solutions. Each experimental solution had a potassium concentration of 74.0 p.p.m. and sodium was supplied at six different levels ranging from 2.8 p.p.m. - 185.0 p.p.m. Calcium was supplied at 200.0 p.p.m. and magnesium at 50.0 p.p.m. The uptake of sodium and potassium by the storage tissue was followed over a ninety-six hour period by making analyses of samples from the media at the commencement of the experiment, and then after 1, 2, 8, 24, 48, and 96 hours, see Table 25.

The absorption of potassium by the potato disks during the course of the experiment can be seen in Fig. 38. In each experimental treatment the tissue disks showed an initial rapid uptake of potassium during the first two hours, followed by a much slower rate of uptake of the cation. The observation of the initial rapid uptake of the cation is in agreement with the findings of Steward and Harrison (1939) using potato tuber tissue, Hanson and Bonner (1954) using Jerusalem artichoke tuber tissue, and Sutcliffe (1957) using beet storage tissue. The above workers suggest that the initial salt uptake is influenced by two processes, the one being physical and

Table 25

Sodium and potassium concentrations (parts per million) external medium during 96 hour experimental period

Time of sampling	Solution 1		Solution 2		Solution 3		Solution 4		Solution 5		Solution 6	
	Na	K	Na	K	Na	K	Na	K	Na	K	Na	K
At commencement	2.8	73.8	7.4	73.8	28.1	74.4	50.1	73.8	83.1	73.8	185.7	74.3
After 1 hour	2.3	69.2	7.4	73.2	26.0	69.7	46.2	69.7	75.9	69.7	176.4	69.7
After 2 hours	2.1	67.7	7.4	66.6	26.5	66.1	61.5	68.2	76.6	68.2	175.9	69.7
After 8 hours	2.0	65.6	7.0	63.3	26.5	62.0	46.7	62.5	76.0	66.1	173.6	67.7
After 24 hours	2.8	65.9	6.7	57.9	26.2	55.4	46.4	54.3	76.6	53.3	173.6	61.0
After 48 hours	2.3	58.4	8.4	57.9	27.4	57.9	48.0	57.4	78.5	52.3	180.1	59.5
After 96 hours	2.6	64.1	6.7	61.5	29.2	58.4	51.1	64.1	81.7	54.3	178.3	53.3



**Fig 38** Uptake of K. by Potato Tuber Disks from solutions containing K: 74.0 p.p.m.  
 + Ca - 200 p.p.m., + Mg. - 50.0 p.p.m. + (○) Na-2.8 p.p.m. (△) Na-7.4 p.p.m.  
 (×) Na-28.1 p.p.m. (◻) Na- 50.1 p.p.m. (▽) Na-83.1 p.p.m. (+) Na-185.7 p.p.m.

frequently termed "passive" uptake, and the other metabolically assisted and termed "active" uptake. The first process is claimed to be primarily responsible for the large initial uptake, and the metabolic or "active" uptake for the salt uptake after the second hour (Epstein, 1956).

It was shown that when the external sodium concentration was increased, there was a general trend for the rate of potassium uptake to increase for the first twenty four hours, followed by a period when there was a gradual diffusion of potassium out of the storage tissue, except in the case of the disks bathed in the solutions with the two highest sodium concentrations.

Like potassium the uptake of sodium was rapid during the first two hours, and this uptake was shown to be dependent upon the concentration of sodium in the external solution. The amount of sodium taken up by the tissues increased as the sodium concentration in the solution was raised. After twenty four hours a gradual diffusion of sodium out of the tissue was shown to take place, see Fig.39.

The amount of potassium absorbed by the potato disks was found to be approximately twice the sodium uptake when the external solution contained approximately equal concentrations of both cations.

#### Experimental 2

In this experiment the solutions were supplied with potassium at a concentration of 54.0 p.p.m. The sodium was supplied at six different levels ranging from 1.6 p.p.m. to 180.0 p.p.m., calcium at 200.0 p.p.m. and magnesium at 50.0 p.p.m. The absorption of sodium and potassium was followed over a ninety six hour period, as in the previous experiment, see Table 26.

The course of the potassium uptake by the carrot root tissue from the different external solutions is shown in Fig.40. As in the previous experiment there was a rapid initial uptake, followed by a much slower, but steady rate of potassium uptake. The concentration of sodium in the external solution was shown to influence the uptake of potassium by the tissue. The uptake of potassium by the root was shown to be markedly reduced as the sodium level in the external solution was raised.

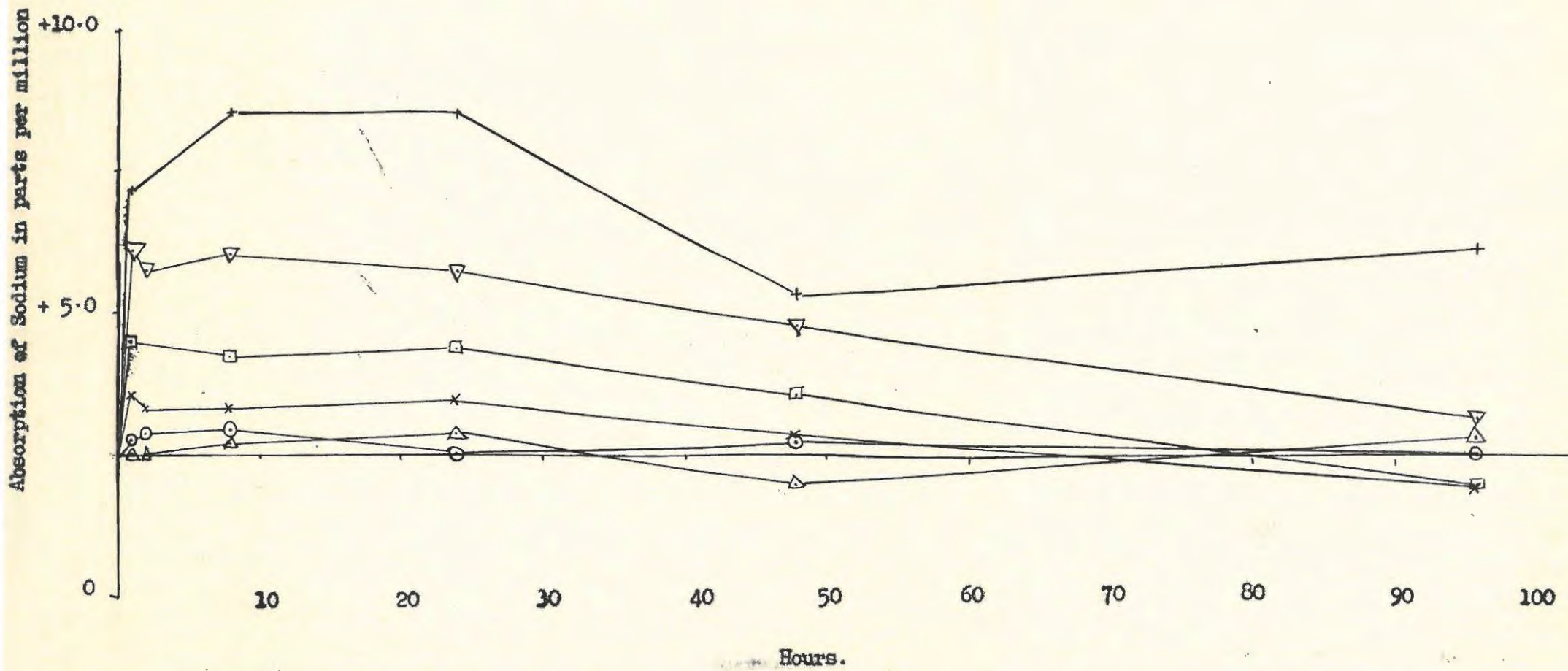


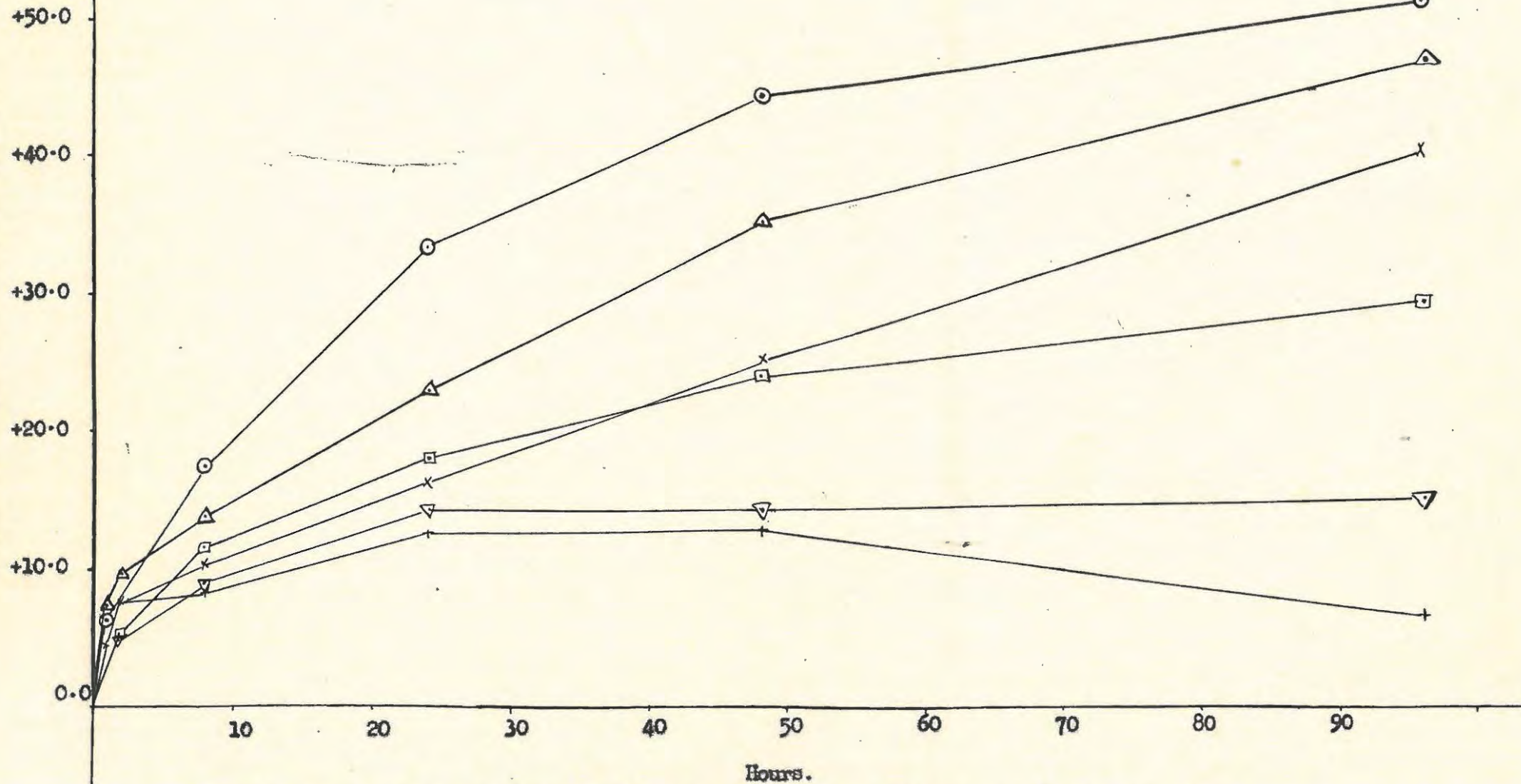
Fig 39. Uptake of Na by Potato Tuber disks from solutions containing K 74.0 p.p.m. + Ca - 200.0 p.p.m. + Mg - 50.0 p.p.m. + (○) Na-2.8 p.p.m. (△) Na-7.4 p.p.m. (×) Na-28.1 p.p.m. (□) Na-50.1 p.p.m. (▽) Na-83.1 p.p.m. (+) Na-185.7 p.p.m.

Table 26

Sodium and potassium concentrations (parts per million) of external medium during the 96 hour experimental period

Time of sampling	Solution 1		Solution 2		Solution 3		Solution 4		Solution 5		Solution 6	
	Na	K	Na	K	Na	K	Na	K	Na	K	Na	K
At commencement	1.6	52.3	13.2	55.4	27.3	53.8	46.9	54.6	78.0	54.6	180.1	56.1
After 1 hour	2.0	46.1	15.3	48.2	29.1	49.2	48.0	49.2	76.1	49.2	176.4	50.2
After 2 hours	1.6	43.6	11.1	45.6	24.6	46.1	45.0	49.2	74.9	49.7	171.8	48.2
After 8 hours	1.7	34.9	12.8	41.5	24.6	43.6	43.9	43.1	74.1	45.6	171.8	47.7
After 24 hours	1.6	18.5	10.2	32.3	23.4	37.3	39.9	36.4	71.2	40.0	169.4	43.1
After 48 hours	1.7	7.9	10.0	20.0	20.1	28.5	37.7	30.2	66.8	40.0	166.2	43.0
After 96 hours	2.3	1.0	8.8	8.3	20.2	13.1	34.8	24.6	64.5	39.0	156.9	48.2

Absorption of Potassium in parts per million.



**Fig. 40** Uptake of K by Carrot Root Disks from solutions containing K - 54.0 p.p.m. + Ca - 200.0 p.p.m. + Mg - 50.0 p.p.m. + (○) Na - 1.6 p.p.m. (△) Na - 13.2 p.p.m. (×) Na - 27.3 p.p.m. (□) Na - 46.9 p.p.m. (▽) Na - 78.0 p.p.m. (+) Na - 180.0 p.p.m.

The carrot tissue showed a non-metabolic uptake of sodium during the first two hours, but this uptake was not as large as that for potassium. The sodium absorption was shown to be dependent upon the concentration of the cation in the solution, the higher the sodium concentration the greater the uptake of this cation by the tissue, see fig. 41.

The amount of potassium absorbed by the tissue disks was found to be approximately double that of sodium, from experimental solution containing approximately equal concentrations of both sodium and potassium ions.

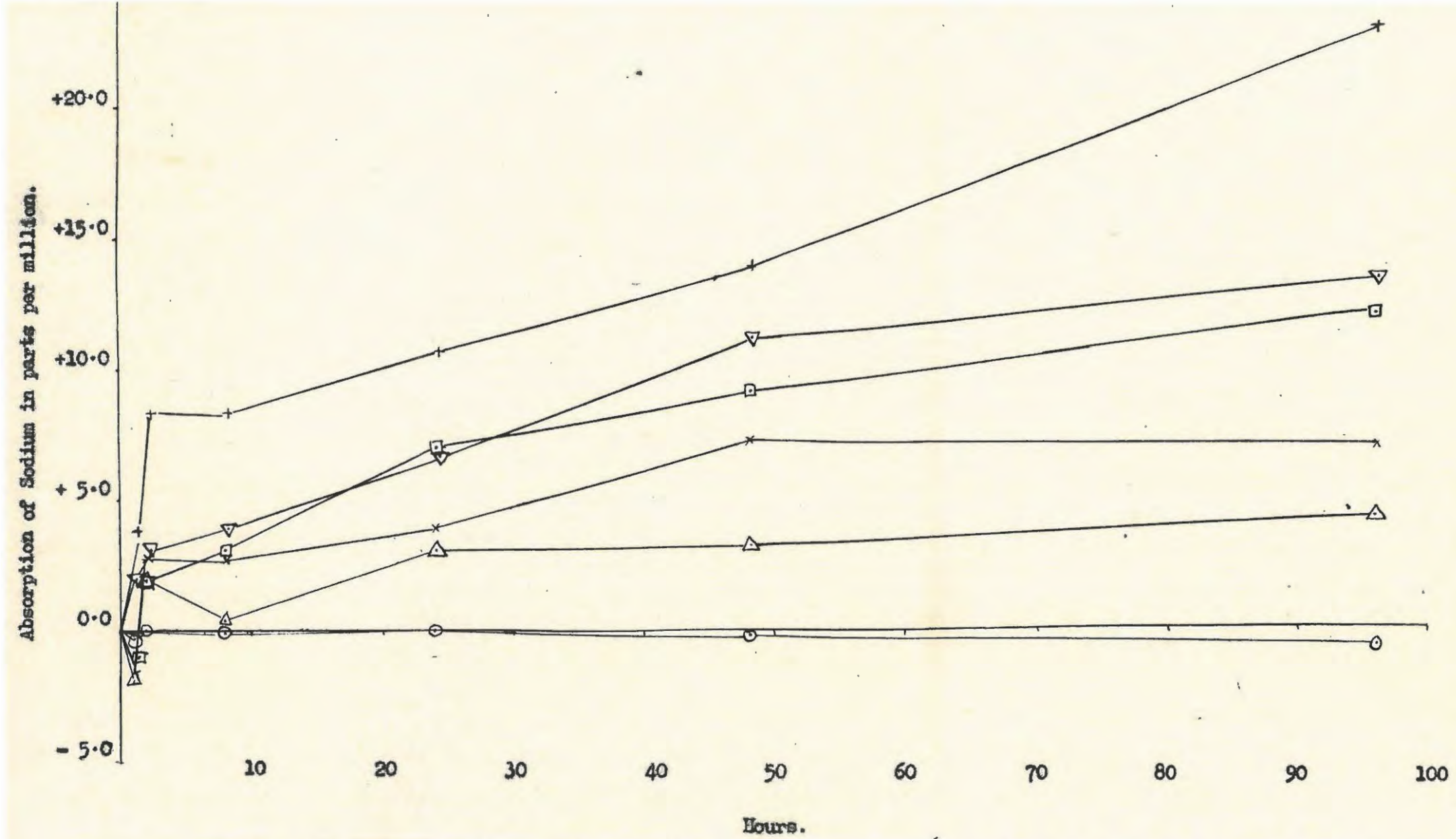
### Experiment 3

In the third experiment the potassium was supplied at 116.0 p.p.m., approximately double the potassium concentration in the previous experiment. Sodium was supplied at six different levels, ranging from 2.9 p.p.m. to 177.0 p.p.m., calcium at 200.0 p.p.m., and magnesium at 50.0 p.p.m. The absorption of sodium and potassium by the tissue disks was followed over a ninety six hour period, see Table 27.

The course of the potassium uptake during the experimental period is shown in Fig.42. The concentration of sodium present in the external solution was shown to markedly influence both the 'active' and 'passive' absorption of potassium by the tissue.

The absorption of potassium by the carrot disks bathed in the solutions with the two highest sodium concentrations was shown to be negligible during the first two hours in comparison with the uptake by the storage tissue in the experimental solutions with lower sodium concentrations. After the first eight hours the disks bathed in the solutions with the high sodium concentrations showed a rapid diffusion of potassium out of the carrot tissue into the external solutions. The tissue disks in the lower sodium concentrations were found to have their potassium uptake only slightly reduced as the level of the sodium concentration was raised.

The uptake of sodium by the root tissue did not appear to be influenced to any appreciable extent by the concentration of sodium in the external solution, see Fig.43. The carrot disks bathed in the higher sodium concentrations showed some absorption of sodium during the first twenty four hours, but after this period there was a diffusion of the cation out of the tissue into the external solution.



**Fig. 41.** Uptake of Na by Carrot Root Disks from solutions containing K - 54.0 p.p.m. + Ca - 200.0 p.p.m. + Mg - 50.0 p.p.m. + (o) Na - 1.6 p.p.m. (Δ) Na - 13.2 p.p.m. (x) Na - 27.3 p.p.m. (□) Na - 46.9 p.p.m. (▽) Na - 78.0 p.p.m. (+) Na - 180.0 p.p.m.

Table 27

Sodium and potassium concentrations (parts per million) of external medium during the 96 hour experimental period

Time of sampling	Solution 1		Solution 2		Solution 3		Solution 4		Solution 5		Solution 6	
	Na	K	Na	K	Na	K	Na	K	Na	K	Na	K
At commencement	2.8	116.9	18.6	119.9	28.8	116.9	49.9	117.9	78.0	114.8	177.3	116.9
After 1 hour	2.3	116.9	13.0	115.8	27.8	116.9	48.0	114.8	76.6	114.8	175.4	114.8
After 2 hours	1.9	112.8	13.0	110.7	27.8	114.8	48.3	113.8	76.1	113.8	174.0	113.8
After 8 hours	2.8	110.7	13.0	107.1	27.6	110.7	46.9	110.7	74.7	113.8	169.0	113.7
After 24 hours	2.1	99.4	13.0	101.5	25.5	105.6	45.5	109.7	73.8	115.8	170.8	118.9
After 48 hours	1.9	95.3	12.1	97.4	24.1	100.5	44.5	114.8	76.1	129.2	174.5	127.1
After 96 hours	2.8	102.5	20.0	94.3	24.1	96.4	40.3	123.0	81.7	139.4	177.2	131.2

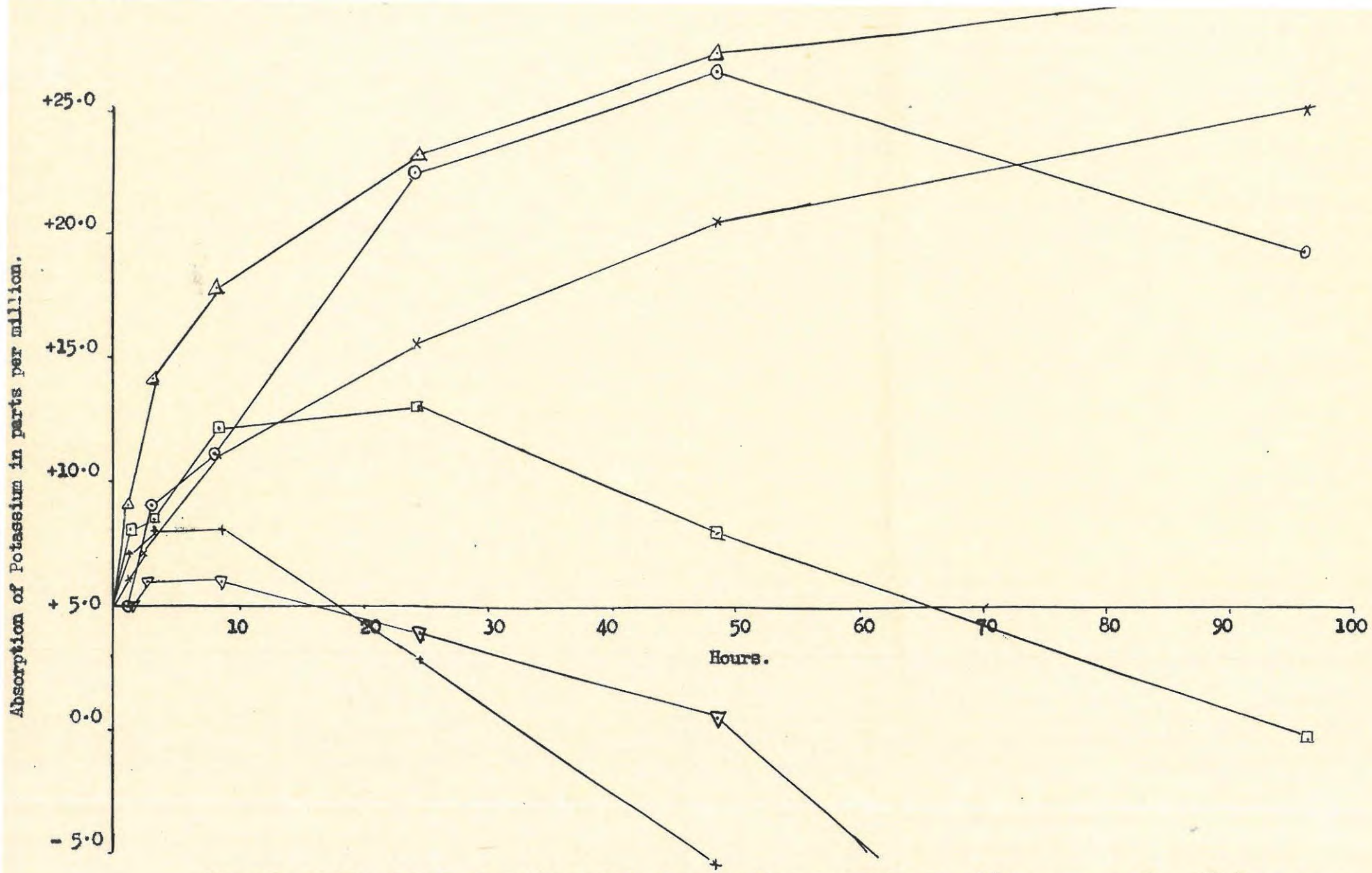
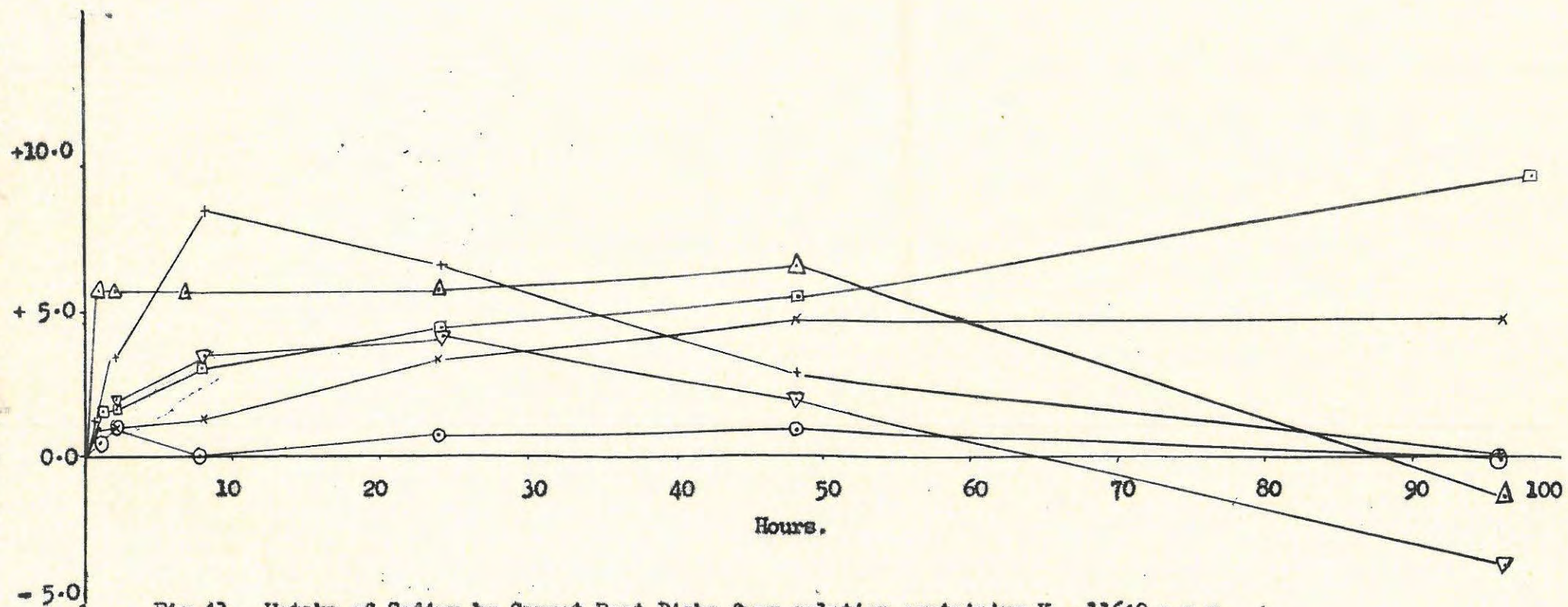


Fig. 42. Uptake of K. by Carrot Root Disks from solutions containing K - 116.9 p.p.m. + Ca - 200.0 p.p.m. + Mg - 50.0 p.p.m. + (○) Na - 2.8 p.p.m. (△) Na - 18.6 p.p.m. (×) Na - 28.8 p.p.m. (□) Na - 49.9 p.p.m. (▽) Na - 78.0 p.p.m. (+) Na - 177.3 p.p.m.

Absorption of Sodium in parts per million.



**Fig. 43.** Uptake of Sodium by Carrot Root Disks from solution containing K - 116.9 p.p.m. + Ca - 200.0 p.p.m. + Mg - 50.0 p.p.m. + (o) Na - 2.8 p.p.m. (Δ) Na - 18.6 p.p.m. (x) Na - 28.8 p.p.m. (□) Na - 49.9 p.p.m. (▽) Na - 78.0 p.p.m. (+) Na - 177.3 p.p.m.

In this experiment there was a larger amount of sodium than potassium absorbed by the carrot disks, from a solution containing approximately equal concentrations of both cations.

#### Experiment 4

In the fourth experiment the experimental solutions had a potassium concentration of 70.0 p.p.m. and sodium was supplied at six different levels ranging from 14.0 p.p.m. to 890.0 p.p.m., this being a wider range of sodium concentrations than used in the three previous experiments. Calcium was supplied at 156.0 p.p.m., together with 50.0 p.p.m. magnesium. In this and the two following experiments, the absorption of calcium, as well as of sodium and potassium by the root tissue was followed over the experimental period, the results being presented in Table 28.

The course of the absorption of potassium by the carrot tissue is presented in Fig.44. The concentration of sodium in the solutions bathing the storage tissue was shown to affect the potassium absorption. The root tissue disks in the solution containing the lowest concentration of sodium, absorbed the largest amount of potassium during the ninety six hours. As the concentration of sodium in the experimental solutions was increased, so the absorption of potassium was shown to decrease. The carrot tissue disks in the solution with the highest sodium concentration were found to absorb potassium to a very small extent during the initial two hours, and after this period there was a diffusion of potassium out of the tissue into the experimental solutions.

The uptake of sodium was shown to be dependent upon the concentration of this ion in the external solution. The storage tissue disks in the solution with the lowest concentration of sodium were shown to lose sodium by diffusion out of the tissue, and as the sodium concentrations in the external media were increased so were both the initial 'passive' and 'active' uptakes found to increase, see Fig.45.

The calcium absorption by the disks of tissue was shown to be small in comparison to the uptake of the other two ions, the general trend being for the calcium uptake to decrease as the sodium concentrations in the external solutions were raised see Fig. 46.

Table 28

Sodium, potassium and calcium concentrations (parts per million) in external medium during 96 hour experimental period

	Solution 1			Solution 2			Solution 3			Solution 4			Solution 5			Solution 6		
	Na	K	Ca	Na	K	Ca	Na	K	Ca	Na	K	Ca	Na	K	Ca	Na	K	Ca
At commencement	14.0	79.6	146.0	44.0	74.0	164.0	100.0	72.0	160.0	185.0	74.0	165.0	370.0	81.0	173.0	890.0	90.0	176.0
After 1 hour	15.0	74.6	140.0	44.0	76.6	154.0	96.0	75.2	154.0	168.0	75.2	161.0	344.0	76.2	163.0	820.0	79.0	175.0
After 2 hours	23.0	70.0	148.0	53.0	75.6	154.0	95.0	76.8	147.0	178.0	76.6	157.0	354.0	78.8	166.0	965.0	80.0	175.0
After 4 hours	18.0	71.6	142.0	42.0	69.8	147.0	86.0	73.8	148.0	176.0	76.8	155.0	353.0	78.0	156.0	885.0	79.2	170.0
After 8 hours	18.0	64.4	140.0	49.0	62.0	146.0	94.0	72.0	146.0	166.0	72.6	148.0	336.0	78.6	157.0	840.0	83.2	174.6
After 24 hours	18.0	25.8	143.0	35.0	35.6	146.4	66.0	48.0	147.0	147.0	65.2	155.0	312.5	77.4	160.0	830.0	88.0	173.0
After 48 hours	13.5	7.4	143.0	33.0	19.0	134.0	62.0	24.4	140.0	144.0	61.6	146.0	307.0	78.6	147.0	790.0	92.8	166.6
After 72 hours	5.5	2.6	149.0	26.0	6.8	138.0	45.0	13.2	146.0	137.0	61.6	146.0	289.0	75.6	160.0	870.0	95.6	173.0
After 96 hours	14.0	7.4	152.0	23.0	5.6	164.0	36.0	8.8	147.0	110.0	45.6	145.6	266.0	72.6	160.0	835.0	101.2	187.4

+100.0

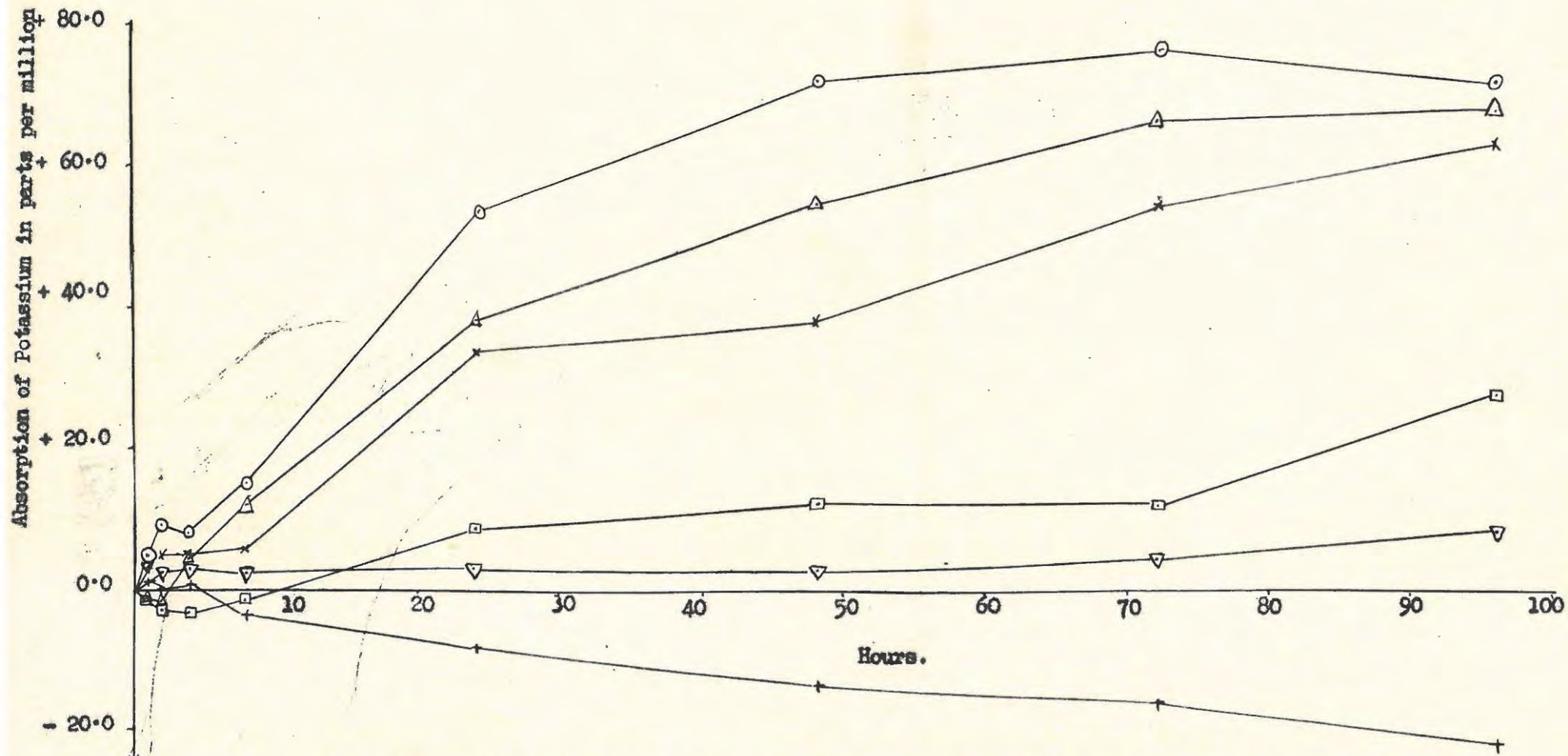


Fig. 44. Uptake of Potassium by Carrot Root Disks from a solution containing K - 70.0 p.p.m. Ca - 156.0 p.p.m. + Mg - 50.0 p.p.m. + (○) Na 14.0 p.p.m. (△) Na 44.0 p.p.m. (×) Na 100.0 p.p.m. (□) Na 185.0 p.p.m. (▽) Na 370.0 p.p.m. (+) Na 890.0 p.p.m.

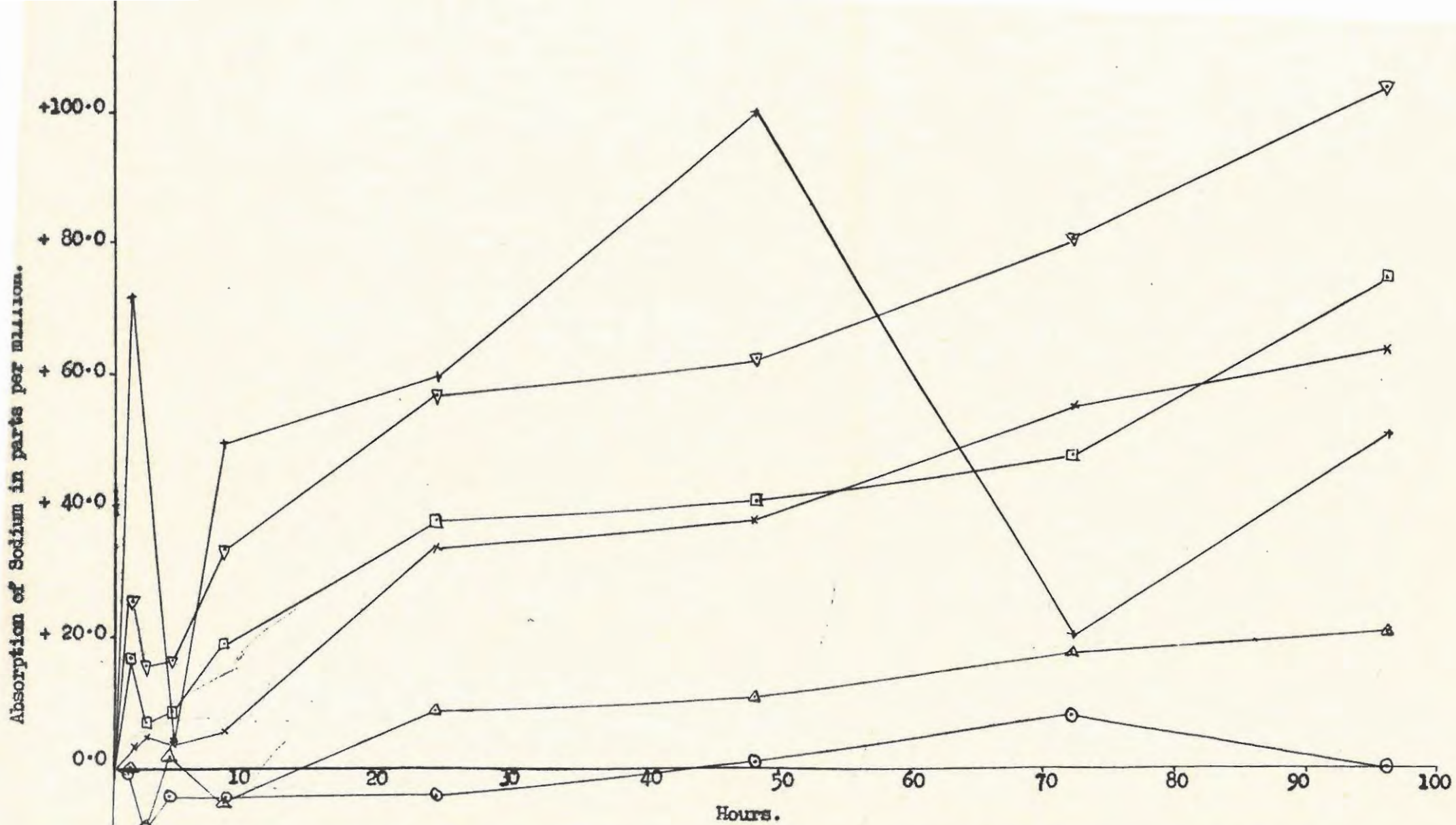


Fig. 45. Uptake of Sodium by Carrot Root Disks from solution containing K-70.0 p.p.m. + Ca-156.0 p.p.m. + Mg-50.0 p.p.m. + (O) Na-14.0 p.p.m. (Δ) Na-44.0 p.p.m. (X) Na-100.0 p.p.m. (□) Na-185.0 p.p.m. (▽) Na-370.0 p.p.m. (+) Na-890.0 p.p.m.

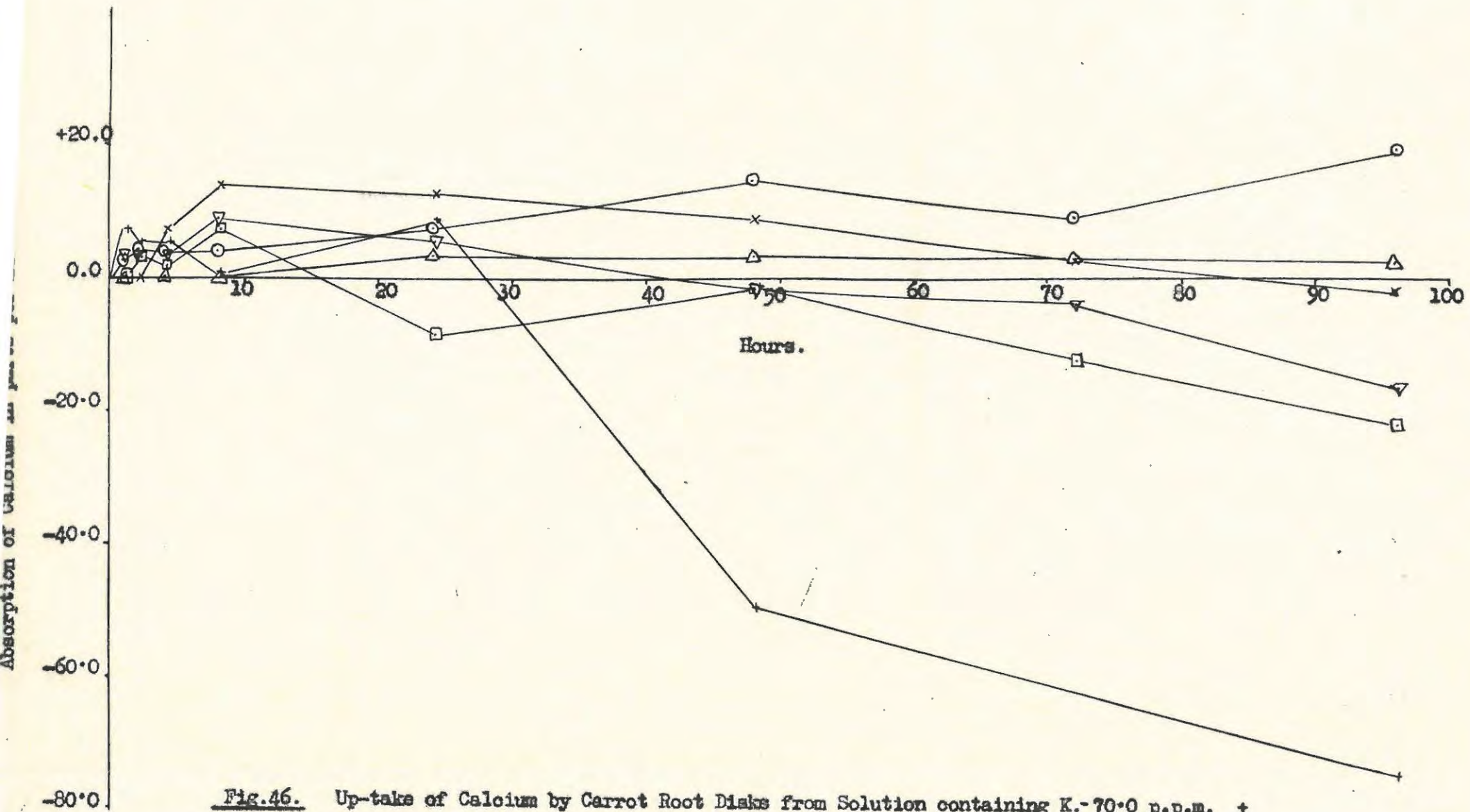


Fig.46. Up-take of Calcium by Carrot Root Disks from Solution containing K.-70.0 p.p.m. + Ca - 156.0 p.p.m. + Mg - 50.0 p.p.m. + (○) Na-14.0 p.p.m. (△) Na-44.0 p.p.m. (×) Na-100.0 p.p.m. (◻) Na-185.0 p.p.m. (▽) Na-370.0 p.p.m. (+) 890.0 p.p.m.

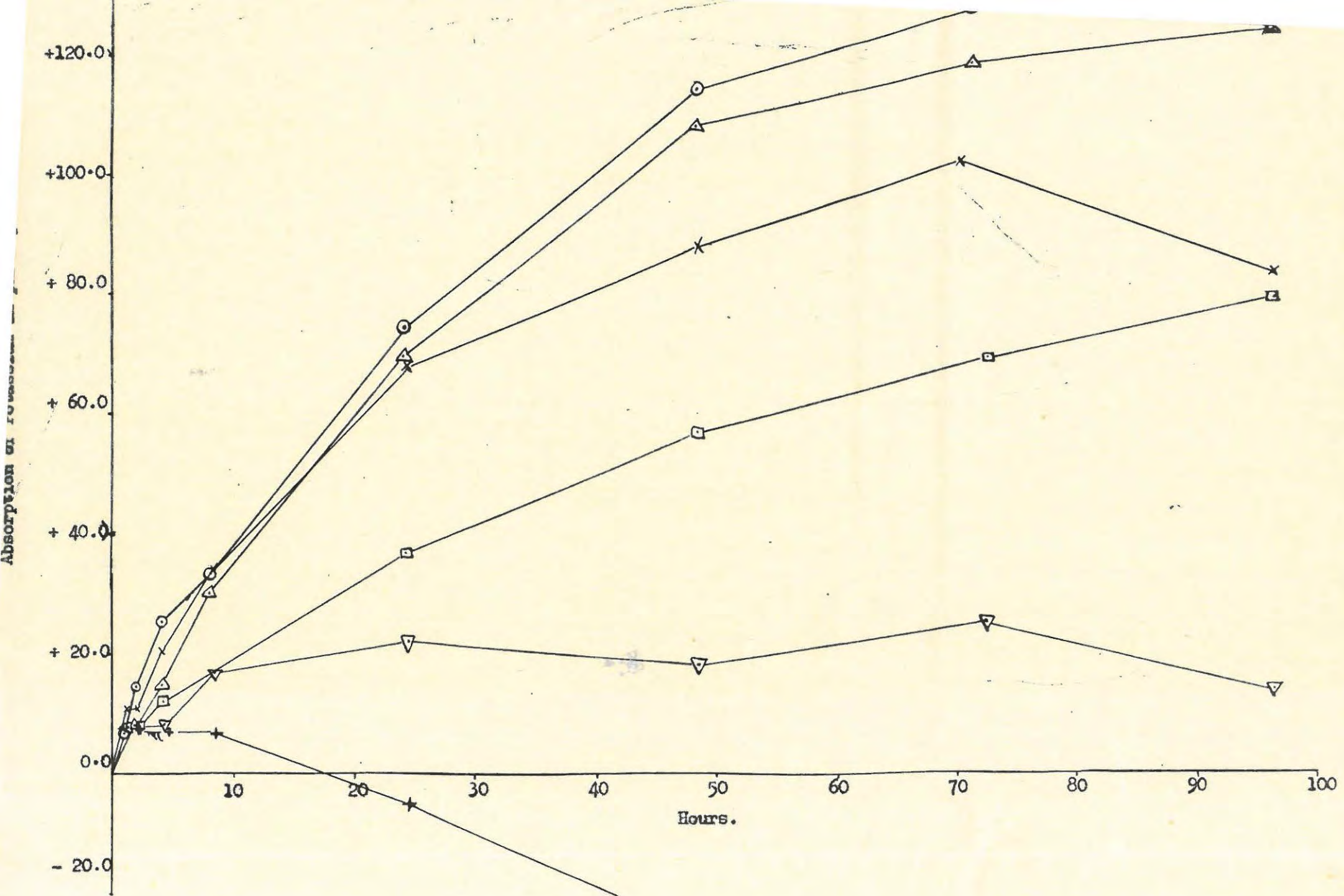
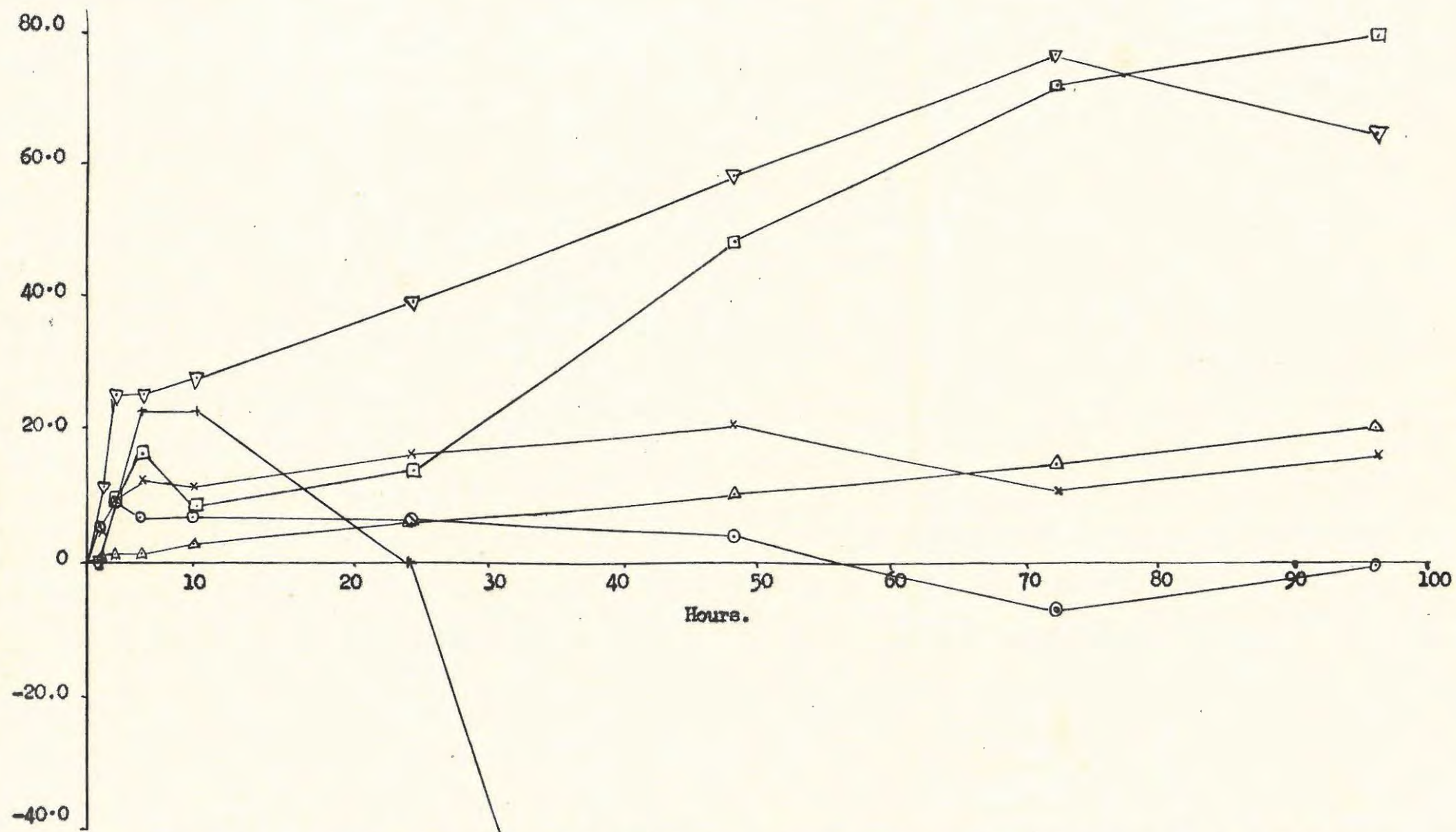


Fig. 47. Uptake of Potassium by Carrot Root Disks from solution containing K: 140.0 p.p.m. + Ca - 152.0 p.p.m. + Mg - 50.0 p.p.m. + (○) Na - 16.3 p.p.m. (△) Na - 38.3 p.p.m. (□) Na - 185.7 p.p.m. (▽) Na - 359.8 p.p.m. (+) Na - 881.9 p.p.m.

absorption of Sodium in parts per million.



**Fig. 48.** Uptake of Sodium by Carrot Root Disks from solution containing K - 140.0 p.p.m. + Ca - 152.0 p.p.m. + Mg - 50.0 p.p.m. + (○) Na - 16.3 p.p.m. (△) Na - 38.3 p.p.m. (×) Na - 102.1 p.p.m. (□) Na - 185.7 p.p.m. (▽) Na - 359.8 p.p.m. (+) Na - 881.9 p.p.m.

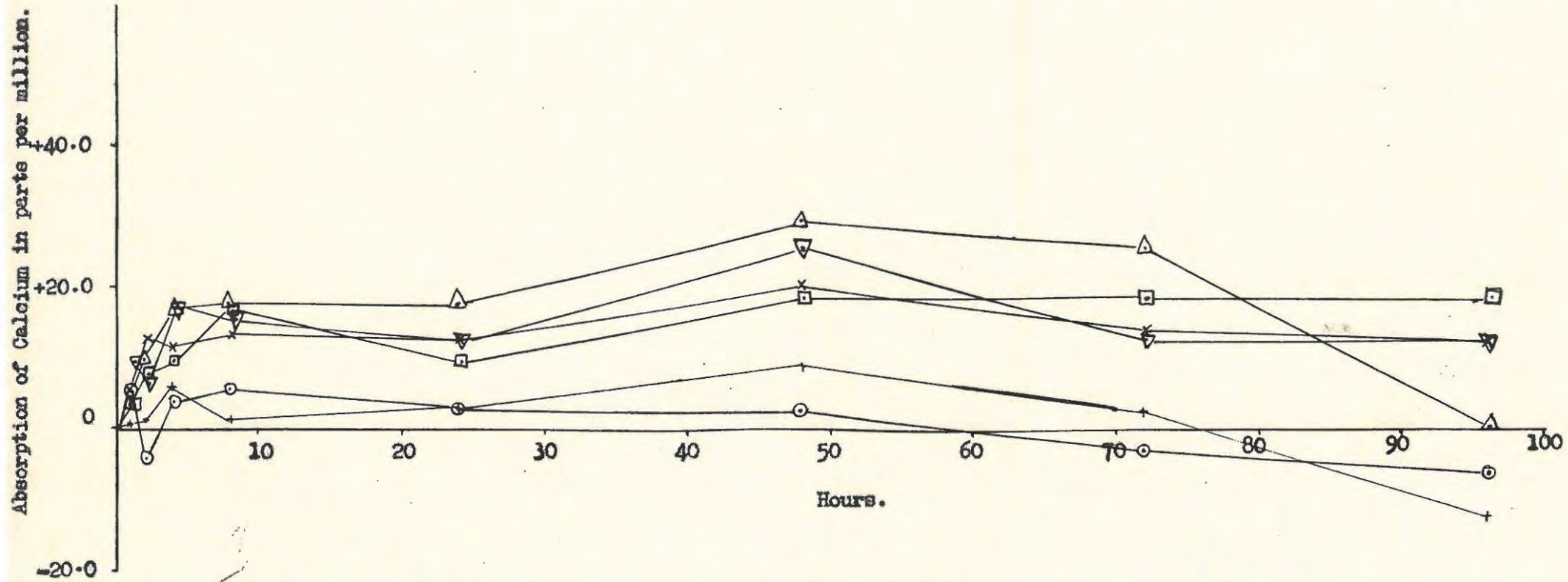


Fig. 49. Uptake of Calcium by Carrot Root Disks from solution containing K - 140.0 p.p.m. + Ca - 152.0 p.p.m. + Mg - 50.0 p.p.m. + (○) Na - 16.3 p.p.m. (△) - 38.3 p.p.m. (×) Na - 102.1 p.p.m. (□) Na - 185.7 p.p.m. (▽) Na - 359.8 p.p.m. (+) Na - 881.9 p.p.m.

magnesium. The uptake of calcium, sodium, and potassium by the carrot disks from the different experimental solutions is presented in Table 30.

The amount of potassium absorbed by the storage tissue was shown to be markedly depressed when the concentration of sodium in the external solutions was increased. High concentrations of sodium were shown to affect both the initial 'passive' uptake, as well as the 'active' uptake of potassium. The disks bathed in solutions with a low sodium concentration showed a large initial uptake of potassium, whereas the disks in a high sodium concentration showed little or no uptake of potassium during the initial period. Diffusion of potassium out of the tissue into the bathing media was shown to result from the presence of a high concentration of sodium in the solutions, see Fig.50.

The amount of sodium absorbed by the carrot disks from the external solution was shown to be dependent upon the concentration of sodium in that solution, an increased uptake occurring when the concentration of sodium in the external medium was raised. In most of the solutions there was no absorption of sodium by the disks of tissue during the initial few hours of the experiment. This differed from the sodium uptake pattern of the previous experiments. Only in the solutions with the two highest sodium concentrations was sodium found to be absorbed during the initial phase. The tissue disks bathed in the solution with the highest sodium concentration were found to have a rapid uptake of sodium during the first twenty four hours, but after this period sodium was shown to diffuse out of the carrot disks into the solution, see Fig.51.

The amount of calcium absorbed by the carrot root tissue was shown to be small in comparison to the uptake of the other two cations studied. There was a general trend for the calcium uptake by the tissue to decrease as the sodium concentration in the external solution increased. In the solution with the highest sodium level there was no uptake of calcium at all, and from the very commencement calcium diffused out of the tissue into the external solution.

Table 30

Sodium potassium and calcium concentrations (parts per million) in external medium during the 96 hour experimental period

Time of sampling	Solution 1			Solution 2			Solution 3			Solution 4			Solution 5			Solution 6		
	Na	K	Ca	Na	K	Ca	Na	K	Ca	Na	K	Ca	Na	K	Ca	Na	K	Ca
At commencement	13.0	439.0	160.0	50.0	438.0	160.0	103.0	437.0	165.0	187.0	434.0	168.0	380.0	442.0	166.0	900.0	440.0	174.0
After 1 hour	13.0	437.0	160.0	50.0	430.0	160.0	105.0	419.0	160.0	189.0	437.0	164.0	372.0	442.0	164.0	930.0	441.0	178.0
After 2 hours	18.0	436.0	160.0	52.0	434.0	160.0	108.0	402.0	163.0	188.0	423.0	163.0	380.0	442.0	160.0	885.0	440.0	182.0
After 8 hours	21.0	402.0	160.0	58.0	409.0	160.0	104.0	395.0	160.0	183.0	402.0	161.0	368.0	434.0	166.0	1075.0	448.0	180.0
After 24 hours	19.0	398.0	160.0	51.0	378.0	160.0	93.0	378.0	160.0	175.0	403.0	166.0	338.0	426.0	167.0	835.0	457.0	185.0
After 48 hours	14.0	363.0	162.0	42.0	345.0	170.0	92.0	362.0	128.0	160.0	394.0	151.0	318.0	430.0	164.8	920.0	488.0	174.0
After 96 hours	20.0	408.0	185.0	45.0	436.0	186.0	95.0	376.0	150.0	158.0	478.0	179.0	308.0	494.0	173.0	885.0	630.0	202.0

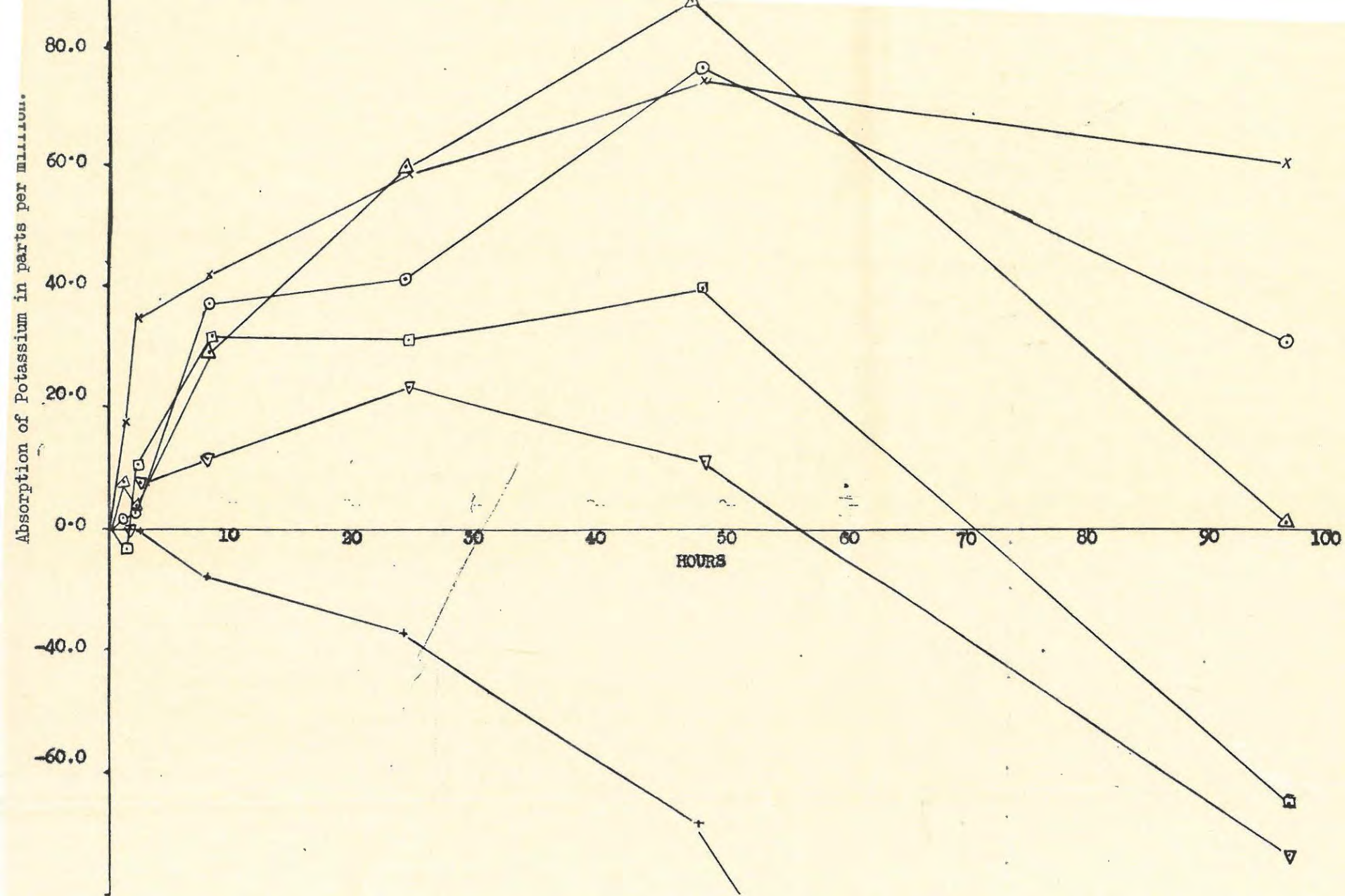
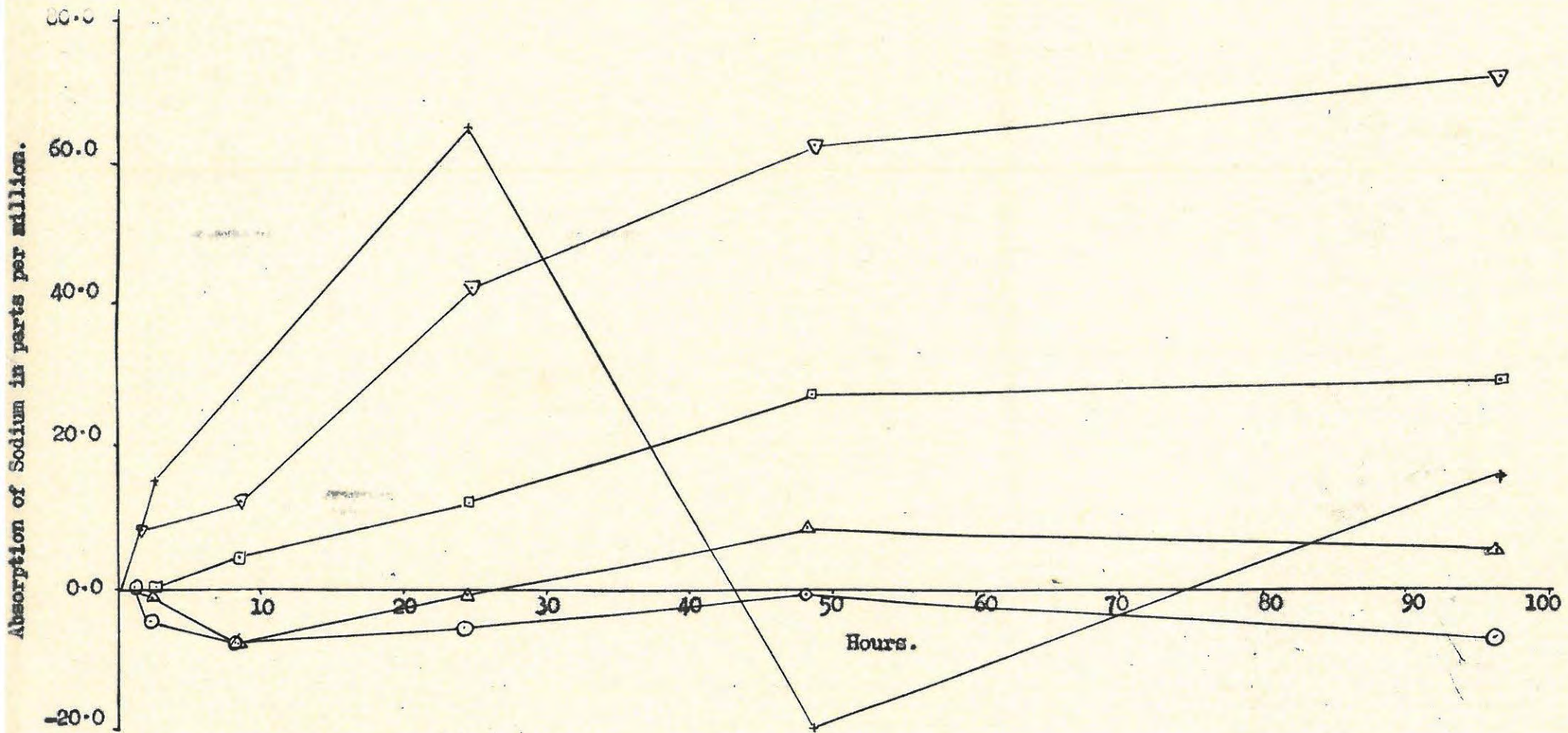


Fig. 50. Uptake of Potassium by Carrot Root Disks from solutions containing K.- 430.0 p.p.m. + Ca-160.0 p.p.m. + Mg-50.0 p.p.m. + (○) Na - 13.0 p.p.m. (△) Na - 50.0 p.p.m. (x) Na - 103.0 p.p.m. (□) Na - 187.0 p.p.m. (▽) Na - 380 p.p.m. (+) Na - 900.0 p.p.m.



**Fig. 51.** Uptake of Sodium by Carrot Root Disks from solution containing K - 430.0 p.p.m. + Ca 160.0 p.p.m. + Mg 50.0 p.p.m. + (○) Na - 13.0 p.p.m. (△) Na 50.0 p.p.m. (x) Na - 103.0 p.p.m. (□) Na - 187.0 p.p.m. (▽) Na - 380.0 p.p.m. (+) Na - 900.0 p.p.m.

### Chapter III

#### Discussion and Conclusions

It has been generally accepted in the past few years that ion absorption by plant material is the sum of two processes namely, (1) 'Passive', or non-metabolic uptake which may be due to cation exchange and diffusion, and, (2) 'Active' transport which is a metabolic process. These two absorption processes have been reviewed in detail by Epstein (1956).

In the experiments carried out in the present investigation the uptake of the cations, both by the potato tuber tissue, and the carrot root tissue showed the two phases of uptake. The first phase being a rapid uptake of cations during the initial two hours, followed by a slower but more stable absorption lasting many hours. The above cation uptake pattern has been attested in investigations by Steward and Harrison (1939), Epstein and Leggett (1954), Hanson and Bonner (1954) and Sutcliffe (1957), and evidence has suggested the non-metabolic uptake to be due largely to cation exchange. Epstein and Leggett (1954) have shown the processes of ion uptake to differ in attributes relating to kinetics, reversibility, ion specificity, and energy requirements, and thus from their evidence it would appear that the exchange spots are not identical with the entities involved in active transport.

In the experiments carried out in the present study, the potassium uptake during the first two hours, was generally found to be unaffected by the level of sodium concentration present in the external media, suggesting there was no competition between the two cations during the non-metabolic phase of uptake by the tissue. The extent of sodium uptake during the same period was shown to increase as the levels of sodium concentration in the external media were raised. The results obtained from the present investigation suggest that the cation uptake during the initial phase is non-selective, and dependent upon the cation concentration in the external media.

There appears to be wide agreement between the present day workers in this field, that active transport requires the operation of "carriers". The essential features of the functioning "carriers" are the attachment of the ions to the carrier molecules, and the movement of the resulting

carrier ion complexes through some barrier intervening between an "outer" and "inner" phase.

From the results of the experiment in which potato tuber tissue was used it was shown that the active absorption of potassium was stimulated as the concentration of sodium was increased. Further when the two cations were mixed in approximately equal proportions, the uptake of potassium considerably exceeded that of sodium, suggesting that the transport system in potato tuber tissue exhibits discrimination with a preference for potassium when a choice is available. The results of the experiments with carrot root tissue, differed from those with potato tuber tissue in that the potassium uptake was found to be reduced when the level of the sodium concentration in the external media was increased. The sodium uptake was shown to be much reduced when the potassium concentration level was increased in the external solution. These results suggest that competition occurs between these cations for absorption.

The selective uptake of cations by plant tissues may be explained on the basis of a carrier hypothesis as suggested by Sutcliffe (1956), in one of two ways. Either (a) there are separate types of carrier molecules for each ion or group of ions, when selectivity depends on the relative numbers and activities of the different sites, or (b) there is a single type of carrier capable of transporting all or a number of ions, but exhibiting a distinct preference when choice is available.

The results of the absorption of sodium by both the potato tuber tissue and carrot root tissue, show the amount of sodium absorbed by the tissue during the ninety six hours was dependent upon the initial concentration in the external media. The results from experiments 2, 4, 5, and 6 present evidence to suggest that the rate of absorption during the first twenty four hours was also dependent upon the concentration of sodium in the external medium. After this time period a steady rate of uptake appeared to follow, irrespective of the external concentration. The disks of tissue bathed in solutions containing high concentrations of sodium and potassium showed a leakage of cations out of the tissue, into the external solution as a result of plasmolysis. In these instances the metabolic absorption by the disks was shown to suffer interference.

The results from the experiments in the present investigation

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substantiate the findings of Sutcliffe (1956) that cation uptake by plant tissue is selective and that competition between the cations may occur. In the experiments with carrot root tissue competition between sodium and potassium, and sodium and calcium has been shown, this phenomenon may be best explained on the basis of the single mechanism hypothesis described above.

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APPENDIX I

CHEMICAL COMPOSITION OF THE EXPERIMENTAL SOLUTIONS USED

IN

PART II

The following tables give details of the composition of the experimental solutions used in Part I. In column I, the ions present in each solution are listed, column II, the weight of each ion per litre of nutrient solution, column III the salt used to supply the required ion, and column IV the amount of stock solution added to 1 litre of water in the preparation of the nutrient solution.

<u>SOLUTION 1</u>	<u>Element</u>	<u>Wt in Sol per litre</u>	<u>Salt Used</u>	<u>Quantity</u>
	Cl	0.18	NH <sub>4</sub> Cl	5.0 ml
	N	0.07	NH <sub>4</sub> Cl	5.0 ml
	Ca	0.16	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
	N	0.11	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
	P	0.15	H <sub>3</sub> PO <sub>4</sub>	13.0 ml
	Mg	0.10	MgSO <sub>4</sub>	4.0 ml
	S	0.13	MgSO <sub>4</sub>	4.0 ml
	N	1.02	NH <sub>4</sub> NO <sub>3</sub>	36.5 ml

SOLUTION 2

Cl	0.18	NH <sub>4</sub> Cl	5.0 ml
N	0.07	NH <sub>4</sub> Cl	5.0 ml
Ca	0.16	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
N	0.11	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
K	0.12	KNO <sub>3</sub>	3.0 ml
N	0.04	KNO <sub>3</sub>	3.0 ml
P	0.15	H <sub>3</sub> PO <sub>4</sub>	13.0 ml
Mg	0.10	MgSO <sub>4</sub>	4.0 ml
S	0.13	MgSO <sub>4</sub>	4.0 ml
N	0.98	NH <sub>4</sub> NO <sub>3</sub>	35.0 ml

SOLUTION 3

Cl	0.18	NH <sub>4</sub> Cl	5.0 ml
N	0.07	NH <sub>4</sub> Cl	5.0 ml
Ca	0.16	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
N	0.11	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
K	0.25	KNO <sub>3</sub>	6.5 ml
N	0.09	KNO <sub>3</sub>	6.5 ml
P	0.15	H <sub>3</sub> PO <sub>4</sub>	13.0 ml
Mg	0.10	MgSO <sub>4</sub>	4.0 ml
S	0.13	MgSO <sub>4</sub>	4.0 ml
N	0.93	NH <sub>4</sub> NO <sub>3</sub>	33.2 ml

<u>SOLUTION 4</u>	<u>Element</u>	<u>Wt in Sol per litre</u>	<u>Salt Used</u>	<u>Quantity</u>
	Cl	0.18	NH <sub>4</sub> Cl	5.0 ml
	N	0.07	NH <sub>4</sub> Cl	5.0 ml
	Ca	0.16	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
	N	0.11	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
	K	0.50	KNO <sub>3</sub>	13.0 ml
	N	0.18	KNO <sub>3</sub>	13.0 ml
	P	0.15	H <sub>3</sub> PO <sub>4</sub>	13.0 ml
	Mg	0.10	MgSO <sub>4</sub>	4.0 ml
	S	0.13	MgSO <sub>4</sub>	4.0 ml
	N	0.84	NH <sub>4</sub> NO <sub>3</sub>	30.0 ml

SOLUTION 5

Na	0.05	NaCl	2.2 ml
Cl	0.08	NaCl	2.2 ml
Cl	0.10	NH <sub>4</sub> Cl	2.8 ml
N	0.04	NH <sub>4</sub> Cl	2.8 ml
Ca	0.16	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
N	0.11	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
P	0.15	H <sub>3</sub> PO <sub>4</sub>	13.0 ml
Mg	0.10	MgSO <sub>4</sub>	4.0 ml
S	0.13	MgSO <sub>4</sub>	4.0 ml
N	1.05	NH <sub>4</sub> NO <sub>3</sub>	ml

SOLUTION 6

Na	0.05	NaCl	2.2 ml
Cl	0.08	NaCl	2.2 ml
Cl	0.10	NH <sub>4</sub> Cl	2.8 ml
N	0.04	NH <sub>4</sub> Cl	2.8 ml
Ca	0.16	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
N	0.11	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
K	0.12	KNO <sub>3</sub>	3.0 ml
N	0.04	KNO <sub>3</sub>	3.0 ml
P	0.15	H <sub>3</sub> PO <sub>4</sub>	13.0 ml

Contd.

<u>SOLUTION 6</u>	<u>Element</u>	<u>Wt in Sol per litre</u>	<u>Salt Used</u>	<u>Quantity</u>
	Mg	0.10	MgSO <sub>4</sub>	4.0 ml
	S	0.13	MgSO <sub>4</sub>	4.0 ml
	N	1.01	NH <sub>4</sub> NO <sub>3</sub>	36.0 ml

SOLUTION 7

Na	0.05	NaCl	2.2 ml
Cl	0.08	NaCl	2.2 ml
Cl	0.10	NH <sub>4</sub> Cl	2.8 ml
N	0.04	NH <sub>4</sub> Cl	2.8 ml
Ca	0.16	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
N	0.11	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
K	0.25	KNO <sub>3</sub>	6.5 ml
N	0.09	KNO <sub>3</sub>	6.5 ml
P	0.15	H <sub>3</sub> PO <sub>4</sub>	13.0 ml
Mg	0.10	MgSO <sub>4</sub>	4.0 ml
S	0.13	MgSO <sub>4</sub>	4.0 ml
N	0.96	NH <sub>4</sub> NO <sub>3</sub>	34.3 ml

SOLUTION 8

Na	0.05	NaCl	2.2 ml
Cl	0.08	NaCl	2.2 ml
Cl	0.10	NH <sub>4</sub> Cl	2.8 ml
N	0.04	NH <sub>4</sub> Cl	2.8 ml
Ca	0.16	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
N	0.11	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
K	0.5	KNO <sub>3</sub>	13.0 ml
N	0.18	KNO <sub>3</sub>	13.0 ml
P	0.15	H <sub>3</sub> PO <sub>4</sub>	13.0 ml
Mg	0.10	MgSO <sub>4</sub>	4.0 ml
S	0.13	MgSO <sub>4</sub>	4.0 ml
N	0.87	NH <sub>4</sub> NO <sub>3</sub>	31.0 ml

<u>SOLUTION 9</u>	<u>Element</u>	<u>Wt in Sol per litre</u>	<u>Salt Used</u>	<u>Quantity</u>
	Na	0.11	NaCl	5.0 ml
	Cl	0.18	NaCl	5.0 ml
	Na	0.08	NaNO <sub>3</sub>	3.5 ml
	N	0.05	NaNO <sub>3</sub>	3.5 ml
	Ca	0.16	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
	N	0.11	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
	P	0.15	H <sub>3</sub> PO <sub>4</sub>	13.0 ml
	Mg	0.10	MgSO <sub>4</sub>	4.0 ml
	S	0.13	MgSO <sub>4</sub>	4.0 ml
	N	1.04	NH <sub>4</sub> NO <sub>3</sub>	37.2 ml

SOLUTION 10

Na	0.11	NaCl	5.0 ml
Cl	0.18	NaCl	5.0 ml
Na	0.08	NaNO <sub>3</sub>	3.5 ml
N	0.05	NaNO <sub>3</sub>	3.5 ml
Ca	0.16	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
N	0.11	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
K	0.12	KNO <sub>3</sub>	3.0 ml
N	0.04	KNO <sub>3</sub>	3.0 ml
P	0.15	H <sub>3</sub> PO <sub>4</sub>	13.0 ml
Mg	0.10	MgSO <sub>4</sub>	4.0 ml
S	0.13	MgSO <sub>4</sub>	4.0 ml
N	1.00	NH <sub>4</sub> NO <sub>3</sub>	

SOLUTION 11

Na	0.11	NaCl	5.0 ml
Cl	0.18	NaCl	5.0 ml
Na	0.08	NaNO <sub>3</sub>	3.5 ml
N	0.05	NaNO <sub>3</sub>	3.5 ml
Ca	0.16	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
N	0.11	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
K	0.25	KNO <sub>3</sub>	6.5 ml
N	0.09	KNO <sub>3</sub>	6.5 ml
P	0.15	H <sub>3</sub> PO <sub>4</sub>	13.0 ml

Contd.

<u>SOLUTION 11</u>	<u>Element</u>	<u>Wt in Sol per litre</u>	<u>Salt Used</u>	<u>Quantity</u>
	Mg	0.10	MgSO <sub>4</sub>	4.0 ml
	S	0.13	MgSO <sub>4</sub>	4.0 ml
	N	0.95	NH <sub>4</sub> NO <sub>3</sub>	34.0 ml

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SOLUTION 12

Na	0.11	NaCl	5.0 ml
Cl	0.18	NaCl	5.0 ml
Na	0.08	NaNO <sub>3</sub>	3.5 ml
N	0.05	NaNO <sub>3</sub>	3.5 ml
Ca	0.16	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
N	0.11	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
K	0.51	KNO <sub>3</sub>	13.0 ml
N	0.16	KNO <sub>3</sub>	13.0 ml
P	0.15	H <sub>3</sub> PO <sub>4</sub>	13.0 ml
Mg	0.10	MgSO <sub>4</sub>	4.0 ml
S	0.13	MgSO <sub>4</sub>	4.0 ml
N	0.85	NH <sub>4</sub> NO <sub>3</sub>	30.5 ml

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SOLUTION 13

Na	0.11	NaCl	5.0 ml
Cl	0.18	NaCl	5.0 ml
Na	0.39	NaNO <sub>3</sub>	17.0 ml
N	0.24	NaNO <sub>3</sub>	17.0 ml
Ca	0.16	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
N	0.11	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
P	0.15	H <sub>3</sub> PO <sub>4</sub>	13.0 ml
Mg	0.10	MgSO <sub>4</sub>	4.0 ml
S	0.13	MgSO <sub>4</sub>	4.0 ml
N	0.85	NH <sub>4</sub> NO <sub>3</sub>	30.5 ml

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<u>SOLUTION 14</u>	<u>Element</u>	<u>Wt in Sol per litre</u>	<u>Salt Used</u>	<u>Quantity</u>
	Na	0.11	NaCl	5.0 ml
	Cl	0.18	NaCl	5.0 ml
	Na	0.39	NaNO <sub>3</sub>	17.0 ml
	N	0.24	NaNO <sub>3</sub>	17.0 ml
	Ca	0.16	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
	N	0.11	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
	K	0.12	KNO <sub>3</sub>	3.0 ml
	N	0.04	KNO <sub>3</sub>	3.0 ml
	P	0.15	H <sub>3</sub> PO <sub>4</sub>	13.0 ml
	Mg	0.10	MgSO <sub>4</sub>	4.0 ml
	S	0.13	MgSO <sub>4</sub>	4.0 ml
	N	0.81	NH <sub>4</sub> NO <sub>3</sub>	29.0 ml

<u>SOLUTION 15</u>	<u>Element</u>	<u>Wt in Sol per litre</u>	<u>Salt Used</u>	<u>Quantity</u>
	Na	0.11	NaCl	5.0 ml
	Cl	0.18	NaCl	5.0 ml
	Na	0.39	NaNO <sub>3</sub>	17.0 ml
	N	0.24	NaNO <sub>3</sub>	17.0 ml
	Ca	0.16	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
	N	0.11	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
	K	0.25	KNO <sub>3</sub>	6.5 ml
	N	0.09	KNO <sub>3</sub>	6.5 ml
	P	0.15	H <sub>3</sub> PO <sub>4</sub>	13.0 ml
	Mg	0.10	MgSO <sub>4</sub>	4.0 ml
	S	0.13	MgSO <sub>4</sub>	4.0 ml
	N	0.76	NH <sub>4</sub> NO <sub>3</sub>	27.2 ml

<u>SOLUTION 16</u>	<u>Element</u>	<u>Wt in Sol per litre</u>	<u>Salt Used</u>	<u>Quantity</u>
	Na	0.11	NaCl	5.0 ml
	Cl	0.18	NaCl	5.0 ml
	Na	0.39	NaNO <sub>3</sub>	17.0 ml
	N	0.24	NaNO <sub>3</sub>	17.0 ml
	Ca	0.16	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
	N	0.11	Ca(NO <sub>3</sub> ) <sub>2</sub>	4.0 ml
	K	0.51	KNO <sub>3</sub>	13.0 ml
	N	0.18	KNO <sub>3</sub>	13.0 ml
	P	0.15	H <sub>3</sub> PO <sub>4</sub>	13.0 ml
	Mg	0.10	MgSO <sub>4</sub>	4.0 ml
	S	0.13	MgSO <sub>4</sub>	4.0 ml
	N	0.67	NH <sub>4</sub> NO <sub>3</sub>	24.0 ml

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SOLUTION 17

Cl	0.18	NH <sub>4</sub> Cl	5.0 ml
N	0.07	NH <sub>4</sub> Cl	5.0 ml
Ca	0.48	Ca(NO <sub>3</sub> ) <sub>2</sub>	12.0 ml
N	0.34	Ca(NO <sub>3</sub> ) <sub>2</sub>	12.0 ml
P	0.15	H <sub>3</sub> PO <sub>4</sub>	13.0 ml
Mg	0.10	MgSO <sub>4</sub>	4.0 ml
S	0.13	MgSO <sub>4</sub>	4.0 ml
N	0.80	NH <sub>4</sub> NO <sub>3</sub>	28.5 ml

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SOLUTION 18

Cl	0.18	NH <sub>4</sub> Cl	5.0 ml
N	0.07	NH <sub>4</sub> Cl	5.0 ml
Ca	0.48	Ca(NO <sub>3</sub> ) <sub>2</sub>	12.0 ml
N	0.34	Ca(NO <sub>3</sub> ) <sub>2</sub>	12.0 ml
K	0.12	KNO <sub>3</sub>	3.0 ml
N	0.04	KNO <sub>3</sub>	3.0 ml
P	0.15	H <sub>3</sub> PO <sub>4</sub>	13.0 ml
Mg	0.10	MgSO <sub>4</sub>	4.0 ml
S	0.13	MgSO <sub>4</sub>	4.0 ml
N	0.76	NH <sub>4</sub> NO <sub>3</sub>	27.0 ml

<u>SOLUTION 19</u>	<u>Element</u>	<u>Wt in Sol per litre</u>	<u>Salt Used</u>	<u>Quantity</u>
	Cl	0.18	NH <sub>4</sub> Cl	5.0 ml
	N	0.07	NH <sub>4</sub> Cl	5.0 ml
	Ca	0.48	Ca(NO <sub>3</sub> ) <sub>2</sub>	12.0 ml
	N	0.34	Ca(NO <sub>3</sub> ) <sub>2</sub>	12.0 ml
	K	0.25	KNO <sub>3</sub>	6.5 ml
	N	0.09	KNO <sub>3</sub>	6.5 ml
	P	0.15	H <sub>3</sub> PO <sub>4</sub>	13.0 ml
	Mg	0.10	MgSO <sub>4</sub>	4.0 ml
	S	0.13	MgSO <sub>4</sub>	4.0 ml
	N	0.71	NH <sub>4</sub> NO <sub>3</sub>	25.2 ml

SOLUTION 20

Cl	0.18	NH <sub>4</sub> Cl	5.0 ml
N	0.07	NH <sub>4</sub> Cl	5.0 ml
Ca	0.48	Ca(NO <sub>3</sub> ) <sub>2</sub>	12.0 ml
N	0.34	Ca(NO <sub>3</sub> ) <sub>2</sub>	12.0 ml
K	0.51	KNO <sub>3</sub>	13.0 ml
N	0.18	KNO <sub>3</sub>	13.0 ml
P	0.15	H <sub>3</sub> PO <sub>4</sub>	13.0 ml
Mg	0.10	MgSO <sub>4</sub>	4.0 ml
S	0.13	MgSO <sub>4</sub>	4.0 ml
N	0.62	NH <sub>4</sub> NO <sub>3</sub>	22.0 ml

SOLUTION 21

Na	0.05	NaCl	2.2 ml
Cl	0.08	NaCl	2.2 ml
Cl	0.10	NH <sub>4</sub> Cl	2.8 ml
N	0.04	NH <sub>4</sub> Cl	2.8 ml
Ca	0.48	Ca(NO <sub>3</sub> ) <sub>2</sub>	12.0 ml
N	0.34	Ca(NO <sub>3</sub> ) <sub>2</sub>	12.0 ml
P	0.15	H <sub>3</sub> PO <sub>4</sub>	13.0 ml
Mg	0.10	MgSO <sub>4</sub>	4.0 ml
S	0.13	MgSO <sub>4</sub>	4.0 ml
N	0.83	NH <sub>4</sub> NO <sub>3</sub>	29.5 ml

<u>SOLUTION 22</u>	<u>Element</u>	<u>Wt in Sol per litre</u>	<u>Salt Used</u>	<u>Quantity</u>
	Na	0.05	NaCl	2.2 ml
	Cl	0.08	NaCl	2.2 ml
	Cl	0.10	NH <sub>4</sub> Cl	2.8 ml
	N	0.04	NH <sub>4</sub> Cl	2.8 ml
	Ca	0.48	Ca(NO <sub>3</sub> ) <sub>2</sub>	12.0 ml
	N	0.34	Ca(NO <sub>3</sub> ) <sub>2</sub>	12.0 ml
	K	0.12	KNO <sub>3</sub>	3.0 ml
	N	0.04	KNO <sub>3</sub>	3.0 ml
	P	0.15	H <sub>3</sub> PO <sub>4</sub>	13.0 ml
	Mg	0.10	MgSO <sub>4</sub>	4.0 ml
	S	0.13	MgSO <sub>4</sub>	4.0 ml
	N	0.78	NH <sub>4</sub> NO <sub>3</sub>	28.0 ml

SOLUTION 23

Na	0.05	NaCl	2.2 ml
Cl	0.08	NaCl	2.2 ml
Cl	0.10	NH <sub>4</sub> Cl	2.8 ml
N	0.04	NH <sub>4</sub> Cl	2.8 ml
Ca	0.48	Ca(NO <sub>3</sub> ) <sub>2</sub>	12.0 ml
N	0.34	Ca(NO <sub>3</sub> ) <sub>2</sub>	12.0 ml
K	0.25	KNO <sub>3</sub>	6.5 ml
N	0.09	KNO <sub>3</sub>	6.5 ml
P	0.15	H <sub>3</sub> PO <sub>4</sub>	13.0 ml
Mg	0.10	MgSO <sub>4</sub>	4.0 ml
S	0.13	MgSO <sub>4</sub>	4.0 ml
N	0.74	NH <sub>4</sub> NO <sub>3</sub>	26.3 ml

<u>SOLUTION 24</u>	<u>Element</u>	<u>wt in Sol per litre</u>	<u>Salt Used</u>	<u>Quantity</u>
	Na	0.05	NaCl	2.2 ml
	Cl	0.08	NaCl	2.2 ml
	Cl	0.10	NH <sub>4</sub> Cl	2.8 ml
	N	0.04	NH <sub>4</sub> Cl	2.8 ml
	Ca	0.48	Ca(NO <sub>3</sub> ) <sub>2</sub>	12.0 ml
	N	0.34	Ca(NO <sub>3</sub> ) <sub>2</sub>	12.0 ml
	K	0.51	KNO <sub>3</sub>	13.0 ml
	N	0.18	KNO <sub>3</sub>	13.0 ml
	P	0.15	H <sub>3</sub> PO <sub>4</sub>	13.0 ml
	Mg	0.10	MgSO <sub>4</sub>	4.0 ml
	S	0.13	MgSO <sub>4</sub>	4.0 ml
	N	0.66	NH <sub>4</sub> NO <sub>3</sub>	23.5 ml

SOLUTION 25

Na	0.11	NaCl	5.0 ml
Cl	0.18	NaCl	5.0 ml
Na	0.08	NaNO <sub>3</sub>	3.5 ml
N	0.05	NaNO <sub>3</sub>	3.5 ml
Ca	0.48	Ca(NO <sub>3</sub> ) <sub>2</sub>	12.0 ml
N	0.34	Ca(NO <sub>3</sub> ) <sub>2</sub>	12.0 ml
P	0.15	H <sub>3</sub> PO <sub>4</sub>	13.0 ml
Mg	0.10	MgSO <sub>4</sub>	4.0 ml
S	0.13	MgSO <sub>4</sub>	4.0 ml
N	0.81	NH <sub>4</sub> NO <sub>3</sub>	29.0 ml

SOLUTION 26

Na	0.11	NaCl	5.0 ml
Cl	0.18	NaCl	5.0 ml
Na	0.08	NaNO <sub>3</sub>	3.5 ml
N	0.05	NaNO <sub>3</sub>	3.5 ml
Ca	0.48	Ca(NO <sub>3</sub> ) <sub>2</sub>	12.0 ml
N	0.34	Ca(NO <sub>3</sub> ) <sub>2</sub>	12.0 ml
K	0.12	KNO <sub>3</sub>	3.0 ml
N	0.04	KNO <sub>3</sub>	3.0 ml

<u>SOLUTION 26</u>	<u>Element</u>	<u>Wt in Sol per litre</u>	<u>Salt Used</u>	<u>Quantity</u>
	P	0.15	$H_3PO_4$	13.0 ml
	Mg	0.10	$MgSO_4$	4.0 ml
	S	0.13	$MgSO_4$	4.0 ml
	N	0.78	$NH_4NO_3$	28.0 ml

SOLUTION 27

Na	0.11	NaCl	5.0 ml
Cl	0.18	NaCl	5.0 ml
Na	0.08	$NaNO_3$	3.5 ml
N	0.05	$NaNO_3$	3.5 ml
Ca	0.48	$Ca(NO_3)_2$	12.0 ml
N	0.34	$Ca(NO_3)_2$	12.0 ml
K	0.25	$KNO_3$	6.5 ml
N	0.09	$KNO_3$	6.5 ml
P	0.15	$H_3PO_4$	13.0 ml
Mg	0.10	$MgSO_4$	4.0 ml
S	0.13	$MgSO_4$	4.0 ml
N	0.73	$NH_3NO_3$	26.0 ml

SOLUTION 28

Na	0.11	NaCl	5.0 ml
Cl	0.12	NaCl	5.0 ml
Na	0.08	$NaNO_3$	3.5 ml
N	0.05	$NaNO_3$	3.5 ml
Ca	0.48	$Ca(NO_3)_2$	12.0 ml
N	0.34	$Ca(NO_3)_2$	12.0 ml
K	0.51	$KNO_3$	13.0 ml
N	0.18	$KNO_3$	13.0 ml
P	0.15	$H_3PO_4$	13.0 ml
Mg	0.10	$MgSO_4$	4.0 ml
S	0.13	$MgSO_4$	4.0 ml
N	0.63	$NH_4NO_3$	22.5 ml

<u>SOLUTION 29</u>	<u>Element</u>	<u>Wt. in Sol. per litre</u>	<u>Salt Used</u>	<u>Quantity</u>
	Na	0.11	NaCl	5.0 ml
	Cl	0.18	NaCl	5.0 ml
	Na	0.39	NaNO <sub>3</sub>	17.0 ml
	N	0.24	NaNO <sub>3</sub>	17.0 ml
	Ca	0.48	Ca(NO <sub>3</sub> ) <sub>2</sub>	12.0 ml
	N	0.34	Ca(NO <sub>3</sub> ) <sub>2</sub>	12.0 ml
	P	0.15	H <sub>3</sub> PO <sub>4</sub>	13.0 ml
	Mg	0.10	MgSO <sub>4</sub>	4.0 ml
	S	0.13	MgSO <sub>4</sub>	4.0 ml
	N	0.62	NH <sub>4</sub> NO <sub>3</sub>	22.0 ml

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SOLUTION 30

Na	0.11	NaCl	5.0 ml
Cl	0.18	NaCl	5.0 ml
Na	0.39	NaNO <sub>3</sub>	17.0 ml
N	0.24	NaNO <sub>3</sub>	17.0 ml
Ca	0.48	Ca(NO <sub>3</sub> ) <sub>2</sub>	12.0 ml
N	0.34	Ca(NO <sub>3</sub> ) <sub>2</sub>	12.0 ml
K	0.12	KNO <sub>3</sub>	3.0 ml
N	0.04	KNO <sub>3</sub>	3.0 ml
P	0.15	H <sub>3</sub> PO <sub>4</sub>	13.0 ml
Mg	0.10	MgSO <sub>4</sub>	4.0 ml
S	0.13	MgSO <sub>4</sub>	4.0 ml
N	0.59	NH <sub>4</sub> NO <sub>3</sub>	21.0 ml

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<u>SOLUTION 31</u>	<u>Element</u>	<u>Wt in Sol per litre</u>	<u>Salt Used</u>	<u>Quantity</u>
	Na	0.11	NaCl	5.0 ml
	Cl	0.18	NaCl	5.0 ml
	Na	0.39	NaNO <sub>3</sub>	17.0 ml
	N	0.24	NaNO <sub>3</sub>	17.0 ml
	Ca	0.48	Ca(NO <sub>3</sub> ) <sub>2</sub>	12.0 ml
	N	0.34	Ca(NO <sub>3</sub> ) <sub>2</sub>	12.0 ml
	K	0.25	KNO <sub>3</sub>	6.5 ml
	N	0.09	KNO <sub>3</sub>	6.5 ml
	P	0.15	H <sub>3</sub> PO <sub>4</sub>	13.0 ml
	Mg	0.10	MgSO <sub>4</sub>	4.0 ml
	S	0.13	MgSO <sub>4</sub>	4.0 ml
	N	0.53	NH <sub>4</sub> NO <sub>3</sub>	19.0 ml

SOLUTION 32

	Na	0.11	NaCl	5.0 ml
	Cl	0.18	NaCl	5.0 ml
	Na	0.39	NaNO <sub>3</sub>	17.0 ml
	N	0.24	NaNO <sub>3</sub>	17.0 ml
	Ca	0.48	Ca(NO <sub>3</sub> ) <sub>2</sub>	12.0 ml
	N	0.34	Ca(NO <sub>3</sub> ) <sub>2</sub>	12.0 ml
	K	0.51	KNO <sub>3</sub>	13.0 ml
	N	0.18	KNO <sub>3</sub>	13.0 ml
	P	0.15	H <sub>3</sub> PO <sub>4</sub>	13.0 ml
	Mg	0.10	MgSO <sub>4</sub>	4.0 ml
	S	0.13	MgSO <sub>4</sub>	4.0 ml
	N	0.45	NH <sub>4</sub> NO <sub>3</sub>	16.0 ml

APPENDIX II

STATISTICAL METHODS OF ANALYSIS OF EXPERIMENTAL DATA USED  
IN THE FOREGOING WORK

The following is an example of the statistical methods employed in the analysis of the experimental data.

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APPENDIX II

The example below illustrates the method employed in statistically analysing the results in the present work.

		Lower	Middle	Upper	Upper, Middle and Lower leaves.				
K <sub>0</sub>	Ca <sub>1</sub>	1.97%	0.43%	0.11%	2.51	6.20	24.82		
	Ca <sub>2</sub>	2.26%	0.88%	0.55%	3.69				
	K <sub>1</sub>	Ca <sub>1</sub>	2.33%	0.62%	0.13%	3.08		7.00	
		Ca <sub>2</sub>	2.27%	1.26%	0.40%	3.92			
	K <sub>2</sub>	Ca <sub>1</sub>	2.05%	0.26%	0.06%	2.36		5.82	
		Ca <sub>2</sub>	2.16%	0.87%	0.42	3.46			
K <sub>3</sub>	Ca <sub>1</sub>	1.82%	0.10%	0.05%	1.98	5.79			
	Ca <sub>2</sub>	2.19%	1.18%	0.44%	3.81				
K <sub>0</sub>	Ca <sub>1</sub>	2.46%	0.45%	0.08%	2.99	7.07		27.20	
	Ca <sub>2</sub>	2.49%	1.18%	0.42%	4.08				
	K <sub>1</sub>	Ca <sub>1</sub>	2.12%	0.56%	0.10%	2.78			6.43
		Ca <sub>2</sub>	2.17%	1.02%	0.45%	3.65			
	K <sub>2</sub>	Ca <sub>1</sub>	1.99%	0.32%	0.05%	2.36			6.65
		Ca <sub>2</sub>	2.12%	1.70%	0.47%	4.29			
K <sub>3</sub>	Ca <sub>1</sub>	1.38%	1.64%	0.07%	2.09	7.06			
	Ca <sub>2</sub>	2.10%	1.59%	0.28%	3.97				
K <sub>0</sub>	Ca <sub>1</sub>	1.05%	0.28%	0.05%	1.38	5.12			23.61
	Ca <sub>2</sub>	2.37%	0.93%	0.43%	3.74				
	K <sub>1</sub>	Ca <sub>1</sub>	1.64%	0.18%	0.06%	2.07	6.47		
		Ca <sub>2</sub>	2.71%	1.19%	0.49%	4.39			
	K <sub>2</sub>	Ca <sub>1</sub>	1.67%	0.21%	0.07%	1.96	5.65		
		Ca <sub>2</sub>	2.10%	1.31%	0.27%	3.69			
K <sub>3</sub>	Ca <sub>1</sub>	1.90%	0.41%	0.06%	2.37	6.57			
	Ca <sub>2</sub>	1.96%	1.62%	0.41%	4.00				
K <sub>0</sub>	Ca <sub>1</sub>	1.74%	0.29%	0.06%	2.09	4.70	21.89		
	Ca <sub>2</sub>	1.27%	1.15%	0.18%	2.61				
	K <sub>1</sub>	Ca <sub>1</sub>	1.99%	0.14%	0.05%	2.17		5.56	
		Ca <sub>2</sub>	1.95%	1.24%	0.20%	3.39			
	K <sub>2</sub>	Ca <sub>1</sub>	1.80%	0.25%	0.05%	2.10		5.82	
		Ca <sub>2</sub>	2.14%	1.20%	0.38%	3.72			
K <sub>3</sub>	Ca <sub>1</sub>	1.96%	0.08%	0.07%	2.11	5.81			
	Ca <sub>2</sub>	1.94%	1.50%	0.26%	3.70				
TOTAL		64.27%	26.06%	7.18%	97.51	Grand Total			

Upper leaves<sup>2</sup> = 2.5573

Middle leaves<sup>2</sup> = 30.0309

Lower leaves<sup>2</sup> = 132.7005

= (Upper + Middle + Lower)

318.9862