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CONTENTS

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THE ABSORPTION OF IONS BY CITRUS AND WALNUT SEEDLINGS*

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I. INTRODUCTION

This paper is a discussion of experiments on the exchange of ions between solutions and citrus and walnut seedlings, and is intended to extend our knowledge of certain problems in nutrition. The highly important rôle of the ions entering the cells of a plant has long been recognized, but the dynamics of the process are far from being understood.

Previous papers from this laboratory have been concerned chiefly with the study of the effects of salts on the growth and composition of the plant; this paper will deal with the ionic exchange as affected by growing plants. Our work has dealt chiefly with citrus and walnut seedlings which we have found to be very favorable for experimentation.

II. TECHNIC OF HANDLING THE CULTURES

A. CITRUS SEEDLING CULTURES

The seedlings were germinated in moist sphagnum moss and allowed to grow until the roots were 5 to 10 cm. long. Before placing the seedlings in the cultures the testa was taken from the cotyledons; the roots were dipped momentarily in distilled water and inserted into holes in tin lids placed on beakers containing the culture solution

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(fig. 1). Since three seedlings were inserted in each perforation they were firmly held without the use of cotton, which tends to promote the growth of injurious fungi. The cultures were grown in a glasshouse and the portion of the roof directly over the cultures was protected by suitable paper to prevent contamination of the cultures by falling substances, especially calcium compounds. The volume of the culture solutions was kept approximately constant by frequent additions of distilled water. Two liters of solution were always prepared, one liter being used in the beakers and the remainder saved for analysis.

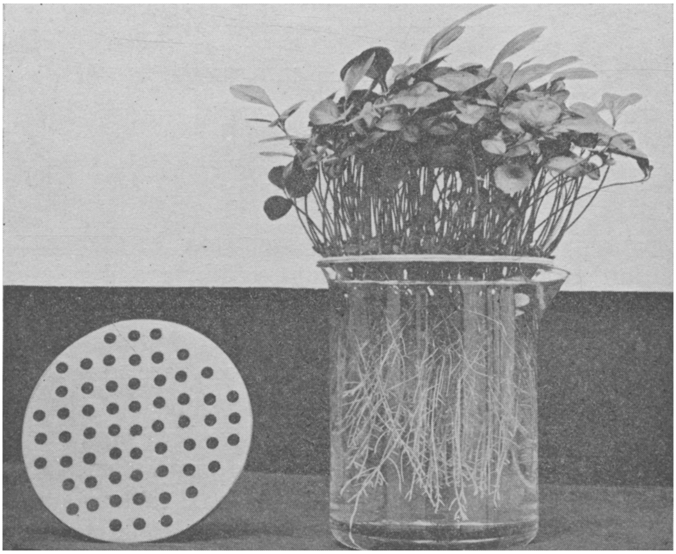

Fig. 1. One of the water cultures used for the study of absorption by citrus seedlings, with the perforated tin lid used shown at the left. The capacity of the beaker is one liter.

B. Walnut Seedling Cultures

Unbleached walnuts (*Juglans regia*) were placed in moist silica sand and kept in the glasshouse until the root just emerged from the shell (usually about four weeks), and then placed in moist sphagnum moss. When the tap root was 5 cm. or more in length, the seedlings were removed from the moss, the entire seedling (epicotyl not emerged as yet) was dipped momentarily in distilled water, and the excess water was blotted off by inverting the seedling and placing the shell against several sheets of filter paper. The roots are very sensitive to lack of moisture and if they become too dry the region just back of the tip develops a light straw color, which disappears if the drying is not too prolonged. The seedlings grew well for several weeks in cultures of the type here described (fig. 2). Distilled water was added frequently to maintain the volume of the solution. In some cases it will be noted that the solutions were renewed, in others they were not changed.

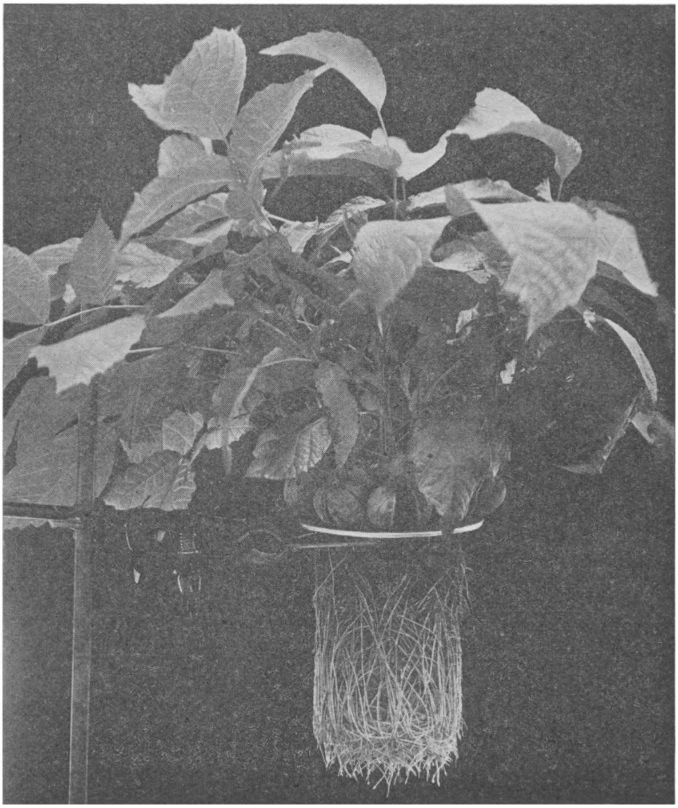

Fig. 2. A group of twelve walnut seedlings which had grown in a beaker like that shown in figure 1. Although the plants had been lifted from the solution, their matted roots held the form of the beaker in which they had grown.

III. EXPERIMENTAL DATA AND DISCUSSION

A. The Absorption of Ions by Citrus Seedlings

1. *The absorption of potassium ions.*

Five cultures of rough-lemon seedlings were grown in solutions which contained 7.7 milliequivalents K for 65 days and two cultures of grapefruit seedlings for 32 days (table 1). The cultures which contained KNO_3 and K_2SO_4 made the best growth, and those with

KH_2PO_4 were better than those with KCl. In the solution low in K, the plants made nearly as good growth as in the KCl set.

In two sets of cultures Cl or SO_4 was added only as KCl or K_2SO_4; thus the milliequivalents of K and anion are directly comparable since the anion was not furnished by any other salt.

TABLE 1

ABSORPTION OF IONS BY CITRUS SEEDLINGS FROM NUTRIENT SOLUTIONS CONTAINING EQUIVALENT AMOUNTS OF POTASSIUM

Salt added to nutrient	Reaction		Total milliequivalents of ions present							
	Initial pH	Final pH	Ca	Mg	Na	K	Cl	SO_4	NO_3	PO_4
A. Rough-lemon seedlings grown 65 days.										
KCl	5.5	4.7	8.323	4.786	1.337	7.613	8.483	5.244	7.734	3.369
K_2SO_4	5.5	5.2	8.114	4.754	1.215	7.726	0.525	13.243	7.499	3.709
KNO_3	5.2	5.7	8.083	4.901	1.484	7.828	0.606	5.312	14.659	2.950
KH_2PO_4	4.6	5.2	8.024	4.450	1.163	7.688	0.646	6.895	7.714	9.896
None	5.0	4.8	8.214	4.967	1.302	0.177	0.567	5.319	7.707	2.977
B. Grapefruit seedlings grown 32 days.										
KCl	7.0	4.8	7.934	4.934	1.866	7.770	8.139	4.905	7.253	1.435
K_2SO_4	7.0	4.8	8.263	4.721	1.662	7.816	4.740	7.885	6.973	1.542

Salt added to nutrient	Reaction		Milliequivalents of ions absorbed							
	Initial pH	Final pH	Ca	Mg	Na	K	Cl	SO_4	NO_3	PO_4
A. Rough-lemon seedlings grown 65 days.										
KCl	5.5	4.7	3.094	1.453	−.417	5.233	1.777	2.101	4.779	1.542
K_2SO_4	5.5	5.2	3.763	1.092	0.503	6.623	0.525	3.415	7.148	2.090
KNO_3	5.2	5.7	3.074	1.059	0.208	4.347	0.606	1.469	6.846	1.487
KH_2PO_4	4.6	5.2	3.114	0.649	−.551	4.887	−.164	1.373	6.054	lost
None	5.0	4.8	5.439	2.709	0.373	−0.046	−.322	1.808	6.438	2.142
B. Grapefruit seedlings grown 32 days.										
KCl	7.0	4.8	2.475	1.026	0.035	3.049	1.458	1.454	2.404	0.288
K_2SO_4	7.0	4.8	2.176	0.630	−.243	2.962	0.302	1.019	1.544	0.551

Table 1 shows that the reaction of the solutions was altered by the growth of the seedlings, but the shift was not always in the direction of neutrality. There was an increase in the acidity of the solutions which received KCl or K_2SO_4 and in that which received no K salt, while those which received KNO_3 or KH_2PO_4 became less acid.

Table 1 also shows the absorption of various ions, as determined by analyses at the end of the culture period. The amount of K absorbed by the rough-lemon seedlings varied considerably but the quantities were not strictly correlated with the growth of the seedlings. A small amount of K was excreted by the roots where K was at a minimum. The amounts of Na absorbed were variable and there was no increased absorption of that ion where K was lacking. The plants absorbed more Ca and Mg when K ions were absent, but the total milliequivalents of kations absorbed was not greater than in the other solutions. The high rate of absorption of K by young seedlings is also shown in table 3. Unless a large amount of Ca was present, the seedling absorbed more K than Ca. These results are in agreement with those obtained by the ash analyses of orange trees grown in sand cultures (tables 12, 13, and 14). There was an increased absorption of the anion of the K salt where the larger amounts were present, although the plants in the K_2SO_4 set absorbed slightly more NO_3 than those in the KNO_3 set. These data as well as those of table 3, show that the citrus seedlings absorbed SO_4 and PO_4 ions in larger amounts than Cl ions, even when the initial concentration of the latter was considerably higher than that of either of the former. In every case except the KNO_3 series, the K absorbed exceeded that of the anion of the K salt. Where much K was supplied the absorption of Mg was greatest when KCl was added.

2. *The absorption of calcium ions.*

Several of our published papers have shown the importance of calcium and the large amounts found in citrus plants. We have made further studies on the absorption of ions from calcium solutions by seedlings of four species of citrus with reference to the reacting values of the ions absorbed. Table 2 shows that the solution became more acid as growth proceeded and that the plants absorbed more of the kations than of the anions that were furnished by the added calcium salt. The results agree with those of the preceding section in showing the greater absorption of the kation. These data show more or less significant differences in the amount of ions absorbed in different experiments. Cultures conducted during the months of March and April have usually produced the best plants and have shown the highest absorption of ions.

The addition of increasing amounts of Ca ions was reflected to a slight extent in the increased absorption, but the increases were by no means proportional to the amounts added. The data show that during certain periods the absorption of NO_3 by *C. limonia* was diminished by the addition of Cl ions. The original concentration of NO_3 in the four cultures was equivalent yet its absorption was lowered where $CaCl_2$ was added.

TABLE 2

THE ABSORPTION OF CALCIUM IONS BY CITRUS SEEDLINGS

Seedlings used	Duration of experiment	Reaction of culture solution		Calcium furnished (milliequivalents)	Milliequivalents of ions absorbed			
		Initial	Final		Ca	Cl	SO_4	NO_3
	Days	*pH*	*pH*					
Poncirus trifoliata Raf.	23	5.8	4.4	7.535	1.587	1.300		
Poncirus trifoliata Raf.	23	6.2	5.2	14.980	2.765	2.304		
Poncirus trifoliata Raf.	23	6.4	<5.0	22.360	2.919	2.622		
Citrus limonia Osbeck	65	6.0	4.0	7.779	4.845			3.864
Citrus limonia Osbeck	48	5.4	4.0	7.685	5.374			4.769
Citrus limonia Osbeck	48	5.7	4.0	15.369	6.013	1.523		3.434
Citrus limonia Osbeck	39	5.2	4.0	7.884	3.518			2.959
Citrus limonia Osbeck	39	5.4	4.0	15.429	3.533	0.818		2.038
C. maxima (Burm.) Merrill	36	5.0	4.0	7.794	2.745			2.346
C. aurantium Linn.	42	5.2	4.4	9.062	4.661			4.518
C. aurantium Linn.	46	6.0	3.8	26.766	10.080		8.776	
C. aurantium Linn.	46	5.2	4.3	7.884	4.491			4.358

3. *The absorption of chlorin ions.*

A series of experiments with rough-lemon seedlings was made to study the absorption of Cl from nutrient solutions to which different chlorids were added (table 3). Since these were added to nutrient

TABLE 3

ABSORPTION OF CHLORIDS BY ROUGH-LEMON SEEDLINGS

Chlorid added to nutrient solution	Duration of experiment	Reaction of culture solution		Milliequivalents of ions furnished							
		Initial	Final	Ca	Mg	Na	K	Cl	SO_4	NO_3	PO_4
	Days	*pH*	*pH*								
None	65	5.0	5.6	7.964	3.177	0.326	4.483	0.443	5.133		3.525
KCl	68	5.3	4.8	8.004	5.074	0.234	12.380	8.483	5.406	10.761	3.553
NaCl	68	5.3	5.3	8.184	5.148	6.662	5.445	8.282	5.206	10.980	3.553
$CaCl_2$	68	5.4	4.6	15.709	5.115	0.512	4.644	8.443	5.408	10.900	3.709
$MgCl_2$	68	5.2	4.7	8.383	12.077	0.456	4.700	8.564	5.418	10.760	3.501

Chlorid added to nutrient solution	Duration of experiment	Reaction of culture solution		Milliequivalents of ions absorbed							
		Initial	Final	Ca	Mg	Na	K	Cl	SO_4	NO_3	PO_4
	Days	*pH*	*pH*								
None	65	5.0	5.6	3.393	0.058	−.338	3.930	−.367	1.855		2.301
KCl	68	5.3	4.8	1.956	0.739	−.386	4.733	0.804	1.639	4.801	1.307
NaCl	68	5.3	5.3	2.335	1.305	0.868	3.154	0.606	1.217	5.319	1.307
$CaCl_2$	68	5.4	4.6	4.770	1.059	−.156	4.001	1.091	2.163	6.693	1.775
$MgCl_2$	68	5.2	4.7	2.136	0.993	−.226	2.496	0.567	1.206	3.718	1.096

solutions there was always a supply of the kation from other salts, except in the case where NaCl was the chlorid added. The amounts of Cl added were very nearly the same in the different cultures but after 68 days the analysis showed the greatest amount of Cl had been absorbed from the cultures which contained $CaCl_2$. This may be due to the fact that these plants made the best growth and hence were able to absorb more Cl without actually increasing its concentration in the tissues. The ratio of Ca to Cl in the solution at the outset was about 2:1, but the ratio of the milliequivalents absorbed was nearly 5:1.

Although the initial supplies of SO_4 and NO_3 were approximately the same in all cultures, the amounts absorbed were greatest in the solution to which $CaCl_2$ was added. This may be due to the favorable ratio between Ca and K in that solution. The absorption of PO_4 ions was greatest where chlorids were at a minimum.

The change in the reaction of the solutions showed the effect of differential absorption of anions and kations. In three cases the acidity developed was close to the limit of tolerance for citrus roots.

4. *The absorption of sodium and chlorin ions.*

With a greater concentration of NaCl than in the preceding experiment the results obtained with *Poncirus trifoliata* seedlings were somewhat different. The absorption of Na ions and that of Cl ions was practically equal (table 4) and the reaction of the solution changed from pH 4.8 to 5.8.

TABLE 4

THE ABSORPTION OF Na AND Cl BY 192 *Poncirus trifoliata* SEEDLINGS GROWN FOR 27 DAYS IN A LITER OF NUTRIENT SOLUTION PLUS APPROXIMATELY 1000 P.P.M. NaCl

	Initial concentration (milliequivalents)	Ions absorbed (milliequivalents)	pH
Na	16.544	2.513	
Cl	17.419	2.544	
Initial			4.8
Final			5.8

The relation of NaCl to the composition of young orange trees grown in sand cultures has been rather extensively discussed in previous papers. The leaves and shoots of such trees usually contained much more Cl than Na, especially if harmful amounts were employed, but in the trunks and roots Na was more abundant than Cl.

TABLE 5

REACTION CHANGES AND ION ABSORPTION INCIDENT TO THE GROWTH OF CITRUS SEEDLINGS IN CULTURE SOLUTIONS

	Chinese-lemon seedlings grown 35 days						Citron seedlings (72 seedlings to a liter) grown 27 days				Grapefruit seedlings (192 seedlings to a liter) grown 32 days			
	Hoagland's solution		Modified Hoagland's plus Na_2SO_4		Modified Hoagland's plus Na_2SO_4 and NaCl		Hoagland's plus 2×NaCl		Modified Hoagland's plus $2\times Na_2SO_4$		Modified Hoagland's plus KCl		Modified Hoagland's plus K_2SO_4	
	Initial	Absorbed	Initial	Absorbed	Initial	Absorbed	Initial	Absorbed	Initial	Absorbed	Initial	Absorbed	Initial	Absorbed
	m.e.	m.e.	m.e.	m.e.	m.e.	m.e.	m.e.	m.e.	m.e.	m.e.	m.e.	m.e.	m.e.	m.e.
Ca	7.804	2.186	8.114	1.906	8.134	1.826	8.104	1.038	8.034	1.088	7.934	2.475	8.263	2.176
Mg	5.008	1.026	4.860	0.624	4.860	0.624	4.844	0.714	4.860	0.534	4.934	1.026	4.721	0.632
Na	1.480	0.165	5.985	0.243	9.487	−.152	8.962	0.517	10.720	0.898	1.866	0.035	1.662	−.243
K	4.429	2.319	9.283	2.908	9.697	3.126	5.545	1.756	9.789	1.697	7.770	3.049	7.816	2.962
Total	18.721	5.696	28.242	5.681	32.178	5.576	27.455	4.025	33.403	4.217	22.504	6.585	22.462	5.770
						−.152								−.243
						5.424								5.527
Cl	0.485	−.040	0.183	−.181	4.526	0.406	9.069	0.767	0.282	−.121	8.139	1.458	4.740	0.302
HCO_3*	0.185	−.187	0.223	0.038	0.243	−.166	0.223	0.112	0.243	0.169	0.581	0.220	0.661	0.381
SO_4	5.495	1.352	7.072	1.302	7.045	1.206	5.514	0.840	12.944	1.105	4.905	1.454	7.885	1.019
PO_4	3.446	1.331	3.185	0.731	2.938	0.432	3.185	0.575	3.054	0.548	1.435	0.288	1.542	0.551
NO_3	9.597	2.826	18.834	4.589	18.753	3.887	10.219	1.703	17.451	1.523	7.253	2.404	6.973	1.544
Total	19.208	5.509	29.497	6.660	33.505	5.931	28.210	3.997	33.974	3.345	22.313	5.824	21.801	3.797
		−.227		−.181		−.166				−.121				
		5.282		6.479		5.765				3.224				
Initial pH	5.0		5.0		5.0		5.1		5.1		7.0		7.0	
Final pH	4.8		5.6		5.4		4.8		4.8		4.8		4.8	

* Determined by titration with methyl orange.

5. *The relation between ion exchange and reaction change of culture solutions.*

In table 5 we have included a few of the representative results of numerous experiments made on the changes which take place in culture solutions as a result of the growth of citrus seedlings. The concentration of the solutions was not excessive and the growth period was long enough for measurable absorption to take place. The cortical tissues of the root of walnut seedlings tend to slough away in time, while with young citrus seedlings this does not take place. Consequently with citrus seedlings grown for short periods there is very little opportunity for acids to arise from the decay of organic matter. The low concentrations employed permit of fair analytical accuracy.

The table shows in milliequivalents the initial concentration of the solution and the amount of absorption. In the first five cultures the sum of the anions originally present exceeded the sum of the kations originally present and the solutions were acid. The last two solutions were apparently neutral to phenol red although the ionic balance would indicate slight alkalinity. In the second and third cultures the final pH showed a decrease in acidity as a result of the growth of the plants. In both these cases the total milliequivalents of anions absorbed was greater than that of the kations absorbed and consequently more kations than anions remained in the solution, which caused greater alkalinity or decreased acidity. In each of the other cultures the final pH of the solutions showed an increase in acidity. In these cultures the totals show that a greater amount of kations was absorbed than of anions, leaving as a result more anions than kations in the solution, with a consequent increase in the acidity. The prevailing conception regarding pH changes in culture solutions is that acid solutions [$(NH_4)_2SO_4$ excepted] tend to change in the direction of neutrality or less acidity and that alkaline solutions tend to change in the direction of neutrality or greater acidity. Our results agree with those of Hoagland[5] in showing that, where no organic secretions enter in as factors, that the changes in reaction of culture solutions may be attributed directly to differential absorption of ions together with negative absorption (excretion) of certain ions.

Table 5 also shows that in the third and seventh cultures there was a negative absorption of Na ions even though in the third considerable Na was present in the solution. In the fourth culture the absorption of Na was approximately the same as that of Cl, and in the fifth culture that of Na approximately the same as that of SO_4 where the initial concentrations were approximately similar. In the second and third cultures, the absorption of Na was considerably less than that of Cl or SO_4 combined; but when KCl or K_2SO_4 were used (sixth and seventh cultures) the K absorbed always exceeded the anion supplied with it, and this factor alone contributed much to the increased acidity of the solution.

The remarkable changes in reaction produced even by young seedlings are shown by an additional experiment with Florida Sour orange seedlings in which the pH changed from 6.8 to 4.0 in 12 days. A parallel set of cultures, in which the initial pH of 6.8 was changed to 5.5 by the addition of citric or tartaric acids, also brought the reaction to pH 4.0 in 12 days. As time went on the roots became

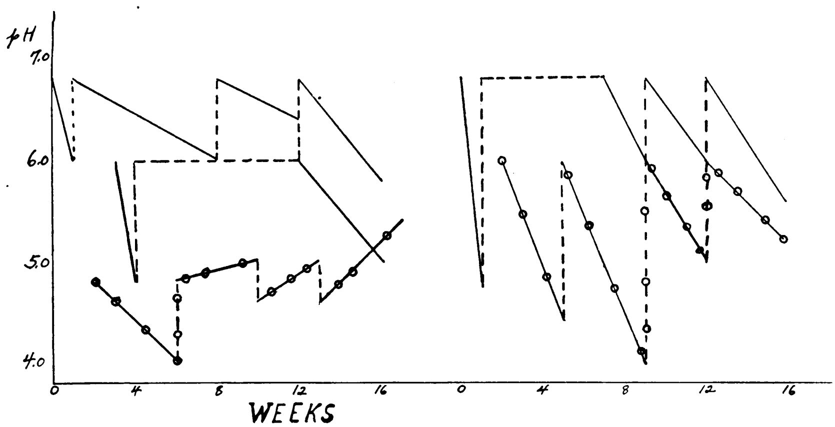

Fig. 3. The changes in reaction of culture solutions incident to the growth of citrus seedlings in them. ———— Sour orange; —o—o—o— Chinese lemon. Oblique lines represent changes brought about by the plants; vertical lines represent changes brought about by renewal of the solutions.

gelatinous but no growth of molds appeared. The results given in tables 4 and 5 show several cases in which the pH changes occurred in the opposite direction.

The changes in the reaction of culture solutions are shown by a series of graphs in figure 3. In one case the initial pH of the culture solution containing Sour orange seedlings was 6.8 and in the other 6.0. In both cases the subsequent changes were in the direction of greater acidity, although the solutions were renewed at frequent intervals. The desired initial reactions of the culture solutions were obtained by taking suitable proportions of KH_2PO_4 and K_2HPO_4.

It appeared that the rate of change of the pH was greatest in the initial stages of growth. The slope of the graphs showing the early changes is greater than that of those showing later stages. A similar effect has been observed in the case of walnut seedlings, especially so long as the cotyledons remained attached. The changes in reaction of Chinese-lemon cultures started at pH 6.0 agreed with those of the Sour orange cultures. Although the culture started at pH 4.8 dropped to 4.0 in the early stage of growth, the subsequent changes in reaction were toward higher pH values.

Since the change of reaction of a nutrient solution depends upon the exchange of kations and anions, it seems impossible to make predictions of the direction in which the reaction will change, unless we know something about the absorption process during that period. In other words, we must recognize a state of equilibrium between the plant, the solution, and the gases of solution and atmosphere.

6. *The absorption of CO_3 by seedlings.*

Breazeale[2] has suggested that when nitrate is present as the anion in a single-salt solution, the plant may take up more NO_3 than of the kation, and that from the bicarbonate formed in the solution, the plant absorbs its supply of what becomes carbonate when the plant is ashed.

We have grown citrus seedlings in solutions of calcium nitrate and found that both the ash of the tops and that of the roots gave a strong effervescence with acid. Low heat was always used in ashing and the plants were not allowed to catch fire. The ash of tops of rough-lemon seedlings taken from $CaCl_2$ cultures as well as the ash of roots of grapefruit seedlings taken from $CaSO_4$ cultures in which the solutions had become either slightly acid or strongly so showed effervescence with acid. Analyses were made of control rough-lemon seedlings which had been grown in moss and whose outer seed coats had been removed prior to being ashed. The ash of these seedlings showed faint, if any, effervescence with acid. We have found that the ash of wheat seed gave no effervescence with acid. When wheat seed was treated with NaCl or $NaNO_3$ in dilute solutions, then dried and ashed, effervescence occurred with acid when ignition temperatures were high. When wheat was treated with dilute calcium nitrate solution, dried and ashed at various temperatures, effervescence always occurred with HCl. It appears therefore that the gentle ignition of any organic calcium compound will yield a carbonate. Hence the presence of CO_3 in plant ash is regulated largely by the relation between the absorption of kations and anions and volatility upon ignition.

7. *The behavior of citrus seedlings in sodium carbonate solutions.*

Breazeale[1] maintains that the toxicity of soil solutions containing small amounts of sodium carbonate is due largely to the action of the sodium carbonate upon the soluble organic matter. He found that neither 400 p.p.m Na_2CO_3 nor a water extract of peat is toxic to citrus seedlings when taken singly but, when the two are mixed, the resulting solution is highly toxic.

In order to ascertain whether Na_2CO_3 up to 400 p.p.m. is stimulating, or at least non-toxic to citrus seedlings, we have grown grapefruit seedlings in cultures of carbon-treated distilled water containing 50, 100, 200, and 400 p.p.m. respectively of Na_2CO_3. The lowest concentration was between pH 8.5 and 10.0 while the others all had pH values above pH 10. After 2 to 3 days, the 400 p.p.m. culture was alkaline to phenolphthalein while those with lower strengths were not. The culture solutions were not renewed. Nine days later most of the seedlings had gelatinous roots.

The experiment was repeated, using Chinese-lemon seedlings and concentrations of Na_2CO_3 equal to 50, 150, and 400 p.p.m. In 6 days most of the roots had become gelatinous. In none of the cultures was there any evidence of elongation of the roots, and the high initial pH and the complete absence of Ca brought about gelatinization of the roots. The results indicate that the toxicity reported by Breazeale[1] may have been due to calcium starvation as well as to OH ions.

Soil containing black alkali, and soil which had been leached with NaCl until it was considered practically free from replaceable Ca, were leached with distilled water and the dark-colored leachate was collected. The solutions gave an alkaline reaction to phenolphthalein. They were then dialyzed against tap water until relatively free from Cl. The dialyzates were not alkaline to phenolphthalein. The dialyzed water-leachate of the soil containing normal carbonates contained 1826 p.p.m. total solids, with an ash content of 1132 p.p.m. The dialyzed water-leachate of the NaCl treated soil contained 2096 p.p.m. total solids and 1147 p.p.m. ash content. When citrus and walnut seedlings were placed in these dialyzed leachates, they grew very well, during the several weeks they were under observaton.

8. *The absorption by young orange trees.*

a. Water cultures. These experiments are largely of an exploratory nature, considerable difficulty having been experienced in the determination of suitable cultural methods. Sometimes trees taken from the field could be started in solution cultures but great difficulty was experienced in removing adhering soil. When all of the lateral roots were removed but not the tops, a fairly clean main root could be secured, but after several days the leaves wilted and the trees usually died. Where the leaves were removed, the buds frequently started but remained at a standstill for long periods.

TABLE 6A

ABSORPTION DURING ONE YEAR BY YOUNG VALENCIA ORANGE TREES IN WATER CULTURES

	Milliequivalents of ions in modified Hoagland's solution						Milliequivalents of ions in modified Hoagland's solution containing sodium chlorid			
	Tree 1		Tree 2		Tree 3		Tree 4		Tree 5	
	Initial	Absorbed	Initial	Absorbed	Initial	Absorbed	Initial	Absorbed	Initial	Absorbed
Ca	255.039	59.077	286.078	129.760	285.104	102.734	281.147	78.388	281.212	78.453
Mg	108.413	11.133	104.062	22.060	94.456	4.442	105.293	9.130	102.141	5.230
K	150.129	31.585	140.053	44.060	147.121	25.224	......	......	......	......
Cl	......	......	......	......	......	......	296.529	16.035	297.730	20.713
NO_3	323.346	113.310	343.461	182.654	348.045	226.381	355.325	140.502	359.505	120.670
SO_4	100.797	10.614	96.533	12.961	96.128	11.611	93.835	8.846	94.910	9.449
*PO_4	76.745	28.121	74.588	62.779	75.184	68.238	......	......	......	......
Transpiration c.c.	31,525		46,115		37,125		41,770		33,000	
Initial pH	5.8		5.8		6.0		5 3		5.3	
Final pH	6.0		6.0		7.2		6.2		6.4	

* Cf. text, p. 81.

Budded Valencia orange trees secured from the nursery were placed in sand cultures after all branches, leaves, and rootlets had been removed. The top consisted of a piece of trunk about two feet in length and the root consisted of an undivided tap root about one foot in length. The root was scrubbed thoroughly with a brush to remove adhering soil. When such trees were placed directly into culture solutions a scum or film usually formed on the surface of the solutions. If the tree first regenerated some of its roots in sand cultures, such films seldom formed. After growing in sand kept moist with Hoagland's nutrient solution (from April until August 18), the trees were removed, the rootlets were freed from the sand and many of the rootlets were pruned away. The trees were then placed in 5-gallon wide-mouthed bottles containing a measured amount of nutrient solution, the partial composition of which is given in table 6A. Trees 1, 2, and 3 were placed in a complete nutrient solution which contained 318 p.p.m. Ca as calcium nitrate and 105 p.p.m. PO_4 derived from KH_2PO_4 and K_2HPO_4. Trees 4 and 5 received the same solution plus 297 m.e. Cl as NaCl. The culture bottles were kept in a box covered with boards and paper so as to exclude light, paper collars being placed about the trunks where they projected through the box cover. Distilled water containing iron was added from time to time but no additional nutrient was added from August 18, 1924, to September 28, 1925.

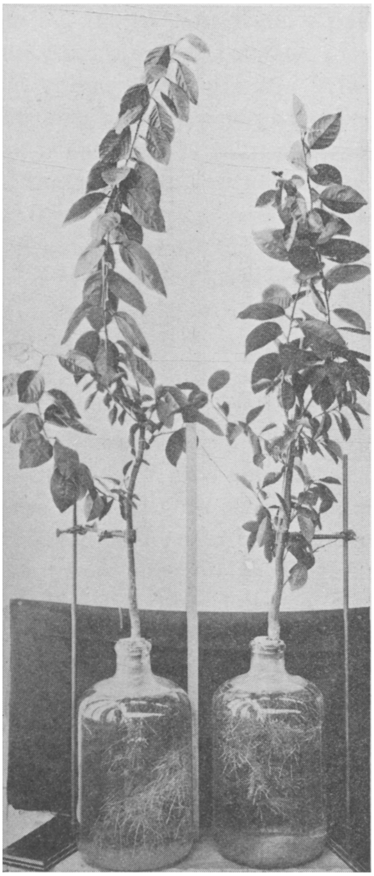

Fig. 4. Cultures used for the study of absorption by young orange trees after one years' growth made in bottles with a capacity of 20 liters.

The method of conducting the cultures and the type of the resulting growth are shown in figure 4. The trees produced about three cycles of growth on each of the larger shoots and the leaves were dark green even though the cultures were grown in the glasshouse. There was some indication of mottle-leaf on the tree in culture 4 which received NaCl. The temperature of the glasshouse was continually maintained above 70° to 75° F and during certain periods of the year it was considerably higher. Some of the roots in the cultures receiving sodium salts died, and absorption by such root systems doubtless may be abnormal. The removal of PO_4 from the solution was due in a large measure to precipitation occasioned by the repeated additions of iron tartrate. The large volumes of water transpired by the trees are indicated in table 6A. The NO_3 absorption and the amount of transpiration furnish indication of the growth made by the various tree cultures. The absorption of NO_3 exceeded that of any other ion determined. Ca absorption, in some of the cultures that grew well (such as 2 and 3) was extremely large. It will be noted, however, that in proportion to the amount furnished, the absorption of K ranks close to that of Ca. The absorption of Mg was not very high, and the absorption of Cl and SO_4 ions was very low in comparison with that of the NO_3 ion. That larger amounts of anion than of kation were absorbed is indicated by the increased pH.

b. Sand cultures. Four sets of Florida Sour-orange seedlings were grown in sand cultures (10 plants to a 10-gallon crock) for about two years, by which time the plants had become approximately four feet high. The cultures received Hoagland's nutrient solution during the early period of development. The sand was then leached with distilled water until practically free from dissolved salts, and each culture was given four liters of complete nutrient solution, the composition of which is given in table 6B. After several weeks the cultures were again thoroughly leached and the per cent of the original ions absorbed was determined (see table 6B).

The nitrate absorption was greatest, practically all of the NO_3 being removed. The absorption of K and that of Ca were practically the same, while that of Mg was considerably less than either. The SO_4 absorption was less than that of PO_4 and NO_3.

9. *The relation of the H-ion to the growth of citrus seedlings.*

Previous experiments[9] on the absorption and growth of rough-lemon seedlings in solutions maintained at pH 6, 7, 8, and 9 showed optima at pH 8 and 9. We have made additional experiments with citrus seedlings and present herewith the results at pH 4, 5, 6, 7, 8, and 9.

TABLE 6B

ABSORPTION BY TWO-YEAR-OLD SEEDLINGS OF FLORIDA SOUR-ORANGE IN SAND CULTURES

	Absorption by two-year-old seedlings of Florida sour-orange in sand cultures		
	Milliequivalents furnished	Milliequivalents absorbed	Per cent absorbed
Ca	62.076	53.274	86
Mg	29.983	16.268	54
K	56.003	47.291	84
NO_3	71.999	70.678	98
SO_4	43.014	26.252	61
PO_4	29.009	20.309	70

Before suitable measurements of the effects of the H-ion concentration upon absorption by citrus seedlings can be adequately studied, we should first know the effects of H-ion concentration upon growth. The method of studying the effects upon growth has been usually that of starting with a certain nutrient solution and then regulating the pH of the culture solutions by the addition of suitable quantities of acid or alkali (usually H_2SO_4 and NaOH).

The present experiments make it evident that the initial pH of the nutrient solution adopted for such studies and the nature of the acid or base used in regulating the pH have considerable influence on the growth obtained at a given pH.

Grapefruit seedlings were grown in white-enamel pails containing 9 liters of Hoagland's solution of pH 5. One of the culture solutions was brought to pH 4 by the addition of HCl and the other solutions to pH 6, 7, 8, and 9 by the addition of NaOH. The pH of the solutions was readjusted two or three times daily and as frequently as changes in the pH of the solutions warranted. The solutions were renewed at intervals of from one to two weeks. Figure 5 shows the growth of plants from the different cultures after 43 days. The growth of the roots was least at pH 5.0. At pH 9 some of the leaves showed burning and some root tips were brown.

When St. Michael orange seedlings were grown in a similar manner for 56 days in Hoagland's solution whose reaction was brought to pH 4 by adding HCl and to 6, 7, 8, and 9 by adding NaOH, we found the poorest root growth again at pH 5 and the best growth at pH 7 with a definite decline at pH 9. In another experiment KH_2PO_4 was added to Hoagland's solution so as to give an initial reaction of pH 7. The pH was then adjusted by the addition of HCl and NaOH. Figure 6 shows the growth made in these solutions during

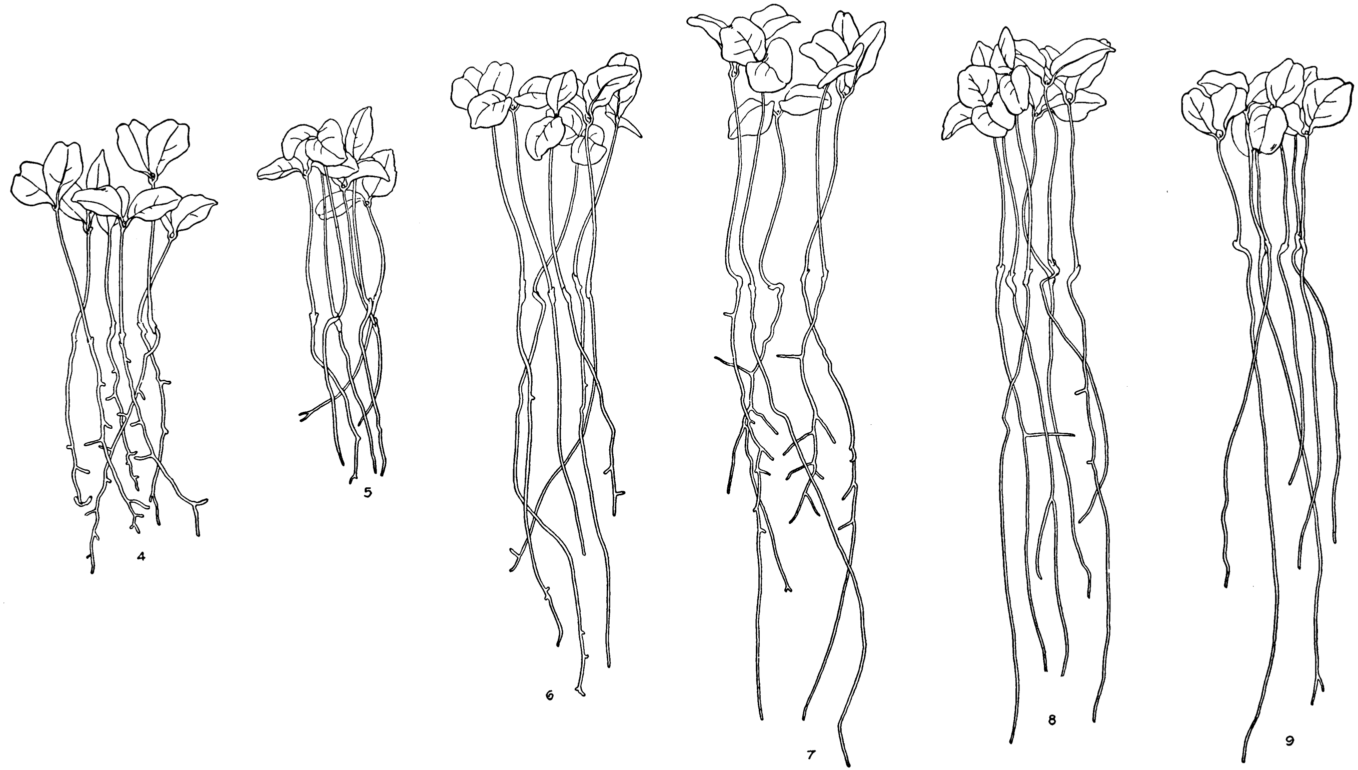

Fig. 5. The relation of hydrogen-ion concentration to the growth of grapefruit seedlings. The original reaction of the solution was pH 5.0 and was adjusted to reactions ranging from pH 4.0 to 9.0 by means of HCl or NaOH. The numerals indicate the pH values of the solutions.

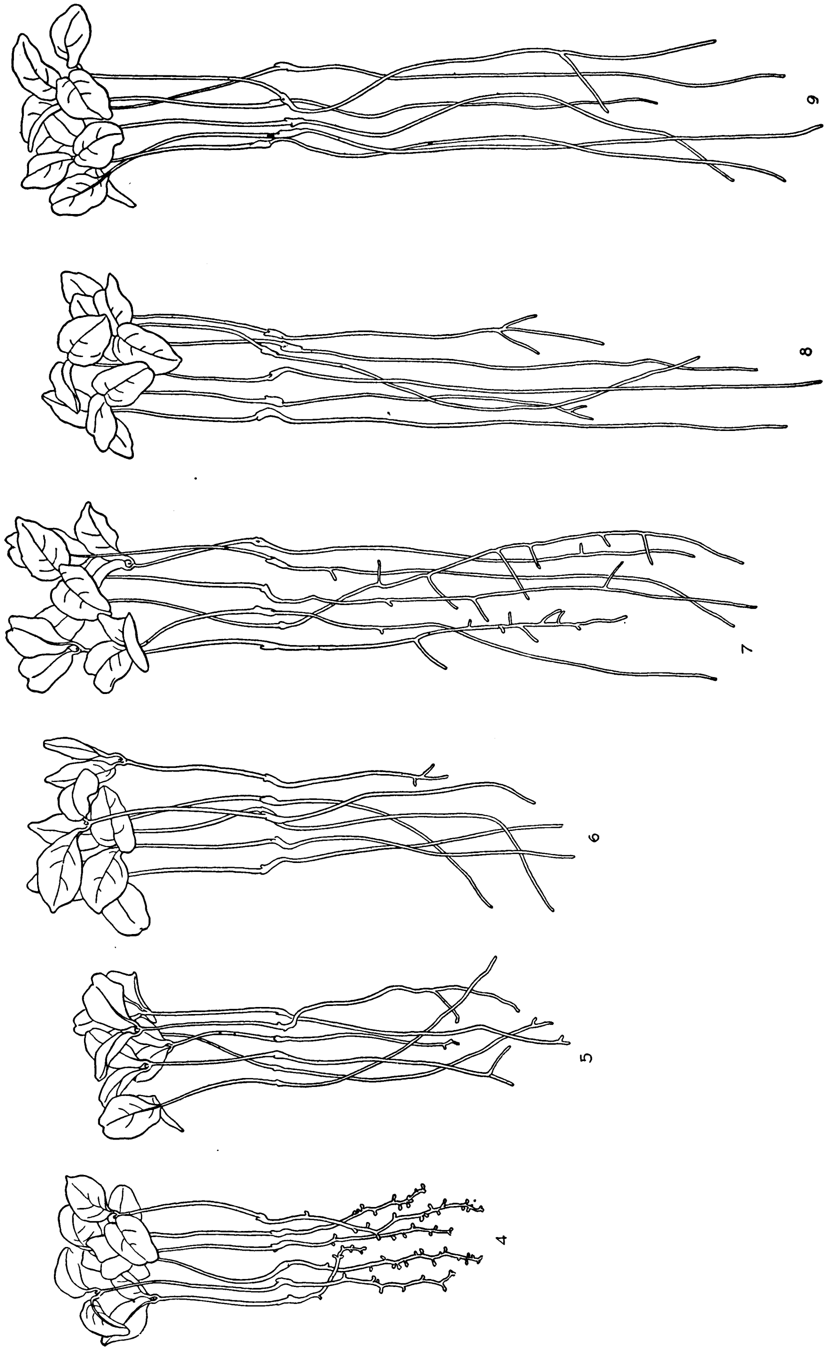

Fig. 6. The relation of hydrogen-ion concentration to the growth of orange seedlings. The original reaction of the solution was pH 7.0 and was adjusted to reactions ranging from pH 4.0 to 9.0 by means of HCl or NaOH. The numerals indicate the pH values of the solutions.

a period of 45 days. The culture of pH 4 was no better than that at pH 5. The marked changes between the results at pH 6 and 7 and between those at pH 7 and 8 are evident.

Grapefruit seedlings were grown for 47 days in Hoagland's solution (reaction pH 5) and in Hoagland's solution regulated to pH 4 with $Ca(H_2PO_4)_2$ and to pH 6, 7, 8, and 9 with $Ca(OH)_2$. The poorest growth was made at pH 5, with greater growth at pH 4 and a marked increase at pH 6. If we use the solution which had an initial pH of 7, and change its reaction to pH 4, 5, and 6 with $Ca(H_2PO_4)_2$ and to pH 8 and 9, with $Ca(OH_2)$, we find a gradually decreasing length of the roots from pH 7 to pH 4 and excellent growth at pH 7, 8, and 9, with slightly the best at pH 8.

The results of these experiments emphasize some matters which have often been overlooked by previous investigators. They show the importance of (*a*) the hydrogen-ion concentration, (*b*) the original pH of the solution employed, and (*c*) the nutrient or toxic action of the reagents *per se* employed in maintaining the desired reaction. For example, the growth in a solution having an original reaction of pH 5.0 was improved for citrus growth either by decreasing or by increasing the hydrogen-ion concentration. On the other hand, growth in a solution having an original reaction of pH 7.0 was better than that in solutions having greater concentrations of hydrogen-ions. $Ca(OH)_2$ was much more favorable for growth than NaOH when used to increase the OH-ion concentration, as shown in a former paper.[9]

B. The Absorption of Ions by Walnut Seedlings

The problems of absorption were further studied by a series of experiments with walnut seedlings grown in culture solutions. On account of their extreme sensitiveness to certain kations walnut seedlings are well suited to the study of many problems akin to those already presented.

1. *The absorption from solutions containing a low concentration of potassium ions.*

The first series of cultures to be described was planned to study the absorption of nutrient ions when potassium was present in small amounts. The question was of no little importance for we have shown elsewhere[9] that walnut seedlings are relatively rich in potassium, and consequently may grow for some time without additional supplies of that element. The seedlings grew in the cultures for two periods of 35 days each. At the end of the first period of 35 days, the plants were in a vigorous condition; consequently they were transferred to another similar solution and grown for a second period of 35 days, at the end of which time the seedlings were still healthy and vigorous. The roots were clear white, the leaves free from tip burn, and the solutions were free from the yellow tinge which often accompanies injury. The seedlings absorbed more of the anions and kations during the first period than during the second, with the exception of PO_4 which was completely absorbed during both periods (table 7). Although the ash of walnut kernels contains approximately 60 per cent of PO_4, the seedlings absorbed all of the PO_4 present in the solution. Although the culture solution contained a small amount of K at the beginning of the first period, no increase of K was evident at the close of that period. In the second period, however, when the initial concentration of K was lower, an excretion of K took place but no retardation of growth or unhealthy appearance of the plants due to lack of K was evident.

TABLE 7

Absorption of Ions by Walnut Seedlings Grown in Culture Solutions Low in Potassium

	First period (35 days)			Second period (35 days)		
	Original m.e.	Absorbed m.e.	Per cent absorbed	Original m.e.	Absorbed m.e.	Per cent absorbed
Ca	15.958	10.798	68	15.429	8.254	53
Mg	4.877	3.908	80	3.908	2.135	54
Na	3.355	1.484	44	1.167	−.100	....
K	0.556	0.149	27	0.261	−.044	....
Cl	0.606	0.606	100	0.364	−.059	....
NO_3	14.381	12.519	88	14.381	9.412	65
SO_4	5.287	−.242	....	5.273	0.478	9
PO_4	2.898	2.898	100	3.213	3.213	100

Additional results (table 8) were obtained from the two sets of cultures in which the absorption from complete and low K solutions was compared. The similarity in the per cent of ions exclusive of potassium absorbed in the two cases is remarkable.

The absorptions of ions by walnut seedlings differed from that of citrus seedlings when the concentration of K ions was low, in that no increased absorption of Ca or Mg took place (compare tables 1 and 8).

2. *The absorption of ions from solutions containing higher concentrations of potassium.*

The first two sets of cultures of walnut seedlings received Hoagland's solution, which contains 8 milliequivalents Ca (table 9). In

TABLE 8

ABSORPTIONS OF IONS BY WALNUT SEEDLINGS GROWN IN CULTURE SOLUTIONS FOR 39 DAYS

	Complete nutrient solution			Culture solution low in potassium		
	Original m.e.	Absorbed m.e.	Per cent absorbed	Original m.e.	Absorbed m.e.	Per cent absorbed
Ca	8.283	6.507	78	8.204	5.808	71
Mg	4.786	3.637	76	4.901	3.752	77
Na	1.016	0.109	10	1.098	−.373	....
K	4.516	3.981	88	0.292	−.161	....
Cl	0.558	0.319	57	0.558	0.358	64
SO_4	5.393	2.363	44	5.368	2.321	43
PO_4	3.473	3.473	100	3.054	3.054	100

cultures 3 and 4, and thereafter when high Ca is noted, we added 15 milliequivalents Ca as the nitrate in making up Hoagland's solution. Solutions 5 and 7 were the basis of a series in which the low K content was augmented by the addition of an appropriate K salt.

When the initial concentration of K was low, there frequently resulted an increase in the concentration of K in the solution. When

TABLE 9

ABSORPTION OF IONS BY WALNUT SEEDLINGS FROM SOLUTIONS CONTAINING HIGHER CONCENTRATIONS OF POTASSIUM

Culture	Modification of Hoagland's nutrient solution	Length of absorption period	Reaction of solution		Initial concentration of K	Ions absorbed	
			Initial	Final		K	Anion
		Days	*pH*	*pH*	m.e.	m.e.	m.e.
1	Nutrient solution (unmodified)	31	5.0	4.6	4.029	2.429	
2	Nutrient solution (unmodified)	39	5.4	4.6	4.516	3.981	
3	High Ca, first period	35	5.0	5.4	4.170	3.840	
4	High Ca, second period	34	5.3	6.0	4.288	3.320	
5	High Ca, low K, first period	35	4.8	7.0	0.556	0.149	
6	High Ca, low K, second period	34	4.9	6.4	0.261	−.044	
7	Low K	39	5.4	4.6	0.292	−.161	
8	High Ca, KCl, first period	35	5.0	5.5	7.839	6.743	2.442
9	High Ca, KCl, second period	34	5.3	6.0	7.839	3.817	1.334
10	KCl	30	5.3	4.6	7.808	7.483	2.183
11	KCl	39	5.2	4.6	47.265	22.339	14.083
12	High Ca, K_2SO_4, first period	35	4.9	5.5	7.790	6.687	2.949
13	High Ca, K_2SO_4, second period	34	5.0	6.0	7.662	4.787	1.909
14	K_2SO_4	30	5.2	5.2	7.734	4.009	3.251
15	K_2SO_4	38	5.0	4.4	39.683	14.121	13.347
16	High Ca, KNO_3, first period	35	5.0	5.6	7.880	5.717	15.083
17	High Ca, KNO_3, second period	34	4.9	6.3	7.759	4.063	14.342
18	KNO_3	30	5.0	6.0	7.767	4.429	8.301
19	KNO_3	39	5.4	6.2	86.502	26.225	
20	High Ca, KH_2PO_4, first period	35	4.6	5.2	7.823	5.985	3.967
21	High Ca, KH_2PO_4, second period	34	4.9	6.0	7.642	2.301	0.208

KCl was added to the culture solution, the absorption of Cl was always less than that of the K. The seedlings in cultures 8 and 9 removed less K from the solution containing 15 milliequivalents of Ca than from that containing 8 milliequivalents (culture 10). The most active absorption of K took place during the first period (culture 8). When the initial concentration of K was 47 milliequivalents a much smaller percentage absorption of K took place than when lower concentrations of K were employed, although the total amount absorbed was greater. The solutions used in cultures 1, 2, 7, 10, and 11 became more acid as a result of growth. Cultures 12 and 13 to which K_2SO_4 was added and which were high in calcium and nitrate were changed toward greater alkalinity, while those with lower Ca and NO_3 (solutions 14 and 15) tended toward greater acidity. There was less absorption of SO_4 than of K, even though the usual SO_4 of Hoagland's solution was present as well as that added with the potassium. Absorption was again greater in the first than in the second period. All the solutions used in these experiments contained calcium nitrate, consequently where KNO_3 was added the NO_3 absorption far exceeded that of the Cl, SO_4 or PO_4 anions. It is of interest therefore to find that, despite the large absorption of NO_3, the absorption of K in the first 35-day period was not greatly different from that of the cultures which received K_2SO_4. The reader may be reminded that a similar relation was found in the case of citrus seedlings (table 1).

The cultures which received KH_2PO_4 with high calcium nitrate showed a decrease in H-ion concentration and a greater absorption of K than of PO_4.

Table 10 gives the results of a determination of all ions (except NO_3) that were absorbed from solutions 11 and 15 of table 9. The milliequivalents of K absorbed in both cases were high, but were less than half the amounts initially present. From these data as well as from those in tables 9 and 13 it will be seen that less Cl and SO_4 were absorbed than K when a potassium salt was added in excess.

A series of cultures with walnut seedlings was conducted in which approximately 7.7 milliequivalents of K was added as a single salt to the nutrient solution. The solutions were analyzed and renewed at the end of 35 days. The plants were removed after growing 35 days in the second solution and the amounts of nutrient remaining in the solutions were again determined (table 11).

We found that, when large amounts of PO_4 were supplied, the total absorption for both periods was not greater than when the usual amount of PO_4 was supplied. It appears that where all the PO_4 was

TABLE 10

IONS ABSORBED BY WALNUT SEEDLINGS FROM TWO CULTURE SOLUTIONS CONTAINING AN EXCESS OF POTASSIUM SALT

	Initial concentration m.e.	Ions absorbed by plants m.e.	Per cent absorbed	Reaction	
				Initial pH	Final pH
Ca	8.134	3.423	42	5.2	4.6
Mg	4.754	1.995	42		
Na	0.074	−.903	....		
K	47.265	22.339	47		
Cl	43.185	14.083	33		
SO_4	5.393	2.244	42		
PO_4	3.186	3.186	100		
Ca	8.144	2.894	36	5.0	4.4
Mg	5.222	1.314	25		
Na	1.332	0.933	70		
K	39.683	14.121	36		
Cl	0.502	0.169	34		
SO_4	48.458	13.347	28		
PO_4	3.342	3.342	100		

TABLE 11

ABSORPTION OF IONS BY WALNUT SEEDLINGS FROM SOLUTIONS CONTAINING EQUIVALENT AMOUNTS OF POTASSIUM

Modification of control culture solution	Period of 35 days	Initial milliequivalents of ions							
		Ca	Mg	Na	K	Cl	NO_3	SO_4	PO_4
Low K	First	15.958	4.877	3.355	0.556	0.606	14.381	5.287	2.898
Low K	Second	15.429	3.908	1.167	0.261	0.364	14.381	5.273	3.213
Plus KCl	First	15.689	4.770	1.697	7.839	8.483	14.628	5.231	2.769
Plus KCl	Second	15.549	4.951	1.424	7.839	8.364	14.628	5.183	2.977
Plus K_2SO_4	First	16.058	5.098	1.632	7.790	0.606	14.767	13.505	3.029
Plus K_2SO_4	Second	15.249	4.811	1.371	7.662	0.403	14.767	13.322	3.002
Plus KNO_3	First	16.048	4.770	1.580	7.880	0.809	22.237	5.115	2.977
Plus KNO_3	Second	15.469	4.918	3.333	7.759	0.567	22.237	5.260	2.662
Plus KH_2PO_4	First	15.788	4.967	1.463	7.823	0.606	14.683	6.725	18.045
Plus KH_2PO_4	Second	15.259	4.844	2.496	7.642	0.403	14.683	6.931	17.209

Modification of control culture solution	Period of 35 days	Milliequivalents of ions absorbed								Initial pH	Final pH
		Ca	Mg	Na	K	Cl	NO_3	SO_4	PO_4		
Low K	First	10.798	3.908	1.484	0.149	0.606	12.519	−.242	2.898	4.8	7.0
Low K	Second	8.254	2.135	−.100	−.044	−.059	9.412	0.478	3.213	4.9	6.4
Plus KCl	First	8.743	2.849	0.104	6.743	2.442	13.899	1.789	2.769	5.0	5.5
Plus KCl	Second	7.216	1.741	−.174	3.817	1.334	12.697	−.347	1.567	5.3	6.0
Plus K_2SO_4	First	8.822	3.021	−.517	6.687	0.606	14.339	2.949	3.029	4.9	5.5
Plus K_2SO_4	Second	6.587	1.905	−.195	4.787	0	8.005	1.909	3.002	5.0	6.0
Plus KNO_3	First	8.313	2.849	−.213	5.717	0.809	15.083	1.510	2.977	5.0	5.6
Plus KNO_3	Second	7.435	1.905	1.428	4.063	0.144	14.342	0.073	1.487	4.9	6.3
Plus KH_2PO_4	First	8.254	3.136	−.538	5.985	0.606	13.252	1.741	3.969	4.6	5.2
Plus KH_2PO_4	Second	8.513	1.486	0.937	2.301	−.203	11.759	−4.832	0.208	4.9	6.0

absorbed from the solution during the first period, the PO_4 constituted a limiting factor as indicated by the continued absorption during the second period. When the amount of PO_4 supplied was high, less K was absorbed during the second period. The total amount of Mg absorbed in each culture for both periods is quite constant. It is of interest to note that the absorption of SO_4 was negative in three cases and practically zero in a fourth. The nutrient solutions in every case were changed towards alkalinity. Except where KNO_3 was added, the K absorbed always exceeded that of the corresponding anion. The nutrient solution ordinarily used contained the primary potassium phosphate. Table 12 gives a comparison of this with solutions containing the secondary phosphate and a mixture of the two. The results are expressed as milliequivalents of ions absorbed by 12 walnut seedlings to a liter of solution during two periods of 33 and 49 days each.

TABLE 12

ABSORPTION OF K AND PO_4 IONS FROM NUTRIENT SOLUTIONS BY WALNUT SEEDLINGS

Potassium salt used in nutrient solution	Reaction			Initial concentration		Ions absorbed by plants		Per cent absorbed	
	Days	Initial	Final	K	PO_4	K	PO_4	K	PO_4
		pH	pH	m.e.	m.e.	m.e.	m.e.		
KH_2PO_4	33	5.0	4.6	4.355	3.081	3.937	3.081	90	100
KH_2PO_4	49	5.2	4.8	9.925	6.867	9.160	6.503	92	95
K_2HPO_4	33	7.0	4.6	5.123	2.690	4.575	2.690	89	100
K_2HPO_4	49	>7.0	4.9	10.985	6.111	10.084	5.588	92	92
K_2HPO_4 + KH_2PO_4	33	6.2	4.4	4.897	3.186	4.506	3.186	92	100
K_2HPO_4 + KH_2PO_4	49	5.6	4.7	10.143	5.248	8.940	4.544	88	87

It will be seen that the solution containing the primary phosphate had an initially lower pH value than the other two but that the final pH value in each case was very nearly the same. The amounts of K and of PO_4 absorbed were practically identical when expressed as percentages of the amount supplied. The large absorption capacity of walnut plants for PO_4, together with the great need of such plants for Ca, makes it desirable to test tricalcium phosphate as a source of Ca and PO_4. Such experiments are now under way but have not been completed yet.

3. *Absorption from solutions of calcium and potassium salts.*

Since it has not been possible to grow walnut seedlings in a solution lacking calcium, we made another experiment in which walnut seedlings were grown 47 days in solutions of KCl and of K_2SO_4

(approximately 7.7 milliequivalents of K) to which a small amount of $CaCO_3$ was added. The object of this experiment was to observe the absorption of K and its accompanying anion from solutions containing few other ions.

TABLE 13

ABSORPTION OF IONS BY WALNUT SEEDLINGS GROWN IN KCl OR K_2SO_4 SOLUTIONS CONTAINING $CaCO_3$

Solution	Initial concentration in milliequivalents				Milliequivalent ions absorbed				Final reaction
	Ca	K	Cl	SO_4	Ca	K	Cl	SO_4	pH
$KCl+CaCO_3$	12.475	7.716	7.930	....	7.595	6.077	2.121	....	5.5
$K_2SO_4+CaCO_3$	12.475	7.619	....	7.850	7.505	6.482	....	1.583	4.8

Table 13 shows that the seedlings absorbed approximately the same amounts of Ca from both solutions, and that although slightly more K was absorbed from the K_2SO_4 than from the KCl solutions, less SO_4 was absorbed than Cl. The results add evidence to that already presented showing the relatively rapid absorption of K by seedling plants.

Further studies were made on the absorption by seedlings of walnut and of St. Michael orange from solutions containing about 7.7 milliequivalents of K ions and about 8 milliequivalents of Ca ions. We have thus an opportunity to observe the absorption of these kations in the absence of all others and to compare the results with those where $CaCO_3$ was furnished.

TABLE 14

RELATIVE ABSORPTION OF K AND Ca IONS BY WALNUT AND ORANGE SEEDLINGS

Salts	Absorption period	Reaction		Initial concentration		Ions absorbed		Per cent absorbed	
		Initial	Final						
	Days	pH	pH	K	Ca	K	Ca	K	Ca
Walnut:				m.e.	m.e.	m.e.	m.e.		
$KCl+Ca(NO_3)_2$	32	4.8	4.4	7.424	7.744	2.726	1.996	37	26
$K_2SO_4+Ca(NO_3)_2$	32	5.0	4.6	7.342	8.044	2.696	2.774	37	34
Orange:									
$KCl+Ca(NO_3)_2$	32	5.0	4.6	7.557	7.924	3.190	2.335	42	29
$K_2SO_4+Ca(NO_3)_2$	32	4.6	4.8	6.774	7.984	2.952	3.094	44	39

Table 14 shows that the amounts of K and Ca absorbed were practically identical when the sulfate and nitrate were used, but when the chlorid and nitrate were used somewhat greater amounts of K were absorbed than Ca. In experiments (table 5) where complete nutrient solutions were used and the initial concentration of K and Ca was approximately the same, the milliequivalents of K absorbed were greater than those of Ca. In table 14, however, we were dealing with incomplete culture solutions.

4. *The absorption of calcium by walnut seedlings.*

In a former paper,[8] we have shown the sensitiveness of walnut seedlings to the presence of calcium. We shall now give some data on the absorption from solutions of single calcium salts.

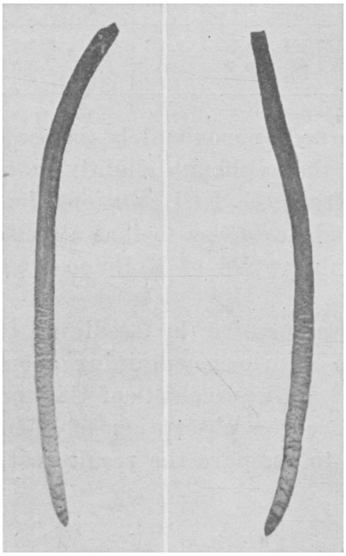

Fig. 7. Walnut-seedling roots showing the banded surface due to elongation after recovery from injury brought about by contact with a solution having unfavorable alkalinity.

The absorption of calcium from solutions of $CaCl_2$ was studied in two experiments (table 15), one of which ran for 20 and the other for 31 days. In each case the culture solution showed a slight increase in acidity due, as will be seen, to the greater removal of kations than anions, confirming the results obtained by Redfern.[6] In the 31-day period the plants absorbed about one-fourth of the calcium present and still less in the 20-day period.

The absorption of calcium from a solution of the primary phosphate (in spite of the higher acidity) was as great as from calcium chlorid solutions of equal strength. On the basis of the ions absorbed from the phosphate culture, we would expect the final reaction of the solution to become more acid instead of alkaline. We can account for the change in the opposite direction only by assuming that kations entered the solution from the plants. There was a certain amount of injury to the roots when first put into the calcium phosphate solution, but they later recovered only to show successive injury and recovery. This was particularly the case when 30 p.p.m. Ca as $Ca(H_2PO_4)_2$ was used. The root cortex of walnut seedlings usually darkens when the solution is markedly unfavorable. For example, if we place seedlings in $Ca(OH)_2$ solution of pH values in excess of pH 10, the root stops growing and the cortex becomes somewhat brown. If we then place the seedlings in a $Ca(OH)_2$ solution of more favorable pH, the root begins to elongate, and as it does so the darkened cortex develops a banded appearance in the zone of elongation (fig. 7). Figure 8 shows the effect of $Ca(H_2PO_4)_2$ solutions upon the root tips of walnut seedlings grown in water cultures. When 30 p.p.m. Ca was used, the

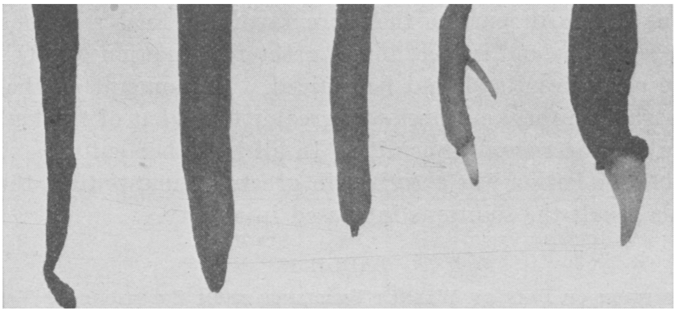

Fig. 8. Walnut-seedling roots showing injury and recovery. The three roots on the left were dead for a short distance back of the apex. The two roots at the right had regenerated new root tips and were beginning to recover.

changes in the effects brought about by this acid solution were readily followed. The roots died at the tip but not very far back. The cortical tissue then decayed away, leaving the central cylinder or stele still protruding. Gradually this disappeared and the root made another effort to grow from the apical end. Figure 9 shows successive injury and recovery and the broken-banded condition of the new roots similar to that shown in figure 7. The seedlings absorbed from a $CaSO_4$ solution about the same proportion of Ca as from the $CaCl_2$ solutions of equal concentration. The development of acidity in the $CaSO_4$ cultures is striking, especially in view of the relatively short duration of the absorption period in the first three cultures, where in some cases the epicotyls had scarcely emerged from the nuts at the end of the period.

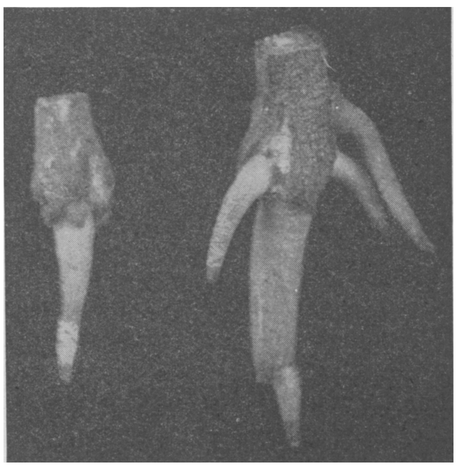

Fig. 9. Walnut-seedling roots showing successive injury and recovery incident to growth in a solution of $Ca(H_2PO_4)_2$. The way in which apical and lateral roots were formed from the stele indicates some difference in the vitality of the tissues of the roots.

The absorption of ions from solutions of calcium nitrate was considerably greater than from solutions of any other salts discussed in this connection. From the weakest solution the plants absorbed in a 20-day period 50 per cent and in a 47-day period 80 per cent of the calcium. The greater absorption of Ca in the case of the nitrate solutions is due in part to the more favorable final reactions of the culture solutions and in part to the greater absorption of NO_3.

Two other results should be noticed. The amount of the nitrate ion absorbed in these cultures was greater than that of the kation and the solutions decreased in acidity. In all the other cultures shown in this table the kation was absorbed in greater amounts than the anion, and as a result the solutions increased in acidity.

TABLE 15

ABSORPTION OF IONS BY WALNUT SEEDLINGS FROM SOLUTIONS OF VARIOUS CALCIUM SALTS

Twelve seedlings to a liter.

Salt	Initial concentration of Ca	Length of absorption period	Reaction		Ions absorbed by plant		Per cent of Ca absorbed
			Initial	Final	Ca	Anion	
	m.e.	days	pH	pH	m.e.	m.e.	
$CaCl_2$	7.764	20	6.8	4.8	1.238	0.742	16
$CaCl_2$	15.100	20	7.8	4.8	2.096	1.689	14
$CaCl_2$	22.914	20	8.3	4.8	2.665	2.160	12
$CaCl_2$	7.685	31	5.4	<4.8	2.071	1.371	27
$CaCl_2$	15.838	31	5.8	<5.2	3.583	2.707	23
$CaCl_2$	23.463	31	5.6	<5.2	5.479	4.436	23
$Ca(H_2PO_4)_2$	7.575	23	3.8	4.2	1.637	0.652	22
$CaSO_4$	6.208	8	5.5	4.6	0.309	0.227	5
$CaSO_4$	12.250	8	5.8	4.8	0.494	0.198	4
$CaSO_4$	24.750	8	5.8	4.8	1.048	−.121	4
$CaSO_4$	20.928	20	5.4	4.0	3.124	1.874	15
$CaSO_4$	30.948	20	5.5	4.2	4.052	4.008	13
$Ca(NO_3)_2$	8.064	20	4.8	5.8	3.952	4.581	49
$Ca(NO_3)_2$	15.773	20	5.2	5.8	4.466	5.152	28
$Ca(NO_3)_2$	23.967	20	5.2	5.8	4.920	5.297	20
$Ca(NO_3)_2$	15.639	47	5.0	7.2	12.575	15.508	80

5. *The effect of other kations on the absorption of calcium by walnut seedlings.*

It has frequently been mentioned that the absorption of ions is modified by the presence of other ions. In their natural habitat the plant roots are in contact with many ions. It may, therefore, be interesting to note the effect of Na and K ions upon the absorption of Ca as shown in table 16.

The absorption of Ca from solutions of $CaCl_2$ was slightly diminished when NaCl or KCl salts were also present, although the absorption of Cl was increased. This diluting effect has been rather fully considered in other studies.[9] The addition of KCl or K_2SO_4 to $Ca(NO_3)_2$ solutions caused a final reaction more acid than the initial one, although when $Ca(NO_3)_2$ alone was used, there was a decrease in the acidity (table 15).

TABLE 16

THE EFFECT OF OTHER KATIONS ON THE ABSORPTION OF CALCIUM BY WALNUT SEEDLINGS

Culture solution	Initial concentration of Ca	Length of absorption period	Reaction		Ions absorbed		Per cent absorbed
			Initial	Final	Ca	Anion	
	m.e.	days	pH	pH	m.e.	m.e.	
$CaCl_2$	12.400	26	5.6	4.6	2.630	2.152	21
$CaCl_2$	24.880	26	6.0	4.6	3.453	2.829	14
$CaCl_2$+21.943 milliequivalents of Na as NaCl	12.744	26	5.4	4.6	2.126	3.706	17
$CaCl_2$+22.381 milliequivalents of Na as NaCl	25.050	26	6.0	4.7	3.024	4.746	12
$Ca(NO_3)_2$+7.424 milliequivalents of K as KCl	7.744	32	4.8	4.4	1.996	......	26
$Ca(NO_3)_2$+7.342 milliequivalents of K as K_2SO_4	8.044	32	5.0	4.6	2.774	......	34

6. *The passage of solutes from walnut seedlings into the solution.*

Walnut seedling roots soon die when maintained in pure distilled water or in calcium-free solutions.[8] On the contrary, they grow for some time if maintained in solutions of a calcium salt. If the seedlings with the cotyledons attached are kept for some time in moist moss or sand they make normal growth, limited only by the amount of material which the cotyledons can supply. The conditions responsible for the sudden injury to roots in distilled water, or in calcium-free solutions, form the pertinent starting point for an inquiry into the question of ionic exchange between the root and its environment.

Walnut seedlings are very sensitive to calcium salts, as was illustrated by an accidental introduction of a small amount of tap water into the distilled water which was being used. The roots made some growth whereas injury was soon apparent when the roots were kept in pure distilled water. It is remarkable that the harmful effects of distilled water upon walnut roots are so quickly evident. Within an hour the color of the sub-apical meristematic region of the root changes from healthy white to pale yellow. During the next few hours (depending somewhat upon the temperature) the sub-apical region changes from yellow to dark brown, probably due to the oxidation of a chromogen, which later appears to diffuse out of the roots, leaving them lighter colored.

While these color changes are occurring, the roots become gelatinous and eventually quite slimy for some distance back of the tip. The condition is altogether typical of that found in previous experiments[8] in calcium-free media. It has been shown that the gelatinous roots will produce laterals if they are transferred to suitable nutrient solutions before injury has involved the vascular system.

Some additional data are here given which bear upon the question of injury in pure distilled water. The experiment was a preliminary test to show whether the amount of ash constituents in a root is reduced when distilled water is used as a culture medium. One set was grown in nutrient solution, another in distilled water and a third in a saturated atmosphere. In distilled water there was opportunity for electrolytes to diffuse out of the roots. Only the seedlings in distilled water showed injury.

TABLE 17

EFFECT OF DISTILLED WATER ON THE COMPOSITION OF WALNUT SEEDLING ROOTS

	Control seedlings from damp moss	Seedlings after 3 days in distilled water
Fresh weight of 25 roots	25.00 grams	28.00 grams
Ash in the dry matter	7.12 per cent	6.40 per cent
Constituents of the ash:		
Na	4.65 per cent	3.20 per cent
K	26.77 per cent	17.39 per cent
Ca	.61 per cent	.36 per cent
Mg	1.07 per cent	.52 per cent
SO_4	3.65 per cent	1.72 per cent
PO_4	38.60 per cent	33.06 per cent

Twenty-five seedlings whose roots were 10 to 20 cm. long were placed in a culture jar containing 20 liters of carbon-treated distilled water. The roots from seedlings grown 3 days in distilled water and from seedlings taken directly from damp moss were cut just below the cotyledons, dried at 60° to 70° C and analyzed. Table 17 shows the composition of the roots of each set. The ash constituents of roots immersed for 3 days in distilled water were less than for those taken directly from the moss. The greatest relative difference was in the case of Ca, Mg, and SO_4. Our interest centers in the loss of Ca, because, as we have shown in another paper,[8] similar injury was noted when walnut seedlings were grown in nutrient solutions lacking Ca.

With our present knowledge of the salt requirements of plants it seems difficult to understand this Ca relation. In general there is no exact minimum requirement; on the contrary, as often shown, there is a more or less wide range in their salt requirements. One would be slow to ascribe the sudden and profound effects observed to a change in Ca content from 0.6 per cent to 0.4 per cent. It is, of course, probable that there is a gradient for these ions in the roots, and that the diminution of Ca in the subapical region may have been much greater than that shown by the analysis of the whole root. The distribution of calcium in the walnut-seedling root was determined on a lot of roots which were about 12 cm. long when removed from the moss in which they had germinated. The roots were cut off immediately below the cotyledons and divided into basal, middle, and apical portions.

The calcium content of these roots is shown in table 18. The apical portion appears to contain more calcium than the others, although it is in this region that injury first appears when the roots are grown in calcium-free solutions.

TABLE 18

THE DISTRIBUTION OF CALCIUM IN THE ROOTS OF WALNUT SEEDLINGS DEPENDENT UPON THE COTYLEDONS

	Fresh weight	Dry weight	Per cent of Ca in	
			Fresh root	Dry matter
	(grams)	(grams)		
1. Basal part	43.68	4.89	.004	.038
2. Middle part	15.30	1.14	.008	.105
3. Apical part	7.87	.59	.009	.119

When walnut seedlings are grown several weeks in the usual nutrient solution the amount of Ca in the roots ranges from 3 to 5 per cent of the ash. Their total Ca content under favorable conditions for absorption is, therefore, not very large. Cranner[3] states that the presence of calcium is necessary to maintain the integrity of the bounding layer of the protoplast. In distilled water enough calcium may have diffused out of this layer to alter the permeability relations to a harmful degree without depleting to a corresponding extent the calcium content of the cell. Further study is necessary to determine whether the loss of this small amount of Ca from the walnut root is the primary cause of the injury noted.

7. *The absorption of sodium ions by walnut seedlings.*

A comparison of the ions absorbed from the nutrient solution and from the same with the addition of NaCl is given in table 19. In each case 12 walnut seedlings were grown for 39 days in 1 liter of solution. When walnut seedlings were grown in nutrient solutions

TABLE 19

ABSORPTION OF IONS FROM NUTRIENT SOLUTION WHEN SODIUM CHLORID WAS PRESENT

	Milliequivalents absorbed	
	Nutrient solution	Nutrient solution+87 m.e. of Na as NaCl
Ca	6.507	3.214
Mg	3.637	2.570
Na	0.109	12.538
K	3.981	3.295
Cl	0.319	12.574
SO_4	2.363	1.937
NO_3	....	....
PO_4	3.473	2.451
HCO_3*	....	−2.56

* Determined by titration with methyl orange.

containing fairly large amounts of NaCl there was usually a characteristic suppression of growth in the epicotyls.

The results obtained from analyses of the residual solutions after the seedlings had grown for 39 days, show that the introduction of NaCl retarded the absorption of Ca more than that of the other kations. This effect has already been shown by the analysis of walnut roots grown in nutrient solutions containing sodium salts. We note also that Na and Cl were absorbed in molecular equivalents.

Additional experiments were made to determine the effect of NaCl in a more concentrated nutrient solution (table 20).

The concentration of the nutrient solutions was 1455 p.p.m. and 7275 p.p.m. respectively. Approximately 1275 p.p.m. of NaCl was added to one set and 2550 p.p.m to another. The four solutions in the order listed in table 20 had total concentrations, therefore, of 2730, 8550, 4005, and 9825 parts per million.

The roots of all seedlings in the four solutions showed no injury from the salts present, but the tops of the seedlings were affected. In solution No. 3 the margins of the leaves were killed in places. In solution No. 4 only two of the 12 seedlings put up epicotyls; the remainder were barely able to emerge from the shells. The plants which grew in the two weaker solutions (Nos. 1 and 2) were good in every respect.

The increased amounts of sodium apparently restricted the absorption of other kations. It seems evident from these data that the presence of an excess of NaCl operates to prevent the absorption of other ions, especially Ca. When the strength of the nutrient solution was increased fivefold there was no substantial difference in the amount of Na and Cl ions absorbed.

TABLE 20

ABSORPTION OF IONS BY WALNUT SEEDLINGS FROM NUTRIENT SOLUTIONS CONTAINING SODIUM SALTS

Twelve seedlings per liter were grown for 42 days.

Solution		Reaction		Initial concentration in milliequivalents						Ions absorbed by plants in milliequivalents						Ions gained by solution in milli-equivalents
No.	Composition	Initial pH	Final pH	Na	K	Ca	Mg	Cl	SO_4	Na	K	Ca	Mg	Cl	SO_4	HCO_3*
1	Nutrient+1275 p.p.m. NaCl	5.4	4.8	19.717	6.633	8.234	7.192	22.219	....	6.185	5.445	5.539	3.965	7.594	....	0.148
2	(Nutrient×5)+1275 p.p.m. NaCl	5.0	6.5	23.314	24.389	39.022	25.476	23.189	....	7.265	9.731	12.874	0.977	6.788	....	3.049
3	Nutrient+2550 p.p.m. NaCl	5.4	6.5	39.598	10.268	7.754	7.430	42.820	....	12.274	6.272	3.962	2.947	11.511	....	1.207
4	(Nutrient×5)+2550 p.p.m. NaCl	5.0	6.5	43.196	24.781	39.571	25.024	45.185	....	13.076	6.966	14.521	0.920	13.350	....	2.453
5	Nutrient+640 p.p.m. Na_2SO_4	5.7	4.9	11.241	....	15.569	8.612	....	12.628	4.978	....	10.130	5.451	....	5.281	....
6	Nutrient+1550 p.p.m. Na_2SO_4	4.9	5.3	22.178	....	8.084	7.233	....	26.121	5.981	....	5.639	4.507	....	5.042	....
7	Nutrient+1550 p.p.m. Na_2SO_4 and 1275 p.p.m. NaCl	5.4	7.0	44.216	....	8.184	7.299	21.029	25.599	6.380	....	4.232	3.744	4.506	4.709	....
8	Nutrient+3300 p.p.m. Na_2SO_4	4.9	5.0	41.451	....	7.884	7.627	....	49.758	11.701	....	3.423	3.251	....	9.108	....

* Determined by titration with methyl orange.

When the amount of calcium was increased fivefold there was increased absorption, though not in proportion to the amount present. The case of magnesium appears somewhat unusual, because there was least absorption from solutions which contained most. It is possible that the absorption of calcium resulted in lowering the absorption of magnesium, which enters less readily into loose combination with the colloidal constituents of the cell. The diminished Mg absorption shown by solutions 5 to 8, accompanying increasing concentrations of Na ions, agrees with the reduced Ca absorption. The chlorin ion was absorbed in amounts substantially equivalent to those of its kation.

The effect of sodium sulphate was also studied (table 20). Some of the walnut leaves in solution 8 showed salt burn at the margins. The seedlings in solutions 5, 6, and 7 made excellent growth.

These results suggest that the roots have come into some sort of equilibrium with the ions in the solutions and that ions lost in the initial stage of the experiment may be absorbed later. The results reported by Hoagland[4] were obtained by placing a mass of roots of healthy plants into solutions for two days. In many cases he found a passage of ions from the plant to the solution where no doubt there would have been a net absorption in the course of time. However, our results agree in the main with those of Hoagland.

8. *The relation of the hydrogen-ion concentration to ionic interchange.*

In several of the foregoing experiments it was shown that the unequal rates of absorption of kations and anions resulted in changes in acidity. Our results agree with those of Hoagland[5] who concluded that the changes in reaction are due to unequal absorption and to the giving off of ions by the root. Many experiments have shown that the pH of the solution between pH 4.5 and 8.5 has practically no effect on the growth of walnut seedlings. This may be due to the large storage of reserve food within the seed. When the walnut plants grew in solutions well supplied with nitrate there was usually an accumulation of HCO_3 in the solution. When $Ca(NO_3)_2$ was furnished this effect was the more pronounced because of the rapid absorption of the NO_3 ions. The methyl orange titration of the nutrient solution calculated as HCO_3 was often as high as .7 milliequivalents at the end of a 33-day growth period and as high as 5.0 milliequivalents at the end of a second 33-day period.

The effect of ion interchanges is well shown by data in table 15. The net effect of absorption from $CaCl_2$ or $CaSO_4$ solutions was an increase in the H-ion concentration, while with absorption from $Ca(NO_3)_2$ solutions the OH-ion concentration increased. The pH of the $CaCl_2$ or $CaSO_4$ solutions usually reached 4.8 or 5.0 without evident injury to the roots. When plants were put into solutions of $Ca(NO_3)_2$ growth continued for a longer time and pronounced increases in pH were observed. For example, the last set of cultures in table 15 grew for 47 days, during which time they absorbed most of the ions from the solution. The solution had a pH of 7.0–7.2 at the end of the period and upon heating to expel CO_2 it changed to pH 8.0. A small variation in pH, however, may be more significant in the more acidic solutions than is a large variation in the less acidic solutions.

C. General Discussion

When a living cell is in contact with a solution, absorption and excretion occur. The process never reaches an end-point because the ions absorbed by the plant promote its growth, thereby increasing its power or capacity further to absorb (or excrete) ions. It presents, therefore, a moving equilibrium. The exchange of ions is influenced by a great variety of conditions, such as the concentration of the solution, the nature and concentration of the ions present, the effect of one ion upon the absorption of another, temperature, light, and other factors. In fact, the absorption of ions is veiled by a host of factors, few of which are as yet understood.

Since energy is undoubtedly expended in the uptake of ions by plants, it is to be expected that the ratio and amount of this uptake should vary with the plant. The results herewith reported may therefore supplement those of Hoagland,[4] obtained from experiments with other plants. There is abundant evidence in this paper to support the idea that the processes of absorption are concerned with ions. The rate of absorption is not related to the velocities, nor to other physical-chemical properties of the ions so far as they are known. The amount of an ion absorbed is not strictly related to the amount already present in the plant. For example, phosphate was rapidly absorbed by walnut seedlings, although their cotyledons are very rich in phosphorus. We can only conclude therefore that their absorption is related to some chemical or physical property of the protoplasm.

Both citrus and walnut seedlings had a greater absorptive power for potassium, calcium, and nitrate ions than for magnesium, chlorin, or sulfate ions.

The effect of one ion upon the absorption of another was very pronounced, and occurred whether the ions had like or unlike electrical charges. Potassium ions retarded the absorption of calcium more than sodium, but sodium did not retard the absorption of potassium. One of the striking features of these experiments is the rapidity with which potassium ions were absorbed. Whenever this ion was abundant it was absorbed, usually to the exclusion of other kations and sometimes anions.

In a soil where the rates of renewal and diffusion are slower than in a water culture there might not be such an accumulation of potassium in the plant. However, if the concentration of potassium were fairly high the absorption process might lead to exactly such a relation between potassium and calcium as we find in "mottle-leaf" and kindred troubles of citrus trees. There is evidence from data upon the absorption of ions by trees that the ratio of potassium to calcium absorbed normally becomes smaller with the growth of the tree, but the possibility of a later reversal and a return to this early condition exists.

Almost without exception the plants absorbed greater amounts of kation than anion, and the rate at which one was absorbed appeared to influence that of the other. The per cent of kations absorbed was always greatest when they were accompanied by a favorable anion like NO_3. The converse of this statement is generally true although the differences are not so striking.

The absorption of chlorin is of particular interest because of its occurrence in certain irrigated soils where it forms one of the constituents of "white alkali" salts. The detrimental effect of the absorption of any considerable amount of chlorin by plants has long been known. Our results indicate that the amounts of chlorin absorbed stand in the following order with respect to the accompanying kation: $Ca > K > Na > Mg$. It is probable that the larger amount absorbed is to a certain extent related to the growth promoting power of the individual kation. The larger plants thus produced have a large capacity to absorb ions. When barley plants of the same size were placed in the salt solutions the order of magnitude of chlorin absorbed was $K > Na > Mg > Ca$, according to Hoagland.[4] One of the effects of sodium chlorid upon the plant seems to be due to an interference with the absorption of calcium. As sodium chlorid accumulates in the leaves and meristematic regions it produces direct injuries, the nature of which is as yet unknown.

The nitrate ion was regularly absorbed in relatively large amounts even when the initial concentration was high. The presence of chlorin retarded the absorption of nitrate, but other ions seemed to have little effect upon its absorption. Where the initial concentration was sufficiently great the absorption of nitrate was sometimes greater than that of the corresponding kation. In the case of walnut seedlings, the absorption of phosphate ions was also large, except where the concentration of the hydrogen ion was too great for satisfactory growth. It appears that the cells of the walnut root have some extraordinary powers of combining with phosphates, because when grown in distilled water there was no excretion of that ion, although the concentration of phosphorus compounds in the root was high.

In all cases studied there was an exchange of ions between the root and the solution in which it grew, resulting generally in a change in the acidity of the latter. The CO_2 excreted by roots tends to maintain a reaction favorable to growth. The most important effect noted was where the rapidly absorbed nitrate anion was furnished with a more slowly absorbed kation. In such a medium a fairly high degree of alkalinity may be developed by the formation of carbonates, but there was no evidence that any carbonate was absorbed by the roots.

The significance of the results of this study seems to lie in giving a clearer understanding of the processes of absorption from a relatively simple medium. From these results we have tried to obtain a clearer idea of the principles governing the process of absorption.

IV. SUMMARY

1. The data presented in this paper deal with the absorption of ions from solutions by citrus and walnut seedlings and by young trees. They deal with the relative amounts of various ions absorbed, the effects of one ion upon the absorption of another, and the changes in the reaction of the solution due to ion absorption and excretion.

2. Rough-lemon and grapefruit seedlings removed relatively more potassium than calcium from solutions containing approximately equivalent amounts of these ions. When the potassium in the culture solution was low in amount, citrus seedlings absorbed more calcium, magnesium, and phosphate from the solution than when potassium was abundant. There was an interchange of ions between the solution and roots resulting in an excretion of potassium into the solutions when the original concentration of potassium was low.

3. Citrus seedlings absorbed more kation than anion from solutions of single calcium salts, causing an increase in acidity of the solutions. Calcium ions were readily absorbed by citrus seedlings when sodium and potassium were absent or low in amount.

4. The presence of chlorin ions reduced the absorption of nitrate ions by citrus seedlings. When various chlorids were added to culture solutions the greatest amount of chlorin was absorbed from those containing calcium chlorid. These citrus seedlings made the best

growth and as a consequence were able to absorb more chlorin without injury than were the other cultures. From culture solutions which contained 1000 p.p.m. sodium chlorid, citrus seedlings absorbed approximately equivalent amounts of sodium and chlorin.

5. The changes in reaction of culture solutions in which citrus seedlings have grown, may be attributed directly to differential absorption of ions, together with an excretion of certain ions. Acid culture solutions do not always change in the direction of neutrality nor do neutral solutions always change in the direction of greater acidity. In complete nutrient solutions citrus seedlings may, in a comparatively short period, bring about so great a concentration of H ions as to be injurious to the roots.

Bicarbonate ions were found in culture solutions from which citrus roots had removed nitrate ions, but effervescence of the ash of citrus seedlings is not conclusive proof that the CO_3 had been absorbed from the solution.

6. No stimulation of the growth of citrus roots occurred in solutions of sodium carbonate. When calcium was completely absent from the solution, a high initial alkalinity due to sodium carbonate was very injurious to the roots of citrus seedlings; so also was the complete absence of calcium from a culture solution containing a favorable amount of the other essential ions.

7. Young Valencia orange trees removed more calcium than potassium from the culture solutions, but in comparison with the initial concentration the absorption of potassium ranked close to that of calcium. The absorption of magnesium was not very high in comparison with that of calcium and potassium. The absorption of the chlorin and the sulfate ions was very low in comparison with that of the nitrate ion, when the concentrations of chlorin and of sulfate were not excessive.

In sand cultures, Florida Sour-orange seedlings, two years of age, removed practically all of the nitrate present, and about equal percentages of the initial concentration of calcium and potassium. The percentage of the initial concentration of magnesium removed was much less than that of either calcium or potassium.

8. The nature of the growth obtained with citrus seedlings in water cultures at different pH values depends not only upon the maintained pH of the solution, but also upon the pH of the original solution as well as upon the nature of the acid or alkali used in maintaining the desired pH.

9. Although the ash of walnut kernels contains approximately 60 per cent of phosphate, the walnut seedlings rapidly absorbed all

of the phosphate from a complete nutrient solution. Walnut seedlings removed less chlorin or sulfate than potassium from culture solutions containing an excess of potassium salt. When walnut seedlings were grown in solutions having equivalent concentrations of potassium chlorid or potassium sulfate, they removed practically the same amounts of calcium from both solutions. Although slightly more potassium was absorbed from the potassium sulfate culture, the absorption of sulfate exceeded that of chlorin. Except in the case of potassium nitrate, the potassium absorbed by walnut seedlings from a nutrient solution to which different potassium salts were added, always exceeded that of the anion added with it.

10. The total amount of magnesium absorbed by walnut seedlings was quite constant and was not affected appreciably by additions of potassium salts to the solution.

11. Walnut seedlings removed more kation than anion from solutions of single calcium salts except in the case of the nitrate. The acidity of the residual solution was entirely dependent upon the absorption rate of the two ions employed. In a solution of calcium acid phosphate having an unfavorable degree of acidity, walnut seedling roots made renewed attempts to elongate, even after the root tips had been killed by the excessive acidity.

12. The addition of the chlorids or sulfates of sodium or potassium to calcium solutions brought about a reduction in the absorption of calcium. The addition of potassium chlorid or sulfate to calcium nitrate caused a final reaction more acid than the initial reaction, although when calcium nitrate alone was used, walnut seedlings brought about a decrease in acidity. The presence of an excess of sodium chlorid in a culture solution prevented walnut seedlings from absorbing large amounts of calcium. Increasing the concentration of the nutrient solution containing large amounts of sodium chlorid caused no substantial differences in the amounts of sodium and chlorin ions absorbed by walnut seedlings.

13. The apical portion of walnut roots grown in calcium-free solutions contained more calcium than the portions further removed from the apex, although the first evidences of injury were visible in this apical portion.

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The titles of the Technical Papers of the California Agricultural Experiment Station, Nos. 1 to 20, which HILGARDIA replaces, and copies of which may be had on application to the Publication Secretary, Agricultural Experiment Station, Berkeley, are as follows:

1. The Removal of Sodium Carbonate from Soils, by Walter P. Kelley and Edward E. Thomas. January, 1923.
3. The Formation of Sodium Carbonate in Soils, by Arthur B. Cummins and Walter P. Kelley. March, 1923.
4. Effect of Sodium Chlorid and Calcium Chlorid upon the Growth and Composition of Young Orange Trees, by H. S. Reed and A. R. C. Haas. April, 1923.
5. Citrus Blast and Black Pit, by H. S. Fawcett, W. T. Horne, and A. F. Camp. May, 1923.
6. A Study of Deciduous Fruit Tree Rootstocks with Special Reference to Their Identification, by Myer J. Heppner. June, 1923.
7. A Study of the Darkening of Apple Tissue, by E. L. Overholser and W. V. Cruess. June, 1923.
8. Effect of Salts on the Intake of Inorganic Elements and on the Buffer System of the Plant, by D. R. Hoagland and J. C. Martin. July, 1923.
9. Experiments on the Reclamation of Alkali Soils by Leaching with Water and Gypsum, by P. L. Hibbard. August, 1923.
10. The Seasonal Variation of the Soil Moisture in a Walnut Grove in Relation to Hygroscopic Coefficient, by L. D. Batchelor and H. S. Reed. September, 1923.
11. Studies on the Effects of Sodium, Potassium, and Calcium on Young Orange Trees, by H. S. Reed and A. R. C. Haas. October, 1923.
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13. Some Mutual Effects on Soil and Plant Induced by Added Solutes, by John S. Burd and J. C. Martin. December, 1923.
14. The Respiration of Potato Tubers in Relation to the Occurrence of Blackheart, by J. P. Bennett and E. T. Bartholomew. January, 1924.
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17. Nutrient and Toxic Effects of Certain Ions on Citrus and Walnut Trees with Especial Reference to the Concentration and Ph of the Medium, by H. S. Reed and A. R. C. Haas. October, 1924.
18. Factors Influencing the Rate of Germination of Seed of Asparagus officinalis, by H. A. Borthwick. March, 1925.
19. The Relation of the Subcutaneous Administration of Living Bacterium abortum to the Immunity and Carrier Problem of Bovine Infectious Abortion, by George H. Hart and Jacob Traum. April, 1925.
20. A Study of the Conductive Tissues in Shoots of the Bartlett Pear and the Relationship of Food Movement to Dominance of the Apical Buds, by Frank E. Gardner. April, 1925.