

THE ABSORPTION OF PHOSPHORUS AND IRON FROM NUTRIENT SOLUTIONS¹

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(WITH FIVE FIGURES)

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Two of the more popular solutions for experimental culture of plants show a striking variation in the amounts of phosphorus used. Shive's so-called best solution, R₅-C₂ (4) contains 2.45 gm. per liter or 0.018 mols of KH₂PO₄, partly as a source of phosphorus, but more perhaps as an acid buffer tending to maintain the solution at pH 4.5-5.0. Hoagland's 1940 solution (2) on the other hand contains only 0.068 gm. per liter or 0.0005 mols of the same salt. The Shive solution thus contains 36 times as much phosphorus as the equally successful Hoagland solution. As a portion of a general study of ion balance in nutrient solutions we have compared the growth, color and phosphorus absorption of several plants in these mixtures and in two experimental solutions; the first ("X") somewhat resembling the Hoagland solution, but carrying more than half of its nitrogen as NH₄NO₃, and the second ("P") being a modification of Zinzadzé's buffered solution (6) carrying a moderately high concentration of phosphorus as a colloidal precipitate of the tricalcium salt and with the nitrogen again added as NH₄NO₃.

Methods

Plants were grown in quart mason jars with cork stoppers (3). Four seedlings in each jar were thinned to two to obtain maximum uniformity. Five replicates were used for each treatment and some of the experiments were repeated as many as five times. Corn (*Zea mays*), broccoli (*Brassica oleracea* var. *italica*), soybeans (*Glycine max*), tomatoes (*Lycopersicon esculentum*), sunflowers (*Helianthus annuus*), cotton (*Gossypium hirsutum*), and rice (*Oryza sativa*) were used in a main test of growth rates in April and May of 1945; corn, soybeans, and rice were used in June and July of the same year for studies of phosphorus absorption. Some of the plants were grown to fruiting, but, because of the small size of the culture jars, most of the work, including the phosphorus analyses, was done with plants 4 to 6 weeks old.

Concentrations of the salts used in grams per liter and of the various ions in millimols are shown in table I. Iron was furnished by 10 ml. of a 0.1% solution of ferric tartrate at each change of solutions and by 1.0 ml. additions of the same solution one to three times a week as required. All solutions received 1.0 p.p.m. of boric acid and 0.5 p.p.m. of ZnSO₄ at each change. After the seedling period of 2-3 weeks, solutions were changed at

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TABLE I

CONCENTRATIONS OF SALTS IN GRAMS PER LITER AND OF IONS IN MILLIMOLS
FOR FOUR NUTRIENT SOLUTIONS

SALT OR ION	HOAGLAND 1940 ("H")	SHIVE R ₅ -C ₂ ("S")	EXP. SOL. "X"	EXP. SOL. "P"
	<i>gm./l.</i>	<i>gm./l.</i>	<i>gm./l.</i>	<i>gm./l.</i>
NH ₄ NO ₃			0.25	0.286
KNO ₃	0.506		0.25	
Ca(NO ₃) ₂	0.591	1.228	0.25	
KCl			0.25	0.50
MgSO ₄	0.247	3.697	0.50	0.50
Ca ₃ (PO ₄) ₂				1.00
KH ₂ PO ₄	0.068	2.450	0.10	
Total	1.412	7.375	1.60	2.286
	<i>m. mols</i>	<i>m. mols</i>	<i>m. mols</i>	<i>m. mols</i>
Ca ⁺⁺	2.5	5.2	1.1	3.2
Mg ⁺⁺	1.0	15.0	2.0	2.0
K ⁺	5.5	18.0	6.6	6.7
NH ₄ ⁺			3.1	3.6
NO ₃ ⁻	7.5	10.4	9.7	3.6
SO ₄ ⁻	1.0	15.0	2.0	2.0
HPO ₄ ⁻	0.5	18.0	0.7	3.2

weekly intervals, except that solutions for larger plants (6-10 weeks) were changed at intervals of 3-5 days.

GROWTH OF PLANTS

The size ranking of plants started April 4-11 was estimated on May 10 and again on May 23. Differences between the best and poorest cultures

TABLE II

RANKING FROM BEST TO POOREST OF SEVEN SPECIES IN FOUR SOLUTIONS.
EXPERIMENT STARTED WEEK OF APRIL 4

PLANT	RANK ON BASIS OF TOP AND ROOT GROWTH			
	FIRST	SECOND	THIRD	FOURTH
May 10, 1945				
Corn	H	X	P	S
Broccoli	P	X	H	S
Soybeans	P	X	S	H
Tomatoes	P	X	S	H
Sunflowers	P	X	H	S
Cotton	P	S	H	X
Rice	P	X	S	H
Scores	P 26	X 19	H 12	S 12
May 23, 1945				
Corn	H	P	S	X
Broccoli	X	P	H	S
Soybeans	P	X	H	S
Tomatoes	P	X	S	H
Sunflowers	P	H	X	S
Cotton	P	H	S	X
Rice	X	P	S	H
Scores	P 25	X 18	H 16	S 11

were slight in tomato to marked in soybean and broccoli (fig. 1). Rankings together with a score obtained by assigning a value of 4 for first rank, 3 for second, etc., are shown in table II, and six of the plants are shown in figure 1.

On the basis of the scores in table II the "P" solution containing NH_4NO_3 and $\text{Ca}_3(\text{PO}_4)_2$ was the best for the plants tested, while Shive's $\text{R}_5\text{-C}_2$ ("S")



FIG. 1. Six species grown in four nutrient solutions. A, soybeans; B, corn; C, tomatoes; D, rice; E, broccoli; F, cotton. Left to right in each photograph: Hoagland's solution ("H"), Shive's $\text{R}_5\text{-C}_2$ ("S"), a buffered solution ("P") and an unbuffered solution with NH_4NO_3 ("X"). Plants started April 4-11, photographed June 15.

rated only one second place in the upper brackets. There was much evidence, however, that the "best" solution changes with the light and temperature conditions (1), with the plant, and perhaps most rapidly with the age of the plant. "X" was a good solution for young corn plants, but they soon developed root rots in the excessive acidity produced. On the other hand, larger plants did reasonably well when returned to this solution (fig. 1B).

Small corn in Hoagland's ("H") solution is chlorotic and may die unless especial attention is given to the iron supply. Even badly chlorotic plants recover quickly, however, if the phosphorus is omitted from the solution for the first half of the week. Figure 2A shows this effect. The solutions were identical except for the time of adding phosphorus. Soybeans also do poorly in "H" solution, but can be carried by delaying the phosphorus (fig. 2B). This principle was used by WEISS (5), working in this laboratory. Chlorosis was induced in genetic lines of soybeans to be tested for efficiency of iron absorption by adding moderate quantities of KH_2PO_4 to a large culture tank. A differential chlorosis developed among the lines which disappeared as the phosphorus was exhausted and more iron was added to the solution.

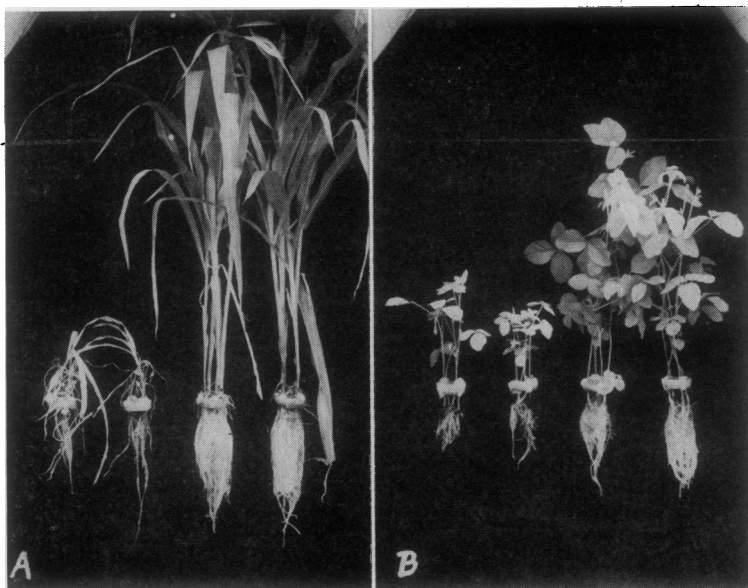


FIG. 2. Effect of alternating phosphorus and iron in Hoagland's solution. A, corn; B, soybeans; left in each photograph, phosphorus and iron added together at solution changes; right, phosphorus addition delayed 2-4 days to permit unhindered iron absorption.

The effect of phosphorus in causing chlorosis in "H" but not in "S" solution, which contains 36 times as much phosphorus, is explainable on the basis of pH. The pH's of fresh and used solutions in table III show that the large quantity of KH_2PO_4 tended to maintain an acid reaction favorable to iron absorption. Modified Shive solutions containing intermediate concentrations of acid phosphate are less successful. In Shive's original paper (4), solutions in the R_1 series contained KH_2PO_4 at 0.0036 mols; series R_2 at twice this concentration, R_3 at three times, etc., to R_7 . None of the R_1 or R_2 solutions was rated good while R_4 's and higher were mostly good to excellent, and three solutions, R_3-C_3 , R_4-C_5 , and R_7-C_2 , were not significantly poorer than R_3-C_2 .

TABLE III

HYDROGEN ION CONCENTRATIONS (pH) OF FRESH AND USED NUTRIENT SOLUTIONS

SOLUTION	FRESH SOLUTION	USED SOLUTION, 3-4-WEEK PLANTS	USED SOLUTION, 5-6-WEEK PLANTS
Hoagland 1940	5.20	5.80-6.35	5.40-6.55
Shive R ₆ -C ₂	4.45	4.85-5.65	4.45-5.70
Exp. sol. "X"	5.25	3.55-4.40	3.70-5.70
Exp. sol. "P"	6.10	5.50-6.30	5.15-5.60

Ammonium nitrate was added to solutions "X" and "P" as a neutral source of nitrogen. We were very considerably surprised, therefore, to find that the pH of the unbuffered solution "X" dropped as low as pH 2.9 and always went below 4.0. The cause of the acidity was shown to be the preferential absorption of ammonia ions by all of the plants studied at all stages of growth up to early fruiting. Ammonia was absorbed faster than NO₃⁻ from the unbuffered "X" solution, the buffered "P" solution or pure NH₄NO₃. Corn and cotton roots (fig. 1F) were injured in these acid solutions but soybeans made excellent growth with a very dark green color (fig. 1A). Young plants sometimes became chlorotic in "X" because they did not absorb nitrogen fast enough to develop an acid reaction. With large plants nitrogen absorption was so rapid that the acid phase was ended on the second or third

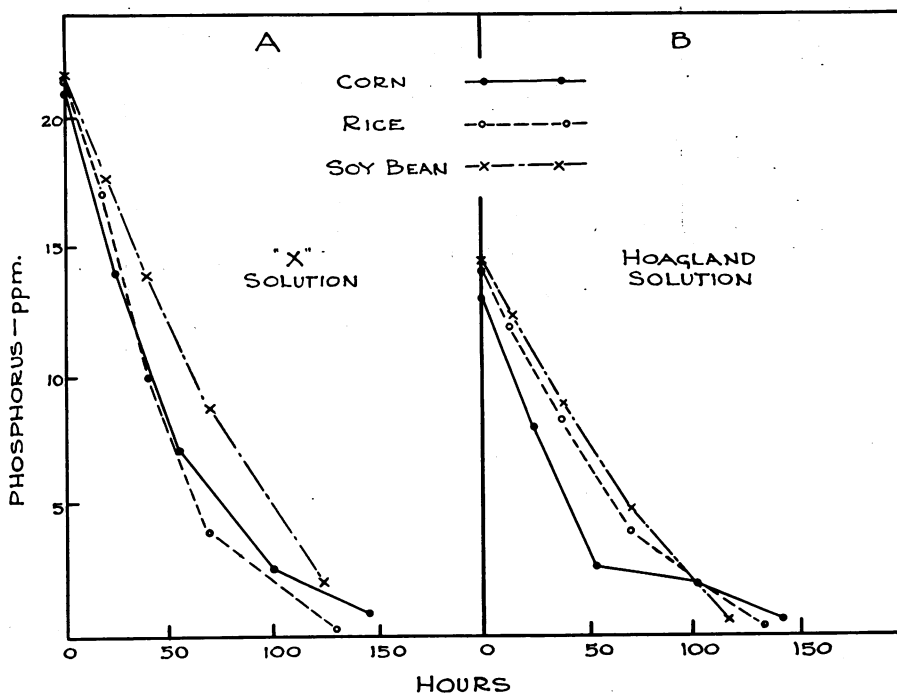


FIG. 3. Phosphorus absorption by three species grown in "X" and Hoagland solutions.

day by nearly complete absorption of both NH_4^+ and NO_3^- . In the "P" solution the $\text{Ca}_3(\text{PO}_4)_2$ prevented low pH values. At the lower pH's shown for this solution in table III, phosphorus became more soluble (fig. 5) and some tendency toward chlorosis was observed.

THE ABSORPTION OF PHOSPHORUS

The apparent interaction of phosphorus and pH in iron chlorosis led to a detailed study of PO_4 absorption from the four solutions. The results are shown graphically in figs. 3, 4, 5. The "H" and "X" solutions (figs. 3A and B) starting respectively with 14 and 21 p.p.m. of phosphorus dropped to 1 p.p.m. or less in 5 or 6 days. Essentially all of the 0.068 or 0.100 gm.

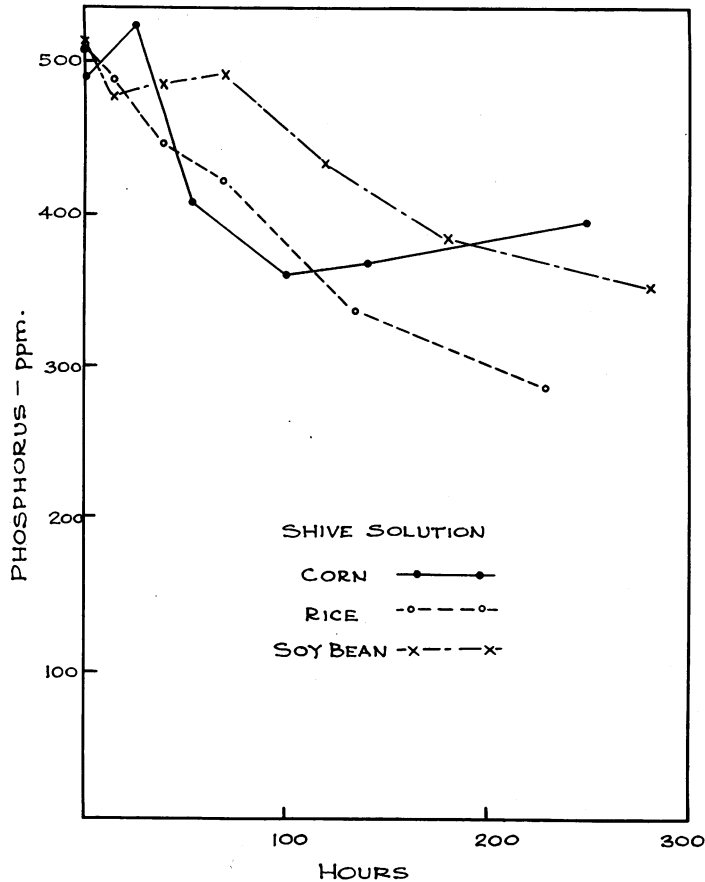


FIG. 4. Phosphorus absorption by three species grown in Shive solution.

of KH_2PO_4 added was absorbed. The 500 p.p.m. of phosphorus in the "S" solution was reduced to about 300 p.p.m. in a week (fig. 4) with a phosphorus absorption 10 or 15 times that in the "X" and "H" solutions. The fact that such heavy absorption did not seriously interfere with iron utilization is evidence that the interference observed with the "H" solution occurred out-

side the plant. Soluble phosphorus concentrations in the "P" solution, which contained a large excess of colloidal $\text{Ca}_3(\text{PO}_4)_2$, started at about 10 p.p.m. and climbed to 20–30 p.p.m. on the second and third days (fig. 5) as some of the insoluble phosphate was brought into solution by accumulating HNO_3 from the differential utilization of NH_4NO_3 . Soluble phosphorus then tended downward but remained above 10–20 p.p.m. after 10 days. The "P" solution has been best for small plants with slow NH_4^+ absorption so that the pH is held near 6.0. Very little iron chlorosis develops in these cultures in

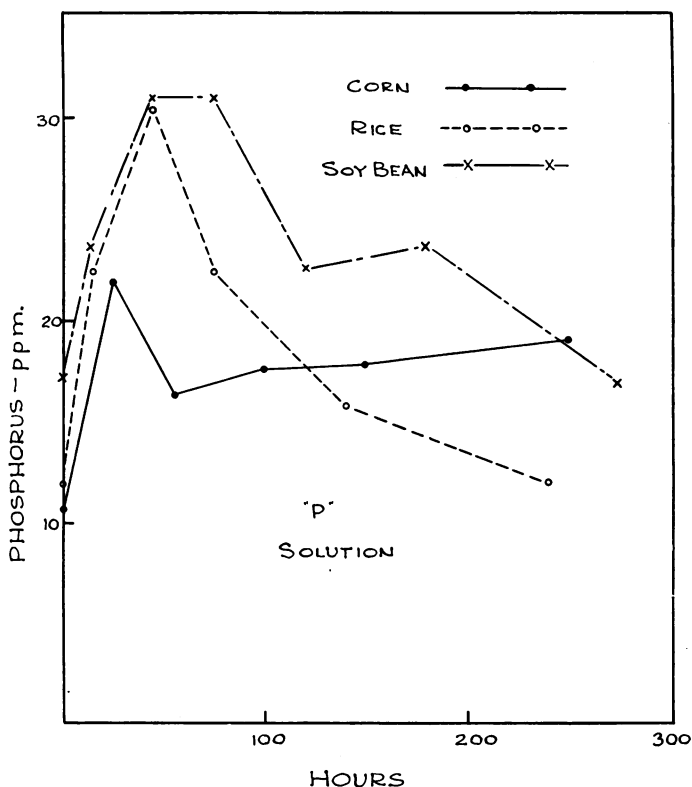


FIG. 5. Phosphorus absorption by three species grown in the $\text{Ca}_3(\text{PO}_4)_2$ buffered "P" solution.

spite of the high pH and phosphorus, suggesting that it is the dibasic or more probably the monobasic phosphate ion which precipitates the iron. The "P" solution would probably be improved for older plants by using some $\text{Ca}(\text{NO}_3)_2$ along with $\text{Ca}(\text{Cl})_2$ in forming the $\text{Ca}_3(\text{PO}_4)_2$. The result would be a partial replacement of KCl by KNO_3 , a higher total nitrogen and perhaps less tendency for excess solubility of the phosphorus.

Discussion and summary

Moderate amounts of KH_2PO_4 (0.5 to 0.7 millimols) caused iron chlorosis of seedlings in solutions less acid than pH 5.5–6.0. The trouble could be

avoided by omitting the phosphorus entirely and adding it separately after 2-4 days. Even with this modification the Knop type of solution (Hoagland, 2) caused persistent chlorosis in soybeans and broccoli.

In the commonly used Shive solution (R_5-C_2) a large excess of KH_2PO_4 holds the pH of the solution between 4.5 and 5.0 and chlorosis is moderate to slight. The Shive solution contains 36 times as much phosphorus as the Hoagland and 6-week-old plants growing in it absorbed 15 times as much to make a slightly poorer growth. The addition of as little as 0.125 gm. per liter of NH_4NO_3 resulted in decidedly acid solutions in every experiment with nine plant species representing seven families. Initial pH's of 5.25 in an unbuffered solution fell to minimums of 3.9 to 2.9 and then rose, to pH 5.0 or 6.0 with large plants. The low pH's were shown to be due to a preferential absorption of NH_4^+ ion and the later rise to the slower absorption of the NO_3^- ion. The use of NH_4NO_3 prevented iron chlorosis with some plants, notably soybeans, but resulted in acidities which were injurious to young corn and to cotton. In work done in this series but not described above, the "X" solution containing NH_4NO_3 gave exceptional growth of young coffee (*Coffea arabica*), but was toxic to older plants.

It is probable that iron chlorosis has more effect on solution culture results than any other single factor, and frequently than all other factors. Iron absorption from cultures is reduced by phosphorus, probably by $H_2PO_4^-$ ions especially, at pH's of about 6.0 or higher. The use of $Ca_3(PO_4)_2$ reduces the trouble as does the use of enough KH_2PO_4 to maintain a pH below 5.5. In tank culture H_2SO_4 may be used more cheaply to accomplish the same result. In miscellaneous work with the Knop type of solution chlorosis can be reduced or prevented by omitting phosphorus from the solution and adding it separately 2-4 days later after iron has been absorbed.

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