in the development of the apparatus and was a continued source of advice and assistance.

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PHOSPHORUS NUTRITION OF SOYBEANS [1,2]

ROBERT W. HOWELL
U. S. Regional Soybean Laboratory, Urbana, Illinois

New strains of crop species in variety development programs are continuously being evaluated with respect to one another and to established varieties. In such a program it is very desirable to know the effect of nutritional factors on the expression of characters utilized for selection. Because of the unique role of phosphorus compounds in metabolic processes and the extensive use of this element in fertilizers, studies of how plants respond to varying levels of phosphorus are particularly interesting. The objective of the present study is to determine the response of several soybean varieties to different phosphorus levels.

The general literature on phosphorus nutrition of plants has been reviewed recently by Mulder (10) and by Hewitt (6). Therefore only those papers pertinent to the present study are reviewed here.

Stumpf (15) discussed at some length the literature pertaining to the accumulation, translocation, and assimilation of phosphorus in plants. Biddulph (3) and Rediske and Biddulph (13), discussed the utilization of iron, and pointed out that iron, although taken up, cannot be utilized by Red Kidney bush beans if a moderately high level of phosphorus is maintained. Weiss (17) found a similar relationship between the iron and phosphorus nutrition of certain varieties of soybeans.

Eaton (4) studied the effects of phosphorus deficiency on the vegetative portions of soybeans of the variety Manchu 3 kept vegetative by an 18-hour photoperiod and harvested 35, 46, and 56 days after planting. A gradual increase in chlorosis was observed in the interveinal areas of the leaves of "minus phosphorus" plants. Similar chlorosis, which will be described and illustrated later, was observed in the work reported here and seems to be characteristic of phosphorus deficiency in soybeans.

Mederski (9) studied the effects of high and low levels of phosphorus supply on dry matter production and on the partition of N and P during the development of the soybean plant. He concluded that if P is supplied at a high level during the first eight weeks, it may be omitted thereafter with no effect on yield.

Materials and Methods

Plants of soybean varieties Lincoln, Chief, Adams, and Illini were grown in nutrient culture apparatus in the greenhouse. Adams was used only in the first two experiments of the series; Illini only in the third.

Seeds were germinated in a sand germination bench and allowed to grow there for about a week. The seedlings were then transplanted to 2-gallon glazed stone crocks. The crocks and the area around them were filled with no. 9 quartz sand. Two plants of a single variety were grown in each crock. The root systems of plants in the different crocks were thus isolated.

Six crocks comprised a unit which was supplied from a common source of nutrient solution. Nutrient solutions described below were supplied from the time of transplanting, except that phosphorus was omitted during the first 10 days. New solutions were supplied at intervals of 10 or 11 days.

In the first two experiments, each variety was grown under two sets of conditions: (1) "competitive," i.e., plants of all varieties were supplied nutrient solution from a single source; and (2) "noncompetitive," i.e., plants of only one variety were supplied from a given source. No difference was found between the performance of the varieties under these two sets of conditions. Only the first of these plans was used for the third experiment be-

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cause it permitted a wider range of phosphorus concentrations.

The nutrient solutions when prepared contained the following salts in the concentrations indicated:

$Ca(NO_3)_2 \cdot 4\,H_2O$	294.0	ppm
$(NH_4)_2SO_4$	44.5	"
KCl	57.8	"
$MgSO_4 \cdot 7\,H_2O$	506.0	"
Ferric tartrate	5.0	"
$CuSO_4 \cdot 5\,H_2O$	0.055	"
$ZnSO_4 \cdot 7\,H_2O$	0.055	"
H_3BO_3	0.61	"
$MnCl_2 \cdot 4\,H_2O$	0.39	"
$Co(NO_3)_2 \cdot 6\,H_2O$	0.055	"

The various elements were present initially in the concentrations shown below:

Element	ppm
Boron	0.11
Calcium	50.0
Cobalt	0.011
Copper	0.014
Iron	1.0
Magnesium	50.0
Manganese	0.11
Nitrogen	44.3
Potassium	30.0
Sulfur	76.0
Zinc	0.012

Phosphorus was supplied as phosphoric acid in the first experiment and as a solution of dibasic and monobasic sodium phosphate in the second and third experiments. The concentration of phosphorus was 2 or 10 ppm in the first two experiments and from 0.4 to 112 ppm in the third.

Additional ferric tartrate was supplied at the rate of 5 ppm five days after the solutions were prepared. There was no other addition to the solutions after their preparation.

A photoperiod of 14 hours was maintained throughout the experiments. Fluorescent lights were used to extend the natural photoperiod and to supplement natural illumination on cloudy days.

Observations were made on the following characters: Agronomic—height; number of days to flowering; number of nodes; number of pods; diameter of third node and third internode; dry weight of stems; dry weight of roots; dry weight of seeds. Chemical—oil, protein, phosphorus and ash content of the seeds; iodine number of the oil and phosphorus content of vegetative parts.

Oil and protein were determined by standard methods (1, 2). Phosphorus in the oil was determined by the method of Stoloff (14). Uptake of phosphorus by the plants was estimated from analyses—using the method of Parks et al (12)—of the nutrient solutions at the time they were discarded.

The agronomic data in experiments one and two were analyzed statistically according to the following plan:

Effect		df
Replications	R	3
Varieties	V	2
Nutrient Levels	L	1
RV		6
RL		3
VL		2
RVL		6
		23

The chemical data for these two experiments were analyzed according to the following statistical plan:

Effect		df
Replications	R	3
Varieties	V	2
Nutrient Levels	L	1
Competition	C	1
VL		2
VC		2
LC		1
VLC		2
Error		33
		47

The main effect "competition" refers to whether plants of only one or of more than one variety were supplied from a single source of nutrient solution. An effect of competition on performance would be indicated by a significant interaction of varieties and competition, since this would show that the varieties did not respond alike to the change in competitive conditions. The chemical data were obtained from analyses of composite samples of all beans from a given treatment and could therefore be included in a single analysis of variance. The agronomic data of the "competitive" and "non-competitive" groups could not be conveniently combined for statistical analysis since there were different numbers of plants involved in the two groups. In a special statistical analysis no significant differences were found in the agronomic characters attributable to the condition of competition so only data from the "non-competitive" groups are presented.

Data pertaining to agronomic characters and to oil and protein in the third experiment were analyzed statistically as follows:

Effect		df
Replications	R	3
Treatments	T	7
TR (Error a)		21
Varieties	V	2
VT		14
Error b		48
		95

Results and Discussion

The difference in growth of soybeans in the early stages on 2 and 10 ppm phosphorus is shown in figure 1. Even at this stage, plants with a restricted phosphorus supply were shorter and had smaller leaves than those of the other group. The appearance of leaves severely affected by phosphorus deficiency is shown in figure 2. The interveinal areas became necrotic very quickly after the first appearance of discoloration on the leaves. Necrosis began on either the primary or first trifoliate leaves and spread rapidly upward if no phosphorus was sup-

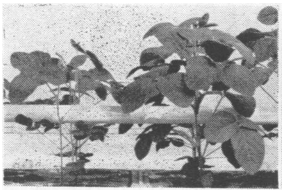



FIG. 1. Adams soybean plants, age 37 days, supplied phosphorus at the rates of 2 ppm (left) and 10 ppm (right).

FIG. 2. Lower leaves of Chief soybeans showing necrotic areas due to phosphorus deficiency. The necrosis developed after transplanting during the cycle when no phosphorus was supplied.

plied. When phosphorus was supplied in subsequent cycles, the appearance of necrosis on the younger leaves was prevented, but the older leaves showed no recovery.

The agronomic and chemical data from the first two experiments are summarized in tables I and II. There was a marked difference in response to the increase in level of phosphorus, using almost any of the agronomic or chemical characters as the measure. The size and yield of plants receiving phosphorus at the rate of 10 ppm were greater and the beans contained more oil, with a lower percentage of protein.

The higher phosphorus level apparently favored shoot over root growth. In five of six comparisons involving the three varieties in the first two experiments, the shoot/root ratio was higher in plants from the 10 ppm cultures than in those from the 2 ppm.

Although there were significant differences between the varieties for most characters under observation, in the first experiment only the oil percentage and in the second only four of the agronomic and none of the chemical characters showed a significant interaction of variety and phosphorus level. It will be shown below that such interactions might occur more frequently when a wider range of phosphorus levels is provided.

In the first experiment there was a swelling of the lower nodes associated with the 2 ppm P level, as is shown by the higher "swelling" index. The swelling was not observed on plants receiving 2 ppm P in the second experiment.

In order to obtain information regarding the response of soybeans to a wider range of phosphorus levels, plants were grown at levels from 0.4 ppm to 112 ppm. The resulting data are presented graphically in figures 3 to 8. Observations of other characters included in tables I and II gave comparable results and are omitted.

TABLE I

EFFECTS OF TWO LEVELS OF PHOSPHORUS ON AGRONOMIC CHARACTERS OF SOYBEANS

		HEIGHT	DAYS TO FLOWER	NUMBER OF NODES	SWELLING INDEX [a]	NUMBER OF PODS	STEM WEIGHT	ROOT WEIGHT	SEED WEIGHT
		MEAN VALUES PER PLANT							
		cm					*gm*	*gm*	*gm*
				Experiment 1					
Lincoln	2 ppm P	65.1	....	12.4	1.55	20.1	1.9	1.6	9.3
Lincoln	10 " "	99.5	....	15.3	1.38	36.9	4.0	2.9	16.5
Chief	2 " "	74.8	....	12.6	1.59	24.4	2.0	1.6	8.7
Chief	10 " "	122.7	....	11.5	1.25	38.6	4.4	3.1	14.3
Adams	2 " "	54.0	....	10.8	1.83	18.4	1.6	1.1	7.2
Adams	10 " "	69.7	....	10.3	1.51	29.1	3.1	2.3	11.8
F values	*df*								
Varieties	2	6.18 **	....	13.88 **	5.61 *	10.38 **	3.90 *	5.40 *	12.26 **
Levels of P	1	14.44 **	....	56.57 **	18.62 **	98.70 **	61.42 **	61.17 **	104.09 **
VL interaction	2	n.s.	....	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.
				Experiment 2					
Lincoln	2 ppm P	117.5	43.5	14.3	1.40	23.6	2.3	1.5	9.7
Lincoln	10 " "	133.7	42.6	15.8	1.45	33.5	3.3	1.9	13.5
Chief	2 " "	138.4	46.0	15.4	1.29	28.7	2.7	1.7	9.7
Chief	10 " "	200.4	47.0	18.5	1.30	38.3	4.4	1.9	13.6
Adams	2 " "	109.0	44.8	13.5	1.38	22.5	2.2	1.4	8.4
Adams	10 " "	156.7	44.3	16.3	1.44	31.7	4.5	2.0	12.7
F values	*df*								
Varieties	2	251.30 **	97.33 **	57.00 **	11.77 **	46.33 **	73.75 **	n.s.	9.14 *
Levels of P	1	599.74 **	n.s.	212.30 **	n.s.	283.30 **	1,017.50 **	18.71 **	281.11 **
VL interaction	2	62.29 **	8.83 *	8.31 *	n.s.	n.s.	46.75 **	n.s.	n.s.

[a] Ratio of third node to third internode is used as an index of swelling of lower nodes.
* Significance at P = 0.05 level.
** Significance at P = 0.01 level.

TABLE II

EFFECTS OF TWO LEVELS OF PHOSPHORUS ON CHEMICAL COMPOSITION OF SOYBEAN SEED

		In percent				Wijs iodine number
		Oil	Protein	Ash	Phosphorus	
		Experiment 1				
Lincoln 2 ppm P		19.9	38.9	4.69	.42	134
Lincoln 10 " "		22.4	32.9	6.03	.85	132
Chief 2 " "		19.6	38.9	4.96	.43	134
Chief 10 " "		21.5	33.5	5.96	.82	132
Adams 2 " "		20.7	38.8	4.82	.43	133
Adams 10 " "		22.0	34.2	5.72	.78	131
F values	*df*					
Varieties	2	23.35 **	n.s.	n.s.	n.s.	4.01 *
Levels of P	1	395.69 **	451.86 **	58.25 **	255.14 **	26.72 **
VL interaction	2	14.91 **	n.s.	n.s.	n.s.	n.s.
Variety-competition interaction	2	n.s.	n.s.	n.s.	n.s.	n.s.
		Experiment 2				
Lincoln 2 ppm P		19.7	43.6	5.00	.52	.....
Lincoln 10 " "		20.6	40.5	6.01	.90	.....
Chief 2 " "		19.1	43.3	5.04	.50	.....
Chief 10 " "		19.9	40.9	5.96	.88	.....
Adams 2 " "		20.1	43.3	4.91	.55	.....
Adams 10 " "		21.0	39.9	5.83	.88	.....
F values	*df*					
Varieties	2	21.29 **	n.s.	n.s.	n.s.	.....
Levels of P	1	47.97 **	182.05 **	273.00 **	1,395.6 **	.....
VL interaction	2	n.s.	n.s.	n.s.	n.s.	.....
Variety-competition interaction	2	n.s.	n.s.	n.s.	n.s.	.....

* Significance at $P = 0.05$ level.
** Significance at $P = 0.01$ level.

Two facts are clear from these data: (1) under the conditions of these experiments, 2 and 10 ppm phosphorus are both intermediate levels, at least for the variety Chief; and (2) the varieties differ markedly in their response to phosphorus. Growth of Lincoln and Illini plants was drastically curtailed by 50 or 112 ppm phosphorus (figs 3, 4, 6). Chief plants continued throughout the entire series of concentrations to show an increase in yield and number of nodes but not in height. Furthermore, the oil percentage in Chief beans increased over the entire series.

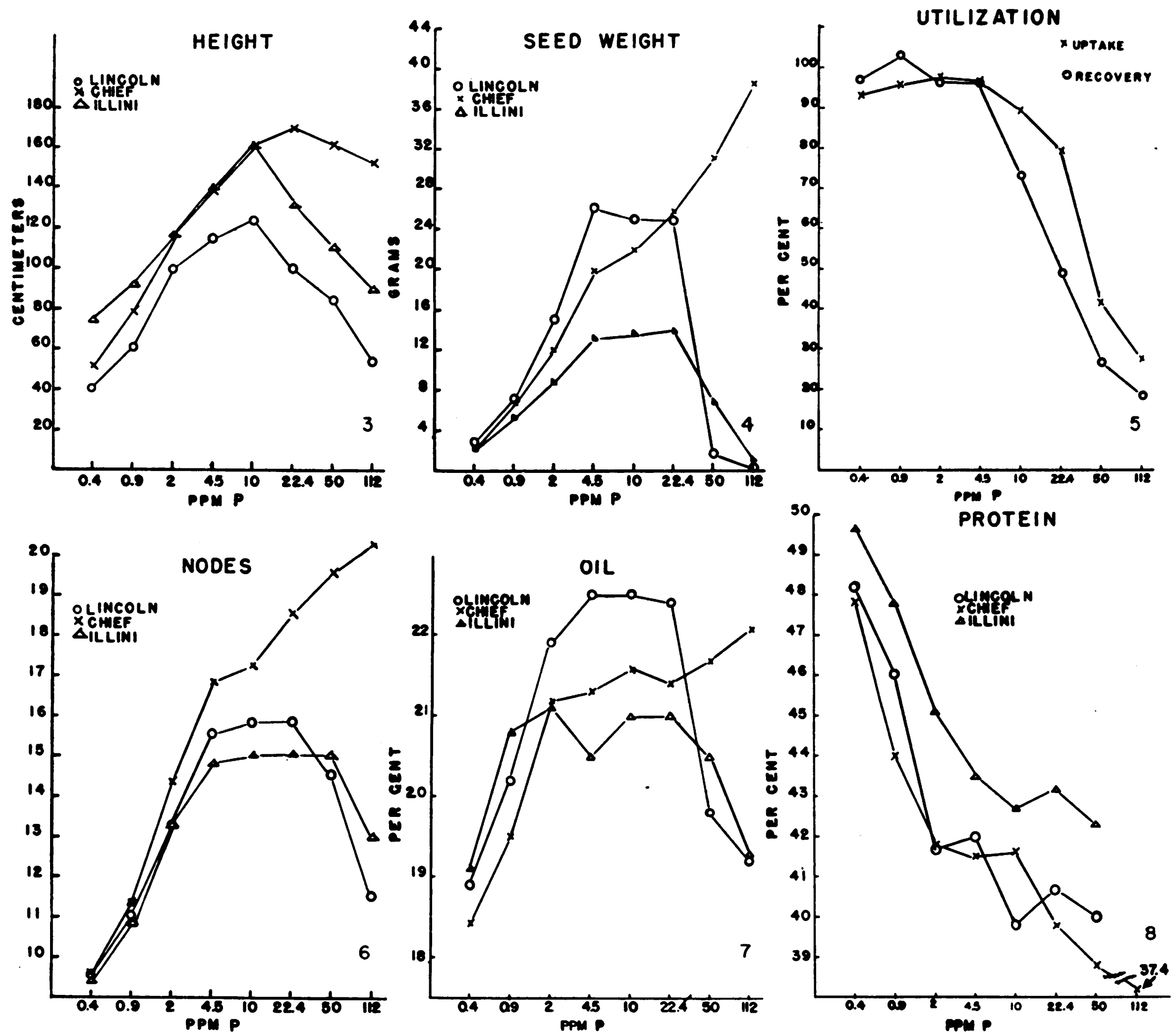
Lincoln plants performed more efficiently at the lower phosphorus concentrations. It will be seen (fig 4) that a seed yield equal to that of Illini at 4.5 ppm P would be produced by Lincoln on about one-third as much phosphorus. Chief was intermediate between the other varieties at this and other low phosphorus levels.

The 5-year average yields (1949–53) of these varieties in experimental plots at Urbana are as follows: Lincoln, 36.5 bu/A; Chief, 32.2; Illini, 29.0. It appears possible, therefore, that the relative productivity of these varieties may be partially determined by their relative efficiency in utilizing phosphorus.

As was found in the first two experiments, the phosphorus level affected the number of days to flowering very little, the principal effect being a slight retardation at the extremely low or extremely high phosphorus levels.

Data on phosphorus uptake and recovery are presented in figure 5. "Uptake" is the phosphorus disappearing from the solution during the periods between changes, including losses through precipitation, as well as phosphorus taken up by the plants. "Recovery" is the sum of the amounts of phosphorus found in the beans and vegetative parts, excluding the leaves. Recovery at levels up through 4.5 ppm was better than 95 %. This shows that phosphorus loss due to precipitation was not serious and that substantially all phosphorus in the leaves was translocated (presumably to the seed) before the leaves became senescent. At higher levels, recovery was progressively poorer, probably indicating that increasing amounts of phosphorus remained in the leaves or were lost through precipitation in the nutrient solution.

There were differences in the amounts of phosphorus recovered from the different varieties (table III), Lincoln plants containing the most at lower to moderate levels of supply. There appears to be an interaction between varieties and phosphorus level, but this could not be tested statistically since stem



FIGS. 3 to 8. Performance of three varieties of soybeans grown on nutrient solutions and supplied phosphorus at specified concentrations at beginning of each nutrient solution cycle of 10–11 days.

FIG. 3. Average height at maturity.

FIG. 4. Average weight of seed produced per plant.

FIG. 5. Utilization of phosphorus (average of all varieties)—"uptake" is the % available phosphorus disappearing from solution as determined by analysis of solutions at end of cycle, "recovery" is the amount of phosphorus found upon analysis of beans, pods, stems and roots as a percentage of "uptake."

FIG. 6. Average number of nodes at maturity.

FIG. 7. Average % oil in beans.

FIG. 8. Average % protein in beans.

and root samples from all replications were composited before chemical analysis.

The ratios of third node to third internode were significantly greater in the lower concentrations in the third experiment, agreeing with the data of the first experiment but not the second in this regard. Since the first and third of these experiments were carried out in the spring when day length and the intensity of natural illumination were increasing, it may be that the absence of swelling on plants in the second experiment was due to unrecognized light effects. Tentatively, however, it is concluded that such swelling may be an indication of phosphorus deficiency.

The data presented above show clearly that the increased vegetative growth on a more nearly adequate phosphorus supply, as reported by Eaton (4), results in increases of both yield (seed weight) and the proportion of oil stored in the seed. The increased synthesis of lipids may simply indicate a more favorable environment at the higher phosphorus level, since it has been found that the oil percentage will also be increased by more favorable temperature (7) and by irrigation under drought conditions (18).

TABLE III

PHOSPHORUS RECOVERED FROM SOYBEAN PLANTS GROWN IN NUTRIENT SOLUTIONS OF DIFFERENT PHOSPHORUS CONCENTRATIONS

VARIETY	P CONCENTRATION IN NUTRIENT SOLUTION (PPM)							
	0.4	0.9	2.0	4.5	10.0	22.4	50.0	112.0
	mg per plant							
Lincoln	12.72 *	30.98	71.44	167.71	241.58	314.42	23.35 **	4.16 **
Chief	12.60	29.31	60.94	137.09	230.99	323.07	441.59	553.49
Illini	10.61 **	27.19	54.12	106.42	181.57	232.72	80.47	19.71 **

* Values are the sums of weight of phosphorus recovered from all plant parts except the leaves.
** These figures include estimates of amount of phosphorus in beans as the total yield was too small to permit phosphorus determination.

There is also a possibility that the phosphorus level, by modifying the supply of organic phosphorus compounds, has a more specific effect on the oil forming mechanism. For instance, Kornberg and Pricer (8) have recently reported the enzymatic esterification of L-α-glycerophosphate and two moles of a 16- or 18-carbon fatty acid with accompanying conversion of two moles of adenosine triphosphate (ATP) to inorganic pyrophosphate and adenosine-5-phosphate. Catalytic quantities of coenzyme A were required. Van Baalen and Gurin (16) have shown that the incorporation of acetate to form fatty acids of longer chain length is dependent on the concentration of diphosphopyridine nucleotide and ATP as well as coenzyme A. The evidence, therefore, implicates phosphorus compounds in both (1) the formation of fatty acids, and (2) their esterification to form oils.

The presence of a similar system in plants may be inferred from the work of Newcomb and Stumpf (11) who found that synthesis of fatty acids by peanut cotyledons was inhibited by dinitrophenol, a compound which has been reported by Green (5) to inhibit the esterification of phosphate.

SUMMARY

The effect of the level of phosphorus in the nutrient solution on several agronomic and chemical characteristics of soybeans was studied.

The relative growth of plants on 2 and 10 ppm P and the characteristic appearance of leaves of plants suffering from phosphorus deficiency are illustrated.

It was found that increase in the phosphorus level from 2 to 10 ppm resulted in taller and heavier plants, greater yield, and a higher oil content in the beans of all of the varieties used.

A marked difference in varietal response was observed when a wider range of phosphorus levels was provided. The variety Chief continued to respond favorably to phosphorus levels as high as 112 ppm, whereas, the varieties Lincoln and Illini were adversely affected by levels of 50 or 112 ppm.

There was little or no effect of the phosphorus level on the number of days to blooming.

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