THE EFFECT OF pH ON THE ABSORPTION OF Sr^{89}, P^{32}, AND Fe^{59} IONS BY LEAVES OF *ZEA MAYS*[1]

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INTRODUCTION

Factors affecting the foliar absorption of cations, anions, and organic molecules have been studied intensively in recent years. The need for such information for studies on the foliar application of herbicides and fungicides, and the availability of radioactive labels for these chemicals have stimulated this research. Detailed information concerning the foliar absorption of specific ions or molecules and their absorption by particular species is still lacking, however. Moreover, the physiological explanation for some phases of the absorption process is largely unknown. The subject of foliar absorption has been recently reviewed by several authors (Boynton, 1954; van Overbeek, 1956; Wittwer and Teubner, 1959).

The hydrogen ion concentration of solutions of both ions and polar solutes applied to the foliage of plants has been considered an important factor in absorption (Orgell, 1954). Orgell (1957) reported that acidic substances were "sorbed" in greater amounts by excised cuticle disks as the pH of the surrounding solution was lowered from 7 to 1. For basic chemicals "sorption" increased as the pH was raised from 3 to 9. Studies on anion absorption by leaves have been largely confined to forms of phosphate. Low pH values of approximately 2 to 3 have brought about increased absorption of phosphate solutions compared to rates of uptake at or near neutrality (Swanson and Whitney, 1953). Little information is available on the effect of pH on the absorption of other anions with the exception of sulfate. Biddulph et al. (1956) found little effect of pH on the absorption of sulfate by bean leaves. Koontz and Biddulph (1957) found that the sodium salt of phosphate was translocated in greatest amounts from the leaf of red kidney bean to which it had been applied at a pH just below 5. For the potassium salt a pH of about 8 was optimum. This study did not give data for pH values of pH 4 or lower. Teubner et al. (1957) reported that the phosphate or citrate salts of K^{42} and Rb^{86} are absorbed most rapidly at pH 8.

This study is a contribution on the effect of pH on the absorption of ions of phosphorus, strontium, and iron by leaves of *Zea mays* L., a species not previously studied in this regard.

MATERIALS AND METHODS

Five grains of corn (Minhybrid 803) were sown per 4-inch pot in soil consisting of 2 parts silt loam to 1 part sand. Following germination and subsequent growth for 10 days in a greenhouse set at 70°F, seedlings were thinned to 3 per pot and treated.

Solutions of phosphorus-32, iron-59, and strontium-89 were prepared having an activity of 0.6 $\mu c/\lambda$ at a pH of 2.5, 3.0, 4.5, 7.0, and 8.2. The radioisotopes were obtained from the Oak Ridge National Laboratory as $FeCl_3$, $SrCl_2$, and H_3PO_4.

A 0.1 M citrate-phosphate buffer with citric acid and dibasic sodium phosphate was used to obtain pH 2.5 and 4.5. The pH of the distilled water is 3.0 and a

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0.1 M phosphate buffer with monobasic and dibasic sodium phosphate was used to make solutions of pH 7 and pH 8.2 (Colowich and Kaplan, 1955).

The plants were treated by placing 10-λ drops of the isotope solution containing 0.6 μc on the upper side of the second leaf of each corn plant. Three pots, each with three plants, were treated with each isotope at each pH. The application of the radioisotope took approximately 4 hours, and the plants were harvested in the reverse order of treatment to insure equal uptake time.

All parts of each plant but the roots were harvested, 24 hours after treatment. Each plant was separated into four morphological parts: the basal leaf was called the first leaf; the one above it the second leaf; the third leaf; and the fourth leaf including the shoot apex (fig. 1, harvest plan). All similar morphological parts from all three plants of each pot were counted together. The area to which the isotope was applied to the second leaf was removed at harvest and was not counted with the rest of the second leaf.

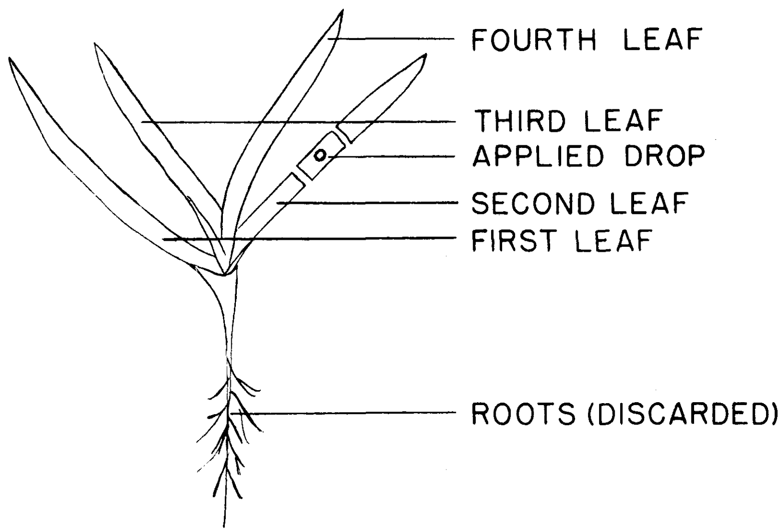

FIGURE 1. The harvest plan. The corn plants were divided into the parts as shown and all parts assayed for radioactivity except the roots. The rectangular area around the applied drop of the radioactive solution was removed before the second leaf was assayed.

The data were analyzed by means of analysis of variance. An analysis was designed to reveal whether there were diffierences in total uptake of isotope as a result of the pH of the applied solution, and a second analysis was made to evaluate the differences in accumulation of each radio-element in individual parts as affected by pH.

RESULTS AND DISCUSSION

Counts in the entire plants (table 1) at the pH values 2.5 and 4.5 are higher than for other pH values and in each case they are part of a nonsignificant group. Mean counts for pH 3.0 are in all cases intermediary, and in general mean counts for the pH values 7.0 and 8.2 are consistently lower than the others. In table 1 are total mean counts per minute per plant for all three isotopes. The counts are arranged in descending order of magnitude. For each of the three isotopes, differences in counts per minute among pH values were significantly different. In all three experiments these differences were significant at both the 0.05 and the 0.01 levels.

A Student-Newman-Kuehls Multiple Range Test was made on each array of means from analysis of variance and nonsignificant groups of means isolated. Brackets in table 1 enclose nonsignificant groups of means.

Relatively little phosphorus-32 moved downward in the plant and from the analysis of variance it is concluded that the difference in this movement due to pH was not significant. Significantly different amounts were taken up and a multiple range test of the means for the second leaf indicates that the pH levels 4.5 and 2.5 in almost all cases are significantly higher than the others.

Table 2 contains counts per minute per plant part for all pH values of the

TABLE 1

Total mean counts per minute per corn plant of P^{32}, Fe^{59}, and Sr^{89} at various pH values together with F ratios from analysis of variance

Phosphorus-32				Iron-59				Strontium-89			
	counts/min	0.05[a]	0.01[a]		counts/min	0.05	0.01		counts/min	0.05	0.01
pH 4.5	5944			pH 4.5	1075			pH 2.5	4249		
pH 2.5	5458			pH 2.5	963			pH 4.5	3703		
pH 3.0	4015			pH 3.0	584			pH 3.0	2185		
pH 8.2	3635			pH 7.0	470			pH 8.2	1319		
pH 7.0	3066			pH 8.2	31			pH 7.0	884		
F=10.816**[b]				F=43.715**				F=28.699**			

[a]Probability levels; brackets enclose groups of nonsignificant means.
[b]Analysis of variance F ratios; ** indicates significance at both the 0.05 and 0.01 levels.

TABLE 2

Mean counts per minute of phosphorus-32 in various parts of 10-day-old corn and at various pH values, together with F ratios from analysis of variance

1st leaf				2nd leaf				3rd leaf				4th leaf and apex			
	counts/min	0.05[a]	0.01		counts/min	0.05	0.01		counts/min	0.05	0.01		counts/min	0.05	0.01
pH 4.5	103			pH 2.5	3003			pH 4.5	698			pH 4.5	245		
pH 2.5	58			pH 4.5	2687			pH 2.5	442			pH 2.5	202		
pH 3.0	41			pH 3.0	2240			pH 7.0	250			pH 3.0	150		
pH 7.0	36			pH 8.2	2057			pH 3.0	233			pH 8.2	133		
pH 8.2	27			pH 7.0	1643			pH 8.2	224			pH 7.0	114		
F=10.816**[b]				F=3.876*				F=14.544**				F=13.006**			

[a]Probability level; brackets enclose groups of nonsignificant means.
[b]*significance at the 0.05 level and ** significance at the 0.01 level.

radioactive phosphorus-32. The table is arranged in descending order of magnitude of counts, with separate values for each morphological entity. Movement of the isotope into the first leaf, following absorption of the isotope by the second leaf, is downward in the plant, while movement into the third and fourth leaves is upward. Analysis of variance for the second leaf indicated significant differences only at the 0.05 level, with the means falling into two large groups of nonsignificant means. In all other instances F ratios indicated significance at both the 0.05 and 0.01 levels and the multiple range tests indicated several smaller

TABLE 3

Mean counts per minute of iron-59 in various parts of 10-day-old corn and at various pH values, together with F ratios from analysis of variance

1st leaf		2nd leaf		3rd leaf		4th leaf and apex	
counts/min 0.05[a] 0.01		counts/min 0.05 0.01		counts/min 0.05 0.01		counts/min 0.05 0.01	
pH 2.5	9	pH 4.5	585	pH 4.5	125	pH 2.5	353
pH 4.5	7	pH 2.5	517	pH 2.5	84	pH 4.5	343
pH 3.0	6	pH 3.0	355	pH 7.0	68	pH 3.0	182
pH 8.2	3	pH 7.0	255	pH 3.0	41	pH 7.0	144
pH 7.0	3	pH 8.2	15	pH 8.2	3	pH 8.2	10
F=1.415		F=22.660**[b]		F=11.192**		F=14.622**	

[a]Probability level; brackets enclose groups of nonsignificant means.
[b]* significance at the 0.05 level and ** significance at the 0.01 level.

TABLE 4

Mean counts per minute of strontium-89 in various leaves of 10-day-old corn and at various pH values, together with F ratios from analysis of variance

1st leaf		2nd leaf		3rd leaf		4th leaf and apex	
counts/min 0.05[a] 0.01		counts/min 0.05 0.01		counts/min 0.05 0.01		counts/min 0.05 0.01	
pH 4.5	4	pH 2.5	4241	pH 7.0	9	pH 7.0	6
pH 2.5	2	pH 4.5	3693	pH 2.5	6	pH 3.0	5
pH 8.2	1	pH 3.0	2175	pH 4.5	5	pH 4.5	2
pH 3.0	1	pH 7.0	1313	pH 3.0	4	pH 8.2	1
pH 7.0	1	pH 8.2	868	pH 8.2	3	pH 2.5	—
F=1.557		F=8.649**[b]		F=0.378		F=2.623	

[a]Significance level; brackets enclose groups of nonsignificant means.
[b]**significance at the 0.05 and 0.01 levels.

groups of nonsignificant means. For the first leaf, pH 4 was isolated as the only mean that differed significantly from the rest, while in the third and fourth leaves more groups appear. It is evident from the array of data that more often than not the pH values 2.5 and 4.5 significantly enhanced uptake and subsequent movement more than the others.

The uptake and movement of strontium-89, as deduced from the data of table 4, differ considerably from those of both phosphorus-32 and iron-59. In terms of counts, more strontium is taken into the second leaf of the plant treated with it than either phosphorus or iron into the second leaves of the plants treated with them, respectively, and less strontium-89, in comparison, is moved out of this locality. The effect of pH on uptake is given in the second leaf portion of table 4 and indicates that differences due to pH are significant. The groups of nonsignificant means for uptake are less well defined than with phosphorus and iron. Not only did a small amount of isotope move into the third and fourth leaves, but the amount of strontium-89 that moved was nonsignificant with respect to pH.

TABLE 5

The effect of pH on the percentage of P^{32}, Fe^{59}, and Sr^{89} translocated from the "applied leaf" to the rest of the corn plant

	Phosphorus-32 activity in counts/min by pH			
	2.5	4.5	7.0	8.2
Total activity in plant minus "applied leaf"	702	1047	400	384
Total activity in plant	5458	5944	3066	3635
% of total activity translocated	12.8	17.6	13.0	10.6
	Iron-59 activity in counts/min by pH			
	2.5	4.5	7.0	8.2
Total activity in plant minus "applied leaf"	446	717	215	16
Total activity in plant	963	1075	470	31
% of total activity translocated	46.3	66.7	45.7	52.2
	Strontium-89 activity in counts/min by pH			
	2.5	4.5	7.0	8.2
Total activity in plant minus "applied leaf"	8	10	16	15
Total activity in plant	916	3703	884	132
% of total activity translocated	0.87	0.3	1.8	0.40

The total amount of each radioisotope absorbed at any given pH varied; the most activity recorded was for phosphorus-32, next, for strontium-89, and least activity for iron-59. Since the actual activity counted depends partly on the counting efficiency, no direct comparisons of relative absorption among these three radioisotopes can be made. The efficiency of counting for the gamma emission of iron-59 is low compared to the counting efficiency for the other two isotopes and this may, in part, account for the lowest values for the activity in the radio-iron group. Some estimation of the relative rates of translocation of the three radioisotopes can be made, however. In table 5 are given the percentage values for the amounts of each radioisotope translocated from the "applied leaf" to the rest of the plant. This value is obtained by dividing the total activity in the plant by the total activity minus the activity in the "applied leaf." The amount of transport of strontium-89 under the conditions of the experiment is very small. With the exception of the plants which received strontium-89 at pH 7.0, less than 1 per cent of the total activity found in the plant was translocated out of the "applied leaf." For the radioisotopes of phosphorus and iron, the greatest translocation occurred among the plants receiving the radioisotope at pH 4. Relatively little difference in translocation occurred at pH 2.5, pH 7.0, or pH 8.2 for both radio-iron and radio-phosphorus.

Since pH may directly affect the amount of radioisotope absorbed by the leaf, this in turn would affect the amount of the element available for movement through the leaf blade. The amount of any particular element which reached the terminal phloem cells would be affected indirectly by the pH-dependence of the absorption mechanism at the leaf surface. Due to the buffer capacity of the non-conducting cells of the leaf as well as those of the phloem, it seems unlikely that the hydrogen ion concentration exerted any effect beyond the cells in the immediate vicinity of the application area. It is assumed here that the phloem cells are the main conductive tissue for translocation of these radioisotopes (Linck and Sudia, 1959; Swanson and Whitney, 1953).

SUMMARY

The absorption and translocation of phosphorus-32, iron-59, and strontium-89 by the leaves of 10-day-old corn plants were studied as functions of pH. The pH of the solution in which the mineral ion is supplied to the corn plants had a significant effect on absorption. For the three mineral ions studied, greater absorption occurred at the lower pH values of 2.5 and 4.5 and significantly lower amounts were absorbed at the higher pH values of 7.0 and 8.2.

All three mineral ions are readily absorbed by the leaves of corn but they differ markedly in the amount of each ion transported from the leaves. For the isotope of phosphorus, between 10 and 18 percent of the total activity in the plant is found in the stem and leaves following transport from the "applied leaf." These figures can be compared to from 45 to 67 percent of the iron translocated. Less than 2 percent of the radio-strontium was transported from the "applied leaf."

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