

Table IV. Acute Oral Toxicity in Rats of Several Diet-Pure Arsenic Trioxide Mixtures

<i>As₂O₃ Dry Diet Mixture</i>	<i>LD₅₀, Mg. As/Kg.</i>
Standard rat diet	
Protein 21%	
Fat 8%	145
Carbohydrate 50-60%	
Protein	
Dried egg albumin	133
Carbohydrate	
Sucrose-starch, equal parts	154
Fat	
Strained bacon fat	225
<i>As₂O₃ solution (distilled water)</i>	15

^a 48 hours.

as milligrams of elemental arsenic per kilogram of body weight.

Results

The acute oral toxicity, expressed as *LD*₅₀, of pure arsenic trioxide incorporated in a diet of carbohydrate or protein, was calculated to be 135 to 150 mg. of elemental arsenic per kilogram of body

weight (Tables II and III). When arsenic trioxide was administered in a diet of bacon fat, the acute oral toxicity was reduced to 225 mg. of elemental arsenic per kg. of body weight (Table III). Calculations were based on the deaths observed up to 48 hours.

Upon autopsy there was no evidence of gastrointestinal hemorrhage, although some slight mucosal irritation was noted in all animals. This confirms previous observations (3).

Comparative acute oral toxicity data summarizing the effect of diet-arsenic trioxide mixtures are presented in Table IV.

Discussion

Incorporation of arsenic trioxide in diets of protein or carbohydrate during this investigation has not significantly altered the acute oral toxicity from the previously observed value. However, when pure arsenic trioxide was fed to rats in a bacon-fat mixture, the acute oral toxicity was markedly reduced from the 145 mg. of arsenic per kg. observed in the standard diet mixture, to 225 mg. of elemental arsenic per kg. of body weight.

One may assume that the feeding of

an arsenic-fat mixture reduces the toxicity by coating the intestinal mucosa, thus decreasing absorption of the arsenic or by physically coating or chemically binding the arsenic compound, decreasing its availability for absorption. Therefore, the coadministration of fat with arsenic trioxide decreases the acute oral toxicity of arsenic.

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PHOSPHATE SOLUBILITY

Evaluation of Water-Insoluble Phosphorus in Fertilizers by Extraction with Alkaline Ammonium Citrate Solutions

T. P. HIGNETT and J. A. BRABSON
Tennessee Valley Authority, Wilson Dam, Ala.

Water-insoluble phosphorus in fertilizers is present mainly as dicalcium phosphate and hydroxyapatite. Agronomic tests show that dicalcium phosphate is an effective phosphorus fertilizer, but hydroxyapatite is much less effective. Official methods of analysis do not distinguish between these two compounds. An alkaline ammonium citrate extraction method was tested, whereby amounts of phosphorus present as dicalcium phosphate and hydroxyapatite can be estimated. Heavily ammoniated superphosphates contained a high proportion of hydroxyapatite; nitric phosphates contained predominantly dicalcium phosphate. This method should be useful in assessing the value of water-insoluble phosphorus in fertilizers.

IN AGRONOMIC tests to evaluate the effect of water solubility of phosphorus in fertilizers, the quality of the water-insoluble phosphorus is often an unknown and uncontrolled variable. This circumstance may give rise to anomalies in agronomic data and may cause different investigators to reach divergent conclusions. The aim of the present work was to provide a convenient method for roughly quantitative characterization of water-insoluble phosphorus in fertilizers.

About 80% of the fertilizer phosphorus used in the United States is in the form of mixed fertilizer. The great majority of the mixed fertilizers are manufactured by processes that involve ammoniation of superphosphate. Ammoniation decreases the water solubility of phosphorus by converting the monocalcium phosphate into ammonium phosphates, dicalcium phosphate, and more basic calcium phosphates. Also, ground limestone or dolomite often is incorporated in mixed fertilizer or added to

superphosphates; these materials also decrease the water solubility of the phosphorus.

As a result of these practices, less than half of the available phosphorus in superphosphates and mixed fertilizers was water soluble in 1955-56 (4). The water solubility of phosphorus in individual samples ranged from 1 to 98%.

The principal water-insoluble phosphorus compounds that are formed by reaction of superphosphate with ammonia, limestone, or dolomite are di-

Table I. Comparison of P₂O₅ Availability Determinations on Dicalcium Phosphate-Basic Phosphate Mixtures by Solubilities in Neutral and Alkaline Ammonium Citrate

Sample Designation	Formulation of Mixture, % of P ₂ O ₅ from		Analysis			Water solubility, % of total P ₂ O ₅
	Basic phosphate	Dicalcium phosphate ^a	Total P ₂ O ₅ , %	P ₂ O ₅ availability, % of Total P ₂ O ₅ , by Indicated Method		
				A.O.A.C. ^b	N.A.A.C. ^c	
Analytical Sample ^d						
1	100	0	42.8	65.4	8.6	1.3
2	75	25	44.2	68.7	27.4	3.3
3	50	50	45.6	72.0	50.0	4.0
4	25	75	47.0	80.0	72.0	3.3
5	0	100	47.9	99.3	98.7	3.3
Analytical Sample ^e						
Basic phosphate	100	0	42.8	29.7	7.5	1.2
Dicalcium phosphate	0	100	47.8	97.7	94.5	1.3

^a Commercial feed-grade material.

^b Neutral ammonium citrate.

^c Alkaline ammonium citrate.

^d Contained 0.15 gram of total P₂O₅ for A.O.A.C. method and 0.30 gram of total P₂O₅ for N.A.A.C. method.

^e Contained 1 gram of material.

calcium phosphate and a more basic calcium phosphate that often is described as tricalcium phosphate (17). Preponderant evidence indicates that the more basic phosphate is usually hydroxyapatite (7). Fluorapatite also may be formed, but this compound is only slightly soluble in neutral ammonium citrate and, hence, little of it is included in the available P₂O₅ as determined by the Association of Official Agricultural Chemists' (A.O.A.C.) method of analysis (3).

There has been much agronomic research to determine the fertilizer effectiveness of the various compounds of phosphorus that may occur in fertilizers. Rogers, Pearson, and Ensminger (8) have summarized results of many such tests. They conclude that dicalcium phosphate is as good a source of phosphorus as is superphosphate on neutral and acid soils, but that basic calcium phosphate often is less effective. Terman, Bouldin, and Lehr (10) studied the fertilizer value of pure, crystalline calcium phosphates. Monocalcium phosphate was slightly more effective in promoting plant growth than dicalcium phosphate on alkaline soils, but was less effective on acid soils. Hydroxyapatite was of very low effectiveness on all soils.

Other agronomic studies have sought to evaluate the effectiveness of commercial or experimental fertilizers containing varying percentages of their phosphorus in a water-soluble form. Results often are interpreted in terms of the effect of water solubility on crop response without regard for the character of the water-insoluble portion. In fact, there is no generally accepted method for estimating, quantitatively, the proportions of dicalcium phosphate and basic calcium phosphate in mixtures

containing both compounds. It appears that such a method is needed in order to provide information that would permit a more meaningful interpretation of agronomic test data.

Ammoniacal ammonium citrate solution (Petermann solution) is used in many European countries to evaluate the acceptability of water-insoluble phosphorus in fertilizers. Dicalcium phosphate is soluble in this alkaline solution, but the more basic calcium phosphates are only slightly soluble. The present work was done to test the suitability of the alkaline ammonium citrate method for estimating the proportions of dicalcium phosphate and basic calcium phosphate in fertilizers and to compare the results obtained by this method with those of the A.O.A.C. method for a representative group of fertilizers.

Official Method for Phosphorus—United States

The official method (3) for evaluation of acidulated materials or mixed fertilizers requires two determinations: total phosphorus and citrate-insoluble phosphorus. Citrate-insoluble phosphorus is the phosphorus in that portion of a sample which remains after a 1-gram sample is extracted with about 250 ml. of water at room temperature and then with 100 ml. of neutral ammonium citrate of specific gravity 1.09, at 65° C. If the sample is a nonacidulated material such as dicalcium phosphate, a 1-gram sample is extracted directly with neutral ammonium citrate.

The water-insoluble portion of an acidulated material or of a mixed fertilizer frequently amounts to less than 10% of the original sample. Nonacidulated materials, on the other hand, may have little or no solubility in water.

Situations may arise in which a water-insoluble phosphate is completely soluble in neutral ammonium citrate if it is a minor constituent of a mixed fertilizer, but it may be relatively insoluble if it is the major phase in a nonacidulated material.

Procedures for Evaluation of Phosphates—Europe

Of the 14 countries comprising the Organisation for European Economic Cooperation, only two, Italy and the Netherlands, use neutral ammonium citrate for evaluation of fertilizers, and in the Netherlands it is used as an alternative procedure (6). Ammoniacal ammonium citrate (Petermann solution) is used by 11 countries, usually as an extractant for the water-insoluble fraction of the fertilizer. The procedure used in the present work was that given for the Netherlands (6). This method is hereafter referred to as the N.A.A.C. method (Netherlands alkaline ammonium citrate method).

The Petermann reagent has a density of 1.082 as compared with 1.09 for the neutral ammonium citrate. The pH of the Petermann reagent is about 9.

The procedures in several of the countries are so designed that the sample size is varied with the amount of P₂O₅ in the sample. In the Netherlands, the directions specify use of 1 gram of double or triple superphosphate, 2 grams of superphosphate or mixed fertilizers containing more than 10% of P₂O₅, or 4 grams of fertilizers containing less than 10% of P₂O₅. The sample is extracted with about 200 ml. of water, and the water-insoluble residue is extracted with 100 ml. of the Petermann reagent for 15 hours at room temperature and 1 hour at 40° C. Aliquots of the water and citrate extracts are combined for phosphorus determination.

Mixtures of Dicalcium Phosphate and Basic Calcium Phosphate

Synthetic mixtures of dicalcium phosphate and basic calcium phosphate were prepared to test the feasibility of estimating the proportion of these materials by the alkaline ammonium citrate method. The materials used were commercial feed-grade dicalcium phosphate and reagent-grade "tricalcium phosphate." The latter material was indicated by microscopic examination and x-ray pattern to be predominantly hydroxyapatite. It had a CaO:P₂O₅ mole ratio of 3.14. Its optical characteristics were similar to material often found in ammoniated superphosphate. For this reason it was considered to be a more appropriate material for the present study than pure crystalline hydroxyapatite. These two materials were mixed together in various propor-

tions, and the mixtures were analyzed by the N.A.A.C. and A.O.A.C. methods.

The A.O.A.C. procedure specifies a 1-gram analytical sample; the N.A.A.C. procedure specifies a 2-gram sample for mixed fertilizers containing more than 10% of total P_2O_5 and a 1-gram sample for double or triple superphosphate.

In the present work, the size of each analytical sample was such that it contained 0.15 gram of total P_2O_5 for the A.O.A.C. procedure and 0.30 gram of total P_2O_5 for the N.A.A.C. procedure. These sample sizes were selected on the basis that they would provide about the same amount of

water-insoluble P_2O_5 as would be present in the prescribed sample size of a fertilizer such as 20-20-0 nitric phosphate. The results of these tests are shown in Table I and Figure 1. For comparison, Table I also shows results of the analysis of each of the two materials when a 1-gram analytical sample was used.

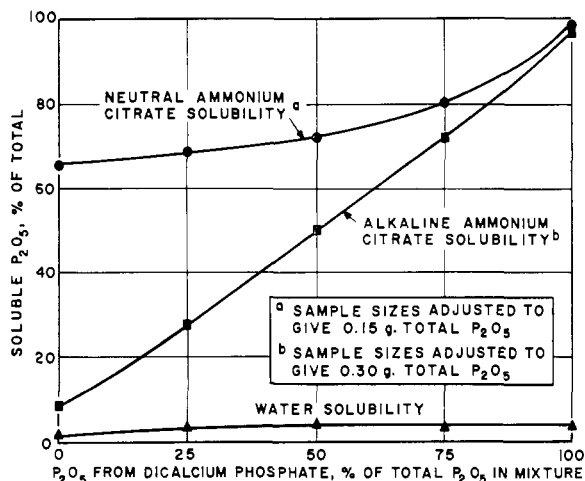


Figure 1. Solubility in alkaline and neutral ammonium citrate of mixtures of basic calcium phosphate and dicalcium phosphate

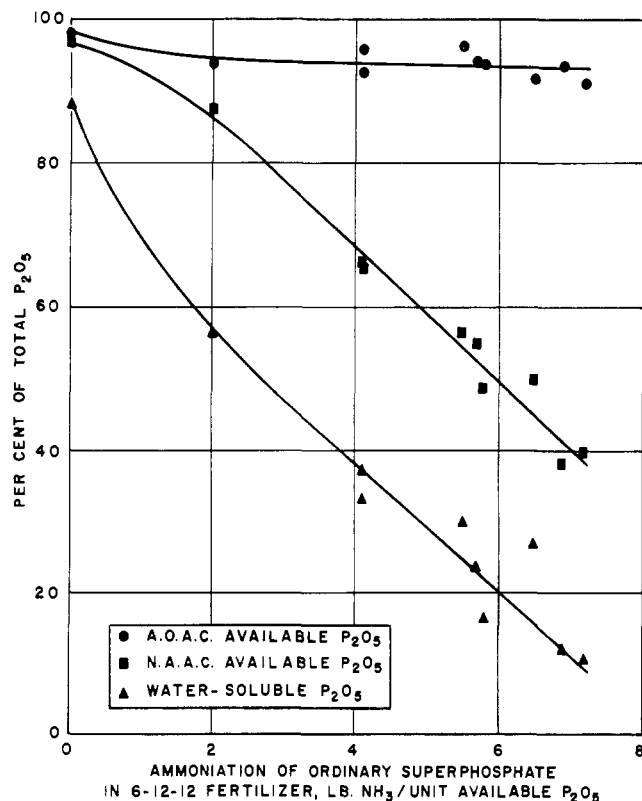


Figure 2. Comparison of A.O.A.C. and N.A.A.C. analytical methods for available P_2O_5 as applied to experimental fertilizers containing ordinary superphosphate ammoniated to various degrees with several sources of free ammonia

Table II. Comparison of Solubility in Neutral and Alkaline Ammonium Citrate of the Phosphorus Contents of Selected Fertilizers

Identification of product	Grade	Analysis, % P_2O_5					Available P_2O_5 , % of Total P_2O_5		Solubility in Alkaline Ammonium Citrate of W.I. P_2O_5 , %	
		A.O.A.C. ^a		N.A.A.C. ^b			A.O.A.C. method	N.A.A.C. method		
		Total	C.I.	Available	W.S.	C.S. ^c	Available ^d			
Nitric phosphate A	20-20-0	20.6	0.2	20.4	6.2	13.6	19.8	99	96	94
Nitric phosphate B	15-15-15	15.6	<0.1	15.6	7.2	8.3	15.5	100	99	99
Nitric phosphate C	6-12-18	12.9	<0.1	12.9	4.8	7.1	11.9	100	92	88
Nitric phosphate D	9-9-18	9.2	<0.1	9.2	0.7	6.3	7.0	100	76	74
Nitric phosphate E	12-12-12	12.3	0.1	12.2	2.0	9.2	11.2	99	91	89
Nitric phosphate F	14-14-14	14.8	0.7	14.1	2.1	10.9	13.0	95	88	86
Nitric phosphate G	12-10-20	10.9	0.1	10.8	5.6	3.8	9.4	99	86	72
Nitric phosphate H	10-8-18	7.8	0.3	7.5	0.1	6.4	6.5	96	83	83
Leached-zone fertilizer	15-15-15	14.9	<0.1	14.9	4.4	9.7	14.1	100	95	92
Ammoniated superphosphate										
Ordinary										
Degree 0.0 ^e	6-12-12	12.9	0.2	12.7	11.3	1.4	12.7	98	98	88
Degree 2.0 ^e	6-12-12	13.7	0.9	12.8	7.7	4.3	12.0	93	88	72
Degree 4.1 ^e	6-12-12	13.5	1.0	12.5	5.0	3.9	8.9	93	66	46
Degree 5.7 ^e	6-12-12	13.7	0.8	12.9	3.9	3.6	7.5	94	55	37
Degree 6.5 ^e	6-12-12	13.9	1.2	12.7	3.7	3.2	6.9	91	50	31
Degree 7.2 ^e	6-12-12	13.2	1.2	12.0	1.4	3.8	5.2	91	39	32
Concentrated										
Degree 3.6 ^e	10-20-20	22.3	1.6	20.7	12.9	4.0	16.9	93	76	43
High analysis										
Degree 4.1 ^e	8-48-0	51.1	2.3	48.8	27.0	20.6	47.6	95	93	85

^a Neutral ammonium citrate method.

^b Alkaline ammonium citrate method for Netherlands as described (6, p. 82).

^c Solubility in alkaline ammonium citrate.

^d Sum of water- and citrate-soluble P_2O_5 .

^e Degree of ammoniation, pounds free ammonia per unit of available P_2O_5 .

The data show that the percentage of P_2O_5 that was soluble in alkaline ammonium citrate was reasonably close to that present as dicalcium phosphate. Very little of the basic calcium phosphate was dissolved. It is concluded that the method would be useful for estimating the percentages of dicalcium phosphate in mixtures containing it and more basic calcium phosphates.

The solubility of basic calcium phosphate in alkaline ammonium citrate was increased only slightly when the size of the sample was decreased. However, the solubility in neutral ammonium citrate increased from 30 to 65% when the sample size was decreased from 1 to 0.35 gram (0.44 vs. 0.15 gram of P_2O_5).

The solubility of the basic calcium phosphate in neutral and in alkaline ammonium citrate agrees very closely with the values determined by Ando for hydroxyapatite (2). MacIntire (5) found that the solubility in neutral ammonium citrate of four preparations of hydroxyapatite varied from 35 to 53% when using 1 gram of charge and 100 ml. of solution as specified by the A.O.A.C. procedure.

Nitric Phosphates and Ammoniated Superphosphates

Several samples of European and experimental Tennessee Valley Authority fertilizers were analyzed by the N.A.A.C. and A.O.A.C. methods. Table II shows a comparison of results.

The solubility in alkaline ammonium citrate of the water-insoluble phosphorus content of eight samples of nitric phosphates ranged from 72 to 99%. The solubilities may be related to process characteristics such as the $CaO:P_2O_5$ ratio, method of ammoniation, and thickness of slurry during ammoniation. In general, the quality of the water-insoluble fraction of nitric phosphates, as determined by solubility in alkaline ammonium citrate, was higher than that of ammoniated superphosphates.

Nitric phosphates E and F had a $CaO:P_2O_5$ mole ratio of about 2.3. Sample E was ammoniated in a solid state; sample F was ammoniated in the slurry state. No appreciable difference in solubility in alkaline ammonium citrate resulted from this difference in ammoniation technique.

The solubility in alkaline ammonium citrate of the leached-zone fertilizer is high, which indicates that the precipitated aluminum phosphate contained in this product is soluble in the alkaline reagent.

In the series of 6-12-12 fertilizers that contained ordinary superphosphate ammoniated to various degrees (Table II), the water solubility decrease as the degree of ammoniation increased. The N.A.A.C. availability, as indicated by the sum of the water-soluble and

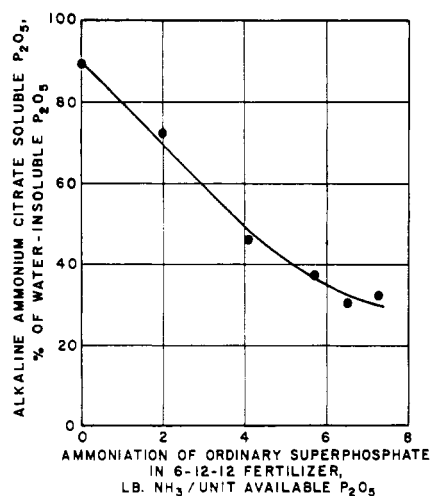


Figure 3. Relationship between degree of ammoniation of ordinary superphosphate and solubility in alkaline ammonium citrate of the water-insoluble P_2O_5 .

alkaline ammonium citrate-soluble P_2O_5 , also decreased as the degree of ammoniation increased, whereas the A.O.A.C. availability was fairly constant. These relationships are shown in Figure 2.

The solubility in alkaline ammonium citrate of the water-insoluble P_2O_5 decreased as the degree of ammoniation was increased, as shown in Figure 3. These data may be interpreted as indicative of increased formation of basic calcium phosphates at the higher degrees of ammoniation, as discussed by Yates, Nielsson, and Hicks (11).

When high-analysis superphosphate containing 54% available P_2O_5 (7) was ammoniated with anhydrous ammonia, the product (8-48-0, Table II) showed a high N.A.A.C. availability although the degree of ammoniation was high. This indicates, presumably, that most of the water-insoluble P_2O_5 was in the form of dicalcium phosphate. On the other hand, the concentrated superphosphate that was ammoniated with solution (10-20-20, Table II) produced a material of relatively low N.A.A.C. availability. It appears, therefore, that the conditions of ammoniation or the type of superphosphate may influence the proportions of dicalcium phosphate and more basic compounds in the water-insoluble fraction of the products.

The 10-20-20 and the 6-12-12 that contained 6.5 pounds of ammonia per unit of P_2O_5 (Table II) were part of a group of fertilizers that has been used in several greenhouse and field experiments to determine the effects of water solubility and granule size on crop response. The group also contained a 7-14-14 material in which all of the phosphorus was present as dicalcium phosphate. Some of the results of agronomic tests with these materials

have been reported by Terman, Anthony, Mortensen, and Lutz (9). In several tests the 6-12-12 material gave poorer results than materials of either higher or lower water solubility. These poorer results probably are related to the high content of basic calcium phosphate of the 6-12-12 as indicated by the low N.A.A.C. availability (50%).

The series of 6-12-12 products in Table II is a part of a group of materials that has been used in field tests. The following tabulation shows results of a typical field test.

Degree of Ammoniation of Ordinary Superphosphate, Lb. Free NH ₃ /Unit of Available P ₂ O ₅	Per Cent of Total P ₂ O ₅ N.A.A.C.		Yield of Wheat, ^a Bu./A.
	W.S. ^b available		
0	88 ^c	98 ^c	20.6
2.0	56	88	20.3
4.1	37	66	19.1
6.5	27	50	15.9
7.2	11	39	13.9
No phosphorus	6.3

^a Data obtained by the Mississippi Agricultural Experiment Station.

^b W. S. = water soluble.

^c Typical value.

These data show that the effectiveness of the phosphorus content of the fertilizer decreased as the degree of ammoniation was increased. The crop yield could be correlated with either water solubility or the N.A.A.C. availability. Further tests are needed to determine which of these factors is the more important.

Discussion and Conclusions

The A.O.A.C. available phosphorus content of ammoniated superphosphates consists of three classes of compounds: water-soluble compounds, mainly monocalcium phosphate and ammonium phosphates; dicalcium phosphate; and basic calcium phosphate, probably hydroxyapatite. The effectiveness of a fertilizer may depend on the percentages of phosphorus that are present in these three classes; an analytical method for providing this information should be helpful in the interpretation of agronomic test data.

The method described in the present paper appears promising for providing a roughly quantitative value for the three classes of phosphorus compounds. Water-soluble phosphorus is determined by a water extraction method as provided by A.O.A.C. or N.A.A.C. procedures. The water-insoluble residue is extracted with alkaline ammonium citrate by the N.A.A.C. procedure. The phosphorus dissolved in this step is assumed to be present mainly as dicalcium phosphate. On another sample

the water-insoluble residue is extracted with neutral ammonium citrate. The phosphorus dissolved in this step is assumed to be the sum of that present as dicalcium phosphate and basic calcium phosphate. The percentage of basic calcium phosphate is obtained by subtraction. While this relationship seems to be valid on most fertilizers, it may not be applicable to those containing unusual proportions of hydroxyapatite or compounds other than those discussed herein.

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FERTILIZER COATINGS

The Effect of Coatings on the Dissolution of Fertilizers and the Uptake of Fertilizer Potassium by Plants

KIRK LAWTON

Department of Soil Science,
Michigan State University,
East Lansing, Mich.

Several fertilizers containing potassium were coated with vinyl acetate, acrylic resin spray, paraffin, and polyethylene emulsion to alter their dissolution properties. Leaching losses of potassium were significantly reduced by several coating materials. Germination of wheat seed in contact with uncoated fertilizer and emergence of seedlings were lower than with similar placement of coated fertilizer in greenhouse trials. In addition, in a field study of alfalfa top-dressed with a large application of mixed fertilizer, coatings reduced luxury consumption of potassium by this legume. Thus, coatings on granular fertilizer can reduce loss of soluble nutrients by leaching in coarse textured soils, minimize salt injury due to seed-fertilizer contact, and level out excess absorption of fertilizer nutrients such as potassium.

NITROGEN and potash salts such as ammonium nitrate or sulfate, urea, and potassium chloride or sulfate dissolve very rapidly in moist soils. Even the more soluble, well granulated phosphorus carriers such as concentrated superphosphate and ammonium phosphates lose a large part of their phosphorus after 12 to 24 hours of exposure to moist soil (5, 6).

Most of the fertilizer used in American agriculture is applied before or at the time a crop is planted. Since the nutrient requirements of seedlings or transplants are small, the components of soluble fertilizers are largely free to react with soil minerals or may be subject to considerable movement within the soil profile as a result of leaching. The concept of a slow release of plant nutrients from fertilizers to be used by crops as they require them has been proposed for slightly soluble materials. Urea-formaldehyde as a nitrogen source (7)

and the meta- and pyrophosphates of potassium (8, 9) have been developed with this idea in mind. It is possible that coatings on soluble or partially soluble fertilizers may accomplish the same objectives.

The benefits that agronomists have speculated might be attributed to such coatings include reduction of leaching and volatilization losses of nitrogen, reduction of leaching losses of potassium applied to sandy soils, and minimization of plant absorption of excessive amounts of mineral elements, such as potassium, beyond normal crop requirements (3). In addition, fixation of soluble phosphate moving out of fertilizer granules or bands might be lessened and salt injury resulting from seed-fertilizer contact might be greatly reduced.

This study examines several materials which could serve as coatings for fertilizers and determines their performance

under laboratory, greenhouse, and field conditions.

Materials and Methods

The following materials were employed as coatings for fertilizers.

Acrylic Resin. Made up of 5% resin in aromatic hydrocarbons and chlorinated solvents with fluorinated hydrocarbons as the spray propellant. This material was sprayed on fertilizer particles as they were rotated on a flat paper surface. These particles were then air-dried, and the spray was repeated. By this procedure, a glassy-like, water-resistant coating was prepared which filled or covered most of the pores and crevices of the particle surface.

Vinyl Acetate. Contained 10 grams of vinylite dissolved in 100 ml. of acetone. This viscous liquid was poured over fertilizer granules in a funnel, and the excess allowed to drain off. Between