Also, with the flask remaining upright during a combustion, the platinum holder formed a bottom beneath the sample. The last fragment of the sample burned away upon it rather than falling into the solution. The perforations in the holder were adequate for admitting oxygen and still allowing for sufficient contact between the metal and sample. If the sample was not compressed in the platinum holder, a flame large enough to contact the glass or metal resulted and carbon was formed. Such samples were discarded. Normally after absorption of gases, the solution in the flask (without color reagents) showed an absorbance of 0.005 to 0.02 in a 2-cm. cell with distilled water as the reference.

The burning characteristics, solvent resistance, sealability, and chlorine content of about 20 plastic films were investigated. Cellulose acetate (S-600) was the most satisfactory. It is resistant to the solvents commonly used for extraction of residues with the exception of acetone which dissolves it. The blank value of the cone and fuse was about 11 γ as chloride. The cones did not leak when made according to the procedure. Heat-sealing was necessary to form a tight seam in order that the solvent seal would be leak-proof. The capacity of the cone is 5 ml.

The flask, platinum holder, and balloon were rinsed thoroughly with distilled water after each combustion. The balloon was replaced after about 25 combustions. The standard curve follows Beer's law as long as the mercuric thiocyanate solution remains saturated. This solution is best prepared by heating, with agitation, an excess of powdered mercuric thiocyanate in absolute ethyl alcohol and allowing it to settle for 48 hours before use. The clear solution and solid mercuric thiocyanate should be mixed, heated, and allowed to settle once every 2 weeks during use. The method should be applicable to the determination of residues of other chlorinated pesticides.

Acknowledgment

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HERBICIDE RESIDUES

Determination of Small Amounts of Arsenic in Potatoes. Extraction and Reduction of Molybdoarsenic Acid

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A method is presented for the determination of small amounts of arsenic in potatoes. Potato tissue is ashed with magnesium nitrate and the ash dissolved in acid. Acid-molybdate is added, followed by extraction with a 1-butanol–chloroform mixture to remove interfering phosphorus as molybdophosphoric acid. The aqueous solution is extracted with 1-butanol to remove arsenic as molybdoarsenic acid. A simple procedure is then used for adjusting acidity and reducing to the heteropoly blue with stannous chloride in ethyl alcohol. The effect of varying the concentration of acid and reducing agent on the blue color is shown. Silicon causes some interference. Silicon interference can be reduced by use of a 1-butanol–ethyl acetate mixture for extraction of molybdoarsenic acid. The method, used to determine possible traces of arsenic in potatoes resulting from the application of sodium arsenite to kill potato vines and weeds, yields an average recovery of 94.1%.

A MODIFICATION of the procedure of Akkseev (*1*) is described by Wadelin and Mellon (*9*) for the removal of phosphorus prior to determining arsenic. Sodium molybdate in hydrochloric acid is added to a solution containing phosphate and arsenate ions to form the respective molybdic acids. Successive extractions of the solution with a 1-butanol–chloroform mixture remove interfering phosphorus as molybdophosphoric acid. Arsenic is then removed as molybdoarsenic acid by extraction with butanol. It is determined by ultraviolet absorptiometric measurement of 12-molybdoarsenic acid. The method is not quite sensitive enough for determining traces of arsenic in biological material.

In this paper, the method of Wadelin and Mellon is used to separate phosphorus and arsenic contained in an acid solution of potato tissue–magnesium oxide ash. A sulfuric acid–ethyl alcohol solution is then added to the 1-butanol followed by a solution of stannous chloride in ethyl alcohol which reduces molybdoarsenic acid to the heteropoly blue. Spectrophotometric measurement is then made of the blue color at 740 mμ.

Method

Preparation of Standard Curve. (0 to 50 γ of arsenic.) Pipet 0-, 2-, 4-, 6-,

Table I. Effect of Phosphorus and Silicon on Recovery of Arsenic from Synthetic Solutions

Added, γ As	P	Si	Apparent as Found
1-Butanol–Chloroform Extraction of Phosphorus. 1-Butanol Extraction of Arsenic			
0	5000		1.3
15	5000		17.1
30	5000		31.0
1-Butanol–Chloroform Extraction of Phosphorus. 1-Butanol Extraction of Arsenic			
0	1000	50	1.0
15	1000	50	17.5
30	1000	50	33.5
1-Butanol–Chloroform Extraction of Phosphorus. 1-Butanol–Ethyl Acetate Extraction of Arsenic			
0	1000	50	0.6
15	1000	50	17.7
30	1000	50	31.6
1-Butanol–Chloroform Extraction of Phosphorus. 1-Butanol Extraction of Arsenic			
0	5000	450	8.2
15	5000	450	21.0
30	5000	450	37.3
1-Butanol–Chloroform Extraction of Phosphorus. 1-Butanol–Ethyl Acetate Extraction of Arsenic			
0	5000	450	5.5
15	5000	450	18.3
30	5000	450	35.5

8-, and 10-ml. aliquots of a solution containing 5 γ of arsenic per ml. [prepared by dilution of an arsenic stock solution (*2*)] into a series of 250-ml. Squibb separatory funnels. Add water to make up the total volume to 50 ml. Treat each sample separately from this point through color development. Add 25 ml. of a 2*N* hydrochloric acid solution containing 0.03 gram of sodium molybdate per ml. Pour into the funnel 50 ml. of a 1 and 3 by volume mixture of 1-butanol–chloroform. Shake for 30 seconds and allow the layers to separate. Discard the lower layer and repeat the extraction with 1-butanol–chloroform three times more, discarding the lower layer each time. Add exactly 20 ml. of 1-butanol (117° to 118.5° C.) to the aqueous solution and shake for 30 seconds. After the layers have separated discard the lower aqueous layer and drain the 1-butanol layer into a 50-ml. Erlenmeyer flask. Add exactly 2 ml. of a 12*N* sulfuric acid solution in 95% ethyl alcohol (U. S. Industrial grade) and mix. Add 0.5 ml. of a solution containing 2 mg. of stannous chloride dihydrate per ml. of ethyl alcohol. Stopper and mix. After 15 minutes, measure the absorbance at 740 mμ using a 1-cm. cell with 1-butanol in the reference cell.

Analysis of Potatoes. The method of Evans and Bandemer (*5*) is used for the ignition of potato tissue with magnesium nitrate. Remove soil, etc., from the surface of the tubers which may contain arsenic. Thoroughly blend a representative sample of potatoes in their own liquid. Fold a disk of 12.5-cm. diameter vegetable parchment paper as for filtering. The second folding should, however, be made so that the resulting cone will just fit inside the crucible and not tip over. Fold the tip of the cone back and fit the truncated cone into a No. 4 Coors porcelain crucible. Then fold a similar disk of No. 40 Whatman filter paper and fit it inside the parchment disk. Weigh into the filter paper 15 grams of potato tissue from the blender. Blend sufficient tissue so that the sample may be taken from the top of the mix, while the blender is operating. This prevents the separation of solid and liquid in the sample. Add 6 ml. of saturated magnesium nitrate solution and mix well with the tissue using a thin stirring rod. For recovery studies, an appropriate aliquot of the arsenic standard solution is also added at this point and mixed in. Rinse any tissue remaining on the stirring rod back into the filter paper with a few drops of water. Evaporate to dryness on a steam bath. Ignite in a muffle furnace by placing in the cold furnace, slowly raising the temperature to 600° C., and heating overnight at this temperature. A light, white ash is obtained.

Wet the ash with water and add sufficient 4*N* hydrochloric acid solution to just dissolve it. By adding the acid dropwise with intermittent stirring, about 12 ml. are required. Transfer the solution quantitatively to a 50-ml. volumetric flask. Adjust the pH of the solution to between 5 and 9 with 1 *M* sodium hydroxide. Make up to volume with water and mix. Pipet a 25-ml. aliquot of this solution into a 250-ml. Squibb separatory funnel and add 25 ml. of water. Proceed as in the preparation of the standard curve.

Results and Discussion

A Model 1740 Temco muffle furnace equipped with a Wheelco Model 404 Capacitrol indicating temperature controller was used in the ignition procedure. The temperature was slowly raised to 600° C. over a 4- to 5-hour period and the crucibles were cooled down slowly after ignition.

If the procedure of Wadelin and Mellon is used, the aliquot of ash solution cannot contain more than 2 mg. of phosphorus. In the above procedure the volumes of reagents are increased by a factor of 2.5 in order that a larger portion, 25 ml., of sample solution can be taken. The upper limit for phosphorus which can be removed is, therefore, 5 mg. An absorbance of 0.123 was obtained when the extraction of 6 mg. of phosphorus was attempted in the absence of arsenic. Wooster (*10*) gives the approximate phosphorus content of raw, white potatoes as 56 mg. per 100 grams. A 25-ml. aliquot of the ash solution therefore represents 7.5 grams of potato or about 4.2 mg. of phosphorus.

Four extractions with 1-butanol–chloroform are necessary to remove virtually all of the phosphorus. An absorbance of 0.016 (Table I) was obtained when a solution containing 5 mg. of phosphorus was extracted four times with 1-butanol–chloroform. The absorbance was 0.041 when only three extractions were used. Three extractions are sufficient if larger quantities of arsenic are determined as in the Wadelin and Mellon procedure. The effect of reagent grade in place of technical chloroform is only that of reducing the amount of interfacial cuff during the extraction of phosphorus.

The sulfuric acid–ethyl alcohol mixture dissolves water droplets giving a clear blue solution. The use of a small volume of sulfuric acid–ethyl alcohol and stannous chloride dissolved in ethyl alcohol is an adaptation of the procedure of Clark (*4*) and Lindsay (*7*) for determining phosphorus in soil solutions after extraction of molybdophosphoric acid with a benzene-isobutyl alcohol solution (*8*). The stannous chloride solution in ethyl alcohol should be prepared fresh daily. The sodium molybdate–hydrochloric acid solution should be prepared fresh monthly.

A Beckman DU spectrophotometer with a slit width setting of 0.03 mm. was used to measure absorbance. Color development is not complete until 12 minutes after the addition of the stannous chloride solution. The blue color is stable for at least 24 hours. Certain lots of 95% ethyl alcohol cause color instability in the determination of phosphorus (*6*). The U. S. Industrial grade did not cause color instability in the determination of arsenic.

The relation between absorbance in the presence and absence of arsenic and

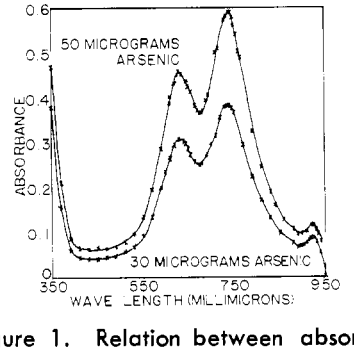

Figure 1. Relation between absorbance of heteropoly blue color and concentration of sulfuric acid in butanol

the concentration of sulfuric acid in the 1-butanol solution is shown in Figure 1. In the presence of arsenic the absorbance is constant when the concentration of sulfuric acid is between 0.2 and 0.8*M*. Higher concentrations cause a decrease in absorbance. The results of Berenblum and Chain (*3*) with phosphorus in aqueous solution are somewhat similar. The concentration of sulfuric acid in 1-butanol in this procedure is 0.54*M*. The absorbance of the control is very small at this acidity.

The absorbance of the blue color was measured and is constant when the 1-butanol solution is between 0.05 and 2.0 m*M* with respect to stannous ion. Between 0.4 and 2.0m*M*, a green color first forms which soon turns blue. The concentration of stannous ion in this procedure is 0.2m*M*.

The absorption spectrum of the heteropoly blue color at two concentrations of arsenic is shown in Figure 2. Maximum occurs at 740 mμ. The standard curve is very reproducible. The method will detect about 1 γ of arsenic using a 1-cm. cell.

Lampitt and Goldenberg (*6*) found the silicon content of potatoes to range between 7.7 and 58.7 p.p.m. Therefore, it is possible to have from about 60 to 440 γ of silicon present in 7.5 grams of potato as represented in this procedure. The method was used to recover arsenic in the presence of phosphorus and silicon in synthetic solutions. The elements were contained in 50 ml. of solution. Phosphorus was present as potassium dihydrogen phosphate and silicon as sodium metasilicate. The amount of silicon added approximately represents the limits found by Lampitt and Goldenberg (*6*). Table I shows the amount of arsenic not corrected for the blank that was recovered in each study.

The presence of 5 mg. of phosphorus causes a small increase in absorbance. The interference by silicon is more serious especially at the 450-γ level. It may be minimized by proceeding through color development with each sample as rapidly as possible after addition of the molybdate reagent (*9*). Use of a 1 and 1 by volume mixture of 1-butanol and ethyl acetate (*7*) in place of 1-butanol for extraction of molybdoarsenic acid is more selective in the presence of silicon (Table I). The absorption spectrum and standard curve of the heteropoly blue color in 1-butanol–ethyl acetate are identical with those in 1-butanol. The same solvent mixture (1-butanol–chloroform) is always used for removal of phosphorus. The recovery of arsenic (Table II) is just as satisfactory when using 1-butanol as compared to 1-butanol–ethyl acetate. There are crops (certain grasses, for example, which contain large amounts of silica) for which the 1-butanol–ethyl acetate extractant might be much more advantageous than with potatoes. For plant materials containing greater than about 8 p.p.m. of silicon, the 1-butanol–ethyl acetate mixture should therefore be used.

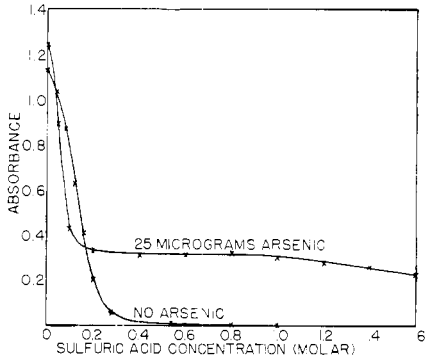

Figure 2. Absorption spectra of heteropoly blue color

The recovery of arsenic added to potato tissue is shown in Table II. Results using 1-butanol and 1-butanol–ethyl acetate are given. The average recovery using 1-butanol extraction is 94.1%. The apparent arsenic content of the potatoes averaged slightly above 1 p.p.m. This accounted, in part, for greater variability in the recovery of arsenic in quantities below 2 p.p.m.

Sodium arsenite is used to kill potato vines and weeds in potatoes. The method described has been used to determine possible traces of arsenic in potatoes resulting from treatment with sodium arsenite just before harvest. About 15 potatoes treated with sodium arsenite were blended in their own liquid. Twelve samples of the blended mixture were taken for the determination of arsenic using 1-butanol extraction. The average content of arsenic found was 1.11 p.p.m. with a range of 0.86 to 1.40. The average content of arsenic found in seven analyses of untreated potatoes was 1.24 p.p.m. with a range of 0.93 to 1.34. There was no significant difference in the arsenic found in the treated and untreated potatoes. The standard deviation of a single determination is 0.18%. The untreated potatoes may have contained arsenic, because the soil in which they were grown had received about five applications of sodium arsenite in the previous 8 years for potato vine and weed control. Also, many soils contain small amounts of arsenic. Interference by silicon contained in the potatoes probably accounted for part of the apparent arsenic found in the untreated potatoes. The method should be applicable to the determination of arsenic in other biological materials.

Table II. Recovery of Arsenic from Potatoes

Added, P.P.M.	Found, P.P.M.	Recovery, %
1-Butanol Extraction		
1.00	1.05	105.3
1.00	0.68	68.0
1.33	0.93	69.5
1.33	1.55	116.0
1.33	0.93	70.0
1.33	1.17	87.5
1.33	1.53	115.0
2.00	1.95	97.7
2.00	2.17	108.3
2.00	1.87	93.3
2.00	2.27	113.3
2.00	1.60	80.0
2.00	1.56	78.0
2.00	2.03	101.7
2.00	1.70	85.0
2.00	1.69	84.7
2.00	2.08	104.0
2.00	1.68	84.0
2.00	1.93	96.7
2.00	1.99	99.3
8.00[a]	8.59	107.3
8.00[a]	8.09	101.2
8.00[a]	7.82	97.8
1-Butanol–Ethyl Acetate Extraction		
1.00	0.76	76.0
1.33	1.41	106.0
1.33	1.31	98.0

[a] A 10-ml. aliquot of ash solution was analyzed.

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