

A METHOD FOR THE DETERMINATION OF SULPHUR LEVELS IN PLANT MATERIAL BY ATOMIC ABSORPTION SPECTROPHOTOMETRY

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SUMMARY

An alternative to a turbidimetric method, involving fewer manipulations, fewer reagents and a much shorter time of analysis, is presented.

Plant material is burnt in a combustion flask containing oxygen at atmosphere pressure in the presence of hydrogen peroxide and sodium hydroxide. The sulphate is precipitated by a known amount of barium chloride and the residual barium is determined by atomic absorption spectrophotometry.

Comparison of recoveries indicates that the proposed method is just as accurate as the turbidimetric method.

I. INTRODUCTION

The turbidimetric method of determining sulphur levels in plant material at present used in the Agricultural Chemical Laboratory of the Queensland Department of Primary Industries is based on two techniques:—

- (a) The oxygen flask combustion technique of Schoniger (1956) to oxidize organic sulphur in the sample to sulphate.
- (b) The development of a suspension of barium sulphate and the measurement of its optical density at 4900 Angstroms (Butters and Chenery 1959).

An alternative method involving atomic absorption spectrophotometry was devised by the authors. It is described in this paper and the results of sulphur analysis measured by each method are given.

II. EXPERIMENTAL

Atomic absorption spectrophotometry.—All spectrophotometric determinations were carried out on a Techtron Type A.A.4 Atomic Absorption Spectrophotometer using an air/acetylene fuel mixture. Absorption peaks were recorded on a Heath Servo Chart Recorder model EUW-20.

Settings used on the Techtron were—

Lamp current	10mA
Wavelength (Ba)	5535.5 A
Slit width	100 microns
Scale expansion	x 5
Damping	C
Air flow	15 lb/sq in
Acetylene flow	2.75 scale divisions

It is noted here that the use of a nitrous oxide/acetylene flame and burner would eliminate the necessity for scale expansion. Barium detection is then reported to be 25 times more sensitive than with an air/acetylene fuel mixture.

Turbidimetry.—Turbidimetric measurements were made on a Unicam S.P. 600 spectrophotometer at 4,900 Angstroms in 4 cm cells.

Apparatus.—One litre pyrex combustion flasks and ground-glass stoppers fitted with platinum combustion cones (ignition heads).

Reagents.—All reagents used were of Analytical Reagent grade:

- (1) 100 volume hydrogen peroxide (sulphate-free).
- (2) 0.2N sodium hydroxide solution.
- (3) 1.0N hydrochloric acid solution.
- (4) 6,000 p.p.m. solution of barium as barium chloride.
- (5) 600 p.p.m. solution of sulphate as magnesium sulphate.
- (6) 7,500 p.p.m. solution of strontium as strontium chloride.
- (7) 600 p.p.m. solution of phosphorus as potassium dihydrogen phosphate.

Standards.—Standards in the range 0–300 p.p.m. barium were prepared from reagent (4). They were adjusted to 1,500 p.p.m. strontium by the use of reagent (6). The range chosen was related to the sensitivity of the atomic absorption spectrophotometer, the amount of sample that could be conveniently ignited, and the expected levels of sulphur present in the plant samples.

III. PROCEDURE

Weigh 300 mg of plant material into a 1½ in. x 1½ in. piece of Whatman No. 54 filter paper leaving a 1 in. x ½ in. taper for ignition. Wrap the sample carefully and secure it by means of a 2 in. piece of platinum wire. Leave one end of the wire free to secure the wrapped sample to the platinum cone of the ignition head of a combustion flask.

Add 5 ml of reagent (2) and 1 ml of reagent (1) to the combustion flask. Flush the flask with oxygen for 10 sec, then stopper with an ordinary Quickfit stopper. Wet the inside of the flask thoroughly with the reagent solutions.

Moisten the ground-glass joint of the ignition head with distilled water, ignite the taper, remove the Quickfit stopper and insert the ignition head firmly into the flask.

Rotate the flask carefully during burning to prevent localized heating of the glass walls, until ignition is completed. Allow the flask to stand 45–60 min.

Into a 100 ml beaker add 2.5 ml of reagent (4) and 3.5 ml of reagent (3).

Wash the contents of the flask and the ash on the combustion cone into the beaker. Use 3–4 washings of approximately 5 ml each, allowing the flask to drain thoroughly after each washing.

Cover the beaker with a cover glass and boil the barium sulphate suspension for 10 min. Allow the beaker and contents to cool and transfer to a 50 ml volumetric flask. Allow 10 ml of reagent (6) and make to volume. It is essential that the strontium chloride solution (6) be added *after* boiling the suspension (see Discussion).

Stopper the flask, shake thoroughly, and filter a portion through a Whatman No. 40 filter paper.

Prepare a calibration curve using the 0–300 p.p.m. barium standards, plot peak height *v.* p.p.m. barium. Read the peak height of the sample filtrate against calibration curve and convert this reading to percent sulphur in the original sample, using the expressions:—

$$\% \text{ sulphur} = \frac{(300 - \text{p.p.m. barium observed}) \times 1.17}{\text{Sample weight (mg)}}$$

Alternatively the percent sulphur may be read directly from a calibration curve constructed by plotting the peak height of each standard against the corresponding percent sulphur, shown in Table 1. This table is based on a 300 mg weight made to 50 ml.

TABLE 1
BARIUM STANDARDS EXPRESSED AS PERCENTAGE OF SULPHUR

Barium in Standards (p.p.m.)	60	120	150	180	210	240	270	300
Sulphur in sample (%)	0.93	0.70	0.58	0.47	0.35	0.23	0.12	0.00

Sensitivity was established as follows:

- (i) The instruments were adjusted to give 90 divisions deflection on the recorder for the 300 p.p.m. barium solution.
- (ii) Since 1 p.p.m. sulphur = 4.28 p.p.m. barium, then for a linear response, 90 scale divisions = $\frac{300}{4.28}$ p.p.m. sulphur.

- (iii) As 300 mg of sample was diluted to 50 ml then

$$\begin{aligned} \text{Sensitivity} &= \frac{300 \times 50 \times 100}{90 \times 4.28 \times 0.3 \times 10^6} \\ &= 0.013\% \text{ sulphur/scale division.} \end{aligned}$$

Interference tests were carried out to measure any interferences to barium absorbance that may be due to the reagents, viz. HCl, NaOH and H₂O₂ solutions. None of these reagents interfered.

Initial determinations on plant samples indicated interference by one or more elements present in the sample. Elwell and Gidley (1966, p. 80) have indicated that the interference could be due to phosphorus. This was verified by tests carried out, the results of which are summarized in Tables 3 and 4.

Recoveries were made as follows:

A 60 p.p.m. sulphur solution was prepared by dilution of reagent (5). Aliquots of 5, 10, 15, 20 and 25 ml of this solution were added to 2.5 ml of reagent (4) and 2.5 ml of reagent (3) in 100 ml beakers. These aliquots were chosen to represent 0.1, 0.2, 0.3, 0.4 and 0.5% sulphur in 300 mg of sample. The barium sulphate suspensions were then boiled for 10 min, cooled, transferred to 50 ml volumetric flasks, made to volume and filtered through a Whatman No. 40 filter paper. Recoveries of sulphur from these aliquots are shown in Table 2.

TABLE 2
RECOVERY OF SULPHUR FROM PURE SOLUTIONS

% Sulphur Added	% Sulphur Recovered			Mean % Sulphur Recovered
	A	B	C	
0.10	0.12	0.13	0.09	0.11
0.20	0.21	0.21	0.23	0.22
0.30	0.31	0.30	0.32	0.31
0.40	0.42	0.41	0.39	0.41
0.50	0.52	0.51	0.52	0.52

The effect of phosphorus on sulphur recoveries was tested by adding aliquots of 0.5, 2.0, 4.0 ml of reagent (7) to solutions prepared as in the preceding paragraph, viz 0.1, 0.3, 0.5% sulphur. These aliquots were chosen to represent 0.1, 0.4 and 0.8% phosphorus in a 300 mg sample. The results of these tests are shown in Table 3.

TABLE 3

INFLUENCE OF PHOSPHORUS ON SULPHUR ESTIMATION

Equivalent % Phosphorus (added) in Sample	% Sulphur Observed		
	For 0.1% Sulphur	For 0.3% Sulphur	For 0.5% Sulphur
0.00	0.12	0.31	0.52
0.10	0.16	0.39	0.59
0.40	0.42	0.58	0.77
0.80	0.69	0.80	0.91

These figures demonstrate that phosphorus produces elevated sulphur figures for the test solutions by depressing the barium absorption peaks. This depression is magnified with increasing phosphorus content. In an attempt to overcome this interference, 10 ml of strontium chloride solution (6) was added to solutions prepared in the same manner as for those summarized in Table 3, to give 1,500 p.p.m. strontium in the final volume of 50 ml.

New standard barium solutions were also prepared containing 1,500 p.p.m. strontium. The added strontium effectively suppressed the interference of the added phosphorus. The results of these tests are outlined in Table 4.

TABLE 4

INFLUENCE OF PHOSPHORUS ON SULPHUR ESTIMATION
IN PRESENCE OF 1,500 p.p.m. STRONTIUM

Equivalent % Phosphorus (added) in Sample	% Sulphur Observed		
	For 0.1% Sulphur	For 0.3% Sulphur	For 0.5% Sulphur
0.00	0.11	0.30	0.50
0.10	0.11	0.30	0.51
0.40	0.12	0.30	0.49
0.80	0.10	0.31	0.48

IV. RESULTS

Two samples were used for analysis—a mixed grass sample of low sulphur content, and a mixed legume of high sulphur content. The sulphur contents of these samples as measured by the proposed atomic absorption spectrophotometer method are shown in Table 5, with the figures derived from the turbidimetric method.

TABLE 5

COMPARISON OF SULPHUR ANALYSES (%S)—
A.A.S. AND TURBIDIMETRIC METHODS

A.A.S. Method		Turbidimetric Method	
Mixed Grasses	Mixed Legumes	Mixed Grasses	Mixed Legumes
0.16	0.39	0.14	0.40
0.15	0.40	0.15	0.40
0.15	0.39	0.15	0.40
0.14	0.40	0.16	0.39
0.15	0.41	0.16	0.41
0.15	0.40	0.15	0.42
0.15	0.42	0.16	0.39
0.15	0.42	0.15	0.43
0.15	0.40	0.15	0.39
0.15*	0.40	0.15	0.40
±.005†	±.012	±.007	±.015

* Mean.

† S.D.

V. DISCUSSION

It was stressed in explaining the procedure that strontium chloride must be added *after* the precipitation of barium sulphate. Low and irregular recoveries of sulphur were obtained when strontium chloride was added prior to precipitation of barium sulphate. This effect was more marked with the high level of sulphur in the mixed legume sample. This was thought to be due to the co-precipitation of some sulphur as strontium sulphate in the presence of the large excess of strontium.

Boiling the suspension serves two purposes—promotion of rapid crystal growth of barium sulphate for ease of filtration, and reduction of volume, when necessary, for ease of transfer to the 50 ml volumetric flask.

The method is suitable for the determination of sulphur at all levels likely to be encountered in plant material. It is much quicker than the turbidimetric method and could be used as the basis for the automation of sulphur analyses of plant material.

The authors are investigating the application of this atomic absorption spectrophotometric technique for the analysis of sulphur in waters, fertilizers and stockfoods.

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CORRECTION: “Chromosome numbers of the exotic *Passiflora* species in Australia”, by P. R. Beal. For “synaptic” in Vol. 26, p. 417, line 32 read “sympatric”.