A METHOD FOR THE ESTIMATION OF THE ACID-BASE BALANCE IN THE ASH OF PLANTS.*

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The common method of determining the excess of acidic or basic elements present in the ash of plant or animal tissue is to make a rather complete analysis of the material, computing the excess from the results of this analysis. Such a procedure is difficult, long, and the results secured are subject to the errors which may appear in the individual determinations.

The advantage of a method which would give an index of the quantity of excess acidic or basic elements present in a plant material without such a laborious procedure as a total analysis is obvious. Extreme accuracy could be sacrificed, if necessary, for speed, inasmuch as small differences in total acidity or basicity in a food material are not as important in many cases as the relative final reaction of the ash elements.

Sherman and Sinclair (1), in a preliminary report, and Sherman and Gettler (2) in a later paper discuss the significance of differing amounts of acidic and basic elements in the ash of foodstuffs, and give results of analyses of a large number of materials used as foods. In this work the authors point out the fact that most of the current compilations of analyses give results in terms of the whole material, whether it be animal or vegetable, and not of the edible portion. The method discussed in the present paper has been applied only to plant materials, and in most cases to the entire plant. However, there seems no reason why this method could not be applied to the edible portions of plants, and perhaps to materials of animal origin.

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It is, then, with the idea that this method may have a wider application than is discussed in this paper, that the procedure described below is presented.

The simplest method for determining the excess of acidic or basic elements in the ash of food materials is to titrate a solution of the ash obtained from a given sample of the material directly with standard acid or alkali. It was early recognized, however, that such a procedure was inaccurate, since a great part of the more volatile elements are lost during ignition. For lack of a better method this procedure is still in use, however, for as late as 1929 Morse (3) titrated the ash of cranberries directly with standard acid, in order to express the alkalinity of the ash of the fruit.

In devising a method which will give a satisfactory measure of the excess of acidic or basic elements, the first consideration is that the more volatile elements be retained while the organic matter is decomposed by ignition. The elements which are lost in considerable quantity seem to be sulfur and chlorine. It is known that the greater part of the former element present in plant or animal tissue is in organic combination, while chlorine has been shown to be liberated from its compounds when ignited in the presence of carbon (4).

It has been shown by Benedict (5) and others that in order to hold organic sulfur during ignition, relatively large amounts of a basic element must be present, together with a suitable oxidizing medium. He has found such a combination of properties in copper nitrate, which, when used in combination with ammonium nitrate and sodium chloride, according to the procedure of Denis (6) allows the retention of organic sulfur in the ignition of many substances. Since it was shown by Frear (7) that such a medium could be used to retain the sulfur of certain plant materials, it was felt that the combination of reagents recommended by Denis (6) and stated above might retain the chlorine as well, and thus give an ash on which a true acid-base ratio could be determined.

Determinations of the content of sulfur and chlorine before and after ignition with Benedict-Denis solution showed practically quantitative retention of these two elements. The precipitate of copper hydroxide formed during the titration, however, readily adsorbs materials in solution. Since this adsorption prevents accurate titration, it was necessary to abandon this line of attack.
A search for a more suitable reagent which would not yield a colored solution or a highly adsorptive precipitate upon neutralization led to the selection of magnesium nitrate as the basis for a solution which would have sufficient oxidizing power, and still contain a suitable alkaline element. The nitrate radical present functions as a strong oxidizing agent, while the magnesium furnishes comparatively strong alkali. A tentative method for the determination of sulfur is given in the Methods of Analysis of the Association of Official Agricultural Chemists (8) which retains that element by the use of magnesium nitrate. Hartwell, Smith, and Damon (9) in work which was the forerunner of the present investigation used calcium nitrate solution in an attempt to determine the acid-base balance in plant material but the reaction upon ignition was extremely violent, and analysis of the resulting ash showed losses of sulfur and chlorine. Work was thus undertaken to establish the suitability of magnesium nitrate as a reagent to retain during ignition the elements sulfur and chlorine present in plant material.

EXPERIMENTAL.

From the standpoint of ease of manipulation, the amount of reagent chosen must be sufficiently large to allow complete ignition at low temperature, but not so large as to cause an explosion or even an extremely violent reaction to take place. Various concentrations of magnesium nitrate solutions were used, and from this preliminary work it was evident that a solution of 25 gm. of magnesium nitrate (Mg(NO₃)₂·6 H₂O) made to 100 cc. with distilled water was suitable, when used in the proportion of 25 cc. of solution to 2 gm. of finely ground plant material. This was entirely from the physical point of view, and it remained to determine the efficiency of the reagent in retaining sulfur and chlorine.

During the following work the procedure given below was followed, and is the method recommended.

Method.

Weigh 2 gm. of finely ground plant material into a porcelain beaker. Add to this about 10 cc. of distilled water, with care, so that no mechanical loss occurs. After the entire mass of the plant material has been wetted, add 25 cc. of a solution containing
25 gm. of magnesium nitrate per 100 cc. Mix, and place on an electric hot plate regulated so that the mix simmers but never boils violently. Allow the liquid to evaporate, heating in all about 3 to 4 hours.

When the residue is entirely dry, but not so dry as to become explosive (this degree of drying is best regulated by experience), remove from the hot plate, cover, and place in an electric furnace heated to about 250°. After 15 minutes, or when it is evident that the first reaction is over, raise the temperature of the furnace to 500°. This heating may be prolonged if black particles of unburned carbon are in evidence, but ordinarily requires not more than 30 minutes. The temperature should never be greater than 500°. Remove from the furnace when the contents of the beaker are white or nearly so, and allow to cool. Add about 10 cc. of water cautiously, allowing the ash to become thoroughly wet. Add 60 cc. of normal nitric acid from a burette, and digest on a hot plate just below the boiling temperature for 3 hours. Transfer the solution and any insoluble residue to an Erlenmeyer flask and titrate the excess acid with normal sodium hydroxide, using methyl red indicator.

Blank determinations are made in the same manner as above, 1 gm. of sugar being used in the place of the plant material.

Calculation.

The amount of normal acid used minus the amount of normal alkali gives the amount of acid required to neutralize the alkalinity of the crop, plus that of the magnesium oxide formed from the oxidizing solution. The amount of normal acid required to neutralize the blank determination is then subtracted from the total quantity of acid used, and the result in cc. equals the milli-equivalent weight of excess alkalinity in 2 gm. of plant material. Should the material have an excess of acidic elements, some of the alkali normally neutralized by the blank will remain. Thus the result of the above calculation will be negative, indicating an excess of acidic elements in the ash.

Qualitative tests for escaping sulfur or chlorine were made by igniting plant material treated with the above reagent and dried, under a close-fitting hood made from a glass funnel. All fumes arising from the ignition were drawn by suction, first through a
inch copper tube heated to redness, and finally through a series of wash bottles filled with distilled water. After the ignition was complete, the contents of the wash bottles were tested qualitatively for sulfates and chlorides. The absence of sulfates was noted in practically every case, while traces of chlorides were found in most of the solutions, although the quantities were very minute.

Quantitative determination of sulfur in four samples of ground plant material were made before and after ignition with the magnesium nitrate. The analysis of the original material was carried out by the adaptation of the Benedict-Denis method described by Frear (7). Results of these determinations are given in Table I. Similar determinations of chlorine before and after ashing were made by the method of the Association of Official Agricultural Chemists and are shown in the same table.

From the data shown, it is evident that the losses in these two elements are small. The discrepancies which appear are all within a reasonable limit of error.

There remained to determine whether the value obtained by the titration of the excess of basic or acidic elements was comparable with that calculated from a chemical analysis of the stronger acid-forming and base-forming elements of each of these crops. Analyses of the four samples of plant materials for the elements chlorine, sulfur, phosphorus, potassium, calcium, magnesium, and sodium were accordingly made. All of these were determined by the official methods of the Association of Official Agricultural Chemists, with the exception of sodium, which was determined by the zinc-sodium-uranyl acetate method of Barber and Koltoff (10).
The results of this comparison are given in Table II. The results secured by the titration method are in good agreement with those obtained by the complete analyses. Only the elements mentioned were considered in the calculation of the acidity or basicity of the ash. Other elements, such as iron, manganese, aluminum, and silicon, are not included in the calculations, since these elements are relatively weakly acidic or basic and with the exception of silicon, are present in very small amounts. Silicon has such extremely weak capacity for neutralizing strong bases that there seems little need for including it in the calculation. To a considerable extent these weak acidic and basic elements balance, thus compensating for any error.

**TABLE II.**

<table>
<thead>
<tr>
<th>Crop</th>
<th>Cl</th>
<th>SO₄</th>
<th>PO₄⁺⁺⁺⁺⁺</th>
<th>K</th>
<th>Na</th>
<th>Ca</th>
<th>Mg</th>
<th>Excess of basic elements by analyses. m.-eq.</th>
<th>Excess of basic elements by titration. m.-eq.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Corn</td>
<td>4.5</td>
<td>6.6</td>
<td>20.3</td>
<td>30.5</td>
<td>2.0</td>
<td>11.6</td>
<td>16.3</td>
<td>29.0</td>
<td>25.0</td>
</tr>
<tr>
<td>Buckwheat</td>
<td>25.4</td>
<td>6.8</td>
<td>18.5</td>
<td>38.8</td>
<td>2.7</td>
<td>53.6</td>
<td>26.9</td>
<td>71.3</td>
<td>71.3</td>
</tr>
<tr>
<td>Cabbage</td>
<td>36.7</td>
<td>0.4</td>
<td>22.4</td>
<td>53.1</td>
<td>27.6</td>
<td>100.4</td>
<td>19.7</td>
<td>151.2</td>
<td>148.8</td>
</tr>
<tr>
<td>Spinach</td>
<td>22.2</td>
<td>28.4</td>
<td>66.3</td>
<td>154.2</td>
<td>82.5</td>
<td>14.6</td>
<td>46.3</td>
<td>180.7</td>
<td>190.9</td>
</tr>
</tbody>
</table>

* H₃PO₄ considered as a dibasic acid.

The determination of the acid-base balance of the ash by the ignition in the presence of magnesium nitrate can be made in approximately 5 hours, and a considerable number can be conducted simultaneously, thus allowing the results to be secured in a fraction of the time required for the analytical procedures.

**SUMMARY.**

A method for the estimation of the excess of acidic or basic elements in the ash of plant materials is presented. The procedure requires much less time than a complete analysis for the elements concerned. It is shown that sulfur and chlorine are retained when plant material is ignited in the presence of magnesium nitrate. The acid-base balance of the plant, determined
by titration of this ash, is in good agreement with the stoichiometric balance of the amounts of strong acidic and basic elements obtained by chemical analysis.

The author wishes to express his obligations to Mr. John B. Smith for his advice throughout the course of this work.

BIBLIOGRAPHY.

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