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Iodometric Determination of Neutral Amino Acids Using Potassium Iodate

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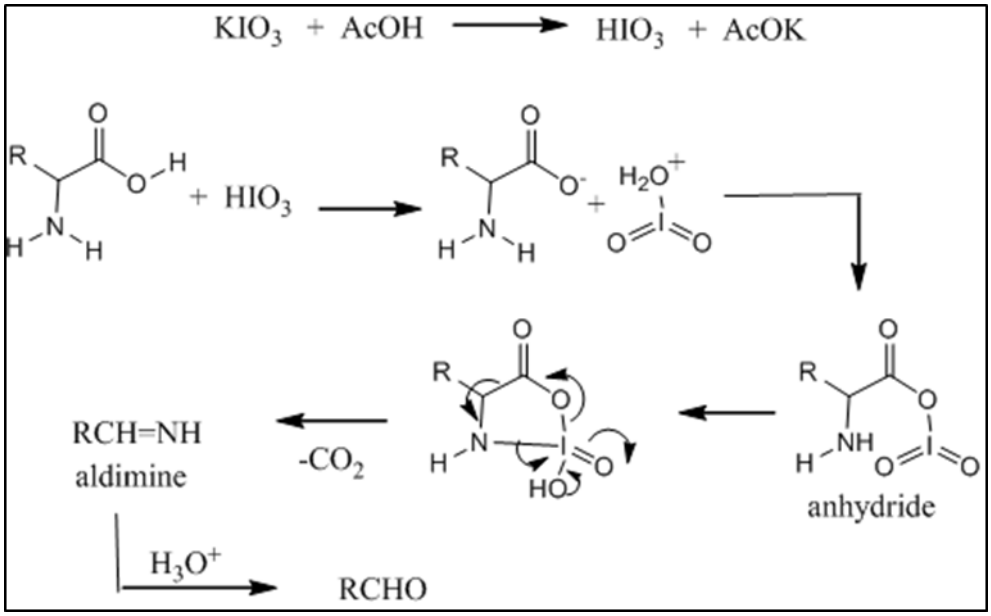
Abstract

In this work, we have reported a simple, cost effective and reliable method for the determination of neutral α-amino acids iodometrically by making use of potassium iodate. This volumetric method determines amino acids instantly, thereby greatly reduces the time of determination.

Introduction

Amino acids and proteins are the building blocks of life. When proteins are digested or broken down, amino acids are left. The human body uses amino acids to make proteins to help the body. Although various methods are reported in the literature for the determination of α-amino acids, few are commonly used. The important colorimetric reagents for the determination of α-amino acids include ninhydrin[1], 3,5-dibromosalicyladehyde[2] and o -diacetylbenzene[3]. During 1999, Rai et al[4] successfully used chloramine-T for the titrimetric determination of neutral α-amino acids.

Literature survey revealed that KIO_3/KI in acetic acid is used as iodination agent at 110^oC[5]. Selective oxidation of *n*-butylbenzene to 1-phenylbutyl acetate was achieved by ammonium iodate and catalytic *N*-hydroxyphthalimide (NHPI) in presence of acetic acid[6]. Recently Rai *et al* used KIO_3, as a novel oxidising agent for the synthesis of isoxazolines[7], for the synthesis of cyclohexenone from cyclohexanone[8] and for the estimation of glucose[9]. In continuation of our work on synthetic and analytical applications of HIO_3, we thought of an operationally simple titrimetric method for the determination of α-amino acids. The method reported here makes use of the fact that α-amino acids is known to undergo oxidation by HIO_3, yielding aldehydes involving one molecule of HIO_3 per mole-cule of α-amino acids. From the mechanism shown



Scheme 1. Mechanism for for the oxidation of amino acid.

below, it is evident that the reactive site involved for the attack of HIO_3 is the carboxyl group. This moiety is more reactive than the other functional groups. The probable mechanism for the oxidation of amino acid involves the protonation of HIO_3 first followed by the attack of carboxylate anion to the protonated HIO_3 forming the anhydride, which then reacts with amino group to form a cyclic intermediate (Scheme 1). This underwent disproportionation to give aldimine with the elimination of carbon dioxide. During work up process; the aldimine gets hydrolysed to form the aldehyde.

Materials & Methods

All reagents and chemicals used were of analytical reagent grade and were procured from SRL, India. Distilled water was used throughout the experiment.

In a typical experiment, a known excess of standard solution of HIO_3 was added to a known amount of α-amino acid. After completion of the reaction, unreacted HIO_3 was determined by iodometry. By carrying out a blank experiment simultaneously, the amount of HIO_3 consumed was determined. As the overall reaction requires one mole of HIO_3 per molecule of amino acid, which is equivalent to one mole of iodine, the molecular weight 'M' of α-amino acid is determined using the equation 1.

One mole of amino acid ≡ one mole of HIO_3 ≡ One mole of iodine ≡ 2000 ml of 1N sodium thiosulphate

i.e. M gm. of amino acid ≡ 2000 ml of 1N sodium thiosulphate

"w" gm. of amino acid ≡ (V_1-V_2) ml of N sodium thiosulpate

$$\text{i.e.,} \quad \frac{M}{2000} = \frac{w}{(V_2-V_1) \times N}$$

$$\therefore \quad M = \frac{2000 \times w}{(V_2-V_1) \times N} \qquad \text{(Equation 1)}$$

Where, M = Molecular weight of α-amino acid

w = Weight of the given sample

V_2 = Volume of sodium thiosulphate consumed (Blank)

V_1 = Volume of sodium thiosulphate consumed (experimental)

N = molarity of sodium thiosulphate

Determination of Molecular Weight of α-Amino Acids

An accurately weighed (20-60mg) sample of α-amino acid was dissolved in distilled water (10ml) in an Erlenmeyer flask. To this, a solution of 0.01 mol of HIO_3 was introduced and it was heated to about 65°C on water bath for 2 hr, to this solution about 5ml of dilute sulphuric

acid and 5ml of 10% potassium iodide was added and the liberated iodine was titrated against the standardised sodium thiosulphate solution using starch as indicator. In a similar way, a blank titration was conducted without adding glucose under identical condition. From the difference in the volume of sodium thiosulphate solution consumed, the molecular weight 'M' was calculated using equation 1.

Results and Discussions

The method reported here makes use of the fact that α-amino acid is known to undergo an oxidative decarboxylation by HIO_3, yielding the aldehyde by consuming one mole of HIO_3 per one molecule of α-amino acid. Generally a known volume of HIO_3 is added to known mass of α-amino acid, after the completion of the reaction, the unreacted HIO_3 is determined iodometrically. By carrying out a parallel blank experiment the amount of the HIO_3 consumed is determined. As the overall reactions require one mole HIO_3 per one mole of the α-amino acid, which is equivalent to mole of iodine, weight of the α-amino acid is determined by using equation 1.

Conclusion

We have developed a reliable, cost effective method for the determination of neutral amino acids using mild conditions and without the use of any sophisticated instruments and also this method requires short time.

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