
Determination of Ascorbic Acid in Fruit Juices by Reducing behaviour of Oxalohydroxamic Acid

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Abstract

The main aim of the present investigation is to determine the concentration of vitamin C by following the mechanism of redox reactions. Oxalohydroxamic acid (OHA), a well known reducing agent is used in this investigation. Concentration of vitamin C in various fruit juice samples such as lemon, orange and lime, also in tablets has been determined by oxidizing its solution with standard Cerium (IV) solution and remainder Cerium (IV) is then back titrated with OHA. The current procedure has been applied for the determination of ascorbic acid minimum up to the concentration of 2 ppm potentiometrically in authentic sample. 90 ppm ascorbic acid was determined in lemon juice, 60 ppm in orange juice and 40 ppm in lime.

Keywords: Redox reaction, Oxalohydroxamic acid, Potentiometry, Vitamin C

Introduction

Vitamin C or L-ascorbic acid is classified as a carbohydrate having chemical structure of 1-keto-L-threo-hexono- γ -lactone-2,3-enediol [1]. Enediol-group [-C(OH)=C(OH)-] is responsible for the acidic and reducing properties of the ascorbic acid [4].

Vitamin C is a water soluble vitamin. It is important in forming collagen, a protein that gives structure to bones, cartilages, muscles, and blood vessel. Vitamin C also aids in the absorption of iron, and helps to maintain bones and teeth. It is the most common and one of the most ubiquitous vitamins ever discovered. Blackcurrant, citrus fruits, leafy vegetables, tomatoes, green and red apples are the rich sources of vitamin C [5]. It has been also used as antioxidant [5]. Ascorbic acid is a mild reducing agent. Its solution is not stable in air for a long time. With the aid of stabilizers which ward off the oxidation of ascorbic acid by atmospheric oxygen, titration can be carried out in open vessels; this establishes the superiority of this useful reagent over the other reducing agents such as titanium and tin. Reducing behavior has been studied for determination of Iron (III) spectrophotometrically [6]. Benzohydroxamic acid has been used for determination of Thorium Manganese [7].

Present work deals with the determination of ascorbic acid with the help of standard Cerium solution and standard solution of Oxalohydroxamic Acid which was used as reducing agent.

Material and Method

Material and Reagents

All the solutions were prepared with millipore water from AR-grade materials. Stock solution of ascorbic acid was prepared on daily basis. Accurately weighed amount of ascorbic acid was dissolved in per 100 mL of millipore water and working solutions of required concentration was prepared by dilution method. Solution of 0.0075M Ceric Ammonium Nitrate was prepared by dissolving required amount in millipore water containing 8N sulphuric acid. Ce(IV) solution was standardized against Fe(II) solution which in turn was standardized

against standard $K_2Cr_2O_7$ solution. Standard solutions of 0.001M Oxalohydroxamic Acid was prepared by dissolving accurately weight amount in millipore water.

Procedure

Accurately measured volume of standard ascorbic acid solution was transferred in titration vessel and excess volume of Ce(IV) solution is added with constant stirring. A portion of Cerium solution was reduced by ascorbic acid whereas remaining Cerium has been back titrated with standard OHA solution potentiometrically. Same procedural method has been applied for fruit juices.

Result & Discussion

Determination of Ascorbic Acid by Iodometric Method using Standard Solution of Ascorbic Acid

Ascorbic Acid : 1mg/ml
 Strength of Ceric Ammonium Nitrate : 0.04945M
 Strength of H_2SO_4 Solution : 8N
 Strength of KIO_3 : 0.0075 N
 Total Volume of Titration Sample : 50 mL

Table I – Determination of Ascorbic Acid in Standard Solution by Iodometric Method

S. No.	Vol. Of Ascorbic acid Taken (mL)	Vol. Of KIO_3 Consumed In Titration (mL)	Amount Of KIO_3 Consumed (mg)	Amount Of Ascorbic acid Found (mg)
1	2.00	1.5185	0.4062	2.0057
2	4.00	3.0314	0.8110	4.0041
3	6.00	4.5493	1.2171	6.0092
4	8.00	6.0578	1.6206	8.0017
5	10.00	7.5963	2.0322	10.0340

Same standard solutions were used for the determination of ascorbic acid by the proposed method.

Determination of Various Quantities of Ascorbic Acid by Proposed Method using Standard Solution of Ascorbic Acid

Standard Solution of Ascorbic Acid : 1mg/ml
 Strength of Ceric Ammonium Nitrate : 0.04945 M
 Strength of H_2SO_4 Solution : 8N
 Volume of Ascorbic Acid Solution Taken : 2 ml
 Strength of OHA : 0.0125M
 Total Volume of Titration Sample : 50 mL

Table II – Determination of Ascorbic Acid in Standard Solution by Proposed Method

S. No.	Ce (IV) Taken (mg)	Vol of OHA required to reduce excess Ce (IV) (mL)	Amount of Ce (IV) Excess (mg)	Amount of Ce (IV) required to oxidize ascorbic acid (mg)	Amount of ascorbic acid found (mg)
1	13.864	0.5072	10.6611	3.2029	2.0128
2	13.864	0.5075	10.6674	3.1966	2.0088
3	13.864	0.5078	10.6737	3.1903	2.0048
4	13.864	0.5078	10.6737	3.1903	2.0048
5	13.864	0.5077	10.6716	3.1924	2.0062
6	13.864	0.5078	10.6737	3.1903	2.0048
7	13.864	0.5078	10.6737	3.1903	2.0048
8	13.864	0.5077	10.6716	3.1924	2.0062
9	13.864	0.5078	10.6737	3.1903	2.0048
10	13.864	0.5078	10.6737	3.1903	2.0048

Mean: 2.0063; SD 0.0026; CV: 0.1296

Standard Solution of Ascorbic Acid : 1mg/ml
 Strength of Ceric Ammonium Nitrate : 0.04945 M
 Strength of H₂SO₄ Solution : 8N
 Volume of Ascorbic Acid Solution Taken : 4 ml
 Strength of OHA : 0.0125M
 Total Volume of Titration Sample : 50 mL

Table III – Determination of Ascorbic Acid in Standard Solution by Proposed Method

S. No.	Ce (IV) Taken (mg)	Vol. of OHA required to reduce excess Ce (IV) (mL)	Amount of Ce (IV) Excess (mg)	Amount of Ce (IV) required to oxidize ascorbic acid (mg)	Amount of ascorbic acid found (mg)
1	13.8614	0.3558	7.4787	6.3827	4.0110
2	13.8614	0.3562	7.4871	6.3743	4.0057
3	13.8614	0.3561	7.4850	6.3764	4.0070
4	13.8614	0.3562	7.4871	6.3743	4.0057
5	13.8614	0.3562	7.4871	6.3743	4.0057
6	13.8614	0.3562	7.4871	6.3743	4.0057
7	13.8614	0.3562	7.4871	6.3743	4.0057
8	13.8614	0.3561	7.4850	6.3764	4.0070
9	13.8614	0.3562	7.4871	6.3743	4.0057
10	13.8614	0.3562	7.4871	6.3743	4.0057

Mean: 4.0065; SD: 0.0017; CV: 0.0417

Standard Solution of Ascorbic Acid	: 1mg/ml
Strength of Ceric Ammonium Nitrate	: 0.04945 M
Strength of H ₂ SO ₄ Solution	: 8N
Volume of Ascorbic Acid Solution Taken	: 6 ml
Strength of OHA	: 0.0125M
Total Volume of Titration Sample	: 50 mL

Table IV– Determination of Ascorbic Acid in Standard Solution by Proposed Method

S. No.	Ce (IV) Taken (mg)	Vol. of OHA required to reduce excess Ce (IV) (mL)	Amount of Ce (IV) Excess (mg)	Amount of Ce (IV) required to oxidize ascorbic acid (mg)	Amount of ascorbic acid found (mg)
1	13.8614	0.2049	4.3069	9.5545	6.0042
2	13.8614	0.2049	4.3069	9.5545	6.0042
3	13.8614	0.2059	4.3279	9.5335	5.9910
4	13.8614	0.2049	4.3069	9.5545	6.0042
5	13.8614	0.2049	4.3069	9.5545	6.0042
6	13.8614	0.2049	4.3069	9.5545	6.0042
7	13.8614	0.2048	4.3048	9.5566	6.0055
8	13.8614	0.2049	4.3069	9.5545	6.0042
9	13.8614	0.2049	4.3069	9.5545	6.0042
10	13.8614	0.2049	4.3069	9.5545	6.0042

Mean: 6.0030; SD: 0.0042; CV: 0.0707

Standard Solution of Ascorbic Acid	: 1mg/ml
Strength of Ceric Ammonium Nitrate	: 0.04945 M
Strength of H ₂ SO ₄ Solution	: 8N
Volume of Ascorbic Acid Solution Taken	: 8 ml
Strength of OHA	: 0.0125M
Total Volume of Titration Sample	: 50 mL

Table V– Determination of Ascorbic Acid in Standard Solution by Proposed Method

S. No.	Ce (IV) Taken (mg)	Vol. of OHA required to reduce excess Ce (IV) (mL)	Amount of Ce (IV) Excess (mg)	Amount of Ce (IV) required to oxidize ascorbic acid (mg)	Amount of ascorbic acid found (mg)
1	27.7228	0.7126	14.9785	12.7443	8.0087
2	27.7228	0.7126	14.9785	12.7443	8.0087
3	27.7228	0.7129	14.9848	12.7380	8.0048
4	27.7228	0.7128	14.9827	12.7401	8.0061
5	27.7228	0.7129	14.9848	12.7380	8.0048
6	27.7228	0.7129	14.9848	12.7380	8.0048
7	27.7228	0.7129	14.9848	12.7380	8.0048
8	27.7228	0.7129	14.9848	12.7380	8.0048
9	27.7228	0.7129	14.9848	12.7380	8.0048
10	27.7228	0.7129	14.9848	12.7380	8.0048

Mean: 8.0057; SD: 0.0017; CV: 0.0207

Standard Solution of Ascorbic Acid : 1mg/ml
 Strength of Ceric Ammonium Nitrate : 0.04945 M
 Strength of H₂SO₄ Solution : 8N
 Volume of Ascorbic Acid Solution Taken : 10 ml
 Strength of OHA : 0.0125M
 Total Volume of Titration Sample : 50 mL

Table VI– Determination of Ascorbic Acid in Standard Solution by Proposed Method

S. No.	Ce (IV) Taken (mg)	Vol. of OHA required to reduce excess Ce (IV) (mL)	Amount of Ce (IV) Excess (mg)	Amount of Ce (IV) required to oxidize ascorbic acid (mg)	Amount of ascorbic acid found (mg)
1	27.7228	0.5610	11.7919	15.9309	10.0112
2	27.7228	0.5616	11.8046	15.9182	10.0033
3	27.7228	0.5616	11.8046	15.9182	10.0033
4	27.7228	0.5617	11.8067	15.9161	10.0020
5	27.7228	0.5614	11.8003	15.9225	10.0059
6	27.7228	0.5617	11.8067	15.9161	10.0020
7	27.7228	0.5617	11.8067	15.9161	10.0020
8	27.7228	0.5617	11.8067	15.9161	10.0020
9	27.7228	0.5617	11.8067	15.9161	10.0020
10	27.7228	0.5617	11.8067	15.9161	10.0020

Mean: 10.0036; SD: 0.0030; CV: 0.0297

Determination of Ascorbic Acid in Various Fruit Juices

Table VII - Determination of Ascorbic Acid in Lemon Juices by Proposed Method

S. NO.	Ce (IV) Taken (mg)	Vol. of OHA required to reduce excess Ce (IV) (mL)	Amount of Ce (IV) Excess (mg)	Amount of Ce (IV) required to oxidize ascorbic acid (mg)	Amount of ascorbic acid found (mg)
1	34.535	0.9610	20.1997	14.3353	9.0085
2	34.535	0.9613	20.2060	14.3290	9.0046
3	34.535	0.9613	20.2060	14.3290	9.0046
4	34.535	0.9613	20.2060	14.3290	9.0046
5	34.535	0.9614	20.2081	14.3269	9.0032
6	34.535	0.9613	20.2060	14.3290	9.0046
7	34.535	0.9613	20.2060	14.3290	9.0046
8	34.535	0.9615	20.2102	14.3248	9.0019
9	34.535	0.9613	20.2060	14.3290	9.0046
10	34.535	0.9613	20.2060	14.3290	9.0046

Mean: 9.0046; SD: 0.0016; CV: 0.0183

Table VIII - Determination of Ascorbic Acid in Orange Juices by Proposed Method

S. NO.	Ce (IV) Taken (mg)	Vol of OHA required to reduce excess Ce (IV) (mL)	Amount of Ce (IV) Excess (mg)	Amount of Ce (IV) required to oxidize ascorbic acid (mg)	Amount of ascorbic acid found (mg)
1	34.535	1.1886	24.9838	9.5512	6.0021
2	34.535	1.1876	24.9628	9.5722	6.0154
3	34.535	1.1880	24.9712	9.5638	6.0101
4	34.535	1.1885	24.9817	9.5533	6.0035
5	34.535	1.1885	24.9817	9.5533	6.0035
6	34.535	1.1885	24.9817	9.5533	6.0035
7	34.535	1.1886	24.9838	9.5512	6.0021
8	34.535	1.1885	24.9817	9.5533	6.0035
9	34.535	1.1885	24.9817	9.5533	6.0035
10	34.535	1.1880	24.9712	9.5638	6.0101

Mean: 6.0057; SD: 0.0045; CV: 0.0748

Table IX - Determination of Ascorbic Acid in Lime by Proposed Method

S. No.	Ce (IV) Taken (mg)	Vol of OHA Required (mL)	Amount of Ce (IV) left un-reacted (mg)	Amount of Ce (IV) required to oxidize ascorbic acid (mg)	Amount of ascorbic acid found (mg)
1	34.6535	1.3453	28.2775	6.3760	4.0068
2	34.6535	1.3454	28.2796	6.3739	4.0054
3	34.6535	1.3454	28.2796	6.3739	4.0054
4	34.6535	1.3453	28.2775	6.3760	4.0068
5	34.6535	1.3453	28.2775	6.3760	4.0068
6	34.6535	1.3453	28.2775	6.3760	4.0068
7	34.6535	1.3453	28.2775	6.3760	4.0068
8	34.6535	1.3453	28.2775	6.3760	4.0068
9	34.6535	1.3453	28.2775	6.3760	4.0068
10	34.6535	1.3453	28.2775	6.3760	4.0068

Mean: 4.0065; SD: 0.0006; CV: 0.0139

Discussion

Standard solutions of ascorbic acid were prepared and analysed by both the methods. In order to prove the validity of the proposed method our results are compared with the earlier reported method i.e. KIO_3 method [11]. The results are recorded in Table - I. Results are found in good agreement Table - X. It proves that proposed method is quite satisfactory for the potentiometric determination of ascorbic acid.

Table-X Determination of Ascorbic Acid by the Proposed Method using Standard Solution of Ascorbic Acid

S. No.	Ce (IV) Taken (mg)	Volume of Ascorbic Acid Solution Taken (mL)	Vol. of OHA Consumed in Titration (mL)	Amount of Ce Titrated (mg)	Amount of Ce Consumed by Ascorbic acid (mg)	Amount Of Ascorbic acid Determined (mg)
1	13.8614	2.00	0.5072	10.6611	3.2029	2.0128
2	13.8614	4.00	0.3558	7.4787	6.3827	4.0110
3	13.8614	6.00	0.2049	4.3069	9.5545	6.0042
4	27.7228	8.00	0.7129	14.9848	12.7380	8.0048
5	27.7228	10.00	0.5617	11.8067	15.9161	10.002

This method has been applied for the estimation of vitamin-C in fruits juices. Using proposed method, 90 mg/100mL in Lemon juice, 60 mg/100mL in Orange and 40 mg/100mL quantities were determined.

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