

Determination of Ascorbic Acid in Pharmaceutical formulations and Citrus Fruits

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ABSTRACT

A simple and sensitive titrimetric method has been developed for the determination of ascorbic acid in pharmaceutical formulations and citrus fruits. The low values of relative standard deviation indicated high precision of the method. Intra and inter day assay relative standard deviations were less than 1 %. The method has been applied successfully to the determination of ascorbic in various pharmaceutical preparations and citrus fruits. There was no interference from drugs commonly administered with ascorbic acid. The method has been shown to be linear, reproducible, specific and rugged.

Keywords: *Ascorbic acid, Pharmaceutical formulations, Citrus Fruits*

I. INTRODUCTION

Ascorbic acid also known as vitamin C is the most important vitamin for human nutrition that is supplied by fruits and vegetables. It is a valuable nutritional component because of its antioxidant and therapeutic properties. It helps the body in forming connective tissues, bones, teeth, blood vessels and plays a major role as an antioxidant that forms part of the body defence system against reactive oxygen species and free radicals, thereby preventing tissue damage¹⁻³

Ascorbic acid is widely distributed in plant cells, which plays many crucial roles in growth and metabolism. It is also a potential antioxidant having the capacity to eliminate several different oxygen species keeping the membrane-bound antioxidant, α -tocopherol (vitamin E) in the reduced state and so acts as a cofactor to maintain the activities of a number of enzymes by keeping metal ions in the reduced state which appears to be the substrate for oxalate and tartrate biosynthesis⁴⁻⁶.

It is widely used in the treatment of certain diseases such as scurvy, common cold, hemorrhagic disorder, wound healings as well as infertility⁷. Ascorbic acid is also reported to be an important ingredient for the synthesis of dopamine, noradrenaline and adrenaline in the nervous system or in the adrenaline glands⁸.

Ascorbic acid is also reported to be an essential antioxidant that helps protect against cancers, heart disease and stress. It is also part of cellular chemistry that provides energy, it is essential for sperm production and for

making the collagen protein involved in the building and health of cartilage, joints, skin and blood vessels. Ascorbic acid helps in maintaining a healthy immune system, it aids in neutralizing pollutants, is needed for antibody production, acts to increase the absorption of nutrients (including iron) in guts, and thins the blood⁹.

Citrus fruits such as lime, lemon, orange, grape fruit and tomato are common sources of vitamin C or ascorbic acid. Other sources include papaya, broccoli, Brussels, sprouts, black berries, cauliflower, spinach, cantaloupe and blue berries.

Most plants and animals have the ability to synthesize ascorbic acid for their needs. The only mammals that are unable to synthesize ascorbic acids are primates including man and guinea pigs. Therefore, humans depend on exogenous sources of ascorbic acid which include fruits and vegetables as well as food supplements and pharmaceutical preparations³.

Lack of ascorbic acid in daily diet leads to a disease called scurvy, a form of vitaminosis that is characterized by loose teeth, superficial bleeding, fragility of blood vessels, poor healing, compromised immunity, mild anemia. The dietary amounts recommended by various authorities are 50 – 150mg of ascorbic acid per day. High doses (thousands of mg) are used but may result in diarrhea. Any excess of ascorbic acid is needed in the diet to prevent scurvy. It also has a reputation for being useful in the treatment of colds and flu. The evidence to support this idea however is ambiguous⁹.

The most important and prominent source of ascorbic acid or vitamin C is the plants and particularly, citrus fruits. However, the availability of these fruits depends on their sources and storage time, which could also affect the levels of concentration of vitamin C in these fruits. It has been reported that the amount of vitamin C in fruits depends on the precise variety, the soil and climate in which it grew and the length of time it was picked⁸.

The human body cannot synthesize ascorbic acid by itself, and so has to get its source of the essential nutrient from its diet. In this research work, the levels of ascorbic acid in pharmaceutical preparation and various citrus fruits were determine. The aim of this work is to determine the levels of ascorbic acid concentration in the citrus species selected and also determine the phase of fruit development containing high concentration of ascorbic acid. This is with the aim of giving a guide as to the phase at which ascorbic acid levels are high in these fruits so as to enhance maximum benefits of the fruits in terms of vitamin C or ascorbic acid yield.

II. MATERIALS AND METHODS

Pharmaceutical grade (> 99%) Ascorbic acid was obtained from Sigma Aldrich, Iodine, starch (s.d Fine-Chem, Ltd., India) and water (Rankem Ltd., India) used were of analytical grade. Calibrated glasswares used for the analysis of ascorbic acid.

Stock solutions: A stock solution of ascorbic acid (1mg /mL) was prepared by dissolving appropriate amount of substance in distilled water and stored at +4°C. The working solution was prepared from the stock solution as and when required.

Pharmaceutical preparation: Twenty tablets of the selected drug were finely powdered. An amount equivalent to 200 mg of the tablet powder was weighed accurately and transferred into a 250 ml volumetric flask. 50 ml of

distilled water was added and the powder was completely disintegrated and was made up to 250 ml with distilled water. Further transferred 10 ml of this solution and made upto 100 ml with distilled water.

For citrus fruits: The bark of the fruits were peeled off with sterile knife and sliced into half, this was manually compressed into a 100cm³ beaker and filtered off in to a 100cm³ volumetric flask to remove the pulps which could block pipette jets. The filtrate is diluted up to the mark with distilled water.

Method: Method used for the determination of ascorbic acid in Pharmaceutical preparation and fruit juice was as specified in the standard AOAC methods, 939.13 and 966.18¹⁰. This method was chosen due to its advantages compared to other methods. It is sensitive, economic, practical and less time consuming⁸.

III. RESULTS AND DISCUSSION

The method was applied to the analysis of ascorbic acid in tablet and the results were shown in Table 1. The precision of the method (within-day variations of replicate determinations) was checked by analysing nine times at the LOQ level. The precision of the method expressed as the relative standard deviations (R. S. D., %) at the LOQ level, were 2.27. The low values of relative standard deviation indicated high precision of the method.

Table 1. Precision parameters

Compound	R. S. D. (%)
Ascorbic acid	2.27

$$R. S. D. (\%) = ((S.D./mean) \times 100)$$

A standard working solution containing ascorbic acid to give final concentration 20mg/ml was prepared. The solution of standard was titrated six times as a test sample. From the results, the concentration of ascorbic acid was calculated. The accuracy, defined in terms of % deviation of the calculated concentrations from the actual concentrations is listed in Table 2.

Table 2 Accuracy parameters (recovery)

Drug	Spiked concentration (mg/ml)	Measured concentration (µg/ml; mean ± S. D.)	R. S. D. (%)	Deviation (%)
ascorbic acid	15.25	15.17	101.2	0.7
	20.80	20.70	100.7	1.4
	25.50	25.28	102.6	0.8

$$R. S. D. (\%) = ((Spiked\ concentration - mean\ measured\ concentration) \times 100) / spiked\ concentration.$$

The values obtained for the concentration of ascorbic acid in various citrus fruits were as given in table 3.

Table 3: Ascorbic Acid Concentration in Citrus Fruits

Citrus Fruits	Botanical name	Ascorbic Acid Concentration (mg/100mL)
Sweet orange	Citrus sinensis	49.27
Lime	Citrus aurantifolia	33.15
Lemon	Citrus lemon	50.41

The values obtained for the analysis of ascorbic acid in various citrus fruits depends upon the ascorbic acid levels in fruits. These factors include; climate, temperature and amount of nitrogen fertilizers used in growing the plants. Climatic conditions such as light and temperature have been reported to affect the chemical composition of horticultural crops. It has been reported that fruits which are exposed to maximum sunlight have been shown to contain higher amount of ascorbic acid than those shaded on the same plant¹¹.

IV. CONCLUSION

The method used for the quantitative analysis of ascorbic acid is linear, accurate, precise, rugged and rapid. The method was fully validated showing satisfactory data for all method validation parameters tested. The method is stability indicating and can be conveniently used by quality control department to determine the related substance and assay in regular ascorbic acid production samples and also in various citrus fruits.

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