Indian Journal of Pure & Applied Physics
Vol. 61, September 2023, pp. 751-753
DOI: 10.56042/ijpap.v61i9.3486

Electronic Spectroscopic Study of Stability of L-Ascorbic Acid (Vitamin C) in Aqueous Solutions of Salts ($NaHCO_3$, NaCl, and KCl)

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Received 7 July 2023; accepted 1 August 2023

Ascorbic acid commonly known as Vitamin C, is a water-soluble anti-oxidant that is essential for the growth, development, and repair of tissues. The stability of this molecule can be affected by several factors, including pH, temperature, and other factors. We aim to study the stability of ascorbic acid in presence of common food additive and ions such as Na^+, K^+, Cl^-, and HCO_3^- that are naturally present in human body via spectroscopic measurement. Physiological concentration of sodium, potassium and chloride ions has no influence on the stability. When ascorbic acid was added to a 0.175 $MHCO_3$- aqueous solution, a strong peak was detected at 265.0 nm due to the presence of ascorbate anion. However, within 75 minutes, this peak disappeared completely, indicating significant decomposition. At a lower, physiological 0.035 M concentration of HCO_3^- , the peak persisted at 265.0 nm, and the rate of decomposition was slower. Our findings suggest that the elevated levels of HCO_3- in serum plasma would rapidly decompose ascorbic acid, underscoring the importance of maintaining proper physiological balance in vivo.

Keywords: L-ascorbic acid; Spectroscopic measurements; Water-soluble vitamins

1 Introduction

Vitamins are an essential part of a healthy diet and are required for the normal functioning of physiological processes in humans[1]. Vitamins do not function as energy currencies or form structural components of the cell; rather, they serve as cofactors for metabolic and maintenance enzymes[2]. Ascorbic Acid also known as Vitamin C, is one of the most indispensable micronutrients for normal metabolic function in the body[3]. In the course of evolution, mutations at the genetic level have rendered the human liver incapable of performing ascorbate biosynthesis[4,5]. Vitamin C, which is an essential nutrient, must therefore be regularly supplied via diet. Traditional culinary methods steaming and boiling can leach water-soluble vitamins and destroy vitamin C[6]. In addition, it has been observed that food processing in the presence of an excess of salts and baking soda ($NaHCO_3$) may also contribute to decreased vitamin C availability.

Reduced ascorbic acid (H_2A) is an organic acid with anti-oxidant role in numerous biochemical reactions occurring within the human body[7,8]. The reducing behaviour of H_2A^- associated with the enediol group in the ring -limits the solution and solid-state stability at room temperature. Ascorbic acid (H_2A) is a dibasic acid (pKa values 4.1 and 11.8). The dissociation of both of its enolic hydroxyl groups is possible. It forms salts, the most important of which are monosodium, disodium, and calcium salts, which form aqueous solutions that are highly acidic. Under mild conditions, ascorbic acid is oxidised to semi-dehydroascorbic acid (AH^-, also known as ascorbate) and then to a free radical intermediate ($AH^{\cdot}$ - ascorbyl free radical), which is subsequently proportional to ascorbic acid and dehydroascorbic acid (A_2). All four forms are depicted in the reversible redox system diagram[9,10]. It is an efficient scavenger of singlet oxygen and super oxide radicals to H_2O_2[11]. In environments with a weakly acidic or neutral pH, dehydroascorbic acid is not ionised; consequently, it is relatively hydrophobic and better able to penetrate the cell membrane. Dehydroascorbic acid (DHA) is unstable in aqueous solution and is degraded by hydrolytic ring opening into 2, 3-diketogulonic acid or 2, 3-dioxo-L-gulonicacid[12]. (Scheme-I).

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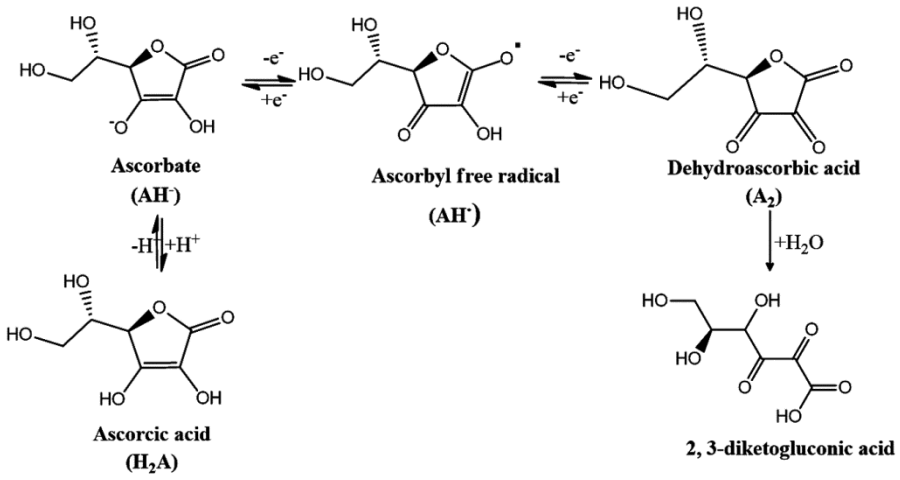

Scheme I —Oxidation of Ascorbic acid.

2 Material and Methods

2.1 Chemicals and Reagents

In our study, we ensured the highest quality and purity of all chemicals used. Ascorbic acid, spectra grade, was purchased from Merck, India. The solvents used in the spectral measurements were also of spectra grade quality.

2.2 UV spectroscopy

In this study, UV spectra of Milliq water was recorded assess the purity. The spectral transmissions of solvents were also checked against Milliq water to ensure accurate measurements. Additionally, the conductivity of the water was recorded and found to be similar to the literature value of demineralized water. Study focused on the effect of the presence of alkali, alkaline earth metals, and various ions on the decomposition of Vitamin C. The Varian Cary 100 spectrophotometer was used for UV measurements, which has a wavelength range of 190-900 nm, resolution of 0.1nm and bandwidth 1nm. To hold the samples, a matched pair of quartz cells with a path length of 1cm was used. Finally, derivative plots were obtained using inbuilt software supplied by Varian to analyse the data. All solutions were prepared just prior to recording spectra.

3 Results and Discussion

Na^+, K^+, Cl^-, and HCO_3^- ions are constituents of human blood plasma that are known to influence the various biochemical reactions in the human body in vivo[13,14]. Concentration of ions may vary depending on the dietary supplements and physiological state of the body. Potassium ions are known to stabilize ascorbate anions. We reasoned that other ionic species might also influence the concentration and physiologically availability Vitamin C and therefore decided to measure the stability in presence of these ions at various concentration.

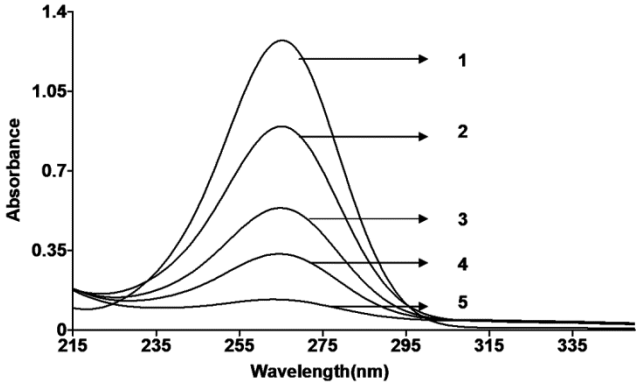

Fig. 1 — UV Spectra of Vitamin C in bicarbonate solution (0.175 M) over a period of time. (1) At 0 minutes; (2) At 30 minutes; (3) At 45 minutes; (4) At 60 minutes; (5) At 75 minutes.

Vitamin C was added to 0.175 M HCO_3^- solution in demineralized water (Fig. 1). Only one peak was observed at 265.0 nm (peak identified with ascorbate anion). The fall in absorbance at this position was quite steep. Within 75 min the ascorbate got decomposed almost completely. When spectra recorded at intervals of time were re-plotted in zero-order derivative mode and made to overlap, no isosbestic point was observed. (However, overlapping of first derivative spectra showed one sharp isosbestic point at 265.0 nm, identified with ascorbate anion). The absence of isosbestic point in zero-order spectra is a good indication more than one equilibrium is involved; the rate of decomposition of all the species involved is very high and a condition

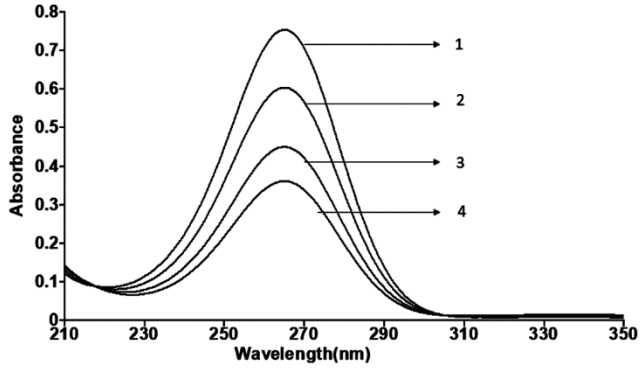

Fig. 2 — UV Spectra of Vitamin C in bicarbonate solution (0.035 M) over a period of time. (1) At 0 minutes; (2) At 30 minutes; (3) At 45 minutes; (4) At 60 minutes.

of steady-state concentration is not attained during decomposition.

- At 0.035M HCO_3^- (the concentration in serum plasma) the peak was still observed at 265.0 nm, but the rate of decomposition was slower. One clear isosbestic point, at 218.0 nm was observed in zero-order derivative mode. This supports our hypothesis (Fig. 2). We tentatively postulate that isosbestic point at this position may be due the equilibrium existing between the DHA and its decomposition product 2, 3-diketogulonic acid (the λ_{max} for this compound is ca. 214.0 nm). This kind of equilibrium we have identified only in this system.
- At further lower concentration of HCO_3^-, the rate of decomposition decreased further. Isosbestic point was observed at 216.0 nm.

  The result of this study points out that, higher concentration of HCO_3^- results in the decomposition of ascorbate irreversibly. Higher conc. of HCO_3^- is usually associated with the salinity of the water. Could it be one of the reasons that saline water is very harmful to health?
- No degradation of Vitamin C was observed at physiological concentration of NaCl and KCl [data not shown].

4 Conclusion

Ascorbic acid is a crucial nutrient for the growth, repair and development of the body tissues. Our study aimed at addressing the stability and decomposition of Vitamin C in presence of ions such as HCO_3^-, Na^+, K^+ and Cl^-. Our findings indicate that Vitamin C is degraded by HCO_3^- in a concentration dependent manner at room temperature and that degradation is irreversible. No significant effect of sodium potassium and chloride ions at physiological concentration was observed in our study. However, higher concentration of Chloride ion did degrade Vitamin C [data not shown].

This finding assumes significance in the context of indiscriminate addition of food additive sodium bicarbonate in the fast-food industry. Excessive consumption of baking soda will significantly reduce uptake of dietary Vitamin C. Overall, our study emphasizes the importance of understanding the effects of various ions and their concentrations on Vitamin C stability and decomposition, particularly in the context of human health.

Acknowledgement

VKG thanks Sanganeria Foundation for financial support.

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