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Research Article

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Nano- Al_2SiO_5 / Graphite Paste Sensor Fabrication: It's Use in Voltametric Estimation of Ascorbic Acid in Locally Consumed Vitamin-C Formulations

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ABSTRACT

The carbon paste modified with aluminum silicate was utilized to develop a new sensor and applied for the study of the electro catalytic oxidation and voltammetric determination of the ascorbic acid in locally used general medications. Differential pulse voltammetric method was used to study the electrochemical properties of AA using electrode. It was found that the anodic peak current increased more than two fold in magnitude in presence of the modifier rather than in pure carbon paste. Optimizations of the parameters have been made at the best electrode composition of 15% AS, 62.5% graphite and 22.5% paraffin oil. A sensitive, simple and time-saving procedure has been developed for the analysis of AA in the linear range 5μ to $100\mu\text{M}$ with a detection limit $1.20\mu\text{M}$ and quantification limit $5.73\mu\text{M}$. The irreversible anodic oxidation process at interface follow both the diffusion controlled as well as surface controlled mechanism at modified electrode surface at high pulse period. The two electrons and two protons oxidation scheme for AA has also been proposed. The validity of this technique was tested on quantification of ascorbic acid in three commercial pharmaceutical products (Limcee, Beconzym C forte tablets and Capsule) directly and after spiking the sample solutions with known quantity of analyte. The observed results showed a good agreement with reported amount of ascorbic acid in these pure and spiked samples.

Keywords: Ascorbic acid, Aluminum silicate, Carbon paste electrode, Differential pulse voltammetry, Electrochemical oxidation

ARTICLE INFO

CONTENTS

1. Introduction	490
2. Experimental.	491
3. Results and Discussion.	491
4. Conclusion.	496
5. References	496

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1. Introduction

Ascorbic acid (AA) as vitamin besides its antioxidant action has found its use for nutritional health of animals and human as supplements and food fortification. Its deficiency leads to scurvy, characterized by weakening of collagenous structures, resulting in widespread capillary bleeding; if left untreated, gangrene and death may ensue. Though, the dilemma is rare in developed countries but still rampant in developing and under-developed nations. It occurs in a variety of natural edibles like citrus fruits, black currants, peppers, green vegetables and soft fruits - strawberries, guava, mango, kiwi etc. These are some of its predominantly rich dietary food sources. It plays a vital role in preventing a variety of diseases and therapeutic utility in wound healing, blood pressure, cancerous and cardiovascular diseases - heart disease and stroke. AA added to foodstuffs during processing or before packing to protect color, aroma, nutrient content and specially wheat flour improves its baking qualities [Bruso -2015; Varvera et. al. -2016]. It is also an essential ingredient of several pharmaceutical products as an anti-oxidants as well as stabilizer for vitamin B-complex. Consequent upon its desirable effects, it is widely used in the treatment of certain diseases such as scurvy, anemia, hemorrhagic disorders etc. It is considered one of the essential components for the development and regeneration of muscles, bones, teeth and skin. The major role of it to cure disease and improve/ maintain human health care- its immune-stimulating effect, as anti-allergic, in regeneration of other antioxidants as vitamin E, as wound healer in combination with zinc and cement for connective tissues. It contributes to the health of teeth and gums by preventing hemorrhaging and bleeding effects besides a crucial factor in the eye's ability to deal with oxidative stress and vision-loss.

The people addicted to alcohol and smoking need more uptake of AA than normal people as these have negative interactions. On the other hand, the presence of antioxidants, as vitamin E and beta-carotene, supports the protective antioxidant action of vitamin-C [Gees et. al. -2015; Lewis et. al. -2013; Suzanne Humphries -2016]. Also, it has been identified as a radical scavenger *in-vivo*. The fate of vitamin-C is still in the first name it received, many years ago: we still ignore much of its actual relevance in cell metabolism, though we are progressively getting aware of many facets of this fascinating molecule and its direct involvement in the regulation of apparently unrelated pathways, much more than just an antioxidant [De-Tullio -2012]. The therapeutic importance of AA has prompted most of the researchers to develop methods for its determination in real samples as well as in pharmaceuticals and some of these methods have been reviewed recently [Arya & Jain -2000; Carmel et.al. -2015; Hossu & Magearu -2004].

Most of the plants and animals have the ability to synthesize vitamin-C. The man and guinea pigs are only mammals that can't synthesize vitamin-C. Therefore, human-beings depend on its exogenous sources like fruits, vegetables as well as food-supplements along with pharmaceutical preparations [Parviainen & Townsend -1995]. The determination of vitamin-C has gained increased significance in several areas of analytical chemistry such as pharmaceutical, clinical and food application. The increasing use of pharmaceuticals and other natural samples containing vitamin-C has necessitated the development of an accurate, specific and easy operable procedure for its determination. It has been demonstrated that AA can undergo mediated oxidation via a homogeneous process by electro-generated hexacyanoferrate / ferricinium derivatives [Pournaghi-Azar & Ojani -1999]. In addition to these, some chemically modified electrodes with various active mediators immobilized at the electrode surface for its mediated oxidation have also been reported [Fang et.al. -2015; Fritea et.al.-2015; Liu et.al. -2011; Xu-li et.al. -2013].

The electrochemical response characteristics of the modified electrode toward AA were investigated by differential pulse voltammetry (DPV) in phosphate buffer solution (pH 2.0) by a modified multiwall carbon nanotube paste electrode using cetrimonium iodide/iodine [Noroozifar et.al.-2012]. A novel nano-structured composite, azidecopperocta (3-aminopropyl) octasilsesquioxane (ASCA) incorporated into a graphite paste electrode and the electrochemical studies of AA were conducted using cyclic voltammetric technique [Umesh et. al. -2010]. Furthermore cyclic and differential pulse voltammetry were employed for electro-catalytic AA determination at a carbon paste electrode modified with 2, 7-bis (ferrocenylethynyl) fluoren-9-one [Raouf et. al. -2007]. Cyclic voltametric (CV) technique using platinum electrode was applied for sensing and estimation of AA contents in citrus juices and soft drinks. The study suggested that the growth of platinum surface-oxides and the anodic response of variety of interferon's (glucose, cysteine, oxalate etc) suppressed by the use of flour-surfactant-modified platinum electrodes [Pisoschi et. at. -2011]. In addition to this AA was also determined in the presence of sulphur dioxide and acetaldehyde by DPV technique at inter-digital microelectrodes [Francioso et. al. -2007]. Ascorbic acid in presence of dopamine at eriochrome black-T modified carbon paste electrode was studied using DPV technique with detection limit of 2.7×10^{-7} M [Chandra et. al. -2010]. Using glassy carbon working electrode DPV method was also developed for the determination of silymarin /vitamin-C acetate mixture in pharmaceuticals ascorbic acid with detection limit 0.03 mg/L for silymarin, and 0.01mg/L for vitamin-C [Hassan et. al. -2008]. A multi-walled carbon nanotubestetradecyl

trimethyl NH_4Br film coated graphite electrode was used to study the electrooxidation of ascorbic acid in differential pulse, cyclic and square-wave voltammetry (swv) [Motahary et. al. -2010]. A linear voltammetric response for vitamin-C was obtained within concentration range 5×10^{-7} - 1.7×10^{-4} M, with detection limit of 1.1×10^{-7} M, using DPV. Cyclic voltammetry and differential pulse voltammetry at a binuclear copper complex modified glassy carbon electrode were also carried to determine ascorbic acid and dopamine. Linear analytical curves were obtained in the ranges 2.0 - 120.0 μM for dopamine and 5.0 - 160 μM for ascorbic acid, using DPV. The detection limits were 1.4×10^{-6} M for dopamine and 2.8×10^{-6} M for ascorbic acid [Beitollahi et. al.-2009; Wang et. al. -2007].

The electrochemical oxidation and selective determination of ascorbic acid in pharmaceutical dosage forms and in some Rosa species was investigated by cyclic, differential pulse and square-wave voltammetry. The linear response was obtained in the range 3.52 - 176.1 $\mu\text{g/ml}$, with detection limit 0.88 $\mu\text{g/ml}$, using DPV and 0.52 $\mu\text{g/ml}$ using SWV techniques [Erdurak-Kili et. al. -2006]. The aim of the present work was to fabricate a new sensor using nano aluminum silicate as the modifier that may be employed as an electrode in present study of electrochemical behavior and quantification of AA levels in locally available pharmaceutical preparations. The accuracy of analyses was evaluated by comparing the amounts of introduced reference substance to the results of voltammetric determinations of real samples.

2. Experimental

Reagents and solutions:

The reagents and the chemicals used were; ascorbic acid (99.5% pure), Limcee, Beconzym-C forte tablets and capsules (local market), graphite powder (England), paraffin oil (India), Na_2HPO_4 (England), NaH_2PO_4 (India) NaOH (Spain), HCl (India) and aluminum silicate (USA). All the used chemicals were of analytical grade. One mM stock solution of ascorbic acid was prepared by dissolving 0.0176g of it in 0.1M phosphate buffer solution and the buffer prepared using double distilled water. The required concentrations of AA were prepared by diluting the stock solution with 0.1M solution of phosphate buffer. Phosphate buffer solutions of different pH within the range from 2.0 - 8.0, were prepared using mixture of NaH_2PO_4 and Na_2HPO_4 solution in double distilled water. The pH of the buffer solutions were adjusted by adding 0.1M HCl or/and 0.1M NaOH dropwise. Graphite powder and paraffin oil of high purity were used in the preparation of a carbon paste.

Apparatus:-

The voltammetric experiments were performed using BAS-50 W potentiostatic / galvanostatic analyzer coupled with Dell Pentium personal computer with conventional three electrode configuration consisting of aluminum silicate modified carbon paste working electrode, a silver-silver chloride reference electrode and a platinum wire serving as a counter electrode. One ml syringe was used for the preparation of the working electrode in the experiment. The pH of the buffer solution was measured with a bench

microprocessor pH meter (HANNA Instruments, Italy) in combination of glass electrode.

Preparation of working sensors:

The procedure deployed for the preparation of normal carbon paste electrode (UCPE) and the aluminum silicate modified carbon paste electrode (ASCPE) by using the components; 70 % (w/w) of graphite powder and 30 % (W/W) of paraffin oil for UCPE whereas for ASCPE, varying amounts of aluminum silicate used in prepared carbon paste, was reported earlier [Alemu et. al. -2015]. These pastes were then packed into electrode bodies, separately, consisting of plastic syringe equipped with copper wire serving as an electric contact.

Preparation of solution of vitamin-C tablets:-

Five tablets of vitamin-C of each brand were weighed and ground into a fine powder. An accurately weighed powder equivalent to 100 mg of the active component (for Limcee), 150mg (for Beconzym-C forte) and 150 mg of capsule were transferred into a 100 mL volumetric flask to dissolve in 0.1M phosphate solution and the mixture was shaken thoroughly till dissolved. Then, it was diluted up to the mark with phosphate buffer and aliquots of this solution were diluted appropriately to get the working concentrations range. Finally the percentage content or concentration of AA in these tablet samples and capsule were determined with the help of the calibration curve. The samples were then spiked with different amounts of known standard solutions of AA and the same standard curve was used for its recovery.

3. Results and Discussion

Electrochemical activity of ascorbic acid:-

The electrochemical behavior of AA was studied in various electrolytes such as Britton-Robinson (BRBS), phosphate (PBS) and acetate (ABS) buffer in the pH range of 2–8 by CV on the ASCPE (results not shown). The most excellent results with the uppermost magnitude, a little background current and good repeatability were obtained in PBS. Hence, it was chosen as suitable working medium for further studies in present work. Generally, AA is an electrochemically active substance which can transfer electrons from solution to the electrode. Its electrochemical characteristics at an electrode-analyte interface could be visualized from Figure-1 where the differential pulse voltammograms (DPVs) are displayed. Figure-1A presented no signal in its I - E curve within the studied potential window which indicated that the sensor and the supporting electrolyte are free from every type of interactions with each other and provide a suitable and peak interference free environment to investigate the electrochemical characteristic of AA at the surface of present sensor. Figure-1B has shown a wider signal of low oxidation current density i.e. 0.15 μA at the higher potential side (480mV) in the DPV which indicated that electro-oxidation of AA at CPE electrode requires a large over potential and associated with a poor catalytic effect of carbon paste on anodic oxidation of AA in 1.0 mM solution of phosphate buffer at pH 7 and scan rate 100 mV/s, hence a poor anodic peak observed. Figure-1C has exposed a strong, sharp and

clear signal of high oxidation current density i.e. 0.33 μA at approximate potential 321 mV.

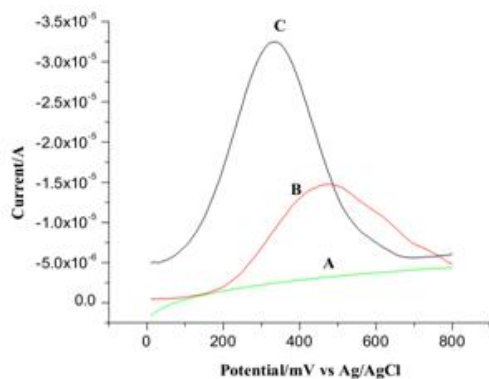
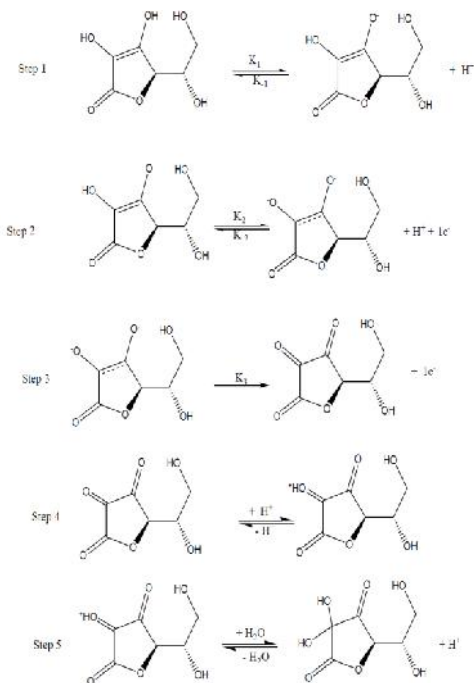


Figure 1: Differential pulse voltammograms of 0.1M phosphate buffer solution at pH 7.0 using ASCPE in absence of AA (A), UCPE in the presence of 1.0 mM AA (B) and ASCPE in the presence of 1.0 mM AA (C) at a scan rate of 100 mV/s and pulse amplitude of 100 mV.

The observed negative shift of peak potential and more than two fold enhancements in anodic current density revealed exceptionally well-built catalytic activity of nanoaluminum silicate towards anodic oxidation of AA and the electron transfer reaction rate accelerated effectively at the electrode- analyte interface as given in scheme-1.



Scheme-1: The proposed mechanism of AA electro oxidation in acidic/neutral solution.

The efficiency/ sensitivity for quantification of an analytes using electrochemical technique depend on the magnitude of parameters of the technique to be used. The systematic studies of various experimental and instrumental parameters that affect the adsorptive/ catalytic voltammogram response were carried out to establish the optimum conditions. So, the different constraints such as pulse-amplitude, scan-rate International Journal of Chemistry and Pharmaceutical Sciences

and pulse-period for using differential pulse voltammetry in present studies were optimized as follow.

Differential Pulse Amplitude

For the investigation of the influence of the pulse amplitude on the analytical signal, this parameter was varied between 10 and 100 mV at 200 ms pulse period and 100 mV/s potential scan rate.

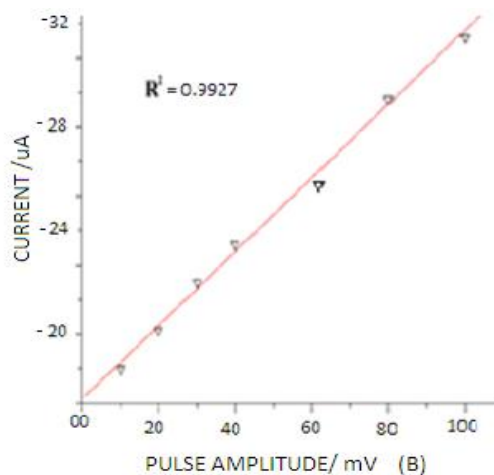
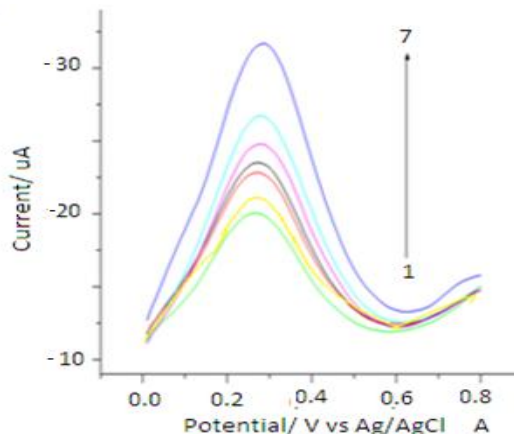


Figure 2: (A) Differential pulse voltammogram of 1.0 mM AA at ASCPE in 0.1M phosphate buffer, pH 7.0, at different pulse amplitude of (1) 10, (2) 20, (3) 30, (4) 40, (5)60, (6) 80, (7) 100 mV and (B) Plot of peak current from DPVs as a function of pulse amplitude.

It is evident from the results presented in Figure 2A that the magnitude of the measured oxidation current intensity increase with the increasing value of applied pulse amplitude. From this figure when anodic oxidation peak current at different pulse amplitude plotted as function of pulse (E), observed a linear behavior as in Figure2B and equation for the same given by:

$$I_p (\mu\text{A}) = 16.156 + 0.104 E (\text{mV}), \text{ with regression coefficient } (R^2) = 0.9927 \dots (i)$$

This activity posture a nearly uniform variation of current density with regularly varied pulse rate was the reflection of uniformly accelerated supply of analyte species at the interface during oxidation process. Pulses of higher amplitude were not considered because of the poor peak resolution that might be due to some intermediates' adsorptive behavior at interface. Thus, the optimum

magnitude 100 mV pulse was chosen for further studies during real sample analysis work in present study.

Effect of pulse period

The investigation of the influence of the pulse period on the analytical signal was displayed in Figure 3. The DPVs were obtained between 150 ms to 450 ms at pulse amplitude 100 mV using potential scan rate 100 mV s⁻¹. The enormities of continuous decrease of peak height and increase in peak width with flattened shape were observed with the increased magnitude of pulse period. This fact has directed that the influence of noise on the anodic oxidation signals elevated with increasing pulse period under the given conditions. So, in order to diminish the influence of noise on the detected analytical signal, lower value of the pulse period was maintained functional. Thus, the optimized parameter for further study was 200 ms in lower pulse period range.

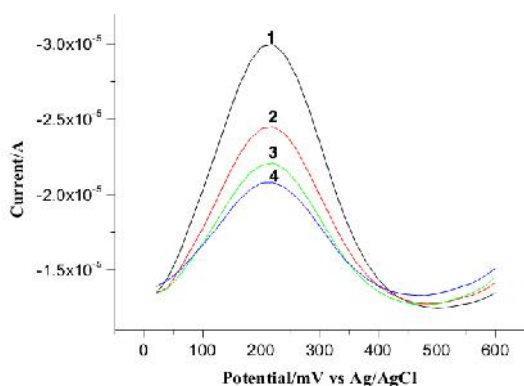


Figure 3: Influence of the pulse period on the analytical signal of AA at ASCPE; (1) 150 ms, (2) 250 ms, (3) 350 ms & (4) 450 ms; using scan rate 100mV/s and pulse amplitude 100mV.

Effect of scan rate

The influence of scan rate on the anodic peak current was investigated by using both techniques i.e. DPV & CV in range of 0–100mV/s. The voltammograms obtained through application of DPV are presented in Figure 4A and the anodic oxidation peak current values at different scan rates as function of scan rate as well as function of square root of scan rate have been portrayed in figures 4B & 4C respectively. For scan rates higher than 100mV/s, AA peaks were found wider and less stable, so the use of greater than 100mV/s scan rates were discarded. Hence, a scan rate 100 mV/s was deemed optimum and utilized for further investigations. Figure 4(B&C) indicated a strong linear increase of anodic peak current with increasing scan rate with highly comparable significant sensitivity of the regression equations. The linear regression equations along with their regression coefficients given as follow:

$$I_p (\mu\text{A}) = 23.78 + 0.104 \quad (\text{mV/s})$$

$$\text{and } R^2 = 0.9924 \quad \dots \text{(ii)}$$

$$I_p (\mu\text{A}) = 19.54 + 0.141 \quad v^{1/2} (\text{mV/s})$$

$$\text{and } R^2 = 0.9948 \quad \dots \text{(iii)}$$

Both characteristics, the peak current (I_{pa}) and peak potential (E_{pa}) of AA increase with increasing scan rate. The oxidation peak currents from these figures i.e. 4B & 4C and linear regression equations (ii) & (iii) suggests that the

electrode process at sensor/analyte interface follow both the surface controlled and the diffusion controlled mechanism during estimation of AA in samples. The electrode reaction was irreversible as depicted from the nonappearance of a reduction peak in the cyclic voltammograms in Figure 5.

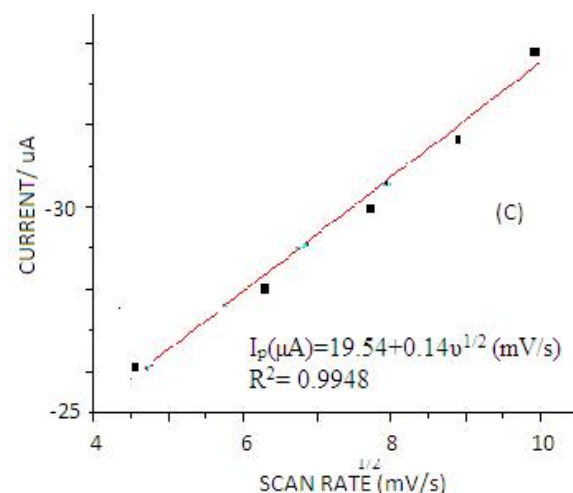
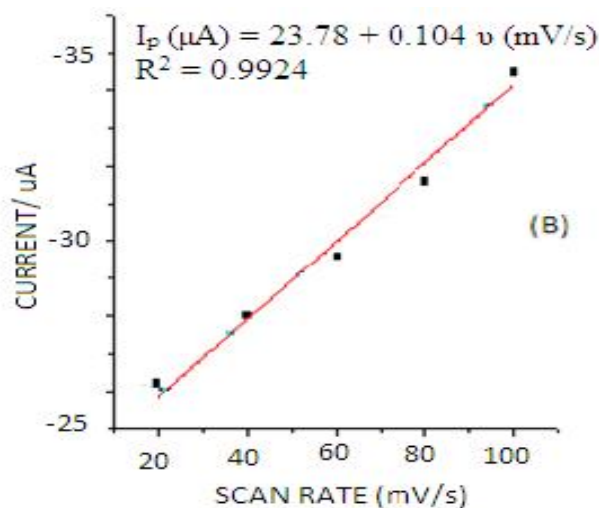
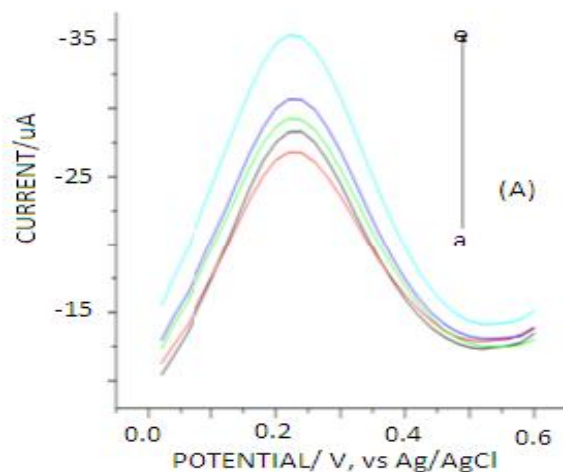


Figure4: (A) DPVs of 1.0 mM AA at ASCPE in 0.1 M PBS of pH 7.0 at scan rates: (a) 20; (b) 40; (c) 60; (d) 80 and (e) 100 mV/s using pulse amplitude of 100 mV. (B) Plot of peak current as a function of scan rate, and(C) Plot of peak current as a function of square root of scan rate.

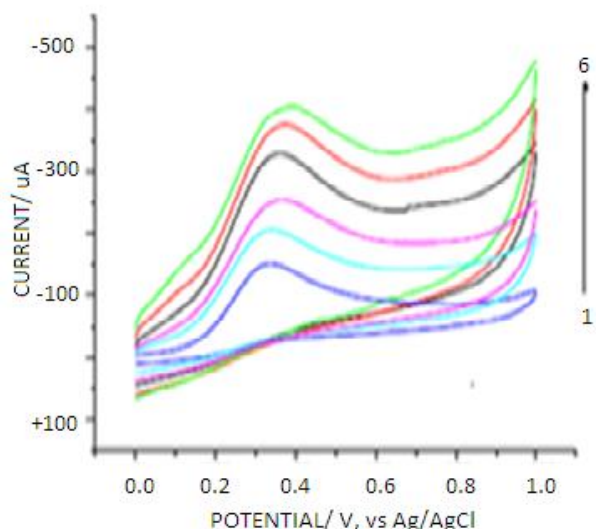


Figure 5: Cyclic voltammograms in the presence of 1.0 mM AA in 0.1 M phosphate buffer solution of pH 7.0 at scan rates (1) 10, (2) 20, (3) 30, (4) 40, (5) 60s and (6) 100mV/s.

The irreversibility of the electrode reaction could also be confirmed by the shift of peak potential (E_{pa}) to more positive values with increasing scan rate. The AA behavior as evidenced from Figures 4B & 4C at the interface of ASCPE, a surface-controlled process in the solution, is mainly diffusion controlled to some extent supported by adsorption which received a good support from literature [Bard & Faulkner -1980; West et. al. 2004]

Effect of AA concentration

The effect of sample’s concentration on peak current has been studied and presented in figure- 6A by using Randles-Sevcik equation used earlier [Erdurak-Kiliet.al.-2006] which correlates the peak current (I_p) with concentration (C) as:

$$I_p = kC + A \dots \dots \dots (iv)$$

Here ‘k’ is a constant which include the contribution of cell parameters like transfer coefficient, diffusion coefficient, electrode area, number of participating electrons in redox process at interface and A is an arbitrary constant. Once the magnitude of ‘k’ and ‘A’ obtained under given conditions, the quantity of AA in crude samples of unknown concentration-i.e. pharmaceutical product, natural edible/ consumable food and fruit juice articles-can be easily estimated by measuring the magnitude of I_p of the sample under investigation.

Because of high sensitivity and excellent resolution power of DPV method under optimized conditions using a potential window 0.0 to 0.8V, the practicability of the technique has been applied using the fabricated sensor for AA quantitative analysis. The dependence of the observed peak current upon concentration of AA has been studied and the peak current of AA at the surface of ASCPE has shown its linear dependence on the AA concentration as in Figure 6B.

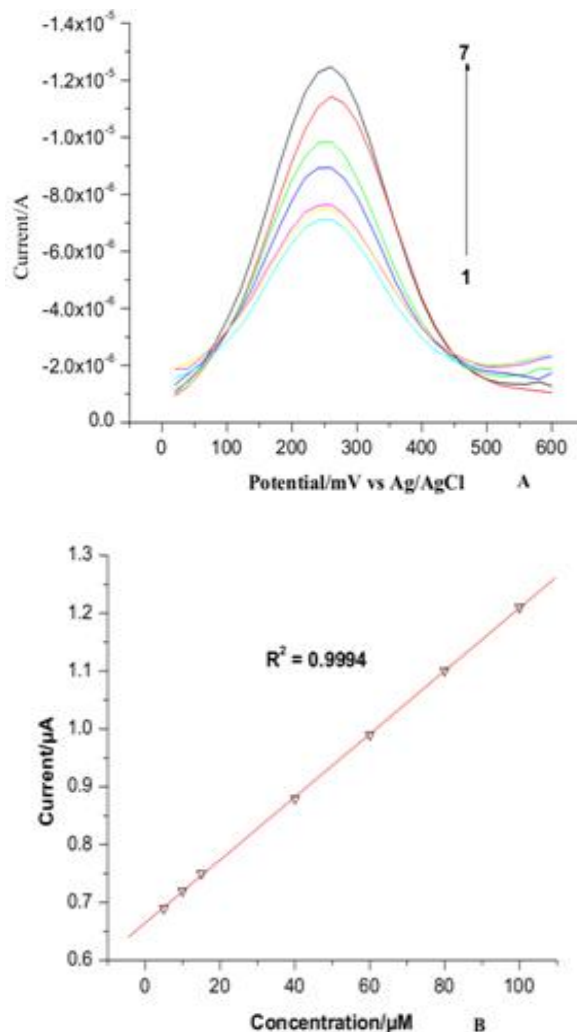


Figure 6: (A) DPVs of different AA concentrations (1) 5; (2) 10; (3) 15; (4) 40; (5) 60; (6) 80; and (7) 100 µM in 0.1 M PBS of pH 7.0 at a scan rate 100 mV/s and pulse amplitude 100 mV. (B) Analytical curve for quantification of AA.

Validation of Method

The validation of the method for the quantitative determination of AA in pharmaceutical products has been carried out after constructing a calibration curve within the concentration range 5.0 µM to 100.0 µM (Fig. 5B) and find out the regression equation as

$$I_p (\mu A) = 0.6647 + 0.054C_{AA}, \text{ with regression coefficient } (R^2) = 0.9994 \dots \dots \dots (v)$$

Where $I(\mu A)$ is the oxidation peak current of AA using ASCPE, C_{AA} is AA concentration and R is the correlation coefficient. The calculations of Limit of detection (LOD) as,

$$LOD = \frac{3s}{m} \dots \dots \dots (vi)$$

and limit of quantification (LOQ) as,

$$LOQ = \frac{10s}{m} \dots \dots \dots (vii).$$

Where ‘s’ is the standard deviation of the blank signals and ‘m’ is the slope of the analytical calibration curve and the calculated values were found to be 1.27µM for LOD and 5.73µM for LOQ.

Determination of AA in real samples

In order to demonstrate the capability of this sensor/modified electrode for catalytic oxidation of AA in real samples, we applied the voltammetric technique to quantify AA in some pharmaceutical preparations, such as tablets and vitamin-C capsule, obtained from India without interference from recipients and other drugs encountered. Therefore, the electrochemical behavior of ASCPE in 0.1 M phosphate buffer at pH 7.0 containing each pharmaceutical preparation by DPV has been carried out. Since, during experimentation the peaks were found sharp and well defined at low concentrations of AA, therefore, in order to fit into the linear range of the method, vitamin-C tablet and vitamin-C capsule engaged for present study, were accurately diluted with the supporting electrolyte. As shown in Table-1, the content of AA- in Limcee tablet was calculated to be 0.09656 g per tablet against the declared content 0.10 g per tablet, in Beconzylm C forte was calculated to be 0.14714 g per tablet against the declared content 0.15 g per tablet and in capsule was calculated to be 0.14635 g per capsule against the declared content 0.15 g per capsule. The determined contents and the declared contents of AA in all real samples are in excellent agreement with each other. Thus, the present studies demonstrate that the proposed method could be efficiently used for the determination of AA.

Table 1: Amount of AA detected from spiked and non-spiked samples of different brands of tablets and capsule by DPV using ASCPE.

Medicine	Amount of ascorbic acid (mg) ^a				Recovery (%)
	Label ed	Spiked	Expected	Obtained	
Limcee	100	0.0	100.0	96.6±0.1	96.56
		4.0	104.0	102.14±0.3	98.08
		10.0	110.0	107.73±0.2	97.94
Capsule	150	0.0	150.0	146.35±0.3	97.32
		7.0	157.0	152.21±0.5	96.95
		15.0	165.0	161.78±0.3	98.05
Beconzylm-C Forte	150	0.0	150.0	147.17±0.1	98.09
		7.0	157.0	153.92±0.3	98.04
		15.0	165.0	160.56±0.1	97.31

^aAverage of five replicates

Interference effect:

The electro-analytical determination of AA is often hampered by other electro-active species. In order to evaluate the selectivity of the method for AA, the influence of potentially interfering substances on the determination of analyte component was investigated. The most common of the interfering substances that can

be possible moiety with the pharmaceutical formulations of AA like benzoic acid, tartaric acid, fructose and glucose were taken for the study. The electrocatalytic peak current of AA Oxidation at ASMCPE was examined using differential pulse voltammetric technique. The forbearance limit of the interfering substances has been taken as the maximum concentration that have resulted nearly two percent relative error for 1.0 mM AA.

The experimental results revealed that the presence of these compounds have not significantly influenced the determination of AA under applied experimental conditions; because no significant change in current response was observed for any of the studied compounds. This suggests that the determination of AA in the pharmaceutical and biological samples at ASMCPE is not affected significantly by the common interfering species present along with molecules of interest. Therefore, this result demonstrated the selectivity of this method for the voltammetric determination of AA.

Table-2: Limit of tolerance of interfering substances on analysis of 1mM AA in 0.1MPBS at 7.0pH using scan rate 100mV/s and pulse amplitude 100mV.

S.No.	Substance	[Interference]/[AA]
1	Glucose	40
2	Fructose	120
3	Benzoic acid	100
4	L-+-tartaric acid	125

Comparison with literature:

This is evident from table-3 that the results of the present investigation are comparable in positive direction to that of the results recently reported in the literature using different techniques. Thus, the present method of analyzing the AA contents in pharmaceuticals formulations and other fields of daily use of humanity is suitable, easyoperable cost-effective and sensitive.

Table 3: Comparison of analytical data from present study to that with literature reported results.

Sensor/method	Linear range	LoD
ASCA/CPE/CV[Chandra et al-2010]	(1-10) 10 ⁻⁴	6.9x10 ⁻⁵
MWCNT/GC/DPV[Noroozifar et al-2012]	(0.56-1.2)10 ⁻⁵	1.2x10 ⁻⁶
SWCNT/WO3/CPE/CV [Koh et al-2012]	(0.2-1.0)10 ⁻⁴	8x10 ⁻⁵
EBT/CPE/DPV[Hassan et al-2008]	(1.0-9.0)10 ⁻⁵	2.7x10 ⁻⁷
Uv-spectroscopic tech. [Santos et al-2016]	(7.4-392.5)10 ⁻⁶	10.9x10 ⁻⁶
nAS/GPE/CV[This work]	(0.5-10)10 ⁻⁵	1.2x10 ⁻⁶

4. Conclusion

This study has developed a simple, fast, reproducible procedure for fabrication of aluminum silicate modified carbon electrode as an electrochemical sensor for detection and determination of ascorbic acid in pharmaceutical formulations both in crude samples as well as final products. The optimum experimental conditions for the oxidation of ascorbic acid were determined. It was found that the oxidation peak current of ascorbic acid was improved significantly and the oxidation peak potential shifted towards less positive value at the ASCPE as compared to that of bare carbon paste electrode. This suggests that ASCPE displays more excellent electrocatalytic property towards ascorbic acid oxidation. The obtained results showed that the present sensor has better analytical performance in sensing and in estimation of ascorbic acid up to $LOD=1.2 \times 10^{-6} M$, $LOQ=5.73 \times 10^{-6} M$.

5. References

- [1] Alemu M, Saini RC, Abraha T and Rishi Pal, A cyclic voltammetric study on the electrochemical behavior and endurance of pyridoxine at cobalt hexacyanoferrate based carbon paste sensor in local available vitamin B6 medications. *Int. J. Adv. Res.*, 3 (2015); 588.
- [2] Arya SP and Jain MM. Non-spectrophotometric methods for determination of vitamin-C. *Anal. Chim Acta.*, 417 (2000); 1.
- [3] Bard AJ and Faulkner LR. *Electrochemical methods, fundamentals and applications*, Wiley, New York, (1980); 243.
- [4] Beitollahi H, Ardakani M, Naeimi H and Ganjipour B. Electrochemical characterization of 2,2'-[1,2-ethanediylbis (nitriolethylidene)]-bis-hydroquinone carbon nano tube paste electrode and its application to simultaneous voltametric determination of ascorbic acid and uric acid. *J. Sol. St. Electrochem.* 13 (2009); 353.
- [5] Brusio J. How Is Ascorbic Acid Used in Food? Last Updated: Dec 26, (2015). <http://www.livestrong.com/article/491522-how-is-ascorbic-acid-used-in-food/>
- [6] Carmel J, Brian H., Terry N., Risa S. and Mark C. Is there a role for oral or intravenous ascorbate (Vit-C) in treating patients in cancer? A systematic review. *The Oncologist*, 20 (2015); 210.
- [7] Chandrashekar B N and Kumaraswamy B E. Electrocatalysis of SDS surfactant modified carbon paste electrode for the simultaneous determination of ascorbic Acid, norepinephrine and folic Acid. *Anal. Bioanal. Electrochem.*, 8 (2016); 345.
- [8] Chandra U, Kumaraswamy BE, Gilbert O and Sherigara BS. Determination of dopamine in presence of ascorbic acid at eriochrome black-T modified carbon paste electrode: A voltammetric study. *Int. J. Electrochem. Sci.*, 5 (2010); 1475.
- [9] De-Tullio MC. Beyond the antioxidant: the double life of vitamin-C. *Subcell Biochem. PubMed.*, 56 (2012); 49.
- [10] Dr. Humphries S. Vitamin-C cures disease. <https://healthimpactnews.com/2015/vitamin-c-cures-disease-but-doctors-and-pharmaceutical-companies-do-not-want-you-to-know-this/>
- [11] Erdurak-Kili CS, Uslu B, Dogan B, Ozgen U, Ozkan SA and Coskun M. Anodic voltammetric behavior of ascorbic acid and its selective determination in pharmaceutical dosageforms and some Rosa species of Turkey. *J. Anal. Chem.* 61 (2006); 1113.
- [12] Francioso L, Bjorklund R, Krantz-Rulcker T and Siciliano, P. Classification of multiple defect concentrations in white wine by platinum microelectrode voltammetry. *Actuator. B-Chem.* 125 (2007); 462.
- [13] Fritea L, Tertis M, Cristea C, Cosnier S and Sandulescu R. Simultaneous determination of ascorbic and uric acids in urine using an innovative electrochemical sensor based on β -Cyclodextrin. *Anal. Letts.*, 48 (2015); 89.
- [14] Gees B, Baets J, De-Jonghe P, Reilly MM, Pareyson D and Young P. Ascorbic acid for the treatment of Charcot-Marie-Tooth disease. *Cochrane Database Syst. Rev.*, (2015) Dec 11;(12):CD011952.
- [15] Hassan EM, Khamis EF, Eman I and Barary MA. Development of a differential pulse voltametric method for the determination of silymarin/vitamin-E acetate mixture in pharmaceuticals. *Talanta*. 74 (2008); 773.
- [16] Hossu AM and Magearu V. Determination of vitamin-C in pharmaceutical products with physicochemical and bio-analytical techniques: Review. *Roumn. Bio-tech. Lett.*, 9 (2004); 1497.
- [17] Koh SN, Wee TT, Zulkarnain T, Ruzniza BMZ and Zidan T. Electrochemical Oxidation of ascorbic acid mediated by single-walled carbon nanotube/ tungsten oxide nanoparticles modified glassy carbon electrode. *Int. J. Electrochem. Sci.*, 7 (2012); 4210.
- [18] Lewis RA, McDermott MP, Herrmann DN, Hoke A, Clawson LL, Siskind, Feely SM, Miller L J, Barohn RJ, Smith P, Luebbe E, Wu X and Shy ME; Muscle Study Group. High-dosage ascorbic acid treatment in charcot-marie-tooth disease type 1a: results of a randomized, double-masked, controlled trial. *Jama Neurol., Pub-Med.*, 70 (2013); 981.
- [19] Liu X, Peng Y, Qu X, Ai S, Han R and Zhu X. Multi-walled carbon nanotube-chitosan/poly (amidoamine) / DNA nanocomposite modified gold electrode for determination of dopamine and uric acid under coexistence of ascorbic acid. *J. Electroanal. Chem.*, 654 (2011); 72.
- [20] Motahary M, Ghoreishi SM, Behpour M and Golestaneh M. Electrochemical determination of ascorbic acid at the surface of a graphite electrode modified with multi-walled carbon nanotubes/tetradecyltrimethylammonium bromide. *J. Appl. Electrochem.* 40 (2010); 841.

- [21] Noroozifar M, Khorasani-Motlagh M and Tavakkoli H. Determination of ascorbic acid by a modified multiwall carbon nanotube paste electrode using cetrimonium iodide/iodine. *Turk. J. Chem.*, 36 (2012); 645.
- [22] Parviainen MT, Townsend A. (Ed.), *Encyclopedia of Analytical Science*. Academic Press, London. (1995); 9.
- [23] Pisoschi AM, Pop A, Negulescu GP and Pisoschi P. Determination of ascorbic acid contents in some fruit juices and wine by voltammetry performed at Pt and carbon paste electrodes. *Molecules*, 16 (2011): 1349.
- [24] Pournaghi-Azar MH and Ojani R. Attempt to incorporate ferrocene carboxylic acid into polypyrrole in chloroform: its application to the electrocatalytic oxidation of ascorbic acid. *J. Sol. St. Electrochem.*, 3 (1999); 392.
- [25] Pournaghi-Azar MH and Razmi-Nerbin H. Electrocatalytic characteristics of ascorbic acid oxidation at nickel plated aluminum electrodes modified with nickel pentacyanonitrosyl-ferrate films. *J. Electroanal. Chem.* 488 (2000); 17.
- [26] Raouf JB, Ojani R and Beitollahi H. Electrocatalytic determination of ascorbic acid at chemically modified carbon paste electrode with 2, 7-bis (Ferrocenyl ethynyl) fluoren-9-one. *Int. J. Electrochem. Sci.*, 2 (2007); 534.
- [27] Santos DA, Lima KP, Marçõ PH and Valderrama P. Vitamin- C determination by ultraviolet spectroscopy and Multi-product calibration. *J. Braz. Chem. Soc.*, 27(10), (2016); 1912-17.
- [28] Varvara M, Bozzo G, Celano G, Disanto C, Pagliarone CN and Celano GV. The use of ascorbic acid as food additive: *Tech. Legal Issues*, 5 (2016): 48.
- [29] Wang M, Xu X and Gao J. Voltammetric studies of a novel bicopper complex modified glassy carbon electrode for the simultaneous determination of dopamine and ascorbic acid. *J. Appl. Electrochem.* 37 (2007); 705.
- [30] West DM, Holler FJ and Grouch SR. *Fundamental of analytical chemistry*, 8th ed.; Thomson; Brooks/Cole: USA, (2004); 696.
- [31] Xu-li M, Shou-bin S, Zhong-de W, Yu-jiao Y, Xiao-gang H, Yang Z, Zhong-lin Z and Shi-bin L. Electrocatalytic oxidation of ascorbic acid on carbon nanotube /cubic nickel cyanoferrate/ polyaniline hybrid films. *New Carbon Materials*, 28 (2013); 26.