JIMMA UNIVERSITY

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DEPARTMENT OF CHEMISTRY

A THESIS ON:

ELECTROCHEMICAL DETERMINATION OF TANNIC ACID AT GLASSY CARBON ELECTRODE MODIFIED BY ANTIMONY FILM

A THESIS SUBMITTED TO SCHOOL OF GRADUATE STUDIES, JIMMA UNIVERSITY, DEPARTMENT OF CHEMISTRY, IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE DEGREE OF MASTERS OF SCIENCE IN PHYSICAL CHEMISTRY

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Abstract

In this study the electrochemical determination of tannic acid (TA) was performed using a glassy carbon electrode (GCE) modified with antimony film. The influences of deposition time, deposition potential, bath concentration and pH on the reduction current enhancement of TA were optimized, and electrochemical method for the determination of TA was reported. Linear sweep voltammetry (LSV) was used for electro-determination of TA. Using antimony film modified glassy carbon electrode (Sb-GCE), the reduction of TA was observed at the potential of −0.8 V vs. Ag/AgCl. Under optimum conditions, the result revealed two linear range regions between 0.5 μM to 2 μM and 5 μM to 40 μM of concentration. The limit of detection (LOD) was found to be 0.2 μM and 3.9 μM. The developed method was used for electroanalysis of TA in tea samples.

Key words: Tannic acid; antimony film modified glassy carbon electrode; electrochemical method; linear sweep voltammetry.

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List of Abbreviation

A	Amper
ASV	Anodic stripping Voltammetry
ASDPV	Anodic stripping differential pulse voltammetry
C	Concentration
CA	Chronoamperometry
E_a	Anodic peak potential
E_c	Cathodic peak potential
GCE	Glassy Carbon Electrode
I_c	Cathodic peak current
I	peak current
LOD	Limit of Detection
LSV	linear sweep voltammogram
mM	Mill Molar
Sb-GCE	Antimony Glassy Carbon Electrode
TA	Tannic Acid
µM	Micro Molar
V	Volt

1. Introduction

1.1. Background of the study

Tannic acid (TA), a water soluble polyphenol compound has been investigated for many years, especially to cure many diseases[1,2,3,4]. Tannic acid (TA) is a natural polyphenolic compound, and it's widely used in food and medicine industry. Due to its wide range of applications, analysis of TA is important not only in food but also in the medical and environmental fields. TA is present commonly in the human diet including fruits and different kinds of vegetables and can be found in several beverages, including wine, beer, coffee, black tea and white tea. It is used as a food additive as clarifying agent, flavor adjunct and flavoring agent[5] as well as additive in medical and veterinary fields due to its antimicrobial activity, anticarcinogenic and antimutagenic potentials and also antioxidant nature protect cellular oxidative damage[6,7]. Antioxidants may be defined as compounds that inhibit or delay the oxidation of other molecules by inhibiting the initiation or propagation of oxidizing chain reactions[8]. Antioxidants can also protect the human body from free radicals and have been widely used as food additives to provide protection against oxidative degradation of foods. The antioxidant activity of phenolic compounds is mainly attributed to their redox properties, which allow them to act as reducing agents, hydrogen donors and quenchers of singlet oxygen.

Phytochemicals are secondary metabolites in plants that play important roles in their metabolism, defense mechanisms, and disease resistance. They are also widely recognized for their bioactive and health-promoting properties. The content of TA in fruits, tea and beer can strongly influence their taste, and hence, is an important parameter to evaluate and control the quality of these products. TA may combine with metals to form tannic acid-metal complexes which have a toxic potential to the root system of plants[9]. For these reasons, the determination of tannic acid has been of great importance. Several methods including spectrophotometry[10], colorimetry[11], liquid chromatography[12] and chemiluminescence[13,14,15] have been applied for the determination of TA content in the waters, pharmaceuticals and foods. Each method has its advantages and drawbacks. For example, chromatographic methods allow the determination of TA, but they are time consuming and expensive. However, these methods are costly, require highly skilled manpower, complicated and time consuming procedures which is difficult to use for TA analysis. Compared to these methods, the electrochemical method has several advantages such as its low cost, the options of analysis without extraction or preconcentration, highly selective and

sensitive and the short time required for analysis. But, only very few reports were accessible in the literature for the electrochemical determination of TA.

The main advantages of electrochemical methods with respect to traditional, more laborious instrumental techniques are described: sensitivity, rapidity, simplicity of the applied analytical procedure which does not require complicated sample pre-treatment etc. For the method presented, the electroactivity and the mechanism of electro oxidation of antioxidant molecules at various electrodes, as well as the influences on the electroactive properties are discussed. Oxidative stress was defined as the organism's status involving cell damage, by enhanced release of radical or non-radical oxygenated species[16]. During cellular respiration, the transfer of unpaired single electrons towards molecular oxygen may occur resulting in free radical generation[17]. Reactive oxygen species may also result in some cell-mediated immune functions[18]. These may be represented by either very unstable radicals containing a minimum of one unpaired electron, or by oxidizing non-radical species, that can promote the peroxidation of membrane lipids with accumulation of lipid peroxides[19,20].

Antioxidants represent a wide class of chemical compounds that fight against the oxidative processes, including the degradation of nutrients found in diet, of materials such as rubber or plastic, of essential molecules found in biological media, etc[21]. Newly, with the continuously growing preoccupation for distinguishing secure food antioxidants, the choice is represented by natural antioxidants, especially plant sourced.

The main biocompounds found in natural sources are phenolics (flavonoids or non-flavonoids), associated to health benefits. With respect to the assessment of the antioxidant ability in foodstuffs, the terminology of "antioxidant activity" or"antioxidant capacity" is employed, although, different meanings are attributed to these terms: the activity is correlated to the rate constant of the reaction of an individual antioxidant with a certain oxidant. Antioxidants are able to deactivate the free radicals and to protect the organism from oxidative damage. The capacity is defined as the amount (expressed as number of moles) of a free radical that can be scavenged by a sample[22].

In electrochemical antioxidant analysis, a glassy carbon electrode has most often been used. However, the electrode surface area is a limiting factor in terms of its sensitivity and capacity to respond to the full polyphenol content. To reduce this factor the Antimony Modified electrodes

films have been studied in order to improve the sensitivity and capacity of electrodes, and conducting polymers have been found to be useful in this regard. Glassy carbon electrode (GCE) is a useful electrode material because of its high electrical conductivity, impermeability to gases, high chemical resistance, reasonable mechanical and dimensional stability and widest potential range of all carbonaceous electrodes[23].

Since foods and beverages have numerous phenolics compounds, including many unidentified compounds, it is not easy to characterize the complete phenolic content by a single method. It was reported that the polyphenolic nature of tannic acid, it's relatively hydrophobic ''core'' and hydrophilic ''shell'' are the features responsible for its antioxidant action[24]. Tannins which refer to tannic acid containing drugs precipitate proteins for the protection of inflamed surfaces of skin and treatment of burns that results for it's used as antioxidant. It prevents cancer by preventing cellular damage and can also be effective in protecting the kidneys. In economic point of view it is used in the manufacture of inks. In this work, the use of glassy carboelectrode modified antimony films as a sensitive electrochemical electrode was established for the determination of TA using voltammetry.

1.2. Statement of the problems

Tannic acid is one of a natural water soluble polyphenolyic compound that is found in some food samples such as white wine, fruit juice, beer and black tea. TA is antioxidants that delay or inhibit cellular damage mainly through their free radical scavenging property. It is considered as antioxidant clarifying agent, and flavoring agent as well as additive in medical and veterinary fields due to its antimicrobial which is able to deactivate free radicals to protect cell damage of human beings[25]. It is used as clarifying agent, anti-carcinogenic and anti-mutagenic potentials and also antioxidant nature protect cellular oxidative damage. Therefore, sensitive analytical methods for the routine determination of TA in samples are highly desired.

Therefore this study may answer for the following questions:

- Is Sb-GCE has higher sensitivity for determination of TA?
- Is bare GCE has higher sensitivity for determination of TA?
- What will be the responsible reason for having different response of electrodes?

1.3. Objectives of the study

1.3.1. General Objectives

To develop simple and sensitive electrochemical method for the analysis of tannic acid.

1.3.2. Specific Objectives

- To optimize parameters for GCE modification with antimony film.
- To optimize parameters for sensitive determination of tannic acid at Sb- GCE.
- To determine tannic acid under optimized condition at Sb-GCE.
- To demonstrate the application in real sample of the developed method.

1.4. Significance of the study

The results of this study could contribute to introducing knowledge and practice about the electrochemical methods for determination of TA Sb-GCE. TA is a natural polyphenolic compound and widely used in food and medicine industry. It can be used as clarifying agent in the brewing and wine industry, and as flavoring agent in baked foods, candy and meat products[26]. It also used as well as additive in medical and veterinary fields due to its antimicrobial activity, anticarcinogenic and antimutagenic potentials and also antioxidant nature protect cellular oxidative damage.

1.5. Scope of the study

This investigation was conducted to determine tannic acid by glassy carbon electrode modified with antimony film. The proposed electrochemical method was used as simple and sensitive analysis for tannic acid at antimony glassy carbon electrode. The optimum conditions were selected based on response to the intensity of tannic acid reduction current during fabrication of antimony film deposition on glassy carbon electrode. Those are;

- Deposition potential
- Deposition time
- Bath concentration
- pH of the Supporting Electrolyte

2. Review of Related Literature

2.1. Tannin Chemistry

The name ‘tannin’ is derived from the French ‘tanin’ (tanning substance) and is used for a range of natural polyphenols[27]. Tannins are commonly defined as water-soluble polyphenolic compounds ranging in molecular weight from 500 to 3000 Daltons that have the ability to precipitate proteins[28]. It is their ability to precipitate proteins, astringency, which is thought to be the primary effect of tannins on biogeochemical cycling. Tannins, the relatively high molecular weight compounds found in complexes with alkaloids, polysaccharides and proteins, are a group of water-soluble polyphenols. They may be subdivided into hydrolysable and condensedtannins. The hydrolysable tannins are esters of gallic acid (gallo- and ellagi-tannins), while the condensed tannins (also known as proanthocyanidins) are polymers of polyhydroxyflavan-3-ol monomers. A third subdivision, the phlorotannins consisting entirely of phloroglucinol, has been isolated from several genera of brown algae. They are found grape (dark/light) seed/skin, apple juice, strawberries, raspberries, pomegranate, walnuts, muscadine grape, peach, blackberry, olive, plum, chick pea, black-eyed peas, lentils, haricot bean, red/white wine, cocoa, chocolate, tea, cider, coffee, immature fruits are the main sources of tannins[29-34].

2.2. Phenolic acids

Phenolic acids are also known as hydroxybenzoates, the principal component being gallic acid. The name derives from the French word *galle*, which means a swelling in the tissue of a plant after an attack by parasitic insects. The swelling is from a buildup of carbohydrate and other nutrients that support the growth of the insect larvae. It has been reported that the phenolic composition of the gall consists of up to 70% gallic acid esters[35]. Gallic acid is the base unit of gallotannins whereas gallic acid and hexahydroxydiphenoyl moieties are both subunits of the ellagitannins. Gallotannins and ellagitannins are referred to as hydrolysable tannins, and, as their name suggests, they are readily hydrolysed by treatment with dilute acid whereas condensed tannins are not. Condensed tannins and hydrolysable tannins are capable of binding to and precipitating the collagen proteins in animal hides. This changes the hide into leather making it resistant to putrefaction. Plant-derived tannins have, therefore, formed the basis of the tanning industry for many years. Tannins bind to salivary proteins, producing a taste which humans recognize as astringency. Mild astringency enhances the taste and texture of a number of foods

and beverages, most notably tea and red wines. Clifford (1997) has reviewed the substances responsible for and the mechanisms of astringency[36].

2.3. Antioxidants in Beverages

Antioxidants are the substances that delay or prevent the oxidation of oxidizable substrates[37]. They are often molecules that are reducing agents and interact readily with free radicals. The most abundant antioxidant molecules in diet are phenolic compounds[38]. Phenolic compounds are very effective antioxidant molecules that is due to their structures and contains more hydroxyl groups bonded directly to an aromatic ring[39]. The antioxidant activity of a phenolic compound is dependent upon its chemical structure. While phenol itself has no antioxidant activity, ortho and para diphenolics show marked activity. It has been found that flavonoids have quite high antioxidant activity. Antioxidant compounds also play an important role in the maintenance of food quality and food shelf-life[40]. For this reason, they are also added to foods and beverages.

2.4. Medicinal and Biological Properties

Tannins containing drugs precipitate proteins for the protection of inflamed surfaces of skin and treatment of burns[41]. These are act as anti-diarrheals and anti-oxidant effect, anti-viral, anti-bacterial, anti-inflammatory and anti-parasitic effects. It prevents cancer by preventing cellular damage. It can also be effective in protecting the kidneys. Tannins have been used for immediate relief of dysentery, skin ulcers[42], sore throats, fatigue, hemorrhaging and diarrhea.

2.5. Economic Properties

It is used in the manufacture of inks. These are used in the laboratory as astringents for the detection of gelatin, proteins and alkaloids[43]. Tannins are used in the tanning process of animal hides to convert them into leather. It's used in oils, dyes, fibers, glues, waxes, perfumes, drugs and flavoring agents. Various tannins are producing different colors with ferric chloride like black, blue, green. These are used as a tanning agent in dying industries and putrefying agents in leather industries[44].

2.6. Antioxidants Determination Methods

Recently, special interest has been developed on the application of electrochemical methods to antioxidant and antioxidant capacity determination, as they have the advantage of sensitivity, fastness, of requiring simple and relatively unexpensive instrumentation, small volumes of samples, with improvement of research resources use. The measured signal is independent on the distance that radiation travels in the analytical cell or on the turbidity, and the dynamic range is quite extended. One aspect in favor of the electrochemical methods is the electro activity of most antioxidants, the electron transfer being involved in antioxidant – free radical reactions, enabling a prompt screening of the antioxidant capacity of a series of organic biocompounds even in complex or colored samples[45,46,47].

2.7. Voltammetry

Voltammetry as potentiodynamic assay is based on the recording of the current intensity at controlled potential variation and exploits the reducing ability of antioxidants or the reversibility of redox active substances which are either single or part of a sample[48]. The confirmed analytical advantages of the various voltammetric techniques are: enhanced sensitivity, a broad concentration range (10–12 to 10–1 M), numerous appliable solvents and electrolytes, wide working temperature ranges, fast analysis, simultaneous determination of analytes (organic and inorganic), the capacity to assess kinetic and mechanistic parameters (including reasonable estimation of unknown ones), the facility of different potential waveforms generation yielding low current intensity values, these features being sustained by a strongly developed theoretical background[49].

2.7.1. Linear Sweep Voltammetry (LSV)

LSV is the simplest voltammetric technique. In LSV the current at a working electrode is measured while the potential between the working electrode and a reference electrode is scanned from a lower limit to an upper limit linearly in time. Oxidation or reduction of species is registered as a peak or trough in the current signal at the potential at which the species begins to be oxidized or reduced.[50]

2.7.2. Cyclic Voltammetry (CV)

CV has become an important and widely used electrochemical technique in many areas of chemistry. In a cyclic CV experiment, the working electrode potential is scanned linearly *vs.* time. Unlike in LSV, after the set potential is reached in a CV experiment, the working electrode's potential is scanned in the opposite direction to return to the initial potential. These cycles of ramps in potential may be repeated as many times as needed. The current at the working electrode is plotted versus the applied voltage to give the cyclic voltammogram. CV is generally used to study the electrochemical properties of an analyte in solution.[51-54]

The rate of voltage change over time during each of these phases is known as the scan rate (V/s). The potential is measured between the working electrode and the reference electrode, while the current is measured between the working electrode and the auxiliary/counter electrode. These data are plotted as current (i) vs. applied potential (E), known as cyclic voltammogram. During the potential sweep in CV experiments, it can be observed that the current starts to increase at a potential where the energy onset at which the electrochemical reaction takes place. As the potential moves from this potential, a continuous depletion of electroactive species near the electrode surface occurs, reaching a point at the peak potential (E_p) in which the electrochemically reactive species has been completely reduced or oxidized. Beyond this potential value, the current response decreases, this shows the mass transport control of the electrochemical reaction. On the other hand during the reverse potential sweep, the same phenomenon but opposite reaction will be happened, *i.e.* the product of the initial oxidation or reduction of the electroactive species is complementary reduced or oxidized. The more reversible the redox couple is, the more similar the oxidation peak will be in shape to the reduction peak. Hence, CV data can provide information about redox potentials and electrochemical reaction rates.[55-56]

3. Materials and Methods

3.1. Materials

3.1.1. Instruments

A three electrode system consisting of the modified GCE (3.0 mm diameter) as the working electrode, a platinum-wire as a counter electrode, and Ag/AgCl electrode as a reference electrode was used for electrochemical measurement. Electrochemical techniques including CV, CA and LSV were performed using Epsilion EC-Ver 1.40.67 voltammetric analyzer (Bioanalytical Systems, USA). All experiments were conducted at room temperature. pH meter (pH -016 pH METER, China), analytical balance (WANT Balance Instrument Co.,Ltd, China) were also used.

3.1.2. Chemicals

Tannic acid, Antimony chloride (($SbCl_3$), Riedel-de Haen), Sodium Acetate ($NaCH_3COO$) (Fine, 99%), Acetic acid (CH_3COOH) (LOBA CHEMIE, 99.5%), Hydrochloric acid (HCl), (Riedel-de Haen, 37%) were used. All chemicals were of analytical grade.

3.2. Methods

3.2.1. Solution preparation

Sb deposition bath consisting of 10 mM $SbCl_3$ in 0.1 M HCl was prepared. A stock solution of 5 mM TA was prepared by dissolving 0.8 gm of TA in double distilled water. From this stock solution of TA other working solution with the desired concentration was prepared by diluting in acetate buffer solution with pH 3.6.

3.3. Electrode Cleaning and Conditioning

A GCE 3 mm in diameter was polished with slurry of alumina then sonicated and rinsed with double distilled water. GCE was conditioned by potential scanning from -1.2 V to 1.2 V in 0.1 M HCl for five complete scans at 100 mVs^{-1}. The prepared electrode was used immediately after mechanical polishing and electrochemical cleaning.

3.4. Preparation of Antimony film modified glassy carbon electrode (Sb-GCE)

Sb-GCE were prepared by chronoamperometric deposition of antimony on GCE in an Sb(III) bath. Briefly, a constant deposition potential of –700 mV for 240 s was applied to the GCE in an antimony deposition bath (5 mM $SbCl_3$ in 0.1 M HCl). The modified GCE was rinsed with distilled water and immediately used for electrochemical studies of TA[57].

3.5. Optimization of Experimental Parameters

Parameters for the modification of the GCE with antimony were studied using linear sweep voltammetry (LSV) and chronoamperometry (CA) methods based on electrochemical response of TA at the modified electrode. Plating solution containing Sb (III) in acidic media was used to avoid hydrolysis of the metal[58].

Initial concentration of the deposition bath, the deposition potential and deposition time for the electrodeposition of antimony films and pH of TA solution were optimized. The optimum conditions were selected based on response to the intensity of TA reduction current.

a) Optimization of deposition potential

The Sb film deposition potential was optimized within the potential range from –400 mV to –900 mV deposited by the CA of, 5 mM of Sb (III) at the GCE. After deposition of the Sb film the selected potentials within this range for 240 s, was used for the modified GCE that used to measure the reduction peak for 1 µM TA (n = 3). The deposition potential that gave the highest reduction peak current was selected.

b) Optimization of deposition time

The effect of deposition time was studied by varying the time of the applied potential for 60 – 360 s. Using the prepared Sb-GCE, the current response for 1 µM TA (n = 3) was recorded. The appropriate deposition time was selected based on the response for the analyte.

c) Optimization of bath concentration

Antimony deposition bath concentrations that in the range from 1 mM to 8 mM Sb (III) at −700 mV deposition potential for 240 s was studied. For each concentration tested, triplicate measurement was made for the reduction of the 1 µM TA at the Sb film modified electrodes and

the average current obtained from different deposition baths was compared to select the appropriate deposition bath concentration.

d) Optimization of pH

For pH optimization, the LSV of 1 μM TA was run at Sb film modified GCE (obtained from deposition bath of 5 mM, Sb at –700 mV for 240 s), using the optimized deposition potential, deposition time and concentration. The resulting current signal (average of triplicate measurement) of the TA was plotted against the pH of the acetate buffer solution from 3-6. The pH which gave the optimum peak current was selected.

3.6. Study of Electrode reproducibility, stability and effects of Interferences

The reproducibility of the antimony film modified GCEs was studied by measuring the same concentration 1 μM of TA with preparing three electrodes under the same modification conditions that was on three consecutive days with triplicate measurement on each day. The relative standard deviation (RSD) of the measured reduction peak current was calculated to determine reproducibility of the modification strategy of the modified Sb-GCE.

Three Sb-GCE were modified with antimony film under the optimized conditions on the same day. The LSV of 1 μM TA was measured on the second day of the experiment and seventh day. On each day, three measurements were made at the modified electrode. The average peak current signals of the three measurements in the second day were compared to those of the seventh day to determine the stability of the modification procedure developed.

The applicability of the developed method of the selectivity of Sb-GCE for the determination of TA, the effect of possible interfering substances was studied under the optimum conditions. Different interfering species like ascorbic acid, uric acid, glucose, folic acid, and vitamin B2, K^+, Na^+, Cl^+ and NO_3^-were added into the solution containing 1 μM of TA.

3.7. Real Sample Analysis

The solution sample of tea was prepared by adding deionized water to 1.5 g of black tea. The black tea was extracted by 100 mL hot water for 10 minutes and the extract was filtered. The filtered sample was transferred to a 250-ml volumetric flask, and diluted with deionized water until the mark was reached[59]. 20 μL of the diluted extract was diluted in 10 mL acetate buffer and directly analyzed by proposed method. Furthermore, three different solutions were prepared

by mixing 20 µL of the diluted extract with 1.00, 5.00 and 15.00 µM of standard TA solutions in 10 mL 0.1 M acetate buffer (pH 3.6) and the % recovery was calculated. The determination of TA in the tea extract was carried out according to the linear regression equation formulated for the calibration curve.

4. Results and Discussion

4.1. Cyclic Voltammetry of Antimony (III) Ion

In order to determine the redox electrochemical signal, cyclic voltammogram of antimony were examined on GCE. As shown in Figure.1, the reduction potential of antimony obtained at −350 mV. This is very important to determine the deposition potential required for antimony film fabrication. Antimony film can be deposited on glassy carbon electrode by adjusting the reduction potential around the maximum reduction peak observed.

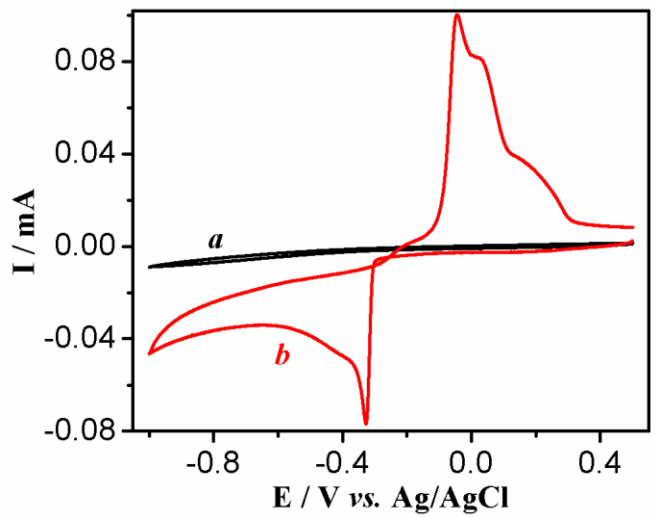

Figure 1. Cyclic voltammogram of (a) 0.1 M HCl and (b) antimony on glassy carbon electrode.

4.2. Electrochemical Behavior of TA at bare GCE and Sb-GCG

The electrochemical behaviour of 1 µM TA was studied using LSV in acetate buffer solution (0.1 M, pH 3.6) at bare GCE and Sb-GCE. Figure 2, shows the linear sweep voltammograms of TA using bare GCE and Sb-GCE.

As shown in Figure 2, the reduction peak of TA was observed at about -0.8 V at Sb-GCE and at about -0.7 V for bare GCE. Comparing the two results, there was excellent improvement in the reduction currents when Sb-GCE was used. The response in the LSV during the experiment revealed that, in case of bare GCE, the reduction peak current was observed about -0.116 mA while, in the case of the Sb-GCE, the reduction peak was observed at 7.8 mA.

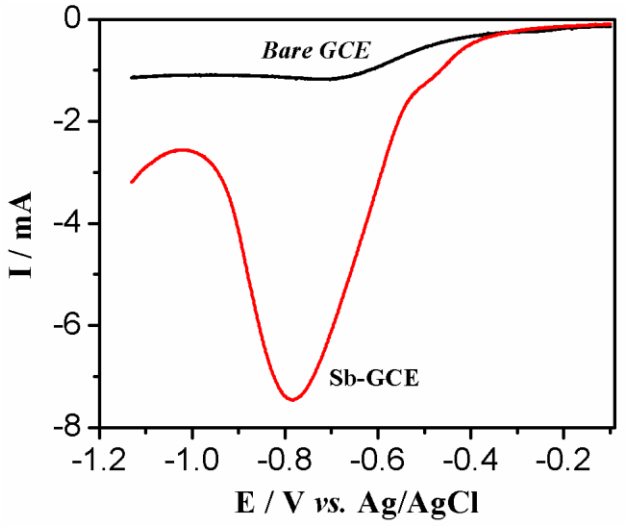

Figure 2. Linear sweep voltammograms of 1 µM TA at bare GCE and Sb-GCE in 0.1 M acetate buffer (pH=3.6); scan rate of 100 mV s^{-1}.

4.3. Optimization parameters for determination of TA

4.3.1. Optimization of deposition potential

The production of thin film by electrochemical deposition for analysis of various analytes is usually done by forced chemical reaction. The deposition proceeds by applying an external potential. So applied potential (deposition potential) has superposition to control the thickness of film during fabrication.

For this study, the optimum potential for antimony film deposition on the GCE from the plating solution containing 5 mM Sb (III) in 0.1 M HCl was studied within the potential range of –450 to –900 mV and shown in Figure 3. The optimum deposition potential was selected by comparing the modified electrode response towards 1 µM TA. The deposition potential of –700 mV gave the highest reduction current and it was selected as optimum deposition potential for further analysis of TA.

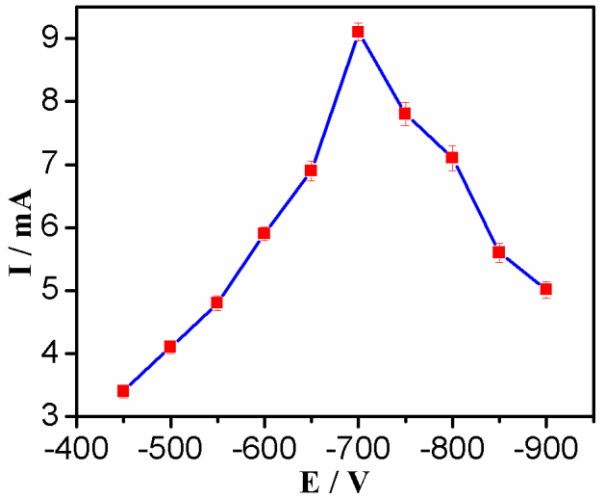

Figure 3. Effect of deposition potential upon LSV response for 1 µM TA. Experiment conditions: 5 mM Sb (III) and deposition time 240 s, each data represent an average of n=3 measurements

4.3.2. Optimization of deposition of time

The amount of metal particles on the surface of the electrode has great effect on the performance of the modified electrode. Therefore, the amount of metal particles on the electrode surface depends on the deposition time. So that, the deposition time should be optimized. The effect of the antimony plating time was studied in the range 60–420 s as shown in Figure 4.

The reduction peak current of TA increases with increasing antimony deposition time up to 240 s and remains constant beyond this deposition time which suggest that the deposited layer of antimony may have covered the surface of the electrode and substantial increment in film thickness prevented as a result of depletion of Sb^{3+} at electrode–electrolyte interface[60].

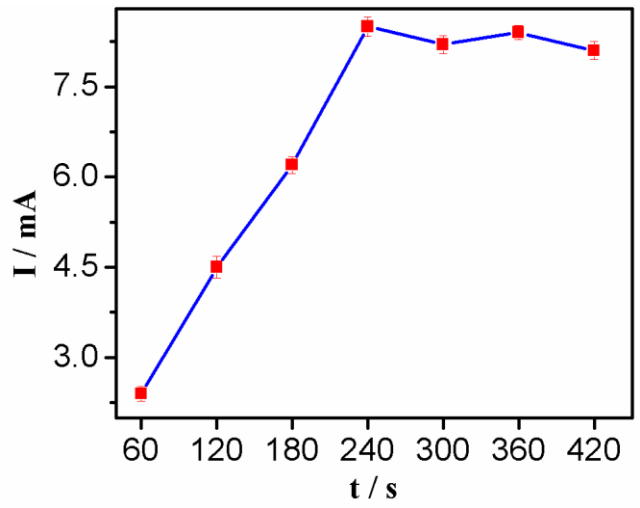

Figure 4. Effect of deposition time upon LSV response for 1 µM TA. Experimental conditions: 5 mM Sb (III) and deposition at -700 mV. Each data represent an average of n=3 measurements.

4.3.3. Optimization of bath concentration

The other important parameter that can influence the thickness of antimony film is the antimony ion concentration on the reduction peak current of the TA. The effect of concentration of antimony ion was studied in the range of 1 mM to 8 mM Sb (III) at −700 mV deposition potential for 240 s. As shown in Figure 5, the reduction peak currents of 1 µM TA in acetate buffer pH 3.6 increases with increasing the concentration of antimony solution from up to 5 mM but further increase in concentration of antimony the reduction current of TA decreases. The increase in peak current for the increase in antimony ion concentration up to 5 mM is attributed to the increase in the antimony film thickness. On the other hand, when the concentration of Sb (III) ion becomes higher, reduction peak currents decreased with increasing antimony film thickness due to cracking of the film at higher concentration. Hence, this result is in agreement with result reported for the optimization of bath concentration[61].

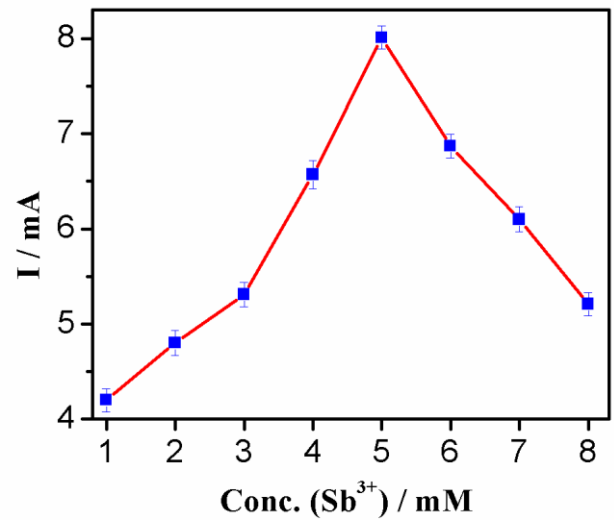

Figure 5. Effect of bath concentration upon LSV response for 1 µM TA. Experimental condition: deposition potential –700 mV and deposition time 240 s.

4.3.4. Optimization of pH of the Supporting Electrolyte

pH of the supporting electrolyte is an important parameter that could have significant influence on the response of the electrode in electroanalysis. It was studied in the pH range of 3 to 6 in acetate buffer. The results obtained are shown in Figure 6 in which the maximum response was observed at pH 3.6.

Since TA is a weak acid and the pH of aqueous solution is an important factor for its ionization and adsorption. Below pH 4.5, TA present in molecular form and shows approximate zero surface charge. As pH of supporting electrolyte increases, TA starting to dissociate at higher pH and it's highly dissociated in ions[62]. It is related to the proton taking part in the electrochemical reaction. In basic solutions the reaction becomes difficult because of the short of proton[63].

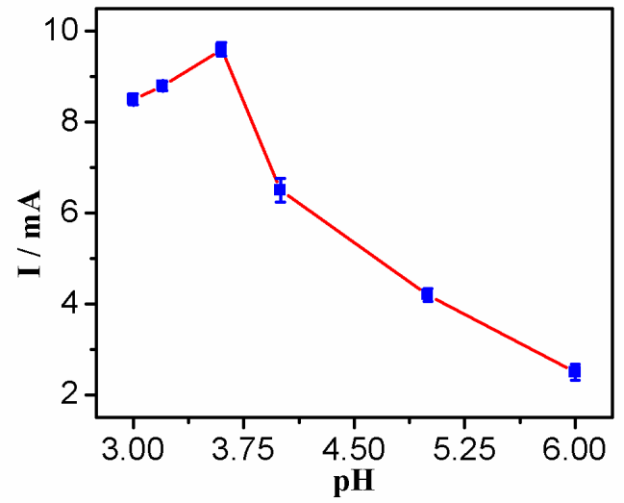

Figure 6. Effect of pH of the supporting electrolyte upon LSV response for a solution containing 1 µM TA. Electrode modification conditions: 5 mM Sb (III) bath, deposition time of 240 s and deposition potential at -700 mV. Each data point represent an average of n=3 measurements

4.4. The effect of the scan rate

The effect of scan rate on the electrochemical behavior of TA was also investigated at different scan rates. LSVs corresponding to TA reduction were recorded at various scan rates ranging from 10 to 300 mV/s using 1 µM TA in 0.1 M acetate buffer (pH 3.6) at Sb-GCE and are shown in Figure 7.

The cathodic peak currents increase with increasing scan rates and are linearly correlated to the scan rates with a high correlation coefficient of 0.990 (inset of Figure 7), implies that the electrochemical process is surface controlled (adsorption) process.

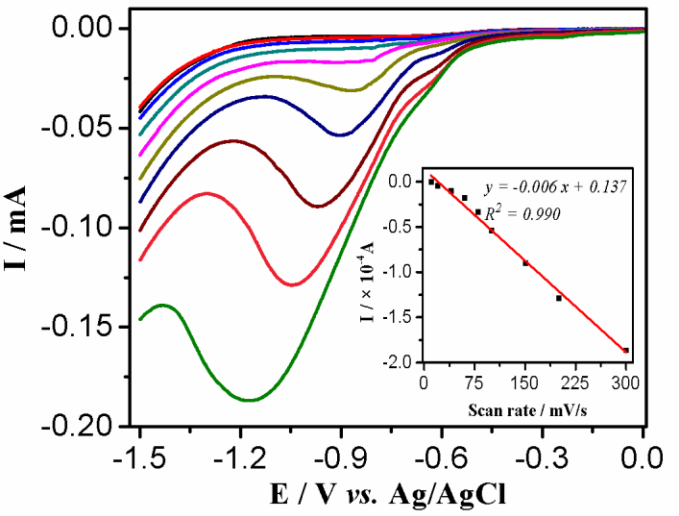

Figure 7: LSVs of 1 μM TA in 0.1 M acetate buffer (pH 3.6) as a function of scan rates ranging from 10 to 300 mV/s (Inset: calibration plot of cathodic peak current vs. scan rate)

4.5. Calibration curve

The Sb-GCE exhibited a well-defined peak with reproducible peak current values for repetitive measurements; and showed a decrease in peak current value with an increase in TA concentration. The result revealed two linear range regions between 0.5 μM to 2 μM and 5 μM to 40 μM.

The obtained linear regression equation for TA was i(mA) = –3.48 C(μM) + 11.49 (R^2 = 0.995) in the concentration range of 0.5 μM – 2 μM and i(mA) = –0.081 C(μM) + 4.36 (R^2 = 0.989) in the concentration range of 5 μM – 40 μM. The detection limit (LOD= 3Sb/m) was found to be 0.2 μM and 3.9 μM respectively.

The reason for the reduction of the peak current with increase in TA concentration is studied. The possible assumption could be either the formation of Sb-TA complex at the surface of the electrode which could result in a competitive surface adsorption or the formation of an electrochemical non-active complex between Sb-TA as a possible reason for the decrease in peak current. The experimental result in section 4.4 above shows electrochemical process is surface controlled process.

Most tannins have several *o*-diphenol groups and are thus capable of forming chelates with many metals. Furthermore, multicatecholate nature of tannins that allows reticulation often favors the formation of metal-tannin precipitates. Different metals and heavy metal ions are reported to complex with TA by adsorption. Because TA consists of different reactive functional groups and binding sites which can bind metals[64-72]. B. H. Cruz *et. al.* reported that the addition of TA to Zn^{2+} produces the simultaneous decrease of differential pulse polarogram peak current and suggests the formation of a labile macromolecule complex.[64] Therefore, it might be competitive surface adsorption or a formation non electroactive complex is a possible explanation for the observed decrease in reduction peak current as a function of TA concentration.

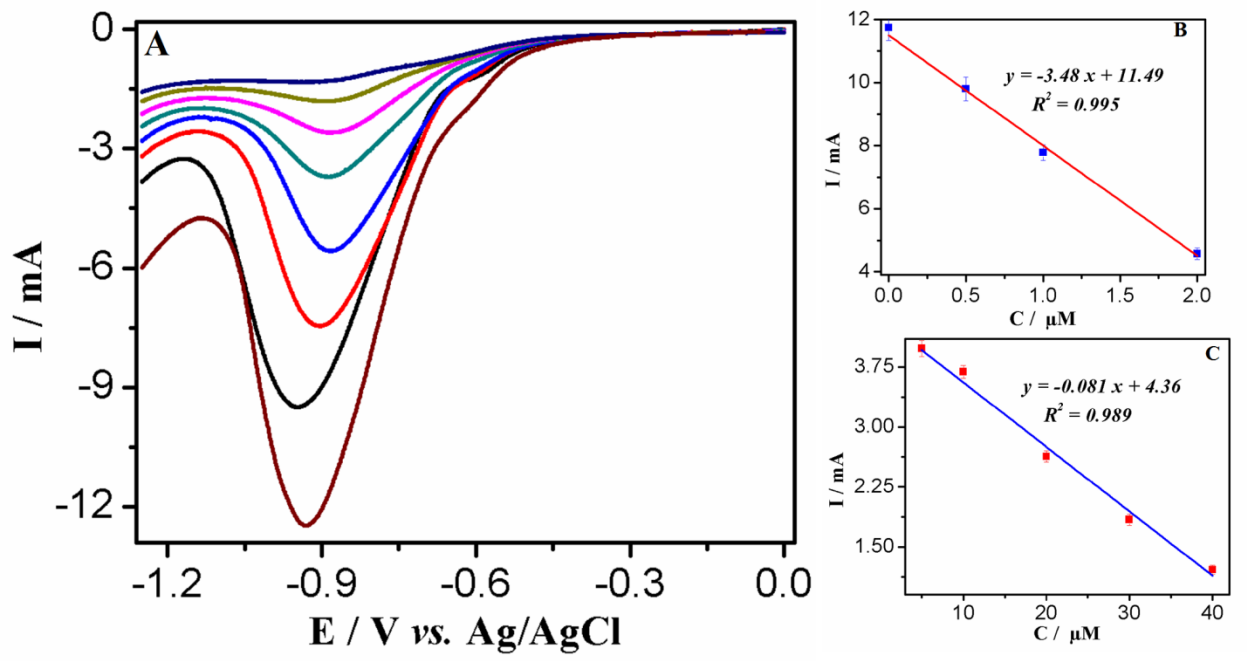

Figure 8. LSVs of varying concentration of TA(A), the corresponding calibration curve from 0 µM to 2 µM (B) and 5 µM to 40 µM (C).

The analytical performance of the proposed electrode was compared with other reported electrodes and results are summarized in Table. 1. It can be seen from the table that the proposed method shows comparable performance with other reported methods.

Table 1: The comparison of the efficiency of Sb-GCE and detection limit with other reported electrodes.

Electrode	Method	LOD(µM)	Ref
Multi wall carbon nanotubes modified GCE	LSV	0.1	[73]
Porous pseudo-carbon paste electrode	ASV	0.01	[74]
Silica gel modified carbon paste electrode	ASDPV	0.0003	[59]
Pre-PGE	ASDPV	0.0015	[75]
Sb−GCE	LSV	0.2	This work

4.6. Electrode reproducibility and stability

The electrode reproducibility was studied for Sb-GCEs by preparing three electrodes under the same modification conditions. Triplicate determination of 1 µM TA with each electrode was

measured to evaluate the reproducibility. The relative standard deviation was found to be 5.06% for the Sb-GCE.

The Sb-GCE showed low stability. The current response of the Sb-GCE decreased for the determination made on the same electrode on consecutive days. The current response had decreased by about 45% on the second day of the experiment. After one week the current response has further declined by 80%. This could be due to the high surface adsorption of TA on the metal film surface. Therefore, it is suggested that a fresh Sb-GCE should be prepared before the experiment.

4.7. Interference Study

In order to evaluate the applicability of the developed method the selectivity of Sb-GCE for the determination of TA, the effect of possible interfering substances was studied under the optimum conditions. Different interfering species were added into the solution containing 1 μM of TA. Figure 9, shows that 0.1 mM ascorbic acid (AA), uric acid (UA), glucose (GL), vitamin B2 (VB2), K^+, Na^+, Cl^-, and NO_3^- have negligible interference with the determination of 1 μM TA.

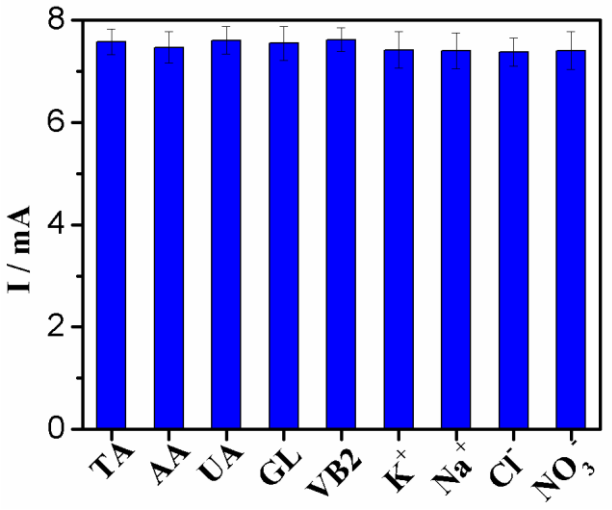

Figure 9. The interference effect of 0.1 mM ascorbic acid, uric acid, glucose, vitamin B2, K^+, Na^+, Cl^- and NO_3^- to 1 μM TA.

4.8. Real sample analysis

For practical application the proposed method was used to analyze TA in black tea extract. This sample was prepared as described in Section 3.7. Briefly, the sample of tea was prepared by

adding deionized water to 1.5 g of black tea. The black tea was extracted by 100 mL hot water for 10 minutes and the extract was filtered. The filtered sample was transferred to a 250 mL volumetric flask, and diluted with deionized water until the mark was reached[59]. 20 µL of the diluted extract was taken and further diluted in 10 mL 0.1 M acetate buffer (pH 3.6) and directly analyzed by proposed method. Furthermore, three different solutions were prepared by mixing 2 mL of the diluted extract with 1.00, 5.00 and 15.00 µM of standard TA solutions in 10 mL 0.1 M acetate buffer (pH 3.6) and the % recovery was calculated. The determination of TA in the tea extract was carried out according to the linear regression equation formulated for the calibration curve.

The result shows (Table 2) that the obtained % recovery is satisfactory for the determination of TA in tea sample extract it was found in the range of 91.02−102.3% demonstrating the selectivity for the detection of TA from complex sample matrices using Sb-GCE.

Table 2. Results of tannic acid determination in real samples

TA added (µM)	TA found (µM)	RSD %	% Recovery
	0.94	4.28	
1.00	1.88	2.50	97.00
5.00	6.12	4.65	102.3
15.00	14.51	3.62	91.02

5. Conclusion

In this work, the electrochemical property of antimony was firstly investigated. Next determination of TA on the surface of modified Sb-GCE by LSV was investigated. The method has shown that, a reduction peak current was linear with TA concentration in two ranges from 0.5 μM to 2 μM and 5 μM to 40 μM TA with the limits of detection were 0.2 μM and 3.9 μM, respectively and the electrode process is adsorption-controlled. The method is exhibited very good electroanalytical properties such as high sensitivity and lower limit of detection. It was shown that the treatment procedure GCE enhanced the reduction peak current of TA compared with the bare GCE. The electrode shows low stability, fresh Sb-GCE should be prepared before the experiment for TA determination. This method was successfully applied to the determination of TA in tea samples. The method has advantages due to its inexpensiveness, simplicity, fast time analysis and lesser toxicity.

References

1. E. C Davidson, Tannic acid in the treatment of burns. *Surg. Malawi Med J* **2005,** *17(1),* 19–20.
2. B. Doug, B. H. Jacobson, Effect of a blend of comfrey root extract (Symphytumofficinale L.) and tannic acid creams in the treatment of osteoarthritis of the knee: randomized, placebo-controlled, double-blind, multiclinical trials. *J. Chiropractic Med.* **2011,** *10 (3),* 147–156.
3. Y. Zhang, Y. Su, J. Peng, X. Zhao, J. Liu, J. Zhao, Z. Jiang Composite nanofiltration membranes prepared by interfacial polymerization with natural material tannic acid and trimesoyl chloride. *J. Membr. Sci* **2013,** *4292*, 35–242.
4. P. Orlowski, M. Krzyzowska. Assessment of in vitro cellular responses of monocytes and keratinocytes to tannic acid modified silver nanoparticles. *Toxicol* **2013,** *27*, 1798–1808.
5. G.H. Gaber Ahmed, R.B. Laíño. Fluorescent carbon nanodots for sensitive and selective detection of tannic acid in wines *Talanta* **2015,** *132*, 252–257.
6. A. Ren, W. Zhang. A tannic acidbased medical food, Cesinex (®), exhibits broad-spectrum antidiarrheal properties: a mechanistic and clinical study. *Dig. Dis. Sci* **2012,** *57 (1)*, 99–108.
7. K.J. Sreeram, T. Ramasami, Sustaining tanning process through conservation, recovery and better utilization of chromium. *Resour. Conserv. Recycl* **2003,** *38*, 185–212.
8. Y. S. Velioglu, G. Mazza, L. Gao, B. D. Oomah, Antioxidant activity and total phenolics in selected fruits, vegetables and grain products. *J. Agri. Food Chem* **1998,** *46*, 4113.
9. T. B. Kinraide, A. E. Hagermann, Interactive intoxicating and ameliorating effects of tannic acid, aluminum (Al3+), copper (Cu2+), and selenate (SeO42–) in wheat roots: a descriptive and mathematical assessment. *Physio Plantarum* **2010,** *139* 68–79.
10. X. J. Qu, J. Zhou, H.Z. Yin, Y.H. Sun, Flow injection-kinetic spectrophotometry for determination of tannic acid in Lupulus. *Chin. J. Anal. Chem* **2000,** *28* 820–824.
11. K. L. Bajaj, A.K. Devsharma, A colorimetric method for the determination of tannins in tea *Microchim. Acta* **1977,** *68*, 249–253.

12. G. Achilli, G.P. Cellerin, P.H. Gamache, Identification and determination of phenolic constituents in natural beverages and plant extracts bymeans of a coulometric electrode array system. *J. Chromatogr*. **1993,** *632*, 111–117.

13. C. Xie, H. Li, Determination of tannic acid in industrial wastewater based on chemiluminescence system of KIO4–H2O2–Tween40. *Luminescence* **2010,** *25*, 350–354.

14. C. Xie, H. Cui, Detection of tannic acid at trace level in industrial wastewaters using a highly sensitive chemiluminescence method *Water Res* **2003,** *37*, 233–237.

15. B. G. Corominas, J. V. G. Mateo, L. L. Zamora, J. M. Calatayud , Calatayud, Determination of tannic acid by direct chemiluminescence in a FIA assembly. *Talanta* **2002,** *58*, 1243–1251.

16. I. Gulcin, Antioxidant activity of food constituents: an overview. *Arch. Toxicol* **2012,** *86*, 345–391.

17. K. J. Davies, Oxidative stress: the paradox of aerobic life. *Biochem. Soc. Symp* **1995,** *61*, 1–31.

18. I. Gulcin, Antioxidant and antiradical activities of L-Carnitine. *Life Sci* **2006,** *78*, 803–811.

19. M. Elmastas, I. Gulcin, S. Beydemir, Ő. I Kufrevioğlu, H.Y. Aboul- Enein, A study on the in vitro antioxidant activity of juniper (Juniperuscommunis L.) seeds extracts *Anal. Lett* .**2006,** *39*, 47–65.

20. I. Gulcin, Antioxidant properties of resveratrol: a structureactivity insigh. *Innov. Food. Sci. Emerg* **2010,** *11*, 210–218.

21. D. Huang, B. Ou, R.L. Prior, The chemistry behind antioxidant capacity assays. *J. Agric. Food Chem* **2005,** *53*, 1841-1856.

22. L. K. MacDonald-Wicks, L. G. Wood, M. L. Garg, Methodology for the determination of biological antioxidant capacity in vitro: a review. *J. Sci. Food Agric* **2006,** *86*, 2046–2056.

23. S. A. Kitte, B. N Olana T. R. Soreta, Electrochemical determination of hydrogen peroxide at a glassy carbon electrode modified with palladium nanoparticles. *J. Serb. Chem. Soc* **2013,** *78 (5)*, 701–711.

24. J. C. Isenburg, N. V. Karamchandani, D. T. Simionescu, N. R. Vyavahare, Structural requirements for stabilization of vascular elastin by polyphenolic tannins. *Biomater* **2006,** *27*, 3645.

25. B. Halliwell, How to characterize an antioxidant- An update. *Biochem Soc Symp* **1995,** *61* 73–101.

26. L. Xu , N. He, J. Du , Y. Deng , Z. Li , T. Wang, A detailed investigation for determination of tannic acid by anodic stripping voltammetry using porous electrochemical sensor. *J.Anal.Chem* **2012,** *28,* 240-244.

27. K. Karamali *a and R. Teunis van, Teunis van Ree b, Tannins: Classification and Definition, In *The Royal Soc Chem*, Academic Press: New York, **2001**, pp 641–649.

28. E. C. Bate-Smith and T. Swain, *Flavonoid compounds. In Comparative Biochemistry*. . . Academic Press: New York., **1962**.

29. N. Balasundram, K. Sundram, and S. Samman, "Phenolic compounds in plants and agri-industrial by-products: Antioxidant activity, occurrence, and potential uses,". *Food Chem* **2006,** *99*, 191-203.

30. M. P. Gonthier, C. Remesy, A. Scalbert, V. Cheynier, J. M. Souquet, K. Poutanen, and A. M. Aura, "Microbial metabolism of caffeic acid and its esters chlorogenic and caftaric acids by human faecal microbiota in vitro," *Biomed & Pharmacoth* **2006,** *60*, 536-540.

31. C. A. Rice-Evans, N. J. Miller, and G. Paganga, "Antioxidant properties of phenolic compounds," *Trends in Plant Sci* **1997,** *2*, 152-159.

32. S. M. Henning, Y. Niu, Y. Liu, N. H. Lee, Y. Hara, G. D. Thames, R. R. Minutti, C. L. Carpenter, H. Wang, and D. Heber, "Bioavailability and antioxidant effect of epigallocatechin gallate administered in purified form versus as green tea extract in healthy individuals," *J. Nutr Biochem* **2005,** *16,* 610-616.

33. S. B. Lotito and B. Frei, "Consumption of flavonoid-rich foods and increased plasma antioxidant capacity in humans: Cause, consequence, or epiphenomenon?" *Free Radi Biol and Med* **2006,** *4*, 1727-1746.

34. N. Rangkadilok, S. Sitthimonchai, L. Worasuttayangkurn, C. Mahidol, M. Ruchirawat, and J. Satayavivad, "Evaluation of free radical scavenging and antityrosinase activities of standardized longan fruit extract,". *Food Chemi Toxicol* **2007,** *45*, 328-336.

35. G. G. Gross, *Enzymes in the biosynthesis of hydrolysable tannins. Plant Polyphenols.* . Plenum Press: New York, **1992**; p 43–60.

36. M. N. Clifford, Astringency. In F.A. . In *Phytochemistry of Fruits and Vegetables* (eds), T.-B. a. R. J. R., Ed. Clarendon Press: Oxford, **1997**, pp 87–107.

37. F. Shahidi, Antioxidants in food and food antioxidants. *Food / Nahrung*, **2000,** *44*, 158-163.

38. A. Scalbert, I.T. Johnson & M. Saltmarsh, Polyphenols: antioxidants andbeyond. *The Amer J. Clin Nutrition* **2005,** *81*, 215S-217S.

39. H. Karaosmanolglus, Travas-Sejdic, Designing PEDOT-based sensors for antioxidant analysis. . *The Inter.J. Nanotechnol* **2014,** *11*, 445-450.

40. D. W. Reisch, D. A. Lillard, Eitenmiller, R. Ronald, *Antioxidants*. **2007.**; Vol. ch 15.

41. J1. Grujic-Vasic, T. M. Bosnic, Jovanovic. The Examining of Isolated Tannins and their Astringent Effect. *J. Planta Medica* **1986** *1(6)*, 548-550.

42. S. L. Athanasiadou, I. Kyriazakis, F. Jackson, R. L. Coop, Direct anthelminthic effects of condensed tannins towards different gastrointestinal nematodes of sheep: in vitro and in vivo studies. *Vet. Parasitol* **2001,** *99*, 205-219.

43. A. A. Degen, K. Becker, H. P. S. Makkar, N. Borowy, Acacia saligna as a fodder for desert livestock and the interaction of its tannins with fiber fractions. *J. Sci.of Food Agric* **1995,** *68*, 65-71.

44. H. Kolodziej, A. F. Kiderlen, Antileishmanial activity and immune modulatory effects of tannins and related compounds on Leishmania parasitised RAW 264.7 cells. *Phytochem* **2005,** *66*, 2056-2071.

45. L. Svork, Determination of caffeine: a comprehensive review on electrochemical methods. *Int. J. Electrochem. Sci* **2013,** *8*, 5755-5773.

46. A. M. Pisoschi, Gh. P. Negulescu, A. Pisoschi. Ascorbic acid determination by an amperometric ascorbate oxidase-based biosensor. *Rev. Chim. (Bucharest)* **2010,** *61*, 339-344.

47. A. M. Pisoschi, A. Pop, Gh. P. Negulescu, A. Pisoschi. Determination of ascorbic acid content of some fruit juices and wine by voltammetry performed at Pt and Carbon Paste Electrodes, . *Molecules* **2011,** *16*, 1349-1365.

48. A. Magdalena, C. Carmen, Electrochemical Methods for Total Antioxidant Capacity and its Main Contributors Determination: A review. *Open Chem* **2015,** *13*, 824–856.

49. S. P. Kounaves, *Voltammetric techniques*. Prentice Hall, Upper Saddle River: New Jersey, **1997**

50. A. Frank. Settle, *Handbook of Instrumental Techniques for Analytical Chemistry*. Prentice Hall PTR (ECS Professional): **1997**.

51. J. Allen. Bard; Larry R.Faulkner, *ELECTROCHEMICAL METHODS: Fundamentals and Applications*. 2nd ed.; John Wiley & Sons: **2001**.

52. R. S. Nicholson, I. Shain, Theory of Stationary Electrode Polarography. Single Scan and Cyclic Methods Applied to Reversible, Irreversible, and Kinetic Systems. *Anal. Chem.* **1964,** *36*, 706-723.

53. W. H. Reinmuth, Theory of Stationary Electrode Polarography. *Anal. Chem.* **1961,** *33*, 1793-1794.

54. J. Heinze, Cyclic Voltammetry - "Electrochemical Spectroscopy". New Analytical Methods (25). *Angew. Chem. Int. Ed.* **1984,** *23*, 831-847.

55. L. R. Faulkner, L. R., Understanding electrochemistry: Some distinctive concepts. *J. Chem. Educ.* **1983,** *60*, 262.

56. M. Sandra, B. Erika, M. Juan, A. Luis, Voltammetric Techniques. In *Agricultural and Food Electroanalysis*, 1st ed.; Alberto, E.; Maria Cristina, G.; Miguel Angel, L., Eds. John Wiley & Sons, Ltd: **2015**, pp 23-44.

57. B. Taddese, K. Siraj, E. Tefera. Determination of Heavy Metal Ions on Glassy Carbon Electrode Modified with Antimony *Adv. Sci. Eng. Med* **2013,** *5*, 275–284.

58. G. Valéria Guzsványa, S. Nobuaki, N. Koji Nakano, Antimony-film electrode for the determination of trace metals by sequential-injection analysis/anodic stripping voltammetry. *J. Anal. Chim. Acta* **2010,** *658* 12–17.

59. L. Dai Vu 1, E. Bensu, C. Libor *, D. Yusuf, Determination of Tannic Acid Using Silica Gel Modified Carbon Paste Electrode. *Int. J. Electrochem. Sci* **2013,** *8*, 9278 -9286

60. M. Makombe, C. Vander Horst, S. Bongiwe, Antimony film sensor for sensitive rare earth metal analysis in environmental samples. *J. Environ. Sci and Health, Part A* **2016,** *VOL. 0*, 1–10.

61. L.T. Tufa, K. Siraj. T. R. Soreta, Electrochemical determination of lead using bismuth modified glassy carbon electrode. *Russian J.Electrochem* **2013,** *49*, 59–66.

62. T.Ummihan, C. E., Selective and sensitive determination of TA using a 1−benzoyl−3−pyrolidine thiourea. *J. Electro. Chem* **2016,** *776*, 1−8.

63. W. Huijun, Y. Rui, Z. Faqiong, and Z. Baizhao, Electrochemistry and voltammetric determination of tannic acid on a single-wall carbon nanotube-coated glassy carbon electrode. *Microchim Acta* **2007,** *159*, 109–115.

64. B. H. Cruz, J. M Díaz-Cruz, C. Ariño, M. Esteban, Heavy Metal Binding by Tannic Acid: A Voltammetric Study. *Electroanalysis* **2000,** *12*, 1130-1137.

65. L. Fan, Y. Ma, Y. Su, R. Zhang, Y. Liu, Q. Zhang, Z. Jiang, Green coating by coordination of tannic acid and iron ions for antioxidant nanofiltration membranes. *RSC Adv.* **2015,** *5*, 107777-107784.

66. Z. Fu, R. Chen, Study of Complexes of Tannic Acid with Fe(III) and Fe(II). *J. Anal. Meth. Chem.* **2019,** *2019*, 6.

67. P. Kraal, B. Jansen, K. G. J. Nierop, J. M. Verstraten, Copper complexation by tannic acid in aqueous solution. *Chemosphere* **2006,** *65*, 2193-2198.

68. H. Luo, S. Zhang, X. Li, X. Liu, Q. Xu, J. Liu, Z. Wang, Tannic acid modified Fe_3O_4 core–shell nanoparticles for adsorption of Pb^{2+} and Hg^{2+}. *J. Taiwan Inst. Chem. Eng.* **2017,** *72*, 163-170.

69. M. McDonald, I. Mila, A. Scalbert, Precipitation of Metal Ions by Plant Polyphenols: Optimal Conditions and Origin of Precipitation. *J. Agri. Food Chem.* **1996,** *44*, 599-606.

70. M. A. Rahim, H. Ejima, K. L. Cho, K. Kempe, M. Müllner, J. P. Best, F. Caruso, Coordination-Driven Multistep Assembly of Metal–Polyphenol Films and Capsules. *Chem. Mat.* **2014,** *26*, 1645-1653.

71. P. K. South, D. D. Miller, Iron binding by tannic acid: Effects of selected ligands. *Food Chem.* **1998,** *63*, 167-172.

72. A. Üçer, A. Uyanik, S. F. Aygün, Adsorption of Cu(II), Cd(II), Zn(II), Mn(II) and Fe(III) ions by tannic acid immobilised activated carbon. *Sep. Purif. Technol.* **2006,** *47*, 113-118.

73. S. Lu, Electrochemical determination of tannins using multiwall carbon nanotubes modified glassy carbon electrode. *Russ. J. Electrochem* **2004,** *40*, 750–754.

74. L. Xu, J. Du, Y. Deng A detailed investigation for determination of tannic acid by anodic stripping voltammetry using porous electrochemical sensor. *Anal. Chim. Acta* **2009,** *634*, 49–53.

75. L. Dai VU 1, D. Yusuf and Č. Libor 3, Voltammetric Determination of Tannic Acid in Beverages using Pencil Graphite Electrode *Czech J. Food Sci* **2015** *33*, (1): 72–76.