

THE USE OF *Amaranthus spinosus* IN THE SYNTHESIS OF SILVER NANOPARTICLES AND THEIR APPLICATION IN THE DETECTION OF GLUCOSE

S. A.Mamuru^{1*}, M. T. Malachy¹, A.E.Eseyin² and G. Ankwai²

¹ Electrochemical NanoLab, Department of Pure and Applied Chemistry, Adamawa State University, Mubi, Nigeria

² Department of Chemistry, University of Jos, Jos, Nigeria

*Corresponding author: mamuru19@adsu.edu.ng

ABSTRACT

Because of biologically active plant secondary metabolites that aid in green synthesis, as well as their specific biological applications, biosynthesized nanoparticles are gaining popularity. This paper describes the electrochemical characteristics and electrocatalytic activity of silver nanoparticles made using *Amaranthus spinosus* aqueous leaf extract for glucose detection. In the biosynthesis of silver nanoparticles, an aqueous leaf extract of *Amaranthus spinosus* was utilized as a reducing and capping agent. UV–visible spectroscopy, Fourier transform infrared (FTIR) spectroscopy, scanning electron microscopy (SEM), and energy-dispersive X-ray studies were used to characterize the biosynthesized *Amaranthus spinosus* silver nanoparticles. The electrochemical properties reveal the redox process of Ag^+ to Ag^0 , while the electrocatalytic activity towards nonenzymatic glucose oxidation occurs at a lower positive potential, 200 mV, implying that the silver nanoparticles synthesized by *Amaranthus spinosus* could be used as a candidate for glucose detection.

Keywords: Silver nanoparticles, *Amaranthus spinosus*, glucose electrooxidation, Biosynthesis

INTRODUCTION

Silver nanoparticles (AgNPs) have increased in popularity in recent years, owing to the wide range of applications for which they can be used. The initial step in the synthesis of AgNPs is to reduce silver ions, Ag^+ to Ag^0 with a strong reducing agent. Synthetic chemical reducing agents are not recommended for synthesizing AgNPs for biomedical applications since unreacted traces of these compounds can be hazardous. Natural reducing agents present in a variety of plants have lately been examined as an alternative to conventional chemical reducing agents for the synthesis of AgNPs [1-5].

Phenolic compounds, terpenoids, alkaloids, and other phytochemicals have been discovered to be effective reducing agents. [6,7]. The plant *Amaranthus spinosus* was chosen for this study. The species is well-known for its wide range of therapeutic qualities [8-12]. The strong antioxidant nature of the ethanolic leaf extract [13] piqued our interest, therefore we decided to carry out the current study and test the plant for silver nanoparticle synthesis.

Because of its use as a fuel, in medicine, and in the food industry, glucose oxidation reactions are an appealing research subject [14]. Glucose electro-oxidation has been studied extensively

using noble metals, modified electrodes incorporating ad-atoms, and alloy metals as electrode materials. However, glucose oxidation on the electrode material is known to result in the formation of an intermediate that can be absorbed on the electrode surface. As a result of the absorbed material's effect on the absorbed electrode surfaces, the active sites on the electrocatalyst surface are blocked, inhibiting glucose oxidation [15]. It is therefore desirable to develop highly effective electrode materials for the electrooxidation of glucose. Because of its electrocatalytic activity in glucose oxidation, silver is a promising electrode material. At +0.3V, a silver nanoparticle-modified electrode used for glucose electrooxidation produced primarily formate, as well as oxalate, gluconate, and other compounds [16-20]. Nonenzymatic glucose sensors have been made using a variety of monometallic and bimetallic nanoparticles [21-23], which have shown good catalytic activity for glucose oxidation. However, there has been little research on non-enzymatic glucose electrodes made of silver nanoparticles and the process of glucose oxidation [24].

The use of *Amaranthus spinosus* leaves aqueous extract as an alternative technique for synthesis of silver nanoparticles and evaluation of their electrochemical behavior for non-enzymatic glucose detection are described in this paper.

MATERIALS AND METHOD

Materials

Silver nitrate; potassium ferrocyanide and potassium ferricyanide 98.5 %; Alumina (Nanopowder); sodium hydroxide; *Amaranthus spinosus* leaves; Perkin Elmer Frontier Fourier Transform Infrared spectrophotometer; ultrasonic bath; Autolab PGSTAT 302N; Hanna Instrument 2211 pH meter; Jenway 6400 UV-visible spectrophotometer.

Method

Preparation of the Leaf Extract

To make the extract, 10 g of fresh *Amaranthus spinosus* plant leaves were washed with distilled water to remove dust before being cut into pieces and placed in an Erlenmeyer flask with distilled water in which they were boiled for 20 minutes before being allowed to cool at room temperature. It was then filtered through Whatman filter paper No. 1 in order to obtain the extracted substance.

Silver Nanoparticle Synthesis

The process for synthesis was adapted from [25] and briefly described. A top load balance was used to weigh 0.042 g of silver nitrate, which was dissolved in 100 mL of distilled water to prepare a 0.001 M silver nitrate solution. 60mL of a 1mM silver nitrate solution was reduced using 5mL of leaf extract at room temperature for 10 minutes; the solution's color changed, indicating the formation of silver nanoparticles. Centrifuged at 3000 rpm for 10 minutes, the product was then

rinsed with ethanol and distilled water and left to dry at room temperature.

Electrode Preparation

In order to prepare the platinum–silver nanoparticles electrode (Pt/AgNP), 10 μ L of an already prepared silver nanoparticles solution was placed on the surface of a platinum electrode (3mm diameter) that had already been polished and ultrasonically cleaned with a micropipette and then allowed to dry at room temperature. With the use of an outer sphere redox probe composed of 0.005M potassium ferricyanide and potassium ferrocyanide in 0.2M NaOH solution, the electrochemical characteristics of the modified electrode were investigated using cyclic voltammetry (CV).

Solution and Surface Characterization

The presence of the synthesized silver nanoparticles was confirmed by sampling the aqueous component using UV-visible spectrophotometer. Distilled water was used as a blank. Absorbance measurement was carried out at a wavelength of 300–800 nm at a scan interval of 5 nm in order to obtain the UV-visible spectrum of the nanoparticles. The biomolecule responsible for the reduction of AgNPs was determined using Perkin Elmer Fourier transform infrared (FT-IR) spectroscopy.

Detection of Glucose

Glucose oxidation was done using electrochemical techniques such as cyclic voltammetry. 0.001M glucose in 0.2M NaOH

solution was prepared and sweep scan from 0.0 V - 0.6V.

RESULTS AND DISCUSSION

Optical Property

The optical properties of silver nitrate solution, *Amaranthus spinosus* leaf extract, and silver nanoparticle are shown in Plate 1. After adding 5 mL of *Amaranthus spinosus* leaf extract, the color of 1mM silver nitrate solution changed from colorless to light brown, then dark brown, indicating the creation of silver nanoparticles. Silver nanoparticles are recognized to have a distinctive brown color. The excitation of the surface plasmon resonance action in silver nanoparticles is responsible for the color variations. The size and shape of metal surface plasma oscillations, as well as their optical properties, are well understood.

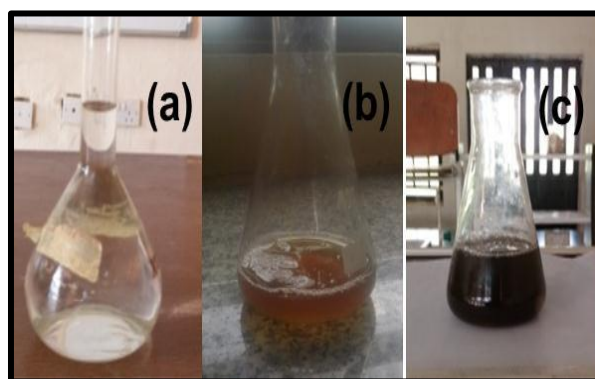


Plate 1: Silver nitrate solution (a), *Amaranthus spinosus* leaves extract (b), silver nanoparticle (c)

UV-visible spectroscopy

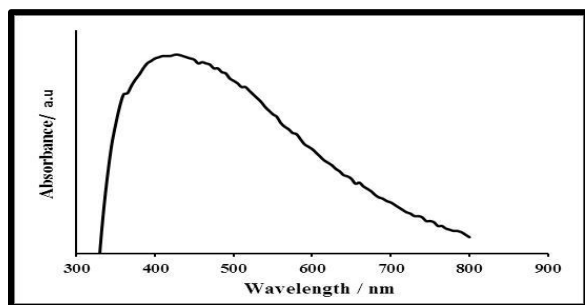


Figure 1: UV-visible spectrum of silver nanoparticle

A greater amount of information about silver nanoparticles was obtained through the use of UV-visible spectroscopy, which was used to observe the reduction of aqueous metal ions after treatment with *Amaranthus spinosus* leaf extract (Figure 1). In the UV-visible absorption spectra of silver nanoparticles, the typical surface plasmon resonance (SPR) is observed with an absorbance of approximately 350–450 nm and peak maxima of 410 nm, which can be attributed to the presence of silver nanoparticles. According to the nearly symmetrical structure of the plasmon band, the nanoparticles are not uniformly distributed and homogeneous. The non-uniformity of the silver nanoparticle is responsible for the large absorption peak observed at longer wavelengths. When it comes to a particle's size, shape, and capping agent, all of these factors influence the position of the SPR band.

Scanning Electron Microscopy (SEM)

Plate 2 depicts the morphology of silver nanoparticles as densely packed spherical particles that are heavily aggregated under a high resolution microscope. The considerable

agglomeration of biosynthesized AgNPs may have been caused by the dehydration utilized during sample preparation for SEM examination. The biosynthesis of silver nanoparticles using *Taraxacum officinale* and *Cucumis prophetarum* produced similar findings[1,3].

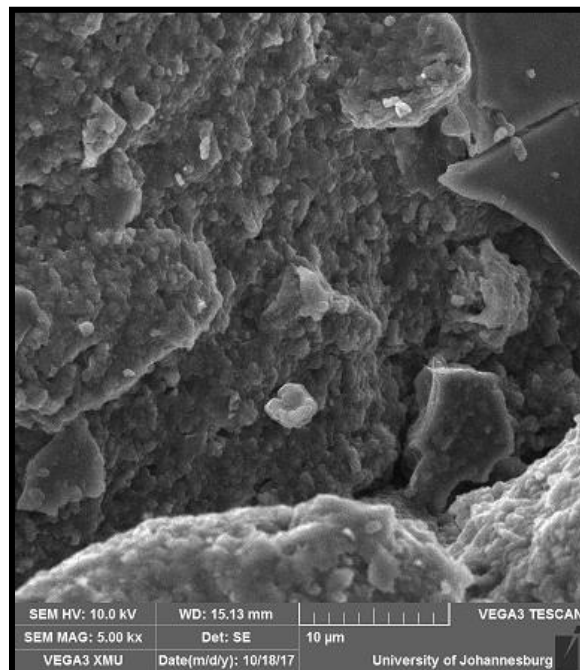


Plate 2: SEM morphology of silver nanoparticles

Fourier Transform Infra-Red Spectroscopy and Energy Dispersive Spectroscopy

Figure 2 depicts the FTIR spectrum of *A. spinosus* aqueous leaf extracts and silver nanoparticles. The former revealed distinct bands for numerous functional groups. IR bands were recorded at 3383, 2918, 2850, 2117, 1623, 1395, 1316, 1242, and 1014 cm^{-1} . The bands at 3383 cm^{-1} and 2918 cm^{-1} , respectively, correspond to hydroxyl ($-\text{OH}$) and primary amine salt ($-\text{NH}_3^+$). Aromatic $-\text{CH}_2-$ is responsible for the bands at 2850 cm^{-1} . The 1395 cm^{-1} and 1316 cm^{-1} IR bands

can be attributed to symmetric bending of $\text{C}(\text{CH}_3)_3$ and CH symmetric rock, respectively. C=C stretching, amide III bonds under N-H bend were seen in the IR bands at 1623cm^{-1} and 1242cm^{-1} , while aliphatic amine was seen in the IR band at 1014cm^{-1} .

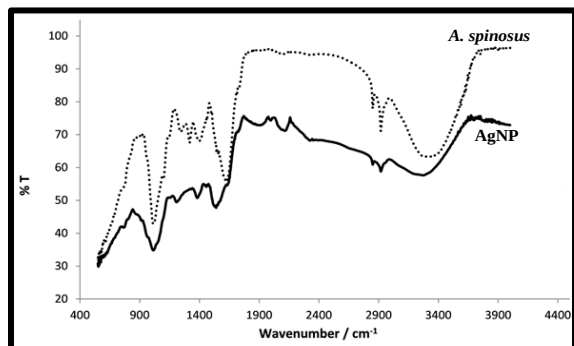


Figure 2: Comparative FTIR spectra of *A. spinosus* and silver nanoparticles

Purified AgNPs samples were submitted to FTIR tests to see if some biomolecules bonded to the silver surface during AgNPs synthesis, particularly those with free carboxylic (-COOH) or amino (-NH_2) groups present in the leaf extract. The hydroxyl (3378 cm^{-1}), aromatic and aliphatic amines (1380 and 1014 cm^{-1} respectively), carbonyl (1623 cm^{-1}), C=C of benzene (1539 cm^{-1}), and C-H (2820 cm^{-1}) functional groups revealed peaks in the IR band of AgNPs. Aqueous leaf extract of *A. spinosus* is recognized to be particularly rich in anti-oxidant characteristics due to the presence of many compounds with functional groups that can donate hydrogen atoms and several free amino or carboxylic moieties capable of binding to free silver surface [13]. As illustrated in the FTIR spectrum of AgNPs, such molecules adhered

securely to the silver surface during the reduction of Ag^+ ions to Ag^0 .

The EDX spot profile (Figure 3) of AgNPs revealed a significant signal for silver at 3KeV, as well as faint oxygen, carbon, aluminum, phosphorus, calcium and potassium peaks, indicating that they are derived from biomolecules bound to the AgNPs' surface.

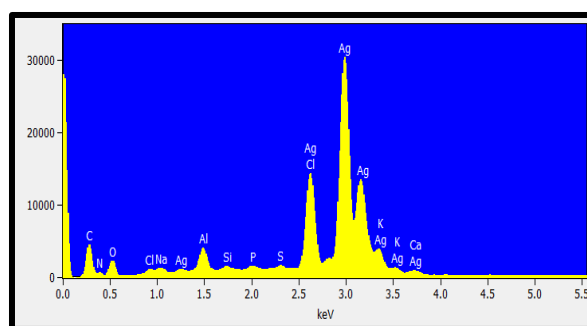
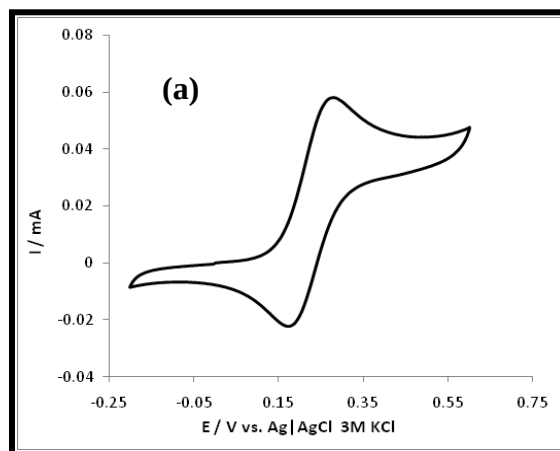


Figure 3: EDX profile of silver nanoparticles synthesized with *A. spinosus* aqueous leaves extract

Electrochemical Characterisation



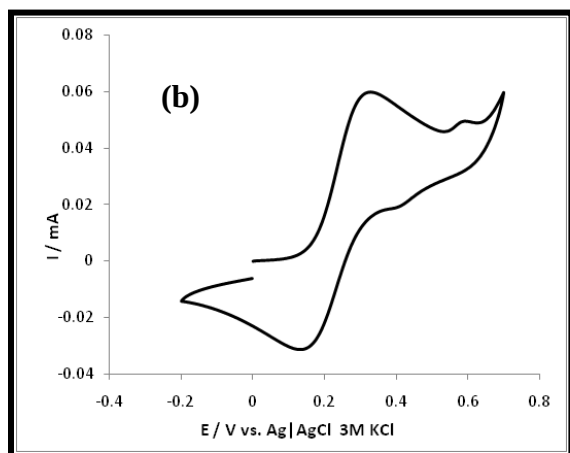
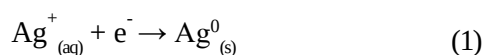


Figure 4: Cyclic voltammograms of bare Pt (a) and Pt/AgNP modified electrode (b) in 0.005M $[\text{Fe}(\text{CN})_6]^{3-}/[\text{Fe}(\text{CN})_6]^{4-}$ 0.2M NaOH solution

Figure 4 shows how bare Pt and Pt/AgNP behave in 0.005M $\text{K}_4[\text{Fe}(\text{CN})_6]/\text{K}_3[\text{Fe}(\text{CN})_6]$ solution. Peak potentials for oxidation (E_{ox}) and reduction (E_{red}) of the $\text{Fe}^{3+}/\text{Fe}^{2+}$ on the Pt electrode were both 256 and 186 mV. The $\text{Fe}^{3+}/\text{Fe}^{2+}$ oxidation and reduction peaks at 290 and 170 mV and 570 and 427 mV are attributed to $\text{Fe}^{3+}/\text{Fe}^{2+}$ and Ag^+/Ag^0 redox processes respectively, on the Pt/AgNP modified electrode. The existence of the silver redox process indicates that the silver nanoparticle is electro active. The formal potential ($E_{1/2}$) for the $\text{K}_3\text{Fe}(\text{CN})_6/\text{K}_4\text{Fe}(\text{CN})_6$ redox pair is 250 mV, and the formal potential ($E_{1/2}$) for the Ag^+/Ag^0 silver redox process is 495 mV, which corresponds to the reaction equation presented in equation (1):



As shown by the silver redox process, silver ion can be converted to silver metal.

Electrocatalytic Behavior towards Glucose Oxidation

Figure 5 depicts the electrocatalytic behavior of Pt/AgNP in a 0.001M glucose + 0.2M NaOH aqueous solution for glucose detection. Glucose electro-oxidation peak was observed at 200mV, according to previous report it occurs at approximately 300 mV this process is directly related to the transfer of two electrons to the gluconic acid formation or similar species, which are adsorbed on the electrode surface [15]. A shift to a lower positive potential indicates that the AgNP detects glucose more quickly than previously thought. The cyclic voltammogram of AgNP in the absence of glucose is shown in Figure 5(inset). There is no peak due to glucose. Ag^+/Ag^0 are responsible for the redox peaks observed at 600mV and 570mV, respectively.

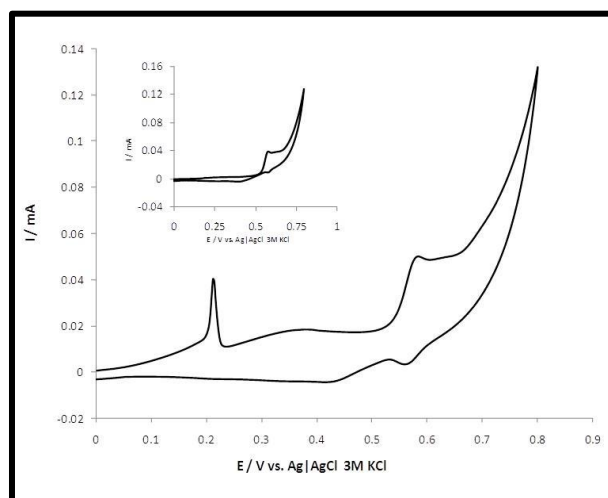


Figure 5: Cyclic voltammogram of Pt/AgNP in 0.001M glucose 0.2M NaOH solution. Cyclic voltammogram of Pt/AgNP in 0.2M NaOH solution (inset)

CONCLUSION

Aqueous leaf extract of *A. spinosus* was used to synthesize silver nanoparticles. The change in color from colorless to brown validated the synthesized AgNP. UV visible spectroscopy, SEM, and EDX were utilized to characterize the nanoparticles, and FTIR spectroscopy was employed to identify the probable biomolecules responsible for the bio-reduction.

When utilized as a modified electrode in the detection of glucose, the silver nanoparticles had an oxidation peak potential of 200 mV. When utilized in the absence of glucose, no peak was seen. As a result of their catalytic function in the oxidation of glucose, AgNPs can be utilized as a sensor to detect the presence of glucose.

ACKNOWLEDGEMENT

The authors would like to thank Adamawa State University, Mubi for providing most of the facilities used in carrying out this research.

REFERENCES

- [1] Hemlata, P., Meena, R., Singh, A.P. and Tejavath, K. K.(2020) Biosynthesis of silver nanoparticles using *Cucumis prophetarum* aqueous leaf extract and their antibacterial and antiproliferative activity against cancer cell lines. *ACS Omega* 5: 5520-5528
- [2] Pirtarighat, S., Ghannadnia, M. and Baghshahi, S. (2019) Green synthesis of silver nanoparticles using the plant extract of *Salvia spinosa* grown in-vitro and their antibacterial activity assessment. *Journal of Nanostructure in Chemistry* 9:1–9
- [3] Rasheed, T., Bilal, M., Li, C. and Iqbal, H. (2018) Biomedical potentialities of *Taraxacum officinale*-based nanoparticles biosynthesized using methanolic leaf extract. *Current Pharmaceutical Biotechnology* 18:1116–1123.
- [4] Shanmuganathan, R., MubarakAli, D., Prabakar, D., Muthukumar, H., Thajuddin, N., Kumar, S.S. and Pugazhendhi, A. (2018) An enhancement of antimicrobial efficacy of biogenic and ceftriaxone-conjugated silver nanoparticles: green approach. *Environment Science Pollution Research* 25: 10362–10370
- [5] Ramkumar, V.S., Pugazhendhi, A., Gopalakrishnan, K., Sivagurunathan, P., Saratale, G.D., Dung, T.N.B. and Kannapiran, E.(2017) Bio-fabrication and characterization of silver nanoparticles using aqueous extract of seaweed *Enteromorpha compressa* and its biomedical properties. *Biotechnology Report* 14: 1–7
- [6] Song, J. Y., Jang, H. K. and Kim, B. S. (2009) Biological synthesis of gold nanoparticles using *Magnolia kobus* and *Diopyros kaki* leaf extracts. *Process Biochemistry* (44)10:1133-1138
- [7] Song, J. Y., Kwon, E. Y. and Kim, B. S. (2010) Biological synthesis of platinum nanoparticles using *Diopyros kaki* leaf extract. *Bioprocess and Biosystems Engineering* (33)1:159-164
- [8] Zeashan, H., Amresh, G., Singh, S. and Rao, C. V. (2009) Hepatoprotective and antioxidant activity of *Amaranthus spinosus* against CCl₄ induced toxicity. *Journal of Ethnopharmacology* (125)2: 364-366
- [9] Hema, E. S., Sivadasan, M. and Anil, K. N. (2006) Studies on edible species of *Amaranthaceae* and *Araceae* used by Kuruma and Paniya tribes in Wayanad district, Kerala, India. *Ethnobotany* (18)1: 122-126
- [10] Odhav, B., Beekrum, S., Akula, U. and Baijnath, H. (2007) Preliminary assessment of nutritional value of traditional leafy vegetables in KwaZulu-Natal, South Africa. *Journal of Food Composition and Analysis* (20)5: 430-435
- [11] Cao, G., Sofic, E. and Prior, R. L. (1996) Antioxidant capacity of tea and common

vegetables. *Journal of Agriculture and Food Chemistry* (44) 11: 3426-3431

[12] Gil, M. I., Ferreres, F. and Thomas-Barberan, F. A. (1999) Effect of post harvest storage and processing on the antioxidant constituents (flavonoids and vitamin C) of fresh-cut spinach. *Journal of Agriculture and Food Chemistry* (47) 6: 2213-2217

[13]. Cai, Y., Sun, M., and Cork, H. (2003) Antioxidant activity of betalains from plants of the *Amaranthaceae*. *Journal of Agriculture and Food Chemistry* (51)8:2288-2294

[14]. Maia Quintino, M.D.S., Winnischofer, H., Nakamura, M., Araki, K., Toma, H.E., and Angnes L. (2005) Amperometric sensor for glucose based on electrochemically polymerized tetra-ruthenated nickel-porphyrin. *Analytica Chimica Acta* 539: 215-222.

[15] Tominaga, M., Nagashima, M. K. and Taniguchi, I. (2007) Surface poisoning during electrocatalytic monosaccharide oxidation reactions at gold electrodes in alkaline medium. *Electrochemistry Communication* 9: 1892-1898

[16] Tominaga, M., Shimazoe, T., Nagashima, M. and Taniguchi, I. (2005) Electrocatalytic oxidation of glucose at gold nanoparticle-modified carbon electrodes in alkaline and neutral solutions. *Electrochemistry Communication* 7:189-193.

[17] Rong, L.Q., Yang, C., Qian, Q.Y. and Xia, X.H. (2007) Study of the nonenzymatic glucose sensor based on highly dispersed Pt nanoparticles supported on carbon nanotubes. *Talanta* 72: 819-824

[18] Safavi, A., Maleki, N. and Farjami, E. (2009) Fabrication of a glucose sensor based on a novel nanocomposite electrode. *Biosensors Bioelectronics* 24:1655-1660.

[19] You, T.Y., Niwa, O., Chen, Z.L., Hayashi, K., Tomita, M. and Hirono, S. (2003) An amperometric detector formed of highly dispersed Ni nanoparticles embedded in a graphite-like carbon film electrode for sugar determination. *Analytical Chemistry* 75: 5191-5196.

[20] Kang, X.H., Mai, Z.B., Zou, X.Y., Cai, P.X. and Mo, J.Y. (2007) A sensitive nonenzymatic glucose sensor in alkaline media with a copper nanocluster/multiwall carbon nanotube-modified glassy carbon electrode. *Analytical Biochemistry* 363: 143-150.

[21] Jin, C.C., and Chen, Z.D. (2007) Electrocatalytic oxidation of glucose on gold-platinum nanocomposite electrodes and platinum-modified gold electrodes. *Synthetic Metals* 157: 592-596

[22] Tominaga, M., Taema, Y., and Taniguchi, I. (2008) Electrocatalytic glucose oxidation at bimetallic gold-copper nanoparticle-modified carbon electrodes in alkaline solution. *Journal of Electroanalytical Chemistry* 624: 1-8.

[23] Hindle, P.H., Nigro, S., Asmussen, M., and Chen, A. (2008) Amperometric glucose sensor based on platinum-iridium nanomaterials. *Electrochemistry Communication* 10:1438-1441.

[24] Quan, H., Park, S-U., and Park, J. (2010) Electrochemical oxidation of glucose on silver nanoparticle-modified composite electrodes. *Electrochimica Acta* 55: 2232–2237

[25] Kanchana, A., Agarwal, I., Sunkar, S., Nellore, J. and Namasivayam, K. (2011) Biogenic silver nanoparticles from *Spinacia oleracea* and *Lactuca sativa* and their potential antimicrobial activity. *Digest Journal of Nanomaterials and Biostructures* (6) 4: 1741-1750.