BIOCOMPATIBLE GRAPHENE OXIDE (GO) NANOBIOSENSOR USED FOR QUANTITATIVE ANALYSIS OF GLUCOSE

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Graphene oxide (GO) nanoparticles based electrochemical nanobiosensor was developed to check the sensitivity response of biological elements. Present work is related to synthesize GO nanoparticles by Hummers method for the detection of glucose using electrochemical nanobiosensor. Two dimensional hexagonal crystal structure and crystallite size was calculated by XRD analysis and randomly crumpled like surface morphology was identified by using SEM micrograph. Furthermore, different rotational and vibrational functional groups (C-O, C=C, CO_2 and C-H) attached to the layer of graphene oxide and peak shift toward longer wavelength was studied by FTIR and UV-visible spectrum. Finally, voltammetry cyclic used for sensitivity of glucose and presence of glucose was recorded in concentration of 5mM at scan rate of 50mVs^{-1}. In future this type of nanobiosensor used for the detection of biological element like DNA.

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1. Introduction

In few decades, different kinds of nanostructure materials (nanofibers, nanotubes, nanoparticles, nanowires and quantum dots) are synthesized variety of methods such as auto-combustion, co-precipitation, sol-gel, chemical vapour deposition and hummer's method [1-3]. These NPs have huge applications in diverse biomedical applications due to stability and biocompatibility such as drug delivery, biosensors, bio-imaging, photo thermal, photo ablation, hyperthermia therapy and MRI contrast agent [4-7]. But recently focus on advance materials like graphene and its derivatives for cancer treatment and electrochemical biosensor to check the sensitivity of glucose, urea and DNA detection in healthy and sick tissues. Graphene is an allotropic form of carbon, which exhibits sp^2hybridization in carbon atoms are organized in such a way having structures like honeycomb lattice [8].

Graphene have unique properties such as structural, thermal conductivity, electrical conductivity and elastic stiffness [9]. Furthermore, due to excellent biocompatibility of 2-D layered material such as graphene oxide was preferred for the fabrication of electrode for nanobiosensor [10].Biosensor is a device that used for the detection of biomolecule and it consists of three major parts bioactive, transducer and detector. Bioactive is a molecule that reacts with analyte results a compound that detected by transducer to produce electrochemical signals and the signal pass through amplifier and reach on detector then output of the sensitivity signal display on

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screen. Biosensor is very high selective device and now used in multidisciplinary areas of research that links the principles of basic sciences with biomedicine [11-14].

There are varieties of biosensor use for different purpose like fluorescence based, enzyme based, photo electrochemical based biosensor, resonant biosensor, optical biosensor, piezoelectric biosensor and potentiometric biosensor. In this current experimental approach, novel and convincing approach of electrochemical based biosensor has been developed by employing desired morphology of nano graphene by adopting very simple, facile, comprehensive and economical technique such as hummers method, The prepared graphene was characterize by using various characterization techniques XRD, SEM, FTIR and finally the quantitative measurements of glucose (volumetric curve) was used electrochemical nanobiosensor.

2. Experimental procedure

2.1. Synthesis of graphene oxide nanoparticles

The modified Hummer's method was used to synthesize the graphene oxide nanoparticles. Graphite powder (0.083M) and $NaNO_3$(0.011M) was dissolved into H_2SO_4(25mL) to prepared a solution and continuous stirring on magnetic stirrer for 50 min at 20°C. After that adding $KMnO_4$(0.189 M) slowly then appear brownish color and continues stirring for 1 day. The prepared mixtures dilute with 400 mL de-ionized water on stirrer, when reaction was completed with $KMnO_4$ then add 35% hydrogen peroxide after that solution was appeared in the form of yellow color. Then the end product washed with de-ionized water for several time and filter. The nanoproducts dry in oven for 5 hours at 200°C and grinding in mortar and pestle to produce nano powder of graphene oxide.

2.2. Glucose test solution

The glucose test solution has made by adding different concentration of glucose in phosphorus buffer solution (PBS). Then heated for 1 hour and cooled until the solution became the clear without any turbidity.

2.3. Fabrication of GO working electrode

In the first step, we have polished the glassy carbon electrode (GCE) by the alumina powder on the polishing material. After that GCE was cleaned in water and ethanol by ultra-sonication. Then finally to obtain the unblemished surface of GCE, we washed the GCE with distilled water and dry. Later on, we made the suspension of GO-NPs ultra-sonication with nafion and ethanol for hours. Then after the suspension was isolated drop wise on the surface of pure GCE and dried in oven at 150°C till the film dried entirely [15].

2.4. Characterizations of graphene oxide (GO)

Prepared GO was characterized by using the different techniques including X-ray diffraction (XRD), Fourier Transform Infrared Radiation (FTIR) and Scanning electron microscope (SEM).X-ray diffraction (XRD) analysis of GO was evaluated by using a PanalyticalX'Pert-Pro apparatus at 30mA, 40 kV with stability of 0.01%/8h.The crystalline layer size of GO nanomaterial was mathematically calculated by using the Scherer's equation. FTIR measurement was engaged to examine the bonding interfaces in graphene and absorption spectra before and after the oxidation progression by using the FTIR spectrophotometer and UV-visible spectroscopy. SEM images were obtained with a field emission gun scanning electron microscope. The extraordinary high energy electron beam in scanning electron microscopy was used to investigate the surface morphology and structure of sample GO. Prepared GO used as electrode in fabrication of biosensor for detection of glucose.

3. Results and discussion

3.1. Structure analysis

XRD was used to study the two dimensional crystal structure and crystallite size of graphene oxide (GO) nanoparticles. Figure (1) indicated that different diffraction peaks at 10.15° and 24.48° represented the hexagonal lattice (flat monolayer) between carbon atoms. These peaks express the miller indices (001) and (002) and d-spacing from 0.39 nm and 0.79 nm. Moreover, GO nano-materials shows highly ordered layer structure and maximum intensity of peak at (001) indicate the formation of graphene oxide with layered structure [16,17].The average crystalline size of GO-NPs were calculated by using scherrer equation and size was expressed in (Table.1).

$$D = \frac{k\lambda}{\beta \cos\theta} \tag{1}$$

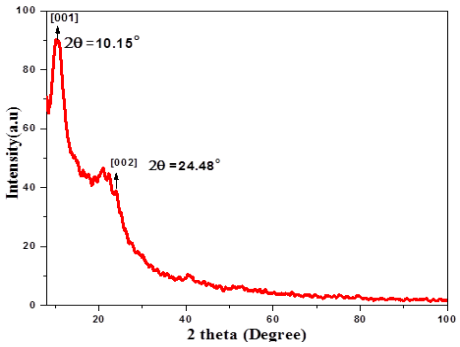

Fig. 1.XRD spectrum of GO nanomaterial layer.

Table 1.Shows crystallite size of GO on different peaks.

Nanomaterials	Peak (001) nm	Peak (002) nm	Average crystallite size (nm)
GO-NPs	7.88	7.89	7.885

3.2. SEM analysis

Fig. 2 shows the surface morphology of 2-D graphene oxide (GO) nanomaterials had multilayered surface and indicate randomly crumpled silk veil waves like surface morphology. Furthermore, its shape randomly crumpled like surface due to oxidation and the layers arranged on one another in the form of rocked area. It can also be noted that the GO nanomaterial layers were highly concentrated (oxygen) at the edges. This is due to oxygen based functional groups mostly attached at the ends of crystal structure [18, 19].

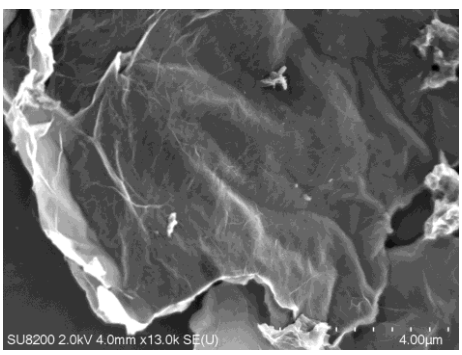

Fig. 2.SEM micrograph of GO-NPs.

3.3. FTIR analysis

FTIR was used to study the transmittance of different rotational and vibrational functional groups attached on the surface of GO layers. Figure (3) indicates the fabricated GO nanomaterial layer have peaks at 864.92 cm^{-1} and 1200 cm^{-1} which was ascribed to the C-O (stretching vibration) bond. The peak at wavelength of 1611cm^{-1}indicated that the C=C (vibrational) bond still continued before and after the oxidation procedure. While peaks at 2066.32 cm^{-1}, 2302.35 cm^{-1} and 2603.62 cm^{-1} indicated the presence of CO_2 and C-H(stretching vibration) functional groups. The absorbed water in GO nano-material layer was shown by a broad peak at 3361.55cm^{-1} to expressed by the O-H functional group due to molecules of water [20, 21]. These provisions the statistic that GO nanomaterial layer was an extremely absorptive material, as confirmed by its capability to become a gel-like solution.

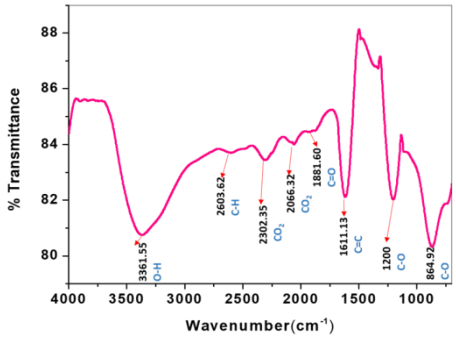

Fig. 3. FTIR spectrum of GO nanomaterial layer.

3.4. UV-visible spectroscopy analysis

The UV-visible spectroscopy was used to study the absorption spectra of GO nanoparticles. Fig. 4, indicates the absorption spectrum of GO near 240 nm wavelength and the absorption peaks shift toward longer wavelength by increasing oxidation. After that this pattern shift toward red shift and the yellow color of solution change into brown color [22].

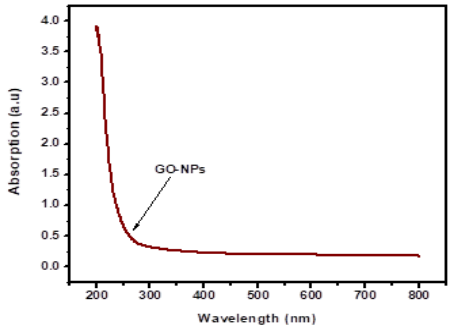

Fig. 4. UV-VIS spectrum of graphene oxide (GO)–NPs.

3.5. Voltammetry cyclic of GO for detection glucose detection

The electrochemical nanobiosensor was used to study the detection of glucose. Figure (5) voltammetry cyclic of (A) Bare in PBS (B) GO in PBS (C) Bare in glucose (D) GO in glucose was observed. Cyclic of bare and modified graphene oxide working electrodes were recorded in the presence of PBS at scan rate of 50 mVs^{-1}.Then there is no prominent change in current observed. After that cyclic of bare and modified graphene oxide working electrodes were recorded in the presence of glucose with concentration 5mM at scan rate of 50 mVs^{-1}. Cyclic current response was increased in the presence of glucose compared PBS which shows that synthesize material was electro active material. Similarly this process is repeated in case of glucose and GO with glucose

[23]. The graphene oxide with glucose shows the broad area as compare to others, then it is clear that GO with glucose indicate the large sensitivity response as compare to other voltammetry cyclic.

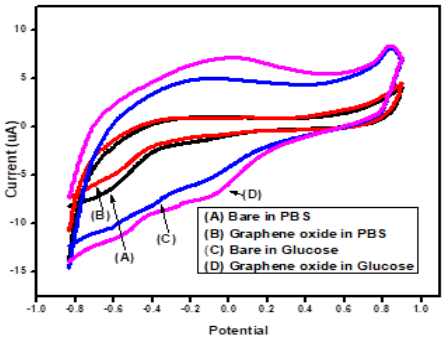

Fig.5.Voltammetry cyclic for the detection of glucose.

4. Conclusion

In this research work, we have prepared the GO-NPs by Hummers method. These NPs were characterized by using various characterization techniques such as XRD, SEM, FTIR and electrochemical biosensor for the detection of glucose. The hexagonal crystal structure and crystallite size (7 to 8 nm) and red shift appear by increasing the oxidation was calculated XRD and UV-Vis spectrum. Randomly crumpled like surface morphology and different functional groups attached on GO-NPs were calculated by SEM and FTIR analysis. Therefore, we used GO nanoparticles to examine potentiometric measurements at different concentration of glucose. We used GO/ethanol/Nafion/GCE as a working electrode and platinum as reference electrode respectively. After the detection of glucose concentration plot a graph between potential vs current. Finally, graphene oxide with glucose indicates the broader area express greater sensitivity of glucose as compare to other voltammetry cyclic.

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