Detection of E.coli O157 in water and food using nanosensor

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Abstract

The bacterial pathogens within the water and meat can be recognized utilizing progressed, expensive and time-consuming apparatuses. In addition to these gadgets, nanostructure-based gadgets can be utilized as both are cheap bacterial sensors. This investigation illustrates the creation of a sensor utilizing ZnO-rGO nano composite to distinguish bacterial pathogens. Escherichia Coli O157:h7, a Gram-negative bacterium are displayed in water and nourishment. The sensor was made of an ostensible composition of ZnO-rGO nano composite. Lean film was arranged by a sol - gel strategy utilizing the turn coating onto glass substrates and tests were carried out at room temperature. At that point the sensor was tried with some known concentration of microscopic organisms blended within the water and meat extricate. When a sample of bacteria mixed with water and meat extricate was dropped onto the sensor, the electrical resistance of the sensor varied proportionally with the concentration of bacteria. They proved as reliable and sensitive sensors for detecting bacteria in water.

1. Introduction

Food and water-borne pathogens, most of which are bacteria, enter our bodies through sullied nourishment and water. This drives to various dangers extending from gentle to critical efficiency misfortunes. Escherichia coli (E. coli) O157: H7 is the foremost serotype of E. coli creating Shiga poisons as detailed (STEC) by food-and-mortality clinics(1). Hamburger in most nourishment disease flare-ups has been criminalized by E. coli O157 as was distinguished by Centres for Disease Control and Anticipation (2). But other sources such as drinking water, dairy products, and vegetables too have been studied as detailed in (3, 4, 5, 6). Wellbeing issues that can be caused by E. coli O157: H7 infection range from mild watery diarrhoea to life-threatening conditions such as uremic hemolytic disorder and hemorrhagic colitis particularly in children and the elderly (7). Given the health risks of E. coli O157: H7 bacteria and their effect on food safety, sensitive detection strategies are fundamental to monitor food and water contamination to protect buyers from the hazard of food borne dangers. As of now available strategies for detecting E. coli O157: H7 that depend on agriculture and after that
biochemical and serological examination more often take two days to complete, where molecular biology-based methods may be required to confirm. However, these traditional methods are completely reliable. They are not simple to utilize since they require well-trained specialists and generally complex research facility hardware, which costs tall (8, 9). The ZnO based nano sensors were found to the capacity to distinguish diverse sorts of microscopic organisms. Besides, ZnO has tall electrochemical steadiness, non-toxic, can be effectively doped and is conservative compared to others semiconductor nanostructures [10]. Subsequently, in this research paper ZnO-rGo were created and tried. Its adequacy in recognizing microscopic organisms was demonstrated to be valuable. A reasonable microscopic organisms locator sensors of microbes were found within the water. Planned nano particles ZnO based biosensor can identify bacteria gram-positive microscopic organisms, as portrayed in this paper.

2. Materials and methods

The materials used for the preparation of ZnO-rGO nanocomposite; zinc nitrate hexahydrate, polyvinyl alcohol, Graphene oxide was supplied by Oxoid, India. Microscope slides and nutrient broth were purchased from Oxoid, India. De-ionized water of the fabrication and testing steps for the bacteria nano biosensor used in this study were also purchased from Oxid, India. The detailed procedures for the fabrication and testing are described in the following sections. Other materials used include zinc acetate.

Fabrication of the bacteria sensor

First a glass piece of 20 mm x 20 mm x 1 mm is taken and then the catcher (assembly) is prepared. And it is washed and cleaned with distilled water and with an ultrasound cleaning device for 20 minutes and washed with ethanol.

![Figure (1): The shape of the base material illustrates the base used in the manufacture of the nano sensor](image)
Preparation of solutions

Zinc salts were dissolved in ethanol of 0.42 grams equivalent to 0.1 molar, and polyvinyl alcohol salts were dissolved in 20 ml of anionic water at a temperature of 70-80 °C according to the following equation. [11]

\[ M = \frac{W_t}{M_{wt} \times \frac{V}{1000}} \]

The solutions were mixed at a constant speed and at room temperature to obtain a homogeneous solution. Then graphene oxide was added at 1% of the total volume of the solution, and then the mixture was placed on the magnetic spinning device for the purpose of obtaining a homogeneous mixture and ensuring the distribution of graphene material in the solution.

A number of pre-prepared glass bases were coated and cleaned with a paint solution by using the Sol-Gel method with a rotating coating system from Laurell Technologies, at a speed of 2000 rpm in 15 seconds for one minute.

The samples were dried at a temperature of 100 °C for a period of 10 minutes using the oven after each coating to get rid of the solvent and to remove the organic residues. By repeating the process (coating) above five times, 5 layers of paint were obtained. After completing the five layers, we would get a more homogeneous and thicker coating layer. The annealing process took place at a temperature of (350) degrees Celsius for one hour in the oven to achieve the crystalline phase, as well as for fixing the coating layer and obtaining a homogeneous coating layer [12].

Manufacture of the sensor component

The copper wires were associated to the silver anode as an association between the lean film and the measuring instrument of the lean film sensor prepared with comb-type cathodes. The wires were settled with silver glue fabric on the anodes stored on the glass pieces.

The process of preparing bacteria

The E. coli were chosen as a bacterial test. 4 gm of supplement broth powder was broken up in 500 ml of distilled water, and blended until a homogeneous arrangement was made. The broth was sterilized utilizing autoclave at 121 °C for 15 minutes. From separated essential agar dishes, unused colonies were vaccinated in supplement broth (10 ml) and brooded for 24 hours at 37 °C. After that, 5 ml of separate culture (10 ml) were weakened to 250 ml utilizing supplement broth and hatched with shaking at 200 rpm for 24 h at 37 °C. CFU was calculated by a practical checking strategy. The concentration of E. coli utilized in this explore was steady (108 cfu / ml) for each sensor.

25 grams of ground meat was bought from the nearby advertise. 10 grams of meat was taken and blended in 90 ml of refined water and sterilized at a temperature of 120 -180 degrees Celsius for one hour. The confines of E. coli were actuated on the agar supplement medium and the segregates were brooded for (24) hours within the incubator. The meat was sifted by channel paper. The sifted fluid of ground meat and water was vaccinated with bacterial segregates of E. coli. 500 ml of the supplement
medium for the microbes Supplement broth was arranged and 250 ml of E. coli microscopic organisms were immunized. At that point the media vaccinated with the bacteria was put within the hatchery for 24 hours at a temperature of 37 ° C for the development of bacterial colonies. After that, 108 bacteria were arranged as a concentration of microscopic organisms within the water and meat.

**Detection for Escherichia coli O157**

The sensor was inspected with a Keithley electrical conductivity device by measuring the electrical conductivity of the thin film at room temperature. Then they were altered within the electrical conductivity properties with Ohm contacts utilized, to get ready the sensor for Escherichia coli. Since the thickness of the vacuum-deposited films is 1000 ° C, estimations at low-voltage voltages (<4 V) were required.

3. Result and Discussion

Structural and surface properties of zinc oxide films with reduced graphene oxide and PVC nanostructures.

The results of X-ray diffraction (XRD) of zinc oxide nano particles prepared on glass substrates as in Fig. (2) shows that zinc oxide is multi crystalline in nature and its crystal structure is hexagonal and in conformity with (JCPDS card No. 36-1451). The main peaks of the substance appeared at 2θ = 31.45°, 34.1°, 36.05°, 45.95°, 56.8°, 60.05°, 66.2°, 67.45°, 70.1° and 71.1°, represented by the levels (100), (002), (101), (102), (110), (103), (002), (112), (201) and (004). This is consistent with previous studies [12].

![X-ray diffraction pattern](image)

**Figure (2):** shows the X-ray diffraction pattern of thin zinc oxide nano films prepared on glass substrates.

It is evident from Fig. (2) that the natural location of the characteristic peaks (100) and other peaks have been driven as reported by [13] the high temperature annealed film will show all sharp peaks of
the printing oxide and no other phase, making it a marker to the last formation. The oxide structure is higher crystallization.

The crystal sizes of the ZnO membrane models were calculated using the Debi-Sharr equation.[14]

\[ D = \frac{k\lambda}{\beta \cos \theta} \]

**Surface structure analysis**

The results of atomic force microscopy showed the surface structure of zinc oxide films with graphene oxide and thin polyvinyl alcohol nano particles with a homogeneous and smooth surface structure. Figure (3) shows atomic force microscopy images of zinc oxide thin films with graphene oxide and prepared polyvinyl alcohol nano particles. On glass substrates, where, from the results of an atomic force microscope examination, the average grain size was obtained (45 nm).

![Atomic force microscopy images](image)

**Figure (3):** shows atomic force microscopy images of zinc oxide thin film with graphene oxide and polyvinyl alcohol nano particles prepared on glass substrates, (A) 2D, (B) 3D, (C) Modulus of granular sizes.

**Optical properties of zinc oxide film with graphene oxide and PVC nanostructures prepared on glass substrates**

The absorbance of zinc oxide films were studied with graphene oxide and polyvinyl alcohol prepared on glass substrates. They were recorded within the wavelength from (200-1000 nm) at room temperature.
Absorption of Zinc Oxide Thin Films on Glass Substrates

The optical absorption depends on the surface and crystalline composition, thickness and surface morphology of the films.

![Absorbance graph](image)

Figure (4) shows the absorbance of a thin zinc oxide film prepared on glass substrates.

Electrical properties of ZnO - rGO, PVA / glass thin films

Current voltage measurements

Figure (5 - 6) shows the current-voltage characteristics (I-V) of ZnO - rGO, PVA / glass thin film preparation.

The interaction of electrical charges on the cell wall of the microbe with suspended bonds of polymeric content, especially in thin films, is the basis for detection of *Escherichia coli* by the polymeric sensor. Since the cell wall of the microorganism is charged, it can be concluded that the microorganism has an electrical charge [15] Figures (7-8) shows the current (IV) voltage characteristics of ZnO-rGO and PVA / Glass thin films with and without exposure to *Escherichia coli* at room temperatures. The results can be explained by shifts in charge carrier concentrations around ZnO-rGO, PVA / thin film. Vitreous after exposure to microorganisms affect the properties of IV. When the sensor membranes are exposed to water and meat containing *Escherichia coli*, the metal ions on the surface of the polymeric film react with the bacteria. If the sensor is exposed to *Escherichia coli* and salmonella, it causes the output current to increase. The positive charge of Zn$^{2+}$ and G$^{2+}$ can be linked to negative *E. coli* [16, 17]. Zn + 2 released from nano particles bind to negatively charged bacteria. Cell wall (*Escherichia coli*) bursts. According to [18] by the release of Zn$^{2+}$ upon surface oxidation, or by electrostatic interactions between the emitted ions and the negatively charged bacterial cell wall, the ZnO nano composite can interact directly with the bacterial cell membrane.
Figure (5) shows the current-voltage characteristics (I-V) of preparing Zinc oxide with reduced graphene oxide and polyvinyl alcohol glass to thin detect E. coli on in water.

Figure (6) shows the characteristics of the current voltage (I-V) for preparing Zinc oxide with reduced graphene oxide and polyvinyl alcohol thin glass on the detection of E. coli in meat.

4. Conclusion

A sol gel process is used to deposit ZnO-rGO doped PVA thin films on glass substrates. The films have outstanding composition, morphology, and electrical properties. The appearance of a peak in the concentration suggests high crystallinity according to XRD. The prevalence of E. Coli in the water and meat was measured by calculating variations in thin film conductivity using I-V tests, which were observed.
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