A review on application of ZnO nano particles as biosensors

A. Barman*1, S. Bhattacharya1, R. Majumder2
1 Asst. Professor, Dept. of Chemistry, JIS College of Engineering, Kalyani, West Bengal, India
2Electronics and Communication, JIS College of Engineering, Kalyani, West Bengal, India

*Corresponding Author Email: ananya.barman@jiscollege.ac.in

Abstract. Recently some of the metal nano composites are used in the semiconductor based electrical, electrochemical and optical bio-sensors. ZnO nano particles have drawn great interest due to its unique properties like low cost, wide range of band gap, biocompatible, easy to synthesize in different size and shapes and catalytic surface activity. The surface of the ZnO nano particles are perfect for immobilisation of bio molecules without hampering their biological activity. This property increase the sensitivity of bio-sensors. In this review we highlight the use of different ZnO nano particles in bio-sensors enhancing the efficiency of Bio-sensors.

Keywords: ZnO nano particles, Bio-sensors, Bio compatibility

1. Introduction

Now a days nano materials are used in bio-sensors which is highly selective and sensitive towards the bio-molecules [1]. The nano-composite materials with different shape and size are remarkably different from conventional bulk materials. The unique molecular recognition of nano materials can enhance the sensitivity of bio-sensors [1]. The metal oxide nano composites have drawn a great attention in the field of bio-sensor due to its specific properties like bio-compatibility, controllable size and shape, strong adsorption ability and ease of fabrication [2].

ZnO nano particles are very useful in different field. Due to its UV-absorption capability and antibacterial effect, it is used in personal care products like cosmetics, sunscreen [3] and also in textile industries [4]. The incorporation of ZnO nano materials in rubber materials, can provide wear proof of the composite materials and also enhance some of the properties like toughness, anti-aging [5, 6].

ZnO is an n-type semiconductors with wide range band gap of 3.37eV and large binding energy of 60meV and high electron mobility. Thus many research work has focused to utilize the ZnO nano particles in the field of biomedical and clinical purpose. Some of the unique property of ZnO nano particles like bio-compatibility large surface area, high film forming capability, improved catalytic activity, chemical stability, corrosion resistant, different grain size and shape and strong adsorption ability, make these nano particles suitable for bio-sensors [7-13].
There are two platforms, electrical [14-21] and electrochemical, [22-32] are used for the developed bio-
sensors. For the electrical method Field Effect Transistor, FET, are used which provides high level
detection. But for this method is expensive and complex as this requires input of lithography and nano-
technology. Electrochemical methods also provide high level of detection and compatible for mass
production. Recently the optical bio-sensors has drawn the attention of many research work, which is
based on absorbance, photoluminescence and Plasmon resonance. For the high precision, it is expected
that optical bio-sensors are the next generation of sensing device.

Now a days, some of the metal oxides are mixed to the ZnO nano materials to make composites for the
use of bio-sensors. These composites are better than the pure ZnO nano materials and show high
catalytic power, surface to volume ratio and other improved properties.

2. Metal oxide modified ZnO nano sheets based biosensor:

The surface of the used ZnO nano composites should be immobilized capture probe for detection of bio-
marker. The higher the stability of this capture probe immobilized on ZnO surface, the higher will be
selectivity and sensitivity .The mechanism of capture probe immobilization follows via covalent bond
formation with organic molecules, called cross-linker or by physical adsorption. High IEP of ZnO
facilitates physical adsorption of low IEP enzymes such as glucose oxidase, cholesterol oxidase, etc., on
ZnO nanostructures through attractive electrostatic force.

2.1. Electrochemical based non-enzymatic biosensor

The surface modification of ZnO nano particles are required with different transition metal
oxides like CuO, Fe₂O₃, TiO₂, Co₃O₄, NiO, NiWO₄, etc as ZnO is not considered as a suitable
candidate for non-enzymatic bio-sensors. Some metal oxides are also used to synthesize nano
composites to catalyse the target analyte.

In a research work, Zhou et al. deposited CuO- ZnO nano composites on tin oxide, doped by
fluorine. It is porous and have 3 dimensional morphology for the fabrication of non-enzymatic
bio-sensors., by using spinning methodology [33]. This ZnO–CuO nano composite/FTO bio-
sensor shows increase in sensitivity of 3066.4 μA mM⁻¹cm⁻² and 0.21 μM detection limit
(Figure. 1). The nano composites which are grown vertically array type, have larger surface area
and improved sensitivity. In another research work, Soejima et al. synthesized CuO-ZnO nano
arrays on the brass plates at low temperature, by using facile one step reaction [34].
Figure 1: Schematic illustration of fabricated electrode and glucose detection mechanism over 3D porous ZnO–CuO HNCs surface. [Reference: Tripathy, N., Kim, D. H., (2018), Nano Convergence, https://doi.org/10.1186/s40580-018-0159-9]

The nano composites, based on non-enzymatic sensor electrodes, are electro-catalytic and responsible for fast response and high sensitivity in glucose oxidation reaction. This catalytic activity can be improved by using NaOH buffer solution. The synergistic effect of CuO nano leaf and ZnO nano rods attributed towards high electro active surface, low working potential and stable responses.

Cai et al. had followed a different reaction path way to synthesize porous ZnO–CuO nano composites to improve the surface area of nano composites. He choose a simple controllable top-down method and synthesized core shell spheres, ZnO-CuO, which was porous [34]. By morphological study, it was revealed that a core shell of CuO was surrounded by porous ZnO. These nano composites showed high catalytic activity of CuO and high electron transfer property of ZnO, responsible for the enhanced sensing performance.

If the ZnO nano rods are grown vertically, in a direct process, at low temperature, it can offer excellent binding sites for CuO nano particles. A method of the synthesis, modification of nano composites, and mechanism of detection is shown in Figure. 2a. The impedance spectra of the synthesized non-enzymatic sensor were measured before bio-sensing characterisation (Figure. 2b).
2.2. Non-enzymatic FET based biosensor:

Nowadays, field-effect transistor (FET) based biosensors are used vastly as they have low fabrication cost, small in size and easy to operate. Surface modified ZnO nano particles with metal oxide are also used in FET based biosensors. In a research work, Jung et al. had used NiO quantum dots (QDs), for the surface modification of ZnO nano rods and used in non-enzymatic FET based glucose bio-sensors [35]. If ZnO nano particles are used only in the bio-sensors, they cannot catalyse the glucose oxidation, resulting in poor response current. So Ahmed et al. [36], modify the surface of vertically oriented ZnO nano particles by Fe$_2$O$_3$ nano particles which helps in the heterogeneous chemical oxidation or reduction of glucose. ZnO nano particles attributes large surface area for catalyst loading.

2.3. Enzymatic FET based biosensor:

Now a day’s, to increase the sensitivity of ZnO nano particles, with the surface modification of ZnO nano particles by metal oxide, also these nano particles are functionalized with some specific enzymes or selective membranes. Ahn et al had chosen valinomycin for immobilization on Fe$_2$O$_3$ modified ZnO surface in a FET based potassium sensor [37]. After this modification with the enzymes, the sensitivity enhanced due to the immobilization of valinomycin in a large
surface area of Fe₂O₃ modified ZnO surface. After the surface modification, the ZnO surface also stable in acidic and basic solutions, which is important for the detection of potassium.

In some other research work, Ahmad et al fabricated sandwich like structure where the layers were highly conductive by using first layer of ZnO seed layer, after that second layer was Ag nanowires and then again ZnO seed layer was used. It was revealed that incorporation of Ag nanowires, the sensitivity of the device was increased.

2.4. ZnO nanostructure-based electrochemical biosensor for Trichinella DNA detection:

ZnO nano particle based biosensors were also used for the detection of Trichinella DNA. Trichinosis is a zooantroponosis. After infection, the parasites were located in the host's intestine and encapsulated in the muscle tissues, which is hard to destroy for any medicine.

In electrochemical based bio-sensors were constructed by two electrodes, one as working electrode and other one as counter electrode. The working electrode was 3mm Ni plated metal, which is coated with ZnO nano particles and the counter part was 0.3mm thick Pt/Ir wire. (Figure. 3).

Experiment has been done to compare which ZnO nano particles are effective as a coating material to enhance the sensitivity and efficiency of a bio-sensor.

1. Thin film of ZnO nano particles
2. The nano rods of ZnO, diameter of which was approximately 80mm
3. Nano tubes of ZnO, which showed best result to enhance the property of bio-sensors [38]. From the morphological study, it has been seen that nano tubes were more suitable due to its porous structure and large defects, produced during the etching process.

According to the SEM images, it was confirmed that the obtained structured coating was dense and homogeneous and suitable for vertically oriented nano structures. The DPV signal curves of primers
immobilization are shown in Figure 4. After comparing the sample containing DNA with the reference one (Figure 4 a–c), it can be concluded that DNA reduced the conductivity of the sample which is also shown in the reference curve of NaCl (dotted line) than NaCl + DNA curves (continuous and dashed lines). If we check the relative augmentation of the signal, for the case of immobilized DNA, we got 78% signal for nanotubes and 29% for nano rods (see Figure 4 d).

Figure 4. Experimental results of immobilization of Trichinella britovi DNA primers on ZnO surface with different morphology: (a) DPV response from device with ZnO nanotube surface, (b) DPV response from device with ZnO nanorod coating and (c) DPV response from device with homogeneous ZnO thin film. Part (d) illustrates the relative augmentation of signal for different surfaces after immobilization of primers. Figure (e) illustrates the relative change of signals between previously immobilized and non-immobilized DNA for different morphologies. (Reference: Gerbredersa, V., Krasovskaa, M., Mihailovaa, I., Ogurcovsa, A., Sledevskisa, E., Gerbredersa, A., Tamanisa, E., Kokinab, I., Plaksenkovab, I., (2019), Sensing and Biosensing Research, 23).

ZnO nano particles attributed towards the development of large surface area of electrodes and also increased the adsorption bonds for the DNA immobilization. In presence of these nano particles, the degree of immobilization increased and hence sensitivity is also increased.

2.5. Glucose biosensor based on ZnO nanoparticles film:

In a research work, an electrochemical glucose bio-sensor was designed, where 1-ethyl-3-thylimidazolium trifluoromethanesulfonate; [EMIM][Otf]), ZnO nano film and egg shell membrane were deposited on glassy carbon electrode. With the help of cross linker, glutaraldehyde, glucose oxidase (GOx), can covalently immobilized on egg shell. Both oxidation and reduction reaction took place while substrate interact with enzymes. Methylene blue was used as redox indicator which increased the electron transfer capacity and also stabilized the both oxidized and reduced form produced in the reaction. From the morphological study, it was seen that ZnO nano particles were deposited onto ESM and the GOx is trapped by ZnO nano particles scattered on the fiber. [EMIM][Otf] deposited on ESM. While GOx was immobilized through the membrane, the rough fiber interacted with the cluster of lumps of GOx.
In this case ionic liquid provided consignment boundary for the immobilization of GOx and ZnO nano particles provided a nano sized environment which was helpful for effective electron transfer.

3. **ZnO-based electrical biosensors for cardiac biomarker election:**

ZnO semiconductor has wide range band gap and presence of two opposite charge stacked along the polar planes. Thus ZnO nano particles can resist high temperature, large electric field and high breakdown voltages and high power operation [39]. In ZnO crystal, presence of interstitials and oxygen vacancy, it shows n-type conductivity [4]. Due to the capability of charge transfer between semiconductor and the electrolyte at the interface, ZnO is vastly used in electrical or electrochemical bio-sensors.

Cardiovascular disease, is one type of heart disorder, where not only the coronary artery is affected, but also the entire arterial circulation is hampered. The sensitivity of ZnO nano rods based bio-sensors depends on the crystalline structure, shape and size of nano particles. The robustness of the electrode depends on the density of ZnO nano rods [41]. At the sensing platform, ZnO nano rods are acting as nano electrodes and facilitates the diffusion of cardiac troponins to its surface. This attributes towards the enhancing of sensing ability [42].

4. **Optical biosensors based on ZnO nanostructures:**

Optical transducer system is used for the optical bio-sensor and the bio-sensing element is connected with the transducer. The mechanism of this type of biosensors are based on adsorption, luminescence or reflectance [43]. Generally, there are two types of bio-sensors are there, direct optical detection and indirect optical detection. ZnO nano particles are used in this case as transducers.

5. **Conclusion:**

The efficiency, analytical performance and the sensitivity of a bio-sensors can be tuned by using nano particles of different structures, size and shape. More over the immobilization of bio-molecules at the surface of nano particles can be obtained with a selective fashion without losing biological activity of that molecule. Due to some unique electrical properties, ZnO nano film, nano rods, nano tubes are promising, to use in bio-sensors. Further modification of surface of these nano particles with some metal oxide or the enzymes, enhance the sensitivity of the bio-sensors. The nano scale ZnO experiences fast electron transport and thus sensitive to any physicochemical changes taking place on the surface even in low biomarker concentration. It has been proved that surface modification of ZnO nano particles by Ag or Au metal are most effective to increase the sensing capacity of a bio-sensor. Now a days, the surface modification of ZnO nano particles with carbon based nano materials like graphene, carbon nano tubes are done to enhance the efficiency of electrical and optical bio-sensors.

6. **Reference:**

[1]. Mohammed, M. T. Desmulliez, M. P. (2011), *Lab Chip, 11,* 569–595.
[2]. Hahn, Y.B., Ahmad, R., Tripathy, N., (2012), *Chem. Commun.*, 48, 10369–10385
[3]. Newman, M. D., Stotland, M., Ellis, J. I., (2009), *Journal of the American Academy of Dermatology*, 61 (4), 685–692.
[4]. Hwaa, k. Y., Subramani, b., (2014), *Biosens Bioelectron.*, 62 127–133.
[5]. Zhao, Y., Deng, F., Nie, Y., Wang, p., Zhang, y., Xing, l., Xue, X., (2014), *Biosens Bioelectron.*, 57, 269–275.
[6]. Choi, A., Kim, K., Jung, H. I., Lee, s. y., (2010), *Sensors and Actuators*, B 148, 577-582.
[7]. Wang, X., Yu, H., Lu, D., Zhang, J., Deng, w., (2014), *Sensors and Actuators B: Chemical*, 195, 630–634.
[8]. Munje, R. d., Jacobs, m., Muthukumar, s., Quadri, b., Shannugam, N. R., Prasad, S., (2015), *Anal. Methods*, 7, 10136–10144.
[33]. Zhou, C., Xu, L., Song, J., Xing, R., Xu, S., Liu, D., Song, H., (2014), Sci. Rep. 4, 7382–7391.

[34]. Soejima, T., Takada, K., Ito, S., (2013), Appl. Surf. Sci. 277, 192–200.

[35]. Jung, D. U. J., Ahmad, R., Hahn, Y. B., (2018), J. Colloid Interface Sci. 512, 21–28.

[36]. Ahmad, R., Ahn, M. S., Hahn, Y. B., (2017), Electrochem. Commun. 77, 107–111.

[37]. Ahn, M. S., Ahmad, R., Bhat, K. S., Yoo, J. Y., Mahmoudi, T., Hahn, Y. B., (2018), J. Colloid Interface Sci. 518, 277–283.

[38]. Krasovska, M., Gerbreders, V., Mihailova, I., Ogurcovs, A., Sledevskis, E., Gerbreders, A., Sarajevs, P., (2018), Beilstein J. Nanotechnol. 9, 2421–2431.

[39]. Elliot, T. B. (2005), Trends in Semiconductor Research, Nova Publishers.

[40]. Jagadish, C., Pearton, S. J., (2011), Elsevier, Amsterdam, the Netherlands.

[41]. Shanmugam, N. R., Muthukumar, S., Selvam, A. P., Prasad, S., (2016), Nanomedicine, 11(11), 1345–1358.

[42]. Jacobs, M., Muthukumar, S., Panneer, S. A., Engel, J. C., Prasad, S., (2014), Biosens. Bioelectron. 55, 7–13.

[43]. Patel, P. N., Mishra, V., Mandloi, A. S., (2010), JERS, 1(1), 15-34.