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Mini Review

# **Graphene Nanoribbons in Electrochemical Sensors and Biosensors: A Review**

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Recently, graphene nanoribbons (GNRs) are narrow strips of graphene sheets with width in nanometers (<50 nm) and they are becoming attractive material in a variety of electrochemical applications owing to their outstanding electronic and catalytic properties. The most fascinating property of GNRs that distinguish them from other members of graphene family is their higher area-normalized edge-plane structures. In addition, GNRs have large surface area, high conductivity, residual oxygen functionalities, and accessible sites for catalysis, biocompatibility and good stability. Therefore, GNRs based nanomaterials are recently getting much popularity in electrochemically sensing and biosensing applications. In this review, we are presenting a summary of recent reports on GNRs based nanomaterials for electrochemical sensors and biosensors.

**Keywords:** Graphene, Layered nanomaterials, Electroanalytical chemistry, Electrochemical sensors and biosensors, Modified electrodes

# **1. INTRODUCTION**

Over the past years, carbonaceous materials, such as graphite, diamond, fullerene, carbon nanotubes, amorphous carbon, active carbon, carbon black, and graphene have been widely employed

in different applications owing to their interesting physico-chemical properties [1, 2]. Most recently, graphene based materials are extensively applied in electrochemical applications including, sensors, biosensors, batteries, supercapacitors, fuel cells, and solar cells [3-6]. Recently, graphene nanoribbons (GNRs), narrow strips of graphene nanosheets with width in nanometers (<50 nm) have become attractive material in a variety of applications particularly in electronics owing to its outstanding electronic and spin transport properties [4, 7]. GNRs possess quasi one dimensional (1D) structure in nature, which lies between 1D structure of carbon nanotubes and 2D structure of graphene and the final structure is predominately depends on the degree of multiwalled carbon nanotubes (MWNTs) unzipping. The band gap significantly varies as the width of GNRs is minimized. GNRs display a finite band gap when their width is less than 10 nm which make it interesting materials for carbon based nanoelectronics [4, 8]. The electronic states of GNRs largely depend on the edge structures, wherein zigzag edges give rise to metallic, while armchair edges give rise to semiconducting/metallic properties [9].

#### 2. PREPARATION OF GRAPHENE NANORIBBONS

GNRs can be prepared by several methods including sonochemical [10], cutting form graphene using lithography [4, 11], chemical vapour deposition [12], chemically oxidizing/longitudinal unzipping CNTs [13], plasma etching [14], ionic liquid-assisted splitting of CNTs under microwave irradiation [15], lithium intercalation/exfoliation of carbon nanotubes [16], longitudinal cutting method using metal clusters as nanoscalpels [17], and microwave assisted synthesis methods [18]. The recent progress in fabrication techniques of graphene nanoribbons on a substrate are reviewed [19]. Although, major portion of GNRs research is focusing on its applicability in nanoelectronics, spintronics, optoelectronics and nanoelectromechanical systems [20], they are also attractive material in electrochemical devices such as, batteries [21], supercapacitors [22, 23], solar cells [1, 8], fuel cells [24], microbial fuel cells [25], sensors [26], and biosensors [27]. Unlike graphene sheets that feature a zero band gap, GNRs own an open band gap. Designed edges of GNRs could effectively modulate the electronic energy gap, which hold significant impact on chemical and biological detection [28]. GNRs have large surface area, high electrical conductivity and electrochemical stability, which make this suitable for electrochemical applications [29]. In any graphitic materials, the electrochemical reactivity at edge planes is several orders of magnitude higher than that at basal planes. Thus, rich edge defects are responsible for the fast electron transfer process. GNRs have the highest edge density over all the other graphene-based materials [30]. Highly expensive high-tech instrumentations are required to prepare high quality GNRs with controlled widths.

Among all the reported methods, longitudinal unzipping through acid-treated oxidation MWNTs is the suitable method for the mass production of GNRs with low-cost. This method of GNRs production creates plethora of structural defects that are actually beneficial for electrocatalytic sensing applications. The functional groups located at the edges of GNRs facilitate the adsorption of analytes by  $\pi$ - $\pi$  stacking, electrostatic, hydrogen bonding, and covalent interactions [31]. Interestingly, GNRs

have higher area-normalized edge-plane structures and chemically active sites compared with either MWNTs or graphene; as a result, they can be better electrocatalysts in electroanalytical methods.

#### **3. GRAPHENE OXIDE NANORIBBONS**

Graphene oxide nanoribbons (GONRs), oxygenated derivative of GNRs are the intermediate compound in the longitudinal unzipping method. GONRs are amphiphilic in nature. The oxygen functionalities present at the basal and edge planes of GONRs allow them to form aqueous dispersion, while aromatic regions of GONRs allow them to form organic dispersion. On the other hand, the aromatic regions offer sites for non-covalent interactions and functionalization and oxygen moieties provide sites for functionalization [32]. GONRs can be reduced to GNRs via electrochemical, thermal and chemical methods. GNRs have high electrical conductivity, chemical stability and high specific surface. In addition, GONRs exhibits good biocompatibility and enhanced sensitivity, which makes them as qualified substrate for enzymatic biosensing applications.



Figure 1. TEM image of MWCNTs (A) and GONRs (B). SEM images of GONRs (C and D). (Reproduced with permission from ref. [35] Copyright 2003, The Electrochemical Society)

The abundant oxygen functionalities, such as hydroxyl, epoxide, carbonyl, and carboxyl groups located on the surfaces and edges of GONRs offer great potential for attaching biorecognition molecules such as antibodies [33]. The carboxyl groups located on GONRs can be cross-linked to the amine groups of proteins through well-established carbodiimide crosslinker chemistry. Recently, our research group described a GONRs modified screen-printed carbon electrode (SPCE) for folic acid detections [34]. Our studies revealed that the GONRs exhibited superior electrocatalytic ability over MWNTs ascribed to their rich edge chemistry and catalytic properties.

In another report, we described GONRs modified SPCE for sensitive determination of methyl parathion (Figure 1) [35]. GONRs/SPCE displayed significantly improved electrocatalytic ability towards methyl parathion in comparison with MWCNTs. The modified electrode was shown excellent real-time applicability in food analysis, which was successfully demonstrated in food samples such as, Ugli and tomato fruits, Beetroot and Broccoli indicating its excellent practical applicability.

## 4. GNRs BASED NANOMATERIALS IN ELECTROCHEMICAL SENSORS

Several GNRs based nanomaterials have been developed in recent times for electroanalytical sensing applications [29]. The hybridization of GNRs with metal nanoparticles can provide nanocomposites of synergic properties and this behaviour is similar to the graphene-based composites [26, 29].



**Figure 2.** (A–C) HR-TEM micrographs of GNRs supported PtPd concave at different magnifications. (C inset) shows the FFT pattern of an specific PtPd nanocrystal. (D) High-angle annular dark-field (HAADF)-STEM image of GNRs/PtPd and corresponding elemental mapping of Pt (E), Pd (F), and the overlay (G). (Reproduced with permission from Ref. [26] Copyright 2015 American Chemical Society) Besides, GNRs can be a good support for making nanoparticles dispersions because of their large surface area, high electrical conductivity, and electrochemical stability in acidic and alkaline electrolytes. For instance, highly stable and catalytically active PtPd concave nanocubes were anchored on GNRs through a hydrothermal process [26]. the TEM image of GNRs/PtPd revealed that the average particle size of PtPd concave cubes was 11 nm and the particles are uniformly placed on the surface of GNRs (figure 2).

Recently, we have described silver particles decorated GNRs prepared through simple wet chemical method and the resulting nanomaterial have shown excellent sensing attributes towards Organophosphorus pesticide methyl parathion [36]. A highly sensitive (sub-nanomolar level) methyl parathion sensor was fabricated using Ag@GNRs/SPCE and its practical applicability was successfully demonstrated in in food samples such as, cabbage, green beans, strawberry, and nectarine samples. The synergic effects between Ag and excellent physicochemical properties of GNRs make the composite highly suitable for pesticide sensing. A nanocomposite of polyaniline (PANI)-GNRs was prepared by *in situ* polymerization of aniline in presence of GNRs [37]. Using this cost-effective method, highly ordered and vertically aligned PANI nanorods can be grown on GNRs (Figure 3).



**Figure 3**. SEM images of PANI-GNRs composite at (a) low resolution and (b) high resolution. TEM image of PANI-GNRs composite (c) and (d). (Reproduced with permission from Ref. [37] Copyright 2013 American Chemical Society)

Several GNRs based nanomaterials have been prepared and demonstrated in electrochemical sensing applications; polyaniline nanorods grown on GNRs [37], GNRs supported PtPd concave nanocubes [26], PdNi nanoparticles and N-doped GNRs [27, 38], graphene quantum dots (GQDs)

supported GNRs [39], Ag/GNRs composite[40], PtPd/GNRs alloy [41], GNR-Pt nanocomposite [24], PdAg alloy nanoparticles modified ionic liquid functionalized GNRs [42], GNR/Co coordination polymer nanohybrids [43], GNRs/polyaniline [44], GNR/polypyrrole [45], GONRs/Au nanoparticle hybrid [46], GONR/poly(diallyldimethylammonium chloride)/Au nanoparticle [47], GNRs/nafion nanocomposite [48], Pd-functionalized GNRs [49], heteroatoms doped GNRs [50], graphene sheet/GNRs [51], Core–shell MWNTs/GONR heterostructure [18], poly(l-arginine)- $\beta$ cyclodextrin/carbon nanotubes@GNRs [52], electrochemically reduced GONR [53], and molecularly imprinted polymer (MIP)–GNR composite [54].



Figure 4. TEM (a-c) and HRTEM (d) images of GQD-GNR hybrid. (Reprinted with permission from Ref. [39] Copyright 2015 American Chemical Society)

Graphene quantum dots (GQDs) Supported GNRs was developed via a one-step simultaneous reduction reaction and the resulting nanomaterial showed ultrahigh electrocatalytic performance for oxygen reduction reaction (ORR) [39]. The uniform distribution of GQDs and hierarchically structured GNRs are evident from the TEM image of the GNRs/GQDs (Figure 4). The resulting GQDs/GNRs

hybrid delivered higher limiting current density and lower overpotential than those of platinum and high selectivity and stability in alkaline media. An ultra-sensitive, reproducible, stable and practically applicable modified electrode based on electrochemically reduced (ER)-GONRs was fabricated for the determination of nimesulide, a non-steroidal anti-inflammatory drug [55]. The sensor has excellent sensor performance and exhibited wide linear range  $(1.0 \times 10^{-8} - 1.50 \times 10^{-3})$ , low detection limit (3.50 nM), high sensitivity (1.20  $\mu$ A.  $\mu$ M<sup>-1</sup>.cm<sup>-2</sup>) and fast response (<5 s) towards nimesulide detection. The assay demonstrated in drug and urine samples revealed the good practical feasibility of the ER-GONRs/SPCE in drug and clinical analysis.

A PtPd/reduced GONRs nanocomposite based modified electrode displayed excellent sensing performance towards trinitrotoluene (TNT) [26]. It exhibited a high sensing performance for TNT detection with a linear range of 0.01 ppm to 3 ppm and a detection limit of 0.8 ppb through adsorption stripping voltammetry (Figure 5)



**Figure 5.** (A) Electrochemical impedance spectra (EIS) obtained at PtPd-rGONRs/GCE, rGONRs/GCE and unmodified GCE in 0.1 M KCl containing 5 mM K<sub>4</sub>[Fe(CN)<sub>6</sub>]/K<sub>3</sub>[Fe(CN)<sub>6</sub>]. (B) Stripping voltammograms obtained at PtPdrGONRs/GCE, rGONRs/GCE, and bare GCE in 0.1 M phosphate buffer (pH =6.5) containing 0.4 M KCl and 3 ppm of TNT. (C) Stripping voltammograms obtained at PtPd-rGONRs/GCE with varied concentrations of TNT in 0.1 M phosphate buffer (pH = 6.5) with 0.4 M KCl. (D) Linear regression plot between peak current density and concentration of TNT concentrations at -0.33 V. The error bars represent the standard deviation of three individual measurements. (Reproduced with permission from Ref. [26] Copyright 2015 American Chemical Society)

A microwave-assisted unzipping process was reported for the rapid synthesis of GONRs [18]. The GONRs able to separate the voltammetric peaks of ascorbic acid, dopamine, and uric acid and the

corresponding peak separations were 229.9 mV (AA to DA), 126.7 mV (DA to UA), and 356.6 mV (AA to UA). The linear range of the resulting amperometric senor for AA, DA and UA were 0.1 to 8.5  $\mu$ M, 0.15 to 12.15  $\mu$ M and 0.15 to 11.4  $\mu$ M, respectively. The detection limits were of 0.06  $\mu$ M, 0.08  $\mu$ M and 0.07  $\mu$ M for AA, DA and UA, respectively, indicating excellent sensing attributes of the GORNs (Figure 6).



Figure 6. Amperometric responses of GONR modified glassy carbon electrode (GCE) after the subsequent addition of (a) AA (0.0 V), (b) DA (0.2 V), and (c) UA (0.3 V) in a 0.1 M phosphate buffer solution. Inset: enlarged curves of (a) 0.10 μM AA, (b) 0.15 μM DA, and (c) 0.15 μM UA. (Reprinted with permission from Ref. [18] Copyright 2011 American Chemical Society)

## **5. GNRs BASED NANOMATERIALS IN ELECTROCHEMICAL BIOSENSORS**

Meeting the demand for practical, cost-effective and portable analytical devices, enzyme-based biosensors have huge potential as useful tools in medicine, biofuel cells, food control, and in biomedical and pharmaceutical analysis [56]. The major limiting step in the performances of biosensor is immobilization of bioreceptors (enzymes, proteins, antibodies, aptamers, nucleic acid, cell, tissue etc.,) which refers proper coupling of bioreceptors to solid electrode surface [57]. In the past years, three kinds of immobilization matrixes were developed, (1) natural polymers, (2) synthetic polymers, and (3) inorganic [56]. Over the past years, carbonaceous nanomaterials are established as promising immobilizing matrix and they contributed enormously to the development of wide range of biosensors and bioelectronics devices [2, 58]. Carbonaceous nanomaterials have excellent mechanical, electrical and thermal properties, high stability, good conductivity, large surface area, antifouling properties and biocompatible. Being extremely miniaturized in size and having large surface areas, carbon nanomaterials are attracted to be excellent immobilization matrix. GNRs have rich edge chemistry and abundant functional groups, which are highly useful for immobilization of biomolecules. The sp<sup>2</sup> domains of oxidized graphene nanoribbons facilitate rapid electron-transfer. The presence of oxygen functionalities is also significantly promoted the electron transfer based reaction mechanisms in GNRs. Similarly, GONRs possessed with large surface area and enriched with abundant oxygen functional groups (epoxide, hydroxyl, and carboxylic groups).

Recently, a field effect transistor (FET) biosensor was developed using solution-processed GONRs and the sensor was applied in sensing methylene blue [28] and adenosine triphosphate [59]. Studies revealed that GNRs possess intrinsic peroxidase-like catalytic activity and hence it is a desired material for biosensing applications [60]. GNRs based nanocomposites are reported to be biocompatible and suitable for immobilizing bio-receptors such as, biomolecules, antibodies, aptamers etc. [61]. The use of PdNi nanoparticles/N-doped graphene nanoribbons for immobilizing adamantine-1-carboxylic acid functionalized primary anti-AFP (ADA-Ab1) was described, which was used to develop an immunosensor for the detection of alpha-fetoprotein. A sensitive sandwich-type electrochemical immunosensor was described using Au nanoparticles/NH2-GNRs as immobilizing matrix for quantitative detection of squamous cell carcinoma antigen [62]. GNRs were used to develop an ultrasensitive electrochemical immunoassay for rapid detection of interleukin-6 and matrix metallopeptidase-9 [63]. An electrochemical DNA biosensor was developed for the detection of genetically modified soybean by anchoring DNA on Au nanoparticles/MWNTs-reduced GONR.[64] Guanine was electrochemically immobilized on GNR modified electrode, which was used as a DNA biosensor for the evaluation of total antioxidant capacities (TAC) in fruit juices [65]. A surface enhanced laser desorption/ionization mass spectrometry probe based on GONRs-polyethylene glycolantibody biomatrix was prepared for chloramphenicol detection [66]. A label-free impedimetric aptasensor that detects femtomolar level of acetamiprid was developed using Au decorated MWNTs/ reduced GONR composite [67]. An electrochemical carbaryl biosensor was developed using acetyl cholinesterase immobilized MWNTs/GONRs nanostructure [68]. A third generation glucose biosensor was developed based on wiring of apo-enzyme of glucose oxidase with GNR bound to FAD at a screen-printed carbon electrode [69]. A magneto-controlled electrochemical immunoassay was developed for the detection of brevetoxin B in seafood using guanine-functionalized GNR molecular tag [31] An ultrasensitive immunoassay was developed to for detect human cardiopathy biomarkers cardiac troponin I (cTnI) and heart-type fatty-acid-binding protein (FABP) using GONRs as immobilization matrix.

## **5. CONCLUSIONS**

In this review, we have summarized remarkable advances in the development of novel ultrasensitive electrochemical sensors based on graphene nanoribbon based modified electrodes. Major preparation routes that can produce GNRs/GONRs are described and discussed. GNRs/GONRs based nanomaterials are ideal immobilization matrix owing to their large surface area, high conductivity, biocompatibility, high stability, good adhesion, inertness, affordability, physical strength, regenerability, ability to increase enzyme specificity/activity and hindrance to product inhibition and nonspecific adsorption. GONRs/GNRs based selective and sensitive sensors are presented and discussed. Sensors ranging from electrocatalytic to affinity-based are discussed. Additional electrocatalyst such as, metal nanoparticles, metal oxides, polymers and other nanomaterials are attached on GONRs/GNRs surfaces to enable additional sensitivity and selectivity.

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