Bull. Mater. Sci. (2022)45:173
https://doi.org/10.1007/s12034-022-02759-5

Developments in graphene-based sensors in diagnostics and other applications

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MS received 11 January 2022; accepted 17 May 2022

Abstract. Miniaturization, portability and lab-on-chip devices, like many features of the sensor, are perceived areas of work by researchers. Advanced nanomaterials like graphene, graphene oxide, reduced graphene oxide and their different forms, such as nanosheet, nanorods, nanotubes, nanoflower, nanodots, have the potential to demonstrate high-performance electrochemical sensitivity and selectivity. In theranostic (diagnostics and biomedicine) and disease monitoring, graphene and composite-based sensors have transformed these areas with new techniques and approaches. It will not be an exaggeration to include graphene as a prominent nanomaterial, which changes the view of nano techniques in sensing. The unique physicochemical properties of graphene have made it worthy of use for nanosensor technology. In this review, the authors inspected recent developments in graphene-based sensing performance and progress in its commercial validations. Herein, we also inculcate some basic aspects of sensors such as biosensors, chemisensor, and physical sensors with their recent developments and critical analysis. We also included the progress in biosensors in health monitoring, care management and prepared a consolidated framework.

Keywords. Sensors; graphene; biosensors; health monitoring; chemosensor.

1. Introduction

Sensors have played an important role in improving standards of the life of common people. The demand for different kind of sensors and research for their improvisation was increased after Novoselov decrypted the sensitivity of graphene [1]. In the medical sector, biosensors have become very famous due to their unavoidable utility in early disease diagnosis and monitoring for post-operational rehabilitation [2]. This inspection aimed to perceive the diverse range of graphene-based sensors, their impact, and their features. The most common forms of a sensor must generate a measurable signal that can quantify the amount of targeted analyte concentration. Herein, we focused on improving sensors and their application-based categories such as biosensors, chemisensor and strain sensors developed by carbon allotrope graphene. A biosensor is a beautifully integrated miniaturized device that concludes assay on its small surface. These devices hold their biological compartment to attach or adhere to biomolecules in one place. On some other plane, it contains ampliative circuits that give some graphical measurements to analyse its concentration. These segments are arranged very precisely on a platform in a very specific manner. The process of synthesis of graphene and its derivatives can be divided into two broad categories, i.e., top down and bottom up techniques, which are further divided into sub-techniques (figure 1) [3]. Graphene has a 2D sheet containing numerous hexagonal honeycomb-like structures with good mechanical strength and low weight. El-Kosasy *et al* [4] have sensed vilazodone with 1×10^{-8} M LOD (table 1) [4,5]. Graphene has high electrical conductivity ($\sim1.0\times10^{8}$ S m^{-1}), high thermal conductivity (2000–4000 W m^{-1} k^{-1}, 5000 W m^{-1} k^{-1}), high bond energy and highest current density ($\sim1.6\times10^{9}$ A cm^{-2}), including high electron mobility (200,000 cm^{2} V^{-1} s^{-1} at electron density $\sim2\times10^{11}$ cm^{-2}) and charge carrier concentration up to 10^{13} cm^{-2}, which ensured its uniqueness and use in developing a good electrochemical biosensor. Graphene has good elasticity and a high surface area ($\sim$2630 m^{2} g^{-1}), making it a favourable nanomaterial for designing a biosensing platform [6]. Srivastava *et al* [7] explored a way for selective substrate growth of ($\sim$3.5 cm $\times$1.5 cm, uniform, and continuous single) few-layer graphene films, and employing chemical vapor deposition approach on polycrystalline Cu foils using liquid hexane as a precursor. Since all these properties in graphene have made it a good contender to design smart devices and sensors, the inertness of graphene also down its feasibility

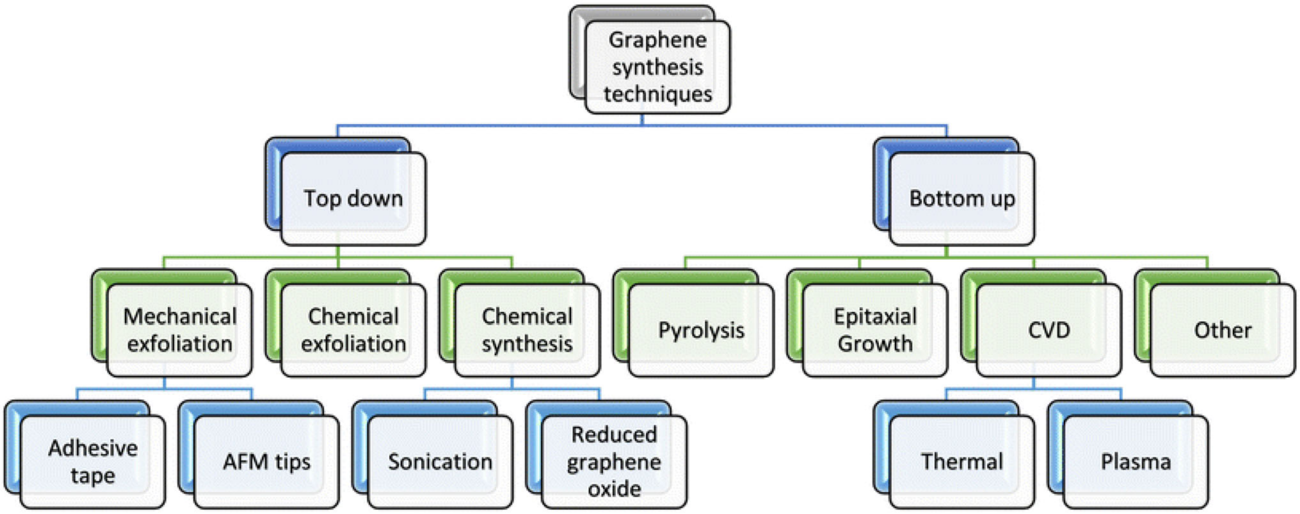

Figure 1. Different modes of synthesis of graphene. Reproduced from the study by Md. Sajibul *et al* [3] (an open access article distributed under the terms of the Creative Commons CC by license).

Table 1. Graphene-based biosensors reviewed in this article.

Electrode	Biomolecule	Limit of detection (μM)	Sensitivity	Reference
CVD graphene/liq Para	Vilazodone	1×10^{-8} M	—	[4]
CVD graphene	CEA	0.23 ng ml^{-1}	563.4 Ω ng^{-1} ml^{-1} cm^{-2}	[14]
GQD/AuNRS (apta)	PSA	0.14 ng ml^{-1}	2.5 ng μA^{-1} ml^{-1}	[15]
GQD/AuNRS (immune)	PSA	0.14 ng ml^{-1}	2.5 ng μA^{-1} ml^{-1}	[15]
CVD graphene/Pt NP	Glucose	2.8 μM	$\sim$2.2 μA μM^{-1} cm^{-2}	[16]
CuNp/Gr/FTO	Glucose	7.2 μM	430.52 μA mM^{-1}	[17]
Graphene/FET	Glucose	2–25 μM	0.46 μM	[18]
LIG/PtNPs	Glucose	300 nm	4.622 μA mM^{-1}	[19]
P-MOF-rGO	ESAT-6	3.3×10^{-5} ng ml^{-1}	706 μA ng^{-1} ml^{-1} cm^{-2}	[20]

in practicality many times. Many advanced studies have been done in this regard to overcome this problem. Different types of graphene composites with metallic and non-metallic substrates have been explored. They have come with intercalation doping that shields the effect of the inertness of graphene and enhances p-type conduct behaviour, sensitivity and selectivity of sensors [8]. Graphene oxide (GO), reduced graphene oxide (rGO), graphene quantum dots (GQD) are also similar forms of graphene. Some forms of graphene contain some functional groups such as epoxy, carbonyl or amino on their margins or amid their structures that make them reactive to another molecule [9]. Kumar *et al* [10] reported Pd@rGO-based label-free electrochemical immunosensor for detection of prostate cancer biomarker (PSA), and this technique shows the high sensitivity and specific detection in the range from 0.01 to 12.5 ng (ml^{-1}) [10]. Doping with different types of metallic elements such as gold, silver and iron has also been studied for better results of graphene, but less expertise in the optimization of doping, making it difficult in commercialization or industrial usability. Similarly, by different fabrication methods, different functional groups have been added to bare graphene to enhance its property resulting in better sensitivity, selectivity and accuracy in the real-world environment [11,12]. In this review, we concluded three forms of sensors: an electrochemical sensor (biosensor), chemisensor and physical sensor that senses physical aspects, such as humidity, pressure, etc., which are mainly discussed [13].

2. Recent breakthrough/advancement in 2D/3D graphene synthesis

Recently, many routes of synthesis of high-quality graphene have been discovered with their pros and cons. For developing an ultrasensitive sensor, a good quality and relatively more surface area having graphene is required [21]. Although mechanical and chemical exfoliation is a very famous method to develop single-graphene sheets, they do not meet the standards for sensor design. The chemical vapor deposition (CVD) method is a relatively better technique for generating an undisturbed, single transparent graphene layer and liquid precursor, making it more cost-effective. Transfer of synthesized graphene on any other desired surface makes it suitable for platform designing. Metallic membranes have been mainly used to transfer graphene on other surfaces by etching the membrane in acidic solutions. A novel roll-to-roll transfer technique to move the graphene sheet entirely without wrinkle from Cu/Ni on another flexible surface has been invented [22]. Studies report stacking order and interlayer distance between bilayer and multilayer graphene. Graphene infers their consistent differences in bandgap and interlayer spacing between bilayer graphene and bulk graphene, revealing the structural information about graphene [23]. A highly stretchable 3D printed graphene-based polymer that is good to use in sensors, soft robotics and wearable electronics with their all original qualities and upgraded quality has been developed [24]. For the upgradation of graphene performance, metallic and non-metallic element hybrids have also been trying to develop. In this continuation, (ZnO) has been a widely studied material for biosensors, and it can be tuned with graphene and its other forms like GO and rGO by doping.

Synthesized graphene/ZnO hybrid is developed as a light sensor having a good photonic and light-harvesting property. It is convenient, economically worthy, and can be used in wearable electronics and biosensing. Defects in graphene can make irreversible changes in the sensing nature of sensors. ZnO-decorated graphene can be used to sense different wavelengths and intensities simultaneously [25]. Researchers are paying enough interest to develop graphene-based sensors that sense and quantify other chemicals, biomolecules, biomarkers, gases, light, metals, and investigate artificial intelligence, robotics, wearable devices, 3D printed stretchable devices, etc.

Moreover, monitoring health is a prime requirement to reduce the unwanted cost of medication and the extra burden of money spent on health issues [26–28]. Likewise, early diagnosis of disease by the invasive and noninvasive techniques pays good results to the economic burden of deadly diseases such as cancer, heart attack, diabetes, etc. So, it is very important to fill the gap between a lab experiment and commercial products. All these above specifications of graphene and the achievements of researchers make it a globally vibrant industry. The graphene-based product industry's market value has grown from 1.5 million US dollars to 310.4 million US dollars in the last 5 years (2015–2019). It is expected to reach 2.1 billion US dollars in 2025 globally [29].

3. Sensors: based on their applications

Sensors are beautifully integrated miniaturized devices that conclude all complex assays on their small surface and generate analytical signals to measure analyte concentration. In general, sensors own conductive platforms of different nanomaterials and linker molecules that make them adhesive or intercalating for different molecules [30]. Generally, there is a wide range of sensors according to their techniques, such as an enzyme-based electrochemical sensor, electrochemical immunosensor, photoluminescence sensor, electrochemiluminescence sensor, fluorescence resonant energy transfer sensor, surface plasmon resonance and printed organofunctionalized graphene biosensor. Many of them can be equipped with graphene or other forms [31,32]. In this review, we have given more attention to CVD-grown graphene-based sensors than the other forms of graphene. Although GO and rGO are more conductive than the CVD-grown graphene, but it is easily transferable on another surface. CVD graphene is inclusive for large-area array configuration and relatively easy fabrication methods, making it suitable for formative devices, lightweight, portable devices and medicinal purposes [33].

3.1 *Biological sensor*

Many sensors have been designed for the detection and concentration measurement of biological molecules, such as protein, immunoglobulin, disease biomarkers, lectins, aptamer, m-RNA, mi-RNA, DNA, gene, virus, bacteria and many more [34,35]. Nanomaterials, such as graphene-based biosensors, have made concentration measurement feasible at the nano, pico and femto levels. Unlike GO and rGO, pure graphene is comparatively less conductive, so the adsorption of targeted molecules on the graphene surface can affect its electrical properties, which can be enhanced by using linker protein [9,36]. Doping with heavy metal is also practical to make graphene conductive, but for the binding of a large biomolecule such as proteins, immunoglobulins, fabrication of the platform is obligatory. Otherwise, antibodies or other proteins will wash out into the electrolyte. The linkage of these molecules to the platform-specific intercalating proteins has been deciphered, and linker proteins give that solution.

Linker proteins have dual intercalating regions by which they attach to the graphene bare layer by π–π interaction mechanism. Polyethylene glycol, EDC-NHS linker protein, 1-pyrenebutanoic acid succinimidyl ester protein are some examples of intercalating agents that are used for the fabrication of the platform [14,34–36]. The flanking site of these linker proteins adheres to the antibody and fixes it on the platform. Figure 2a describes consecutive developments in platform synthesis, while figure 2b shows different layers of the platform, and image 2c represents the electrochemical analysis by cyclic voltammetry (CV; table 1) [14]. Conclusively, graphene biosensors can accomplish a veteran sensor demand, electric impedance spectroscopy, differential pulse voltammetry, CV and other voltammetric techniques are potential and reliable with a real human fluid sample. Vijay *et al* [14] measured a carcinogenic biomarker CEA by using a graphene platform and revealed sensitivity of 563.4 Ω ng^{-1} ml^{-1} cm^{-2} with a limit of detection (LOD)

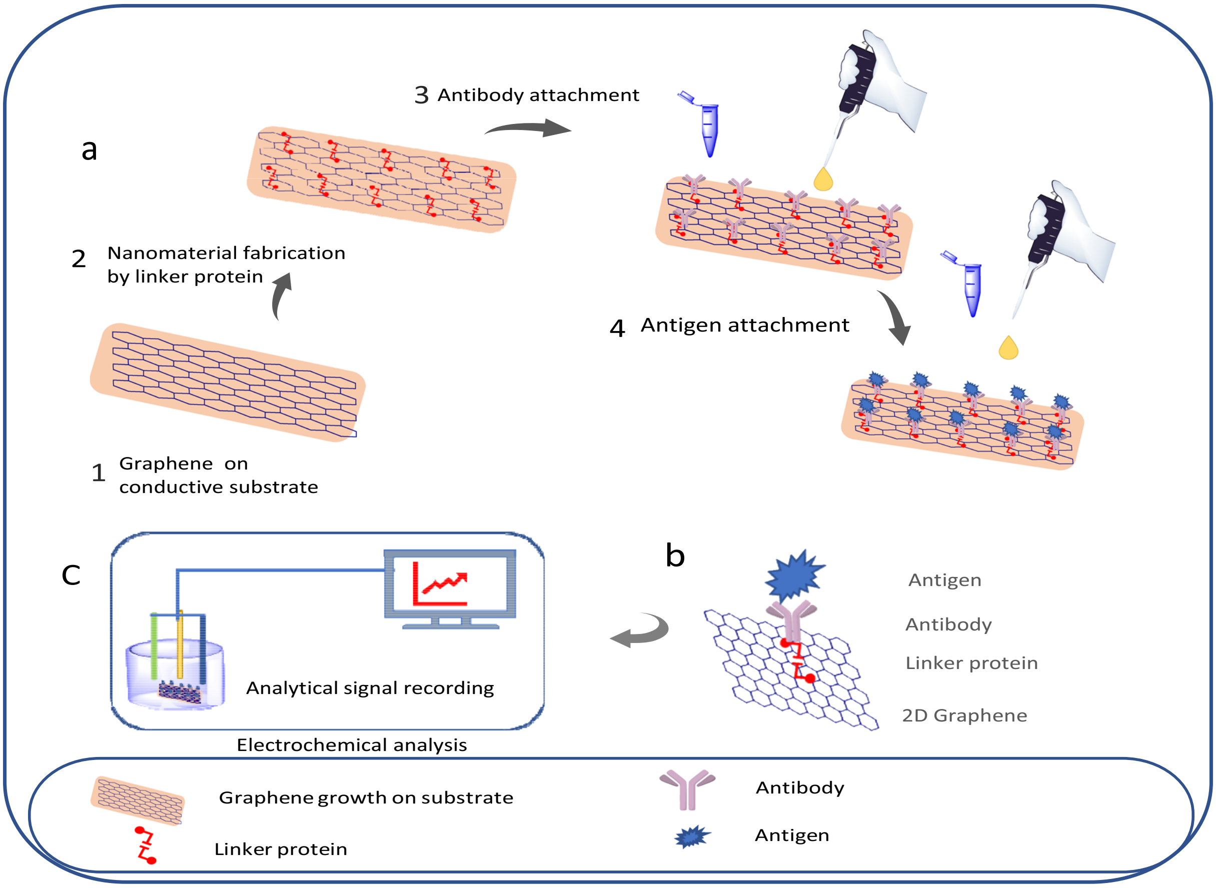

Figure 2. (**a**) Schematic illustration of the fabrication of different layers in nano biosensor for detecting antigen: (1) CVD-grown graphene on a substrate, (2) fabrication of linker protein layer by π–π stacking mechanism, (3) attachment of antibody to linker protein, (4) binding of the desired antigen molecule for detection or sensing. (**b**) A vertical section showing different layers on a nano biosensor. (**c**) Electrochemical analysis.

of 0.23 ng ml^{-1} (table 1). Likewise, Srivastava *et al* [15] reported a comparative study on label-free PSA aptasensor and PSA immune sensor platforms using CV, differential pulse voltammetry and electric impedance spectroscopy techniques. They suggested designing the PSA aptasensor for better stability, simplicity and cost-effectiveness. Biomarkers have been discovered at the time when their detection and measurement with accuracy have become a huge task for researchers. Hence, different techniques-based biosensors have been designed, which show good LOD and linearity. But recently, SERS (surface enhanced Raman microscopy), SPR (surface plasmon resonance) and CV techniques are gaining more interest due to their realistic approach, and good response with human serum samples and CVD graphene corresponds with these techniques with good efficiency [37]. Graphene-based HRP electrochemistry reagent-less amperometric immunosensor was studied by Du *et al* [38] for cancer biomarkers (CA). Graphene has been explored in health management such as the diabetic monitoring industry, mostly improvisation in NEGS (non-enzymatic electrochemical glucose sensors) sensitivity and its cost-effectiveness [39] came with a needleless, noninvasive method for diabetes to glucose monitoring through the interstitial fluid of the body by pixel-based array targeting the transdermal hair follicle pathway. In the same way, Lee *et al* [40] published a CVD graphene-based wearable sweat-based diabetic monitor that efficiently delivers thermally actuated metformin to mice's bodies and reduces blood glucose levels [16,17,40,41]. Srivastava *et al* [15] sensed and compared PSA by GQD/AuNRS aptasensor and immunosensor and reported sensitivity with 0.14 ng ml^{-1} (table 1) [15]. Graphene and their composite-based sensors sense some biomolecules such as glucose [16–20] with nano range sensitivity (table 1). Table 1 depicts the detection limit and the sensitivity of sensors towards different biomolecules.

3.2 *Chemical sensor*

Solid-state gas and chemical sensors have been broadly used as *in-situ* gas detection, ménages monitoring and industries for sensing different chemical molecules, such as H_2, NO_2, CO_2, methane, SO_2, hydrogen sulphide, mercury, volatile organic compound, explosive, chemical warfare agent detection by graphene-based chemisensor [42–44]. In a chemical sensor, graphene can perform an excellent role due to its specific structure and high conductivity, and ballistic transport ensures less signal interference.

Graphene can work efficiently without any auxiliary electric heating device and remains chemically stable at ambient temperature. Figure 3a presents chemisensor, while image 3b shows chemisensor with different layers of sensing mechanism. Bipolar charge carrier due to single atom thick 2D structure of graphene has been a favourable nanomaterial for field effect transistor (FET)-based gas sensor [19,42,44]. Gas\vapor sensors generally can be divided based on their reaction type into chemiresistor, silicon-based field-effect transistor (FET), capacitance sensor, surface work function change transistor, surface acoustic wave change transistor, optical fibre sensor, refractive index sensor and many more [45,46]. Likewise, a wide investigation has been done for Hg^{2+} (active mercury) detection. Most recent Hg^{2+} sensors relied on colorimetry, fluorometry, SWE, differential pulse voltammetry, anodic stripping voltammetry, SERS and FET-Hg sensors with many traditional Hg^{2+} sensors with their LOD, working range and response time [27]. In a chemiresistor, changes in the electrical resistance of the sensing layer of the device produce an electrical signal from the appropriate transducer, which can be easily read by the reader [47]. Shang *et al* [48] developed an enantiomeric differentiating chemosensor system that was developed by spontaneous chiral functionalization of the surface of graphene field-effect transistors (GFET). In continuation, Popov *et al* [18] developed a multilayered graphene humidity sensor with RH range 15–18 and <1 s response time [18]. This decade, graphene-based sensors and nanomaterials have been widely used in the applied field of research, such as anode in batteries, cells and supercapacitors due to their large area and high mechanical strength [6]. We can evaluate this chemisensor by measuring detection response time, recovery time, resistance measurement and selectivity. Nowadays, to enhance the conductance of graphene after exploring defects and vacancies [49], binding of specific functional groups, researchers are seeking to assemble 2D graphene sheets into 3D hydrogel/aerogel structure. Shao *et al* [50] reported a chemiresistive gas sensor that is a 3D assembled interconnected polypyrrole (PPy)-coupled graphene/$W_{18}O_{49}$ nanowire aerogel on-chip by polymerization reduction mode [50]. In this work, researchers developed a direct integration of graphene in aerogel and fabricated it in a sensor that can detect NO_2 gas at very low concentrations [49,50]. Zhang *et al* [51] worked on metal oxide-decorated graphene-based sensors to sense ammonia and formaldehyde from the same mixture. For NO_2 sensing, graphene-based RTA and UV illumination treatment-based sensors have been studied by Yang *et al* [52], displaying 97% recovery with a 20% response at 1 ppm. Likewise, Chen *et al* [53] formulated rGO/TEBAC electrode and sensed NO_3 gas molecule with 1.1 $\mu g\ l^{-1}$ LOD limit (table 2) [1]. Chen *et al* [53] tried to mimic the dog's nose sensor maxillo

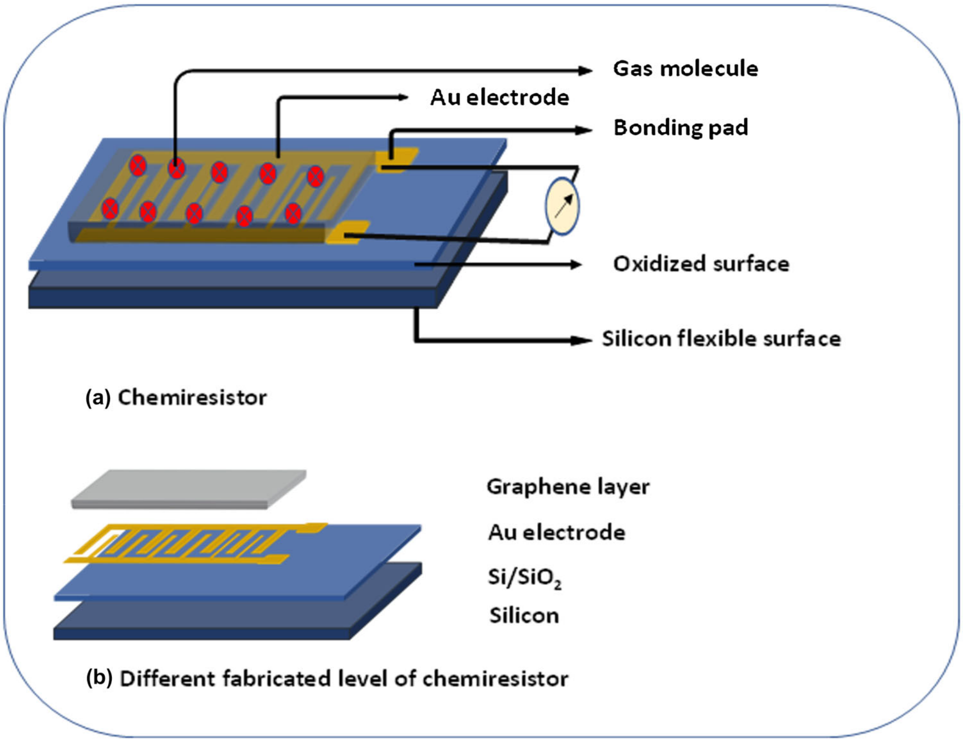

Figure 3. (**a**) A typical chemiresistor with graphene sensing film and Au electrode. (**b**) Different fabricated levels of chemiresistor.

turbinates with a simple fabrication method (table 2) [52,53]. They transform 2D plane graphene into 3D crumpled graphene and fabricated NO_2 sensors with excellent selectivity, stability and sensing linearity (table 2) [53]. Lee *et al* [2] introduced defect engineering NO_3 graphene-based NO_2 chemisensor with a 33% improvement in NO_2 and 614% in NH_3 sensing [2]. Sensing of different analytes and their monitoring is an imperative area of research for the future and graphene-based sensors like chemiresistor and FET are tunable sensors that can also be used for doped analytes. This work could help speed down water-borne disease, especially in remote areas. Still, we have seen a huge gap in lab experiments that convert the commercial product at the ground due to lack of border range analysis, reusability, less lasting and cost-effectivity [54]. Table 2 depicts the limit of detection and the sensitivity of sensors towards nitrate and nitrogen dioxide.

3.3 *Physical sensors*

High mechanic stiffness ($\sim$1 TPA) and intrinsic breaking strength (130 GPa) considered graphene as the most robust and powerful material, which made it usable in a strain sensor, and easy transfer process on other stretchable and non-stretchable surfaces makes it useful in conformal application [6,55,56]. This category of sensors is mostly based on physical applications such as measuring pressure, humidity, light, and many robotics devices. These sensors are light weighted and mostly used in detection as photodetectors, optoelectronics, inverters and strain sensors. Graphene in lithium-ion batteries enhances its charging and discharging capability rather than a conventional lithium battery and is used as a li-Gr hybrid foam electrode. Uzlu *et al* [36] introduced a novel fabrication method on top-gated tunable graphene hall sensor on a flexible substrate. They demonstrated its signal-to-noise ratio (by a factor of $\sim 10^5$), which is significantly lower than that allowed to measure in a much smaller magnetic field [36]. For the encapsulation of graphene, polymethyl methacrylate (PMMA) has been used for a long time, and now hexagonal boron nitride (hBN) encapsulation has also been studied. Wang *et al* [57] found that current and a voltage normalized graphene sensitivity up to 2270 V/AT and 0.68 V/VT by hBN encapsulation instead of PMMA in graphene hall sensor, which improves the performance of hall sensor in terms of higher sensitivity in magnetic field detection. Resonance, interferometry, fibre Bragg gratings, light intensity modulation and fluorescence are some key features of graphene that support the development of optical fibre sensors [20,57]. The structural strength and load transfer capacity of graphene permit it to combine with other conductive materials and polymers to improve [55]. The main criteria are the zero bandgap of graphene which limits its usability in digital applications due to its slow on/off ratio. Still, researchers are working to overcome this hurdle and design analog devices such as nanogenerators and graphene attenuators [55,57]. Graphene platelets and polydimethylsiloxane composite can bear both cryogenic and high-temperature environments and be used to fabricate a microfluidic device that can be developed as an electrochemical electrode for amperometry detection [58].

Generally, it is believed that the rigid structure of graphene made it less sensitive and less stretchable for banding to design strain sensors but converting graphene into a 3D format by a practical approach can decrease its gauge factor. Co-printing graphene with PLA and thermoplastic polyurethane (TPU) enhanced its stretchability up to 4-fold and 30% strain without compromising its sensitivity [24,59,60]. For the better performance of the strain sensor, an advanced composite GO-liquid crystal-konjac glucomannan (GO/KGM) synthesized by Wu *et al* [61] showed auto-assembling and annealing at some level with 0.28 KPa^{-1} sensitivity [61]. Strain sensors have been mostly employed in many different applications such as healthcare devices to monitor different physiological parameters, sound-signal acquisition and recognition devices [16,62]. In the physical sensor, gauge factor is a prominent term in the case of the strain sensor, which determines the electric shift and efficiency of fabricated strain sensors. Polydimethylsiloxane and epoxy are well-studied matrices for flexible and stretchable electronics that also show good percolation and thermal conductivity with composite [33,55,58,63]. Many technologies have been worked out to increase the conductivity of graphene by doping with Au, Pt, Ni, etc. Batignani *et al* [64] reported a radiation sensor based on GFET technology that can sense ionizing radiation by inducing electric fields and developed a novel radiation detector on a silicon layer [64]. By tuning wavelength and amplitude, the air hole size and thickness of graphene and Ag layer can affect the performance of PCF-based SPR sensors [65]. Li *et al* [66] sensed ESAT-6 with 3.3×10^{-5}

Table 2. Graphene-based chemical sensors reviewed in this article.

Electrode	Molecule	Limit of detection (μM)	Sensitivity	References
rGO/TEBAC		1.1 μg l^{-1}	~	[1]
Graphene	NO_2	1–5 ppm	20% at 1 ppm	[52]
3D–CNN	NO_2	10 ppm	74%	[53]

LOD limit that is impressive [66]. Despite these, all experimental work reviews also give insights about electrochemical events during the sensing process, such as Coroş *et al* [67] provided a detailed study on the electrochemical behaviour of graphene-based platform by explaining through CV, and Shahdeo *et al* [68] described the SPR-based biosensor modified by gold, and graphene could detect the biomolecule with 81% increased efficiency and also the detailed version of graphene and its different forms such as graphene oxide, graphene quantum dots, reduced graphene oxide with their properties and strength in the field of sensors [67–69].

4. Conclusion and further perspective

This review focused on consequent advancement and application-based critical analysis of the graphene-based platform from recent year publications. Herein, we comprehensively outlined CVD-grown graphene-based platforms but included some other graphene derivatives of high impacted work in this review. When we have seen the progression in graphene-based product market value, industrial demands have been hiked in this decade. Many researchers have exploited more qualities and features of graphene and its derivatives. The commercialism of graphene made Pandora boxes for all sectors of industries, mainly disease monitoring, health management, artificial intelligence devices, robotics, a new generation of electronic products and many other sectors. Worldwide static growth data of the graphene industry have shown a good future for further research in this area, but much more scalable work remains to be done. There is also much need to do work in the direction of reusability of the graphene biosensor platform. Although electrochemical biosensors are cost-effective, they are one-time, one-use platforms and must be disposed of, so improvisation should be done for the reusability of these platforms. In the future, more detailed and analysed research is important in reusability and to increase the expiry duration of biosensors. Commercial production of these platforms should be cost-effective for wide usability. In medicinal science, this approach is beneficial for diagnostic purposes to get countable results in disease diagnosis.

Acknowledgements

We thankfully acknowledge the Council of Scientific and Industrial Research (CSIR), India, for JRF Grant (JRF Grant No: 09/013(0811)/2019-EMR-I) to Poonam Jangir; the Indian Council for Medical Research (ICMR), India, for SRF Grants. (File No. 5/3/8/16/ITR-F/2020, IRIS CELL NO. 2020-6904) to Kirti Singh and CAS of the department for providing financial support to Preeti Suman Saxena.

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