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Graphene-based nanocomposites for sensitivity enhancement of surface plasmon resonance sensor for biological and chemical sensing: A review

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ABSTRACT

Surface plasmon resonance (SPR) offers exceptional advantages such as label-free, *in-situ* and real-time measurement ability that facilitates the study of molecular or chemical binding events. Besides, SPR lacks in the detection of various binding events, particularly involving low molecular weight molecules. This drawback ultimately resulted in the development of several sensitivity enhancement methodologies and their application in the various area. Among graphene materials, graphene-based nanocomposites stands out owing to its significant properties such as strong adsorption of molecules, signal amplification by optical, high carrier mobility, electronic bridging, ease of fabrication and therefore, have established as an important sensitivity enhancement substrate for SPR. Also, graphene-based nanocomposites could amplify the signal generated by plasmon material and increase the sensitivity of molecular detection up to femto to atto molar level. This review focuses on the current important developments made in the potential research avenue of SPR and fiber optics based SPR for chemical and biological sensing. Latest trends and challenges in engineering and applications of graphene-based nanocomposites enhanced sensors for detecting minute and low concentration biological and chemical analytes are reviewed comprehensively. This review may aid in futuristic designing approaches and application of grapheneous sensor platforms for sensitive plasmonic nano-sensors.

1. Introduction

From its inception, surface plasmon resonance (SPR) technique plays a prevailing role in the field of optical sensors. The SPR has evolved from a moderately impenetrable physical phenomenon to an optical tool that is widely used in chemical and biological investigations (Slavík et al., 1999; Yamamoto, 2008; Zeng et al., 2014) to study the binding events between two molecules of interest. Since its first intervention in 1990 by a Biacore group (GE Healthcare), the technology has established exponential growth in the last years, which is evident from the increase in the number of publications as well as the number of the methodology developed, till 2019, total of 24,148 papers are published as per PubMed search database (Fig. 1).

SPR technique is advantageous in terms of an *in-situ,*label-free methodwitheconomical and ease of fabrications as compared with the electrochemical and other methods (Merwe, 2001). The SPR phenomenon occurs in between the metal surface of sensorgram with specific molecule recognition element and a medium either vacuum/air or liquid.Whenever there is recognition of the particular molecule specific to the site/scaffold/receptor of this element, it results in the change of the surface of the metal, causing an angle shift as shown in Fig. 2(i).The shift resulted due to the changes in the refractive index (RI) at the surface of the metal. A usual SPR sensor either works in the angular interrogation mode or the wavelength interrogation mode. At the resonance wavelength or angle, the dispersion relation of the incident light matches with that of the surface plasmon, at which the reflectance shows a dip as seen in Fig. 2 (ii). The reflectance dip is attributed to the transfer of energy possessed by the photons incident to the surface plasmon and is more sensitive to the changes in the dielectric medium adjacent to the sensor surface (Ekgasit et al., 2004; Vasić et al., 2013).

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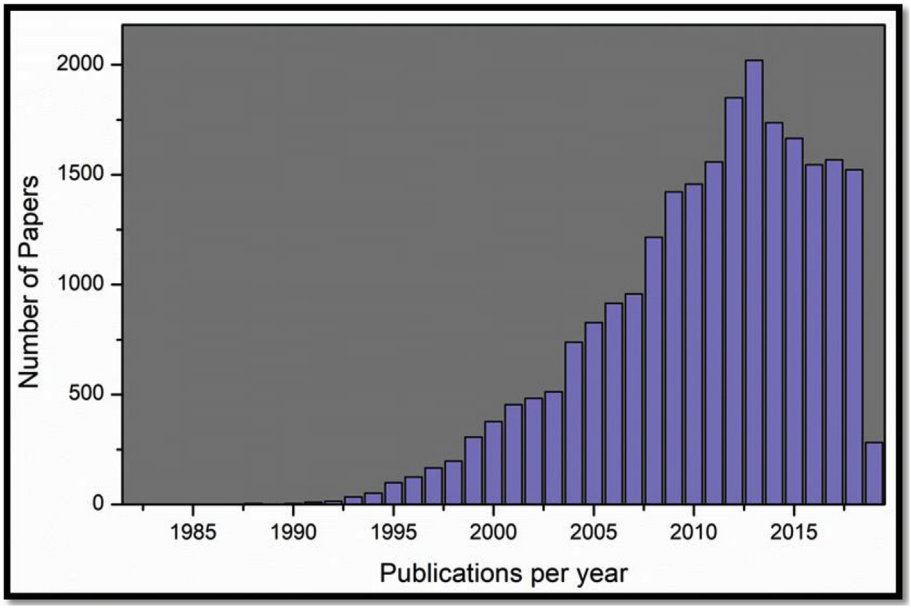

Fig. 1. Bar chart showing the number of papers published on Surface Plasmon Resonance (SPR) technology on a yearly basis as of given in PubMed.

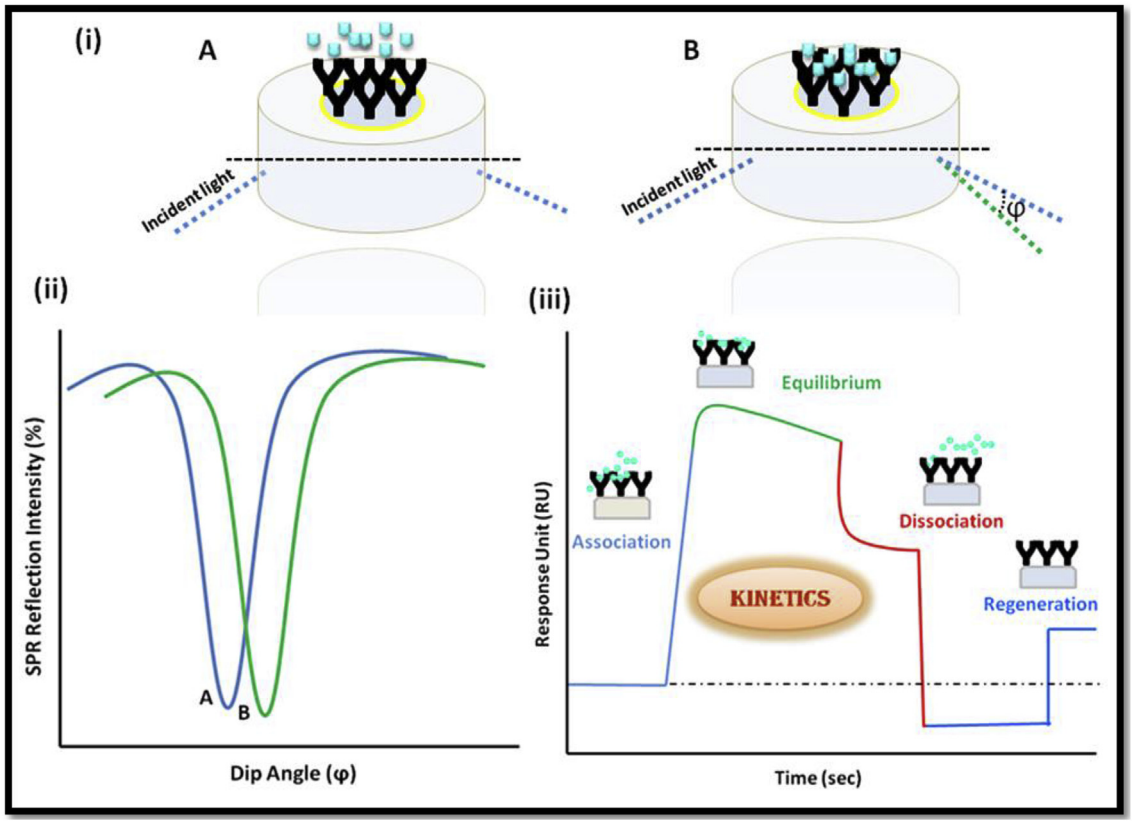

Fig. 2. Schematic representation of surface plasmon resonance. A sensor with metal surface with molecule recognition site binds with the molecule of interest (i) a change in sensorgram is observed at a certain incident angle (ii) as there is the change in RI of a surface which is sensed by the detector (iii) Association-dissociation kinetics when the impulse is sensed on real-time (Reaction Kinetic).

The ultimate advantage of SPR is the real-time sensing of a molecular phenomenon which is helpful in rapid diagnostic sensing. Real-time sensing could be achieved by changing the dip sensing events into adsorption and desorption kinetics and measuring the changes in dip angle with respect to time. This so-called sensorgram obtained will show the change of dip angle as the change in time (Fig. 2 iii). The sensitivity of the SPR is mainly based on the surface of the sensor which changes RI when association events take place on the surfaces by altering phase, wavelength, etc. of incident light. SPR sensitivity is closely related to excited electric field. Larger the electric field, the more sensitive SPR sensor will be, to the change of its surrounding medium.Working of a SPR system is based on two techniques, namely Otto configuration and Kretschmann configuration, which involve different positioning of prism and sensogram. This configuration is useful in the

study of the SPR in solid phase media. However, the Kretschmann (Kretschmann and Raether, 1968) configuration is more efficient due to the fact that the metal layer is directed on top of the TIR (Total Internal Reflectance), giving it an edgeover Otto(Courtois, 1992) configuration, wherein a distance between TIR surface and the metal is filled with low RI medium. Hence, it hampers the sensitivity of sensorgram, which ultimately results in less sensitivity towards solutions than solid.

On the other hand, an SPR could be further differentiated into two types, Localized SPR (LSPR) and Propagating SPR (PSPR). The LSPR is an emergent technique, a phenomenon observed due in metal nanoparticles (MNPs), where plasmon oscillation undergoes localization within the particles that act as an oscillating dipole induced by incident light. The SPR field enhancement majorly depends upon the nanoparticle on the surfaces and incident light. Coupling of LSPR with SP (Surface Plasmon) waves generated from the metal thin film on a conventional PSPR sensor could be an efficient way to obtain a larger field enhancement and results in higher sensitivity. However, SPR alone is not that much sensitive to detection of the analyte molecules, especially small molecules ($\leq$800 kDa) which is a bottleneck and holding back the scope of SPR for biological application. As low molecular weight molecules reflect low changes in dielectric constant or extremely low RI on the surface of the sensor chips. Persistent refinement for improvement in signal to noise ratio has made it possible to measure the binding of such small analytes. One of the refinements is an optimal condition like high active ligand concentration which is difficult to achieve and limiting the widespread use of SPR in biological sensing applications. Moreover, kinematical studies are not possible with a high ligand concentrationashigh molecular densities affect accurate kinetic analysis because of mass-transport limitations(Singh, 2016) and rebinding(Merwe, 2001).

These drawbacks can be prevailed by the effective tuning of nanocomposites and their successful fabrication.In an attempt, Li et al. (2018) designed and demonstrated a high level of sensitivity enhancement by attaching PDA (Polydopamine)-Ag@Fe_3O_4 onto rGO (reduced graphene oxide), the nanocomposite was synthesized by applying Schwartzberg's (Schwartzberg et al., 2006) and Liu (Liu et al., 2015) developed hollow gold nanosphere (HGNS) and Ag@Fe_3O_4/rGO synthesis. This platform gave promising results not only in a sensitivity enhancement, but also for lower concentration detection. The interactions between the targeted antigen-antibody substances and the multifunctional nanocomposite at the surfaces were studied in the range of 0.019–40.0 μg/mL which was 132 times lower than traditional gold-3-mercaptopropionic acid (3-MPA) SPR and 8times lower than antibody sandwich assay establishing it as highly potential over other conventional methodologies. The author clearly marked nanocomposites for sensitivity due to its high mass which plays a crucial role in signal amplification. Thus, the sensitivity of SPR mainly can be increased by the materials used particularly for the interfaces (Banerjee and Ray, 2019). More specifically, employing the nanomaterial interfaces increased the limits of detection and a wider sensing range could be obtained by means of the miniaturized SPR system. The material for sensor platform not only fundamentally increases sensitivity towards low weight molecule (less than 1000 Da) but also mean for detection of low concentration analyte ($\geq$1 ppm). The material thus imposed should have the potential to change the RI on a slight interaction between the molecules of interest, leading to the sensitivity and specificity. Graphene due to its high RI could serve as potential material for this purpose. Hence, the selection of material for the sensor platform has an impact on the working and sensing ability of the SPR sensor. Also, the catalytic activity of functionalized nano-materials further triggers secondary signal amplification. From the past few decades, the different methodology has been in use to increase the sensitivity of SPR. Many of which resided in increasing the interaction between plasmons and excitons and similarly in-between the analyte molecules and the interfaces. The interaction between plasmons and excitons can be increased by physical means. For example, utilization of optical fibers in sensor systems (Z. Chen et al., 2018; Jorgenson and Yee, 1994; Tabassum and Gupta, 2015), attracted the researcher's attention due to the technical abilities of remote sensing and online monitoring, resistant to electromagnetic field interference and most of all *in-vivo* applications (Bhandari et al., 2017). The later one could be successively achieved by incorporating two or more than two material either in bulk or in nano size. The nano-size composites have various advantages as compared to bulk such as high surface area, giving large access to molecules for interaction. The term nanocomposites as defined are the collective assimilation of two or more materials where the material exhibits their unique properties in conjugation for their multiple applications as compared to single entity alone. Based on the above assumption different nano-composites have been implied for SPR sensing enhancement. Such as plasmon metal-based nanoparticles, carbon-based nanoparticles, liposome nanoparticles, and magnetic nanoparticles to name a few. The fabricated nanocomposites so prepared can enhance the SPR signal amplification either primary or secondary amplification and thereby enhancement of sensitivity, by evanescent layer or LSPR. Out of which graphene-based nanoparticle has been ascribed mainly for the detection of bio-molecules. Nevertheless, graphene with other material in the form of a nanocomposite is found to be an excellent material for delivering outstanding amplification and sensitivity by large into femtomolar concentration.

Many reports have reviewed various aspects of SPR techniques, especially for nanomaterial-based SPR sensing (Masson, 2017; Mohammadzadeh-asl et al., 2018; Tabasi and Falamaki, 2018; Zeng and Baillargeat, 2014). Graphene-based optical fiber techniques were also discussed by authors recently (Bhandari et al., 2017; Gupta and Kant, 2018; Hernaez et al., 2017). In this review recent reports are discussed with the intention to link the grapheneous nanocomposites and SPR sensitivity enhancement for biological and chemical science. Current state-of-art grapheneous nanocomposites for enhanced sensitivity for several chemical and biological surface plasmon resonance sensors are discussed in this review. The probable mechanism behind the sensitivity enhancement directed by different nanocomposites is also described. Materialistic properties of graphene are illuminated that are necessary for incorporation into the development of SPR based biosensor. Literature for graphene synthesis, fabrication, nanocomposites based sensitivity enhancement methodologies is also reviewed in detail. Updated application of graphene nanocomposites for the detection of disease biomarker, environmental factors, nucleic acid, harmful chemicals, etc. are explained. This discussion may encourage the scientific community to pursuit further utilization of nanocomposites paving the way for clinical applications.

2. Graphene: An excellent material for SPR

Graphene is a two-dimensional crystalline allotrope of carbon with sp^2 hybridization and one atom thickness (1.42 Å) yet a strong semimetal with tensile strength (130.5 GPa) (C. Lee et al., 2008), containing carbon in a densely packed hexagonal structure(Allen et al., 2010), low band conductance (Ahmad et al., 2011) and theoretical surface area of 2630 m^2/g (Stoller et al., 2008).From the first successive practical demonstration of isolation of free-standing graphene sheets in 2004, (Novoselov et al., 2004), graphene has been utilized in several research domains owing toits outstanding electronic (Neto, 2009), optical (Kwiecinska et al., 1977; Liu et al., 2009), thermal conductivity and mechanical properties (Balandin et al., 2008; Jung, 2008; Kuzmenko et al., 2008). Due to the remarkable low-energy electronic structure of single-layer graphene, white light can be absorbed by ~2.3% (Kuzmenko et al., 2008).Nonetheless, graphene/graphene oxide nanosystems exhibit electrochromic behavior, allowing the tuning of both linear and ultrafast optical properties(Jung, 2008; Loh et al., 2010). In monolayer graphene, all the carbon atoms can directly interact with the analyte promising ultimate sensitivity in superconductor (Bouchiat, 2018; Komatsu et al., 2012), ultracapacitors (Stoller et al., 2008),

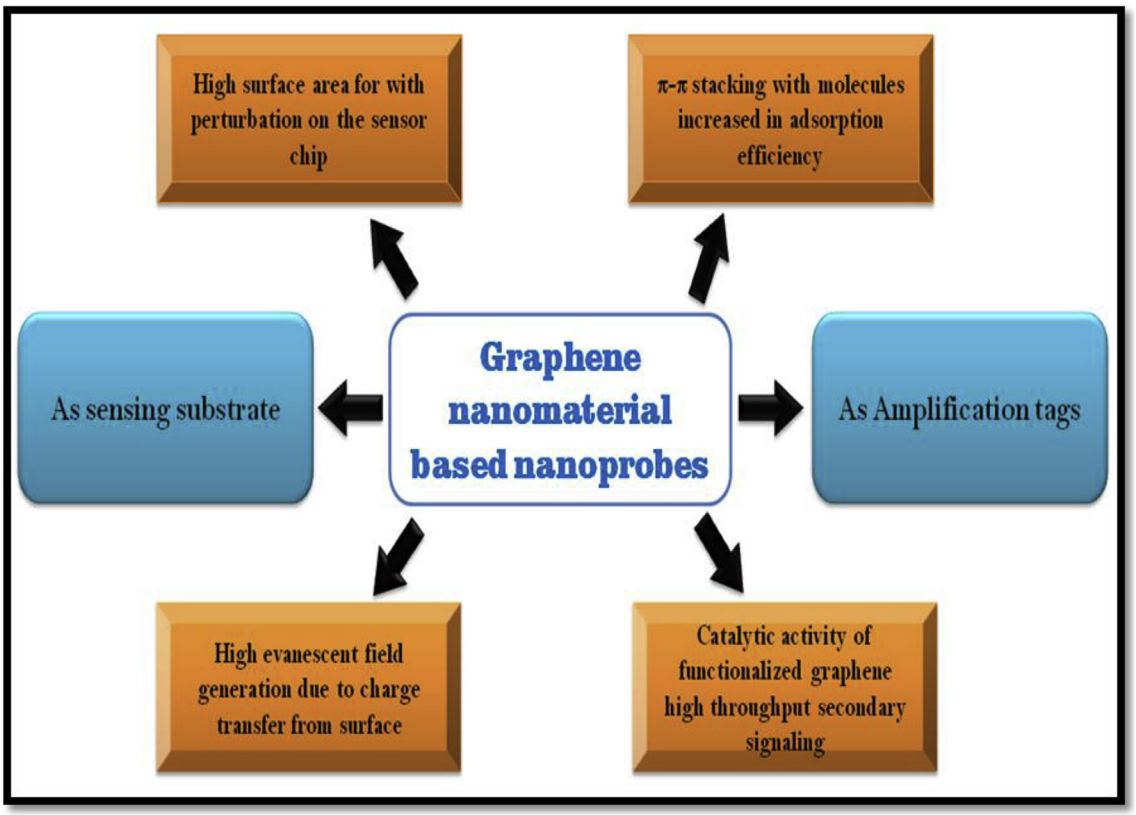

Fig. 3. Graphene nanomaterial-based nanoprobes for SPR: methods by which graphene-based nanoprobes can increase the overall performance of the SPR system, both as a sensing substrate and as an amplification tag.

sensors-biosensor (Shao et al., 2010), nanoelectronics (Berger et al., 2004; Xuan et al., 2008), biomedical research (Chung et al., 2013; Shen et al., 2012; Yin Zhang et al., 2012a,b) and industrial application (Ren and Cheng, 2014).

2.1. Enhanced SPR sensing based on grapheneous nanocomposites

Grapheneous materials or its derivatives have been commonly used with metals for inducing larger SPR signal changes compared to those with bare metal thin films (Aksimsek et al., 2018; Alharbi et al., 2017; Jafari et al., 2018; Shushama et al., 2017; Vikas and Verma, 2018; Xiong et al., 2018).Graphene prepared by incorporation with other composite material provides a sensitivity enhancement by inducing large field enhancement at the substrate interface (Cittadini et al., 2013; Han and Wu, 2018).The optical property of graphene shifts the SPR curves and increase the sensitivity to RI (Rahmani et al., 2017; Verma et al., 2011; L. Wu et al., 2010a,b). Investigators have elaborated the utility of GO in SPR sensing for the GO and dye interaction studies (Xue et al., 2014a). Also, a change in the oxidation state of GO influences the reflective property(Xue et al., 2013) and subsequently the sensitivity for ultra-small sensing.Graphene is a non-plasmonic material that can be conjugated with the plasmonic material like gold (Au) and silver (Ag) metals. More specifically, graphene layer on gold or silver thin films can induce large SPR signal changes compared to the bare metal film basically due to charge transfer from porous graphene to the surfaces. The oxygen of graphene usually undergoes desorption in the view of the plasmon field assisted absorption (Tan et al., 2012). This metal is a part of a conventional SPR system and has a property of easiness for undergoing oxidation (Choi et al., 2011a; Dash and Jha, 2014). Although, bimetallic interfaces like gold with silver surface could eventually lead to the prevention of silver oxidation as well as evanescent wave generation(Reckinger et al., 2013). However, the slow and spontaneous transformation of the morphology of gold further downgrades bimetallic as an SPR surface. The other drawback of using these metals is poor adsorption of biomolecules (L Wu et al., 2010a,b). Graphene is much stronger and stable that can be used instead of precious metals. For SP wave generation, it is vital to generate a powerful electric field in between the interface of two materials. Graphene having the highest electronic structural configuration serve the purpose for the generation of SP waves. In addition, the graphene layer could be used to prevent the sulfidation of silver(Reed et al., 2012).This graphene-passivated silver (Gr−Ag) interface was found to be extremely higher in sensitivity aspect (2600%) as compared to silver alone, by employing graphene, with the SPR excitation wavelength varied the imaginary part of the graphene and silver dielectric constant (ε_i), increases significantly as the excitation wavelength (λ_{ex}) increases. In this manner, the excellent optical properties and large spectral range of silver can be functionally utilized in a variety of nanoscale plasmonic devices and applications. For sensing of lower molecular weight biological and chemical analytes with graphene enhanced SPR sensor, the sandwich detection format is usually employed, where the graphene sheets in graphene-aluminium-graphene sandwich interface are utilized for prevention of oxidation which resulted in increased sensitivity (Xu et al., 2016). Whereas, the graphene and other analogous 2-D metals had been ascribed as a sensitivity enhancement substrate based on the parameters like material type and metal layer thickness/number(X. Zhao et al., 2018). However, most nanomaterials are subject to certain limitations due to weak interactions with biomolecules or poor chemical stability. Graphene, a 2D material with a stronger molecular-level interaction is the material of choice for overcoming such problems. In the view of recent advancement based optical fiber based SPR, the graphene gives two folds sensitivity as compared to conventional gold film, due to a stronger light-matter interaction (An et al., 2019; Wei et al., 2018). Graphene based sensitivity enhancement and graphene based RI sensitivity enhancement using gold film covered with graphene deposited on the surface of the polished plane of D-shaped

photonic crystal fiber (PCF) are studied extensively and reported recently (Nikbakht et al., 2018; An et al., 2019). The hexagonal ring structure of graphene and a good surface to volume ratio are extremely beneficial for adsorption of bio-molecules byπ-π stacking and thereby increases the absorption of biomolecules on the graphene layer(Akca et al., 2011). Grapheneous derivatives, such as GO, make it the material of choice for further functionalization with other metals, inorganic and polymer structures (Georgakilas et al., 2012).Chemistry of GO and metal colloids in liquid phase forming a stable and miscible interface was elaborated by researchers (Bei et al., 2011). Furthermore, these interfaces could effectively undergo functionalization due to complimentaryfreebinding sites of graphene, which provide the researcher'sflexibility for further fabrication of graphene-based interfaces.Thus, all these studies indicate that by carefully designing the nanocomposites as well as the successful fabrication, one can get hold of the most favorable set of investigational sensing parameters to detect extremely diluted bio and chemo-molecules.

Sensitivity enhancement methods for grapheneous nanoprobes are depicted in the scheme below (Fig. 3).Graphene could turn up as sensing substrate or as an amplification tag or sometimes both. This scheme would facilitate the researchers to various mechanisms of graphene-based sensitivity enhancement and its applications ranging from clinical to environmental monitoring. This scheme provides the most appropriate enhanced mechanisms for engineering the specific type of nanomaterial-based enhanced SPR sensing system. Graphene and its derivatives could be efficiently used for biomedical and clinical diagnosis. Moreover, functionalization of graphene and its derivatives enables the synthesis of newer nanocomposites material, owing to its rigid, large molecular size, functional groups and high surface area. The presence of the free sites of graphenecan be used for molecule or functional group doping(Katsnelson et al., 2018). By interacting in an ionic, covalent or non-covalent manner, it could serve as a site for enzyme immobilization with proven highest extraction efficiency for biomolecules per unit area(Tang et al., 2010). One of the most important properties of graphene materials is its ability to transfer electron(Zhang et al., 2005) between the interface of the sensor chip and fabricated probe mainly due to the perfect arrangement of electronic configuration(Weitz et al., 2007). Likewise, 2-D planar structure and hollow voids (Yanhui Zhang et al., 2012a,b)acts as a bridge between the sensor surface and fabricated probe for rapid and faster electron exchange(Gomez-Navarro, 2007) and contributing to signal amplification (Huang et al., 2013).Graphene and GO based sensing surface as a substrate have attracted much attention. Its specificity relies upon charge transfer ability, which ultimately leads to strong excited electric field enhancement. Also, high surface area and π-stacking allow the adsorption of biomolecules and aromatic ring structure. Thus, it works as a substrate which eventually increases the sensitivity(Choi et al., 2011b; Giovannetti et al., 2018; Grigorenko et al., n.d.; Liang et al., 2017; Salihoglu et al., 2012).

2.2. Methodologies for graphene synthesis, fabrication, and graphene nanocomposites-based sensitivity enhancement

Over the past few decades, functionalized carbon-containing materials such as graphene(Suvarnaphaet and Pechprasarn, 2017), GO (Kubesa et al., 2017; Wijaya et al., 2012), carbon nanotube (CNT)(Lee et al., 2011), GQDs(Ramdzan et al., 2019), carbon dots (Vivek Borse et al., 2017) etc., have been used in sensor-biosensor studies.Graphene nanoparticles have been commonly used as signal amplification labels in conventional gold and silver substrate PSPR sensors(Choi et al., 2011b; Jinfa et al., 2014; Kim et al., 2015). The sensitivity of the graphene embedded SPR chips mainly relied on their size, shape, number of layers and the dielectric constant of the surrounding medium(Li and Zhu, 2014; Vasić et al., 2013)Thus, picking the accurate number of graphene layer with optimum particle size and couple its corresponding high adsorption carrier mobility to the surface plasmon waves of PSPR sensors for obtaining the largest field enhancement effect thereby increasing the sensitivity of the sensor. For instance, numbers of graphene layers are directly interlinked with the overall sensitivity, which after certain numbers of graphene gives downtrends in the signal amplification(L. Wu et al., 2010a,b).

Graphene by virtue of allotrope of carbon can be easily sourced from graphite. However, till the year 2004, no reliable and convenient method was in existence for making graphene. Novoselov et al.obtained single atom thick graphene by mechanical exfoliation using graphite (Novoselov et al., 2004). Graphene can be easily converted to GO by Hummer and Offemen method(Hummers and Offeman, 1958). However, it is now replaced by the series of washing with HCl followed by drying at 60 °C to make dispersion and filtering the graphene suspensions popularly known as a modified Hummers' method(Athawale and AuthorAnonymous, 2014; Rahmawati et al., 2009). Most widely used synthesis protocol is chemical vapor deposition (CVD)(Li et al., 2009). It involves the reaction of reactant gases (CH_4) using a metal catalyst breaking the C–H bond carrying the reaction at a lower temperature, which ultimately gives the deposition of solids onto a suitable substrate (Li et al., 2009). A tubular furnace is used and reaction kinetics involves the several steps like the heating of furnace, annealing for resurfacing of a metal catalyst, growing the graphene over a metal catalyst, cooling of the furnace, and the passing of inert gas. However, metals are rate governing factor involved in the reaction kinetics and one must consider appropriate metal for the reaction to proceed. Direct electrodeposition of GO at a predetermined time is also a suitable technique, in point of fact that it gives a several nanometer-thin layers(Subramanian et al., 2013). Epitaxial growth, synthesis of colloidal suspensions, and chemical/electrochemical exfoliation (Singh et al., 2012; Zhu et al., 2010)are also preferred routes for the synthesis.

The graphene-based sensor chips can be successfully fabricated by the amalgamation of graphene with other substrate in nanosized scale. Graphene may also be functionalized in order to enhance amplification signals, loading capacity, specificity, sensitivityand biocompatibility. However, in another way, fabrication strategy provides enhancement of surface loading capacity, increasing the efficiency in selectivity and sensitivity.This can be also optimized during the cross-linking, carbodiimide coupling and cytidine linking. Grapheneous material for conjugation with other material can be achieved in most of the cases through carbodiimide chemistry (EDC/NHS)(Williams and Ibrahim, 1981). It works as a site-directed modifier such as for carboxyl group (C = O) modification where the amine group (-NH_2) acts as nucleophile or vice-versa. Water soluble carbodiimide acts as a biological tool for attachment of biological to solid or polymer support, for fixing it, sometimes as a cross-linker for generating intermolecularly thermostable proteins, etc. Nevertheless, other techniques like cytidine linker are also been used widely, but GO can easily circumvent this process due to the availability of numerous sites for fabrication on its surface.A strong acids or oxidant treatment provides free oxygen and carboxyl (-O and C = O) groups on the GO surface helping in effortless grafting of molecules through covalent binding. For instance, proteins and GO can be coupled via an amide bond (CO–NH), esterification could be chosen as a path in case of carbohydrates and polymers. However, owing to 2D planar geometry and high adsorption coefficient of graphene provides extravagance of non-covalent attachment of most of the aromatic structure(Stebunov et al., 2015)increases the sensitivity of sensors (Singh et al., 2015). More profoundly, grapheneous material fabrication can be straightforwardly performed by a self-assembled monolayer (SAM) (Fig. 4).

The method is attractive and extensively used by researchers because of two reasons: (1) they provide dense, well-ordered, tightly bonded films and (2) they form a simple motif for selective tailoring of surface chemical properties(Malinsky et al., 2001). The most used SAM is alkanethiols on Au or Ag and organosilanes on oxides. However, other convenient processes have been used by different researchers. For instance, Zhang et al.(J. Zhang et al., 2013b) flowed GO-Au composites

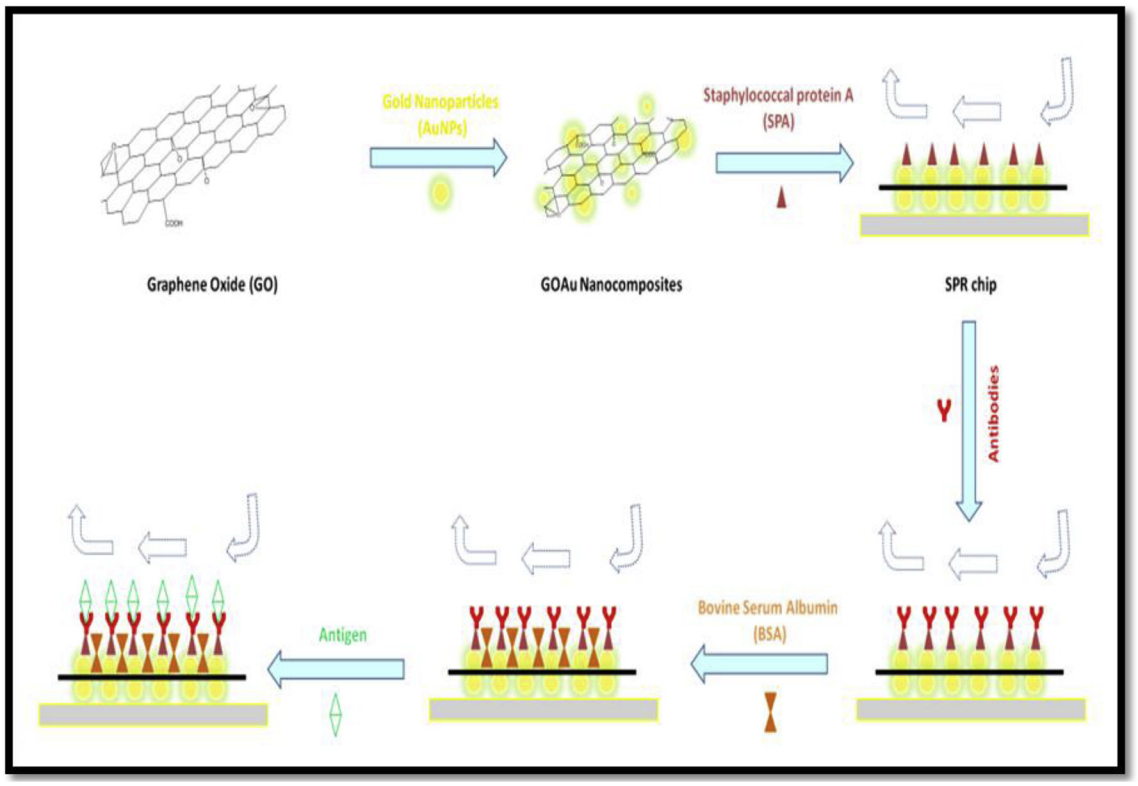

Fig. 4. Self Assembled Monolayer (SAM) Fabrication on graphene-oxide-gold using *stayphaloccal protein A* and BSA where arrow showing the direction of flow of substrates. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

in a flow cell and incubated for 3 h to form GO modified Au film. Moreover, the more common form of SAM is Layer-by-layer (LbL) assembly (Chung et al., 2015; Mao et al., 2011). The group demonstrated the successful fabrication of cationic polyelectrolyte-functionalized ionic liquid decorated graphene sheets (PFIL–GS) with Prussian blue (PB). The cleaned gold slide was dipped into positively charged PFIL-GS followed by dipping in negatively charged PB nanoparticle, a significant increase in peaks has been observed in UV-spectra with respect to LbL layer (Fig. 5). It is a powerful technique, advantageous because of simple operation and retention of bulk properties of the material, in which a surface alteration is done by an alternatively adding thin film or positive and negative charged polymer or polymer with H-bond donor and acceptors onto the substrate of choice(H. Lee et al., 2008; Lutkenhaus et al., 2005).

The researchers successfully achieved the desired functioning of nanoprobes such as Zeng et al. (2015) synthesized gold film coated by graphene. Gold metal would be in the perturbed form, commonly referred as “metasurface” that could be further functionalized with aromatic biomolecules through π-stacking for efficient detection. For instance, treated optical fibers with dip coating and different sensing over-layer could results in sensitivity enhancement(Mishra et al., 2014). Ng et al. (2017)plasticized fabrication on cleaned glass slide and thermally annealed at about 550 °C for 3 h establishing the formation of self-assembled gold nanoislands (SAM-AuNIs) further spin coated at 2000 rpm with graphene to fabricate the chip and used for 3-Nitro-L-Tyrosine (3-NT). Similarly, the nano-composite thin film could effectively be fabricated using the spin coating technique at high speed and used for optical fiber based sensing(Daniyal et al., 2018).

Theoretical and simulation predictions suggested extremely high carrier mobility ofgraphene in metal surface or on nanocomposites (Gonzalez de la Cruz, 2019; Karimi and Ahmadi, 2016; Li and Zhu, 2014; Pang et al., 2019; Said et al., 2017; Zhang et al., 2015).In addition, earlier research findings reported the guided-wave SPR (GWSPR) biosensor has a higher sensitivity than that of the SPR biosensor. Sensitivity of GWSPR can be extended higher using MoS_2–graphene hybrid structure which could cover on the surface of the thin silicon film. This nanocomposites sensitivity can be increased further with the additional layer TiO_2–SiO_2composite layer between the prism base and metal layers(Maurya et al., 2015). In this regard, recently,Wu et al. (2018) demonstrated MoS_2 layers to enhance the sensitivity, while the graphene monolayer was applied as the bimolecular sensing element. The anticipated biosensor was sensitive to the change in RI in the range from 1.33 to 1.80 of sensing medium which helps to detect the biomolecules.The graphene itself possess the distinctive properties involving in the signal amplification of SPR sensor(Verma et al., 2015), which can be effectively tuned with other material composites for enhancing SPR sensing (Mishra et al., 2016; Reckinger et al., 2013; Vasić et al., 2013). In a similar context, effect of graphene derivatives at the gold surface for protein detection using bovine serum albumin (BSA) as a model protein was studied (Emiliano N Primo et al., 2018a,b). The researchers demonstrated the correlation between the nature and amount of grapheneous nanomaterial, amount of immobilized protein and electrochemical as well as plasmonic effect of the nanohybrids. For this purpose, researchers have chosen three grapheneous material and the non-covalent immobilization of (A) PDDA-GO (B) GO-CHIT and (C) rGO-CHIT at Au/MPS, with the covalent attachment of BSA at each of the resulting platforms. A rapid increase in δ-ΘSPR after immobilization of graphene-based material was observed on both positive PDDA and negative MPS, where the carbonyl group of played a role in the covalent attachment of the BSA, by thoroughly studying the changes seen prior and after the washing, the result for surface coverage of the protein at different platforms was drawn.The obtained data showed that GO could be utilized asSPR sensors based on direct quantification of protein. Likewise, Kumar et al.(R. Kumar et al., 2018)optimized Kretschmann configuration with a layer of zinc oxide, gold, graphene presented on a prism. The thickness of ZnO, Ag, and graphene was kept at 32 nm, 32 nm, and 0.34 nm, respectively, by varying the Au thickness (1, 3, 5 nm). each modified layer was further coated with biomolecules in this

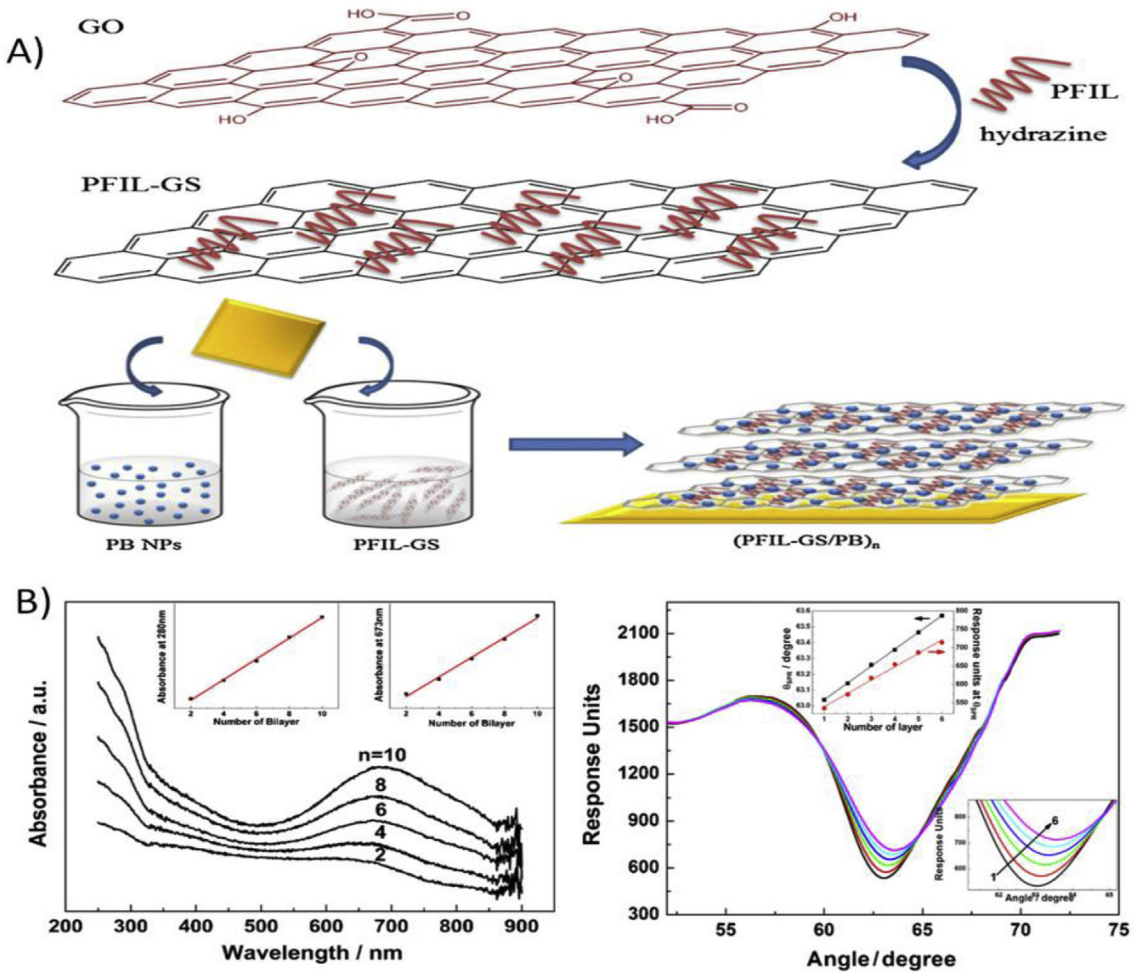

Fig. 5. A) LbL method B) Sensitivity enhancement of SPR with respective to number of layers. Reprinted with permission from (Mao et al., 2011), Copyright 2011 Elsevier.

case ssDNA (100 nm) the formed sensor chip was mathematically analyzed for variation in reflectance by varying thickness of sensor chip and Au layer, a number of the graphene layer, RI and the thickness of biomolecules. The outcomes of this theoretical studies were, for 1 nm thickness of Au at L = 0 and L = 1 (where L is no. of graphene layer) the sensitivity was 72 and 76 deg/RIU respectively, as compared to conventional biosensor 52 deg/RIU. Kushwaha*et al* (Kushwaha et al., 2018). theoretically investigated nanocomposites containing Graphene-ZnO/Au, it is worthy to note that nanocomposites possessed monolayer graphene due to the fact that SPR sensitivity is indirectly proportional to the number of graphene layer (R. Kumar et al., 2018) as each graphene layer involve in the absorption of light multiple of 2.3% (Kuzmenko et al., 2008), but could help in the absorption of biomolecules (Akca et al., 2011). While zinc oxide with gold metal shown sensitivity enhancement due to the absence of centrosymmetry that increased the second order susceptibility at the interface and thereby second harmonic waves responsible for the long range shift of resonance angle leading to a soaring performance of SPR system. The theoretically drawn data for detection accuracy and sensitivity were 2.05 deg^{-1} and 187.4 deg/RIU respectively.GO sheet (GOS)-based SPR sensors was developed for protein immobilization detection(Chiu et al., 2015). Where, applicability of GO sheets as the intermediate, due to its high covalent binding affinity for proteins was studied. Well-established real-time monitoring of the binding phenomenon was achieved in the fabricated system. The prepared GO-based sensor was able to detect bovine albumin serum (BSA) directly in GOS films. The sensor exhibited sensitivity up to a 4.3-fold higher angle shift than that of the conventional Au/Cr-based sensor up to the 10 ng/mL of BSA concentration. The BSA detection limit as low as 100 pg/mL and the average affinity binding value (K_A) for the GOS-based sensor was observed about 80.8 × 106 M^{-1} with respect to BSA. Moreover, fabricated GOS-functionalized Au–Cys composite SPR biosensor showed high sensitivity and decent detection limits for protein analysis immobilization via GOS–protein binding. Graphene derivative can increase the overall sensitivity as well as can prevent the oxidation of LSPR (Ag, Au) substrate (Dash and Jha, 2014; Khalili Fard et al., 2018). Since the first use of graphene-coated gold sensing film for improving the SPR sensitivity was mentioned by Wu et al. (L. Wu et al., 2010a,b). The reasons behind increase in sensitivity of sensor platform is its high adsorption ability and electronic and optical property (Bonaccorso et al., 2010).The theoretically drawn numerical calculation reveals that RI was increased by 25% for $L = 10$ (L = number of graphene layer). Similarly, the sensitivity (S) was increased by γ times to the $(1 + 0.025\ L)$, where γ is greater to one and L is a number of the graphene layer. For instance, when the γ is equal to 4 the sensitivity (S) will be increased by 5 folds. Gold nanoparticles are part of the traditional SPR system and commonly applied for signal amplification. The AgNPs/GO nanocomposites embedded with hCG (Human chorionic gonadotropin) protein were successfully fabricated. In this work, carbonyl group using EDC/NHS chemistry exhibited the detection of anti-BSA in 10 ng/mL of hCGand it was found that C = O was responsible for BSA protein conjugation. The group sensed the anti-BSA in the range of 1.45 nM–145 fM with limit of detection (LOD) of 145 fM. In another report, GO sheets were decorated with AuNPs; a significant increase

(~5.5 folds) in K_A (binding affinity) of between antigen and antibody was found (Chiu et al., 2017), when Au/GO was further modified with a carboxyl group. It is important to note that high binding affinity is a great tool for real-time monitoring. The GO-COOH when combined with BSA results in better bio-molecular interaction in solution by providing the hydrophilic environment. This carboxyl-functionalized GO composites sensor chip had a high affinity for specificity assessment owing to high specific surface area and a very low LOD compared to traditional SPR.

Graphene modified conventional silver substrate has demonstratedthe high sensitivity compared to simple old age methodology of using the traditional SPR, the sensitivity of sensorgram was decreased with the increase in graphene insulating the silver/gold layer.Graphene layer of two to three atom thicknesses with deposition on silver substrates exhibits better performance than that of the gold substrate at the wide range of silver film thickness. On the other hand, the high impermeability of graphene could potentially be used as a protective layer that prevents undesired oxidation of silver.More importantly, the graphene-on-silver substrate presents approximately three times greater imaging sensitivity than that of the traditional gold substrate.Without graphene sheet, the peak imaging sensitivity was obtained to be as high as 3.82 with the silver thickness of 60 nm, presenting 5.6 times larger sensitivity than for the gold film. More significantly, the graphene-on-silver substrates with extremely thin graphene sheets exhibited an extremely high sensitivity. Choi*et al*(Choi et al., 2011a). attempted up to ssDNA detection, which is merely possible with gold sensor alone using a modified gold layer with graphene(Song et al., 2010). High adsorption property of grapheneous material and LSPR effect provided by Au can be combined for evanescent SPR which could lead to a new way forward for high sensitivity sensor chips. Wang et al.(Wang and Wang, 2018)comparatively studied the sensitivity of silver film SPR against the newly fabricated Ag-coated polymer cladding silica fiber containing a layer of GO had shown a sensitivity of RI 3311 nm/RIU in the RI range of 1.3334–1.3731 compared with 2875 nm/RIU in the range of 1.3328–1.3739 of the silver film SPR sensor.Cen et al. (Liang et al., 2018) obtained results proving the plasmonic resonance absorption characteristics of H-shaped graphene arrays in far infrared and tetrahertz bandwidth. This H-shaped graphene had provided shape and size based shift in wavelength. Computational investigation using the FDTD algorithm (Said et al., 2017)(Finite-Difference Time-Domain)(Kane, 1966)was carried out for graphene on different metal substrates (Au and Ag), the acquired numerical value had proved on both analytical and experimental stage.The graphene layer was applied on all metal substrates and compared to conventional SPR (with only metal surfaces) and analyzed at two different wavelengths 670 and 785 nm with emphasis on TAR (Total attenuation reflection) and FWHM (Full width half maximum). Gold interface (60 nm) primarily shown high reflectivity and FWHM at 92.4% and 0.88. Similarly, graphene alone was able to vary the changes with narrower FWHM of 0.88° and reflection spectra of 89.2%, whereas, the graphene-gold interface had shown the same reflectivity of 91.7% and FWHM of 1.32°.It suggests better shift of incident angle which is helpful to study sharp changes with respect to a low change in RI; particularly useful for DNA and other small biomolecules interaction studies.The following table (Table 1) provides fundamental information on grapheneous nanocomposites based sensitivity enhancement.

3. Grapheneous nanocomposites applications in biological and chemical domains

3.1. Sensing of disease biomarkers

The development of a precise sensor for real-time detection of disease biomarkers is currently thrust area of work in SPR. Extensive research on biosensing has been carried out in the clinical domain to provide cutting-edge technology for reliable, easy to use, cost-effective and early prognosis or day to day monitoring of chronic disease and the point of care diagnosis as well (Borse et al., 2016b, 2016a; V. Borse et al., 2017a,b; Borse and Srivastava, 2019; Makkar et al., 2018). Subramanian et al. (2013) detected the urine and serum levels of lysozymes, an enzyme, which is a biomarkerfor leukemia as well as renal disease. The rGO layer with a proper thickness on-chip was fabricated by electrophoretically deposition (150 V) within 15 min. The selectivity of the rGO-SPR chip for lysozyme was achieved by pi-stacking of anti-lysozyme DNA aptamer. The chip-based system has provided the sensitivity in nanomolar concentration with LOD of 0.5 nM. Chemotherapy is a crucial step in the eradication of cancerous cells not only in initial stages, but also before to radiation therapy to minimize the cell spreading. Hence, a studying the cell-drug interaction is important steps for determination of efficacy of a particular drug. The whole cancerous cell response on a graphene sensor chip was studied (Y. Wang et al., 2018), similarly anticancer drug paclitaxel was treated with two colorectal cell lines HCT116 and LoVo. The fabricated graphene based biosensor was effective with signal to noise ratio of 5.3 and sensitivity up to 1.2×10^8 mV/RIU, for the single molecule detection.Recently, sensitive and selective detection of biomarkers is reported as an effective tool for the successful diagnosis of early-stage cancer and follow-up treatment(He et al., 2017). SPR-based combination with different strategies could be used for a point-of-care sensor for the detection of folic acid protein (FAP) using graphene-based SPR chips. The selective recognition of FAP was based on the interaction between folic acid receptors integrated through π-stacking on the graphene-coated SPR chip and the FAP analyte in the serum. A simple post-adsorption of human serum:BSA mixtures onto the folic acid modified sensor resulted in a highly anti-fouling interface while keeping the sensing capabilities for folate biomarkers. This sensor allowed femtomolar 5–500 fM detection sensitivity of FAP and a detection limit 5 fM.Lysozyme could also be owing to its affinity guided desorptive interaction with *M. Lysodeikticus* in undiluted serum(Fig. 6) (Vasilescu et al., 2017).Besides this, nickel doped graphene(NDG) was first time applied for early detection of neurodegenerative biomarker named 3-nitro-l-tyrosine (3-NT), the direct absorption mediated detection verified by both AFM and SPR, the SPR had given the sensitivity at 0.5 pg/mL to an1 ng/mL with LOD of 0.13 pg/mL(Ng et al., 2017). Hu et al. (2014a) demonstrated signal amplified detection of alpha-Fetoprotein (AFP) known as a tumor biomarker in a 0 ng/mLto 100 pg/mL by using GO.

Lung cancer biomarker cytokeratin-19 (KRT-19) was detected in a study (Chiu et al., 2018). The KRT-19 protein biomarker in the human plasma spiked sample has shown better sensitivity in an extremely diluted sample (0.05 pg/mL).Whereas, interacting with immobilized KRT-19 antibodies (10 μg/mL) carboxyl fabricated GO- SPR chips, field energy propagation intensity with ease of fabrication, selectivity and leads to better sensitivity and proposed sensor reliable for KRT-19 detection. Hence, detection of non-small cell lung carcinoma (NSCLC) could be done with an excellent accuracy. Graphene with gold nanorods provides ultrasensitive detection by improving the sensitivity (Zeng et al., 2013). On a similar track, a gold nanorod (AuNR) decorated GO for sensing of transferrin, a biomarker for different diseases was proposed (J. Zhang et al., 2013a). An increase in concentration of transferring can be marked for a disease like acute hepatitis, anemia and pregnancy. Additionally, decrease in concentration of transferring can be an excellentkey for diagnosis of rheumatism, malignant tumorand acute leukemia.The fabrication of GO/AuNR-AB was accomplished using incubation of Au glass film with MEA (2-Mercaptoethylamine) followed by electrostatic binding of EDC activated GO as shown in Fig. 7.

After which PBS was injected as a baseline, an AuNR previously synthesized by seed-mediated growth and attached with transferrin antibody (rabbit) using EDC/NHS chemistry was injected and incubated for 12 h prior to giving PBS wash and blocking unreacted sites with BSA.An antibody-modified Au film with 10 mg/mL solution of transferrin yields 2.7 nm shift in resonant wavelength, while the exposure of

Table 1
Grapheneous nanocomposites based improved SPR sensitivity.

Nanocomposite	Fabrication	Analyte	Methods of sensitivity enhancement	Sensitivity	Limit of Detection	Refer.
Graphene	Prussian (LbL)	H_2O_2	LbL assembled graphene/prussian	5–50 μM	1 μM	(Mao et al., 2011)
rGO	Au chip	Dye Cy5 and TPA	The higher binding ability of rGO with dyes	–	10^{-8} to 10^{-6} M	(Xue et al., 2014b)
rGO-gold chips	Anti-lysozyme DNA aptamer	Lysozyme	rGO thin layer/π-stacking	0.5–200 nM	0.5 nM	(Subramanian et al., 2013)
Graphene	–	Paclitaxel	Graphene film based ultrasensing depth with 2 μm	1.2×10^{8} mV/RIU	2.6×10^{-8} mV/RIU	(Y. Wang et al., 2018)
graphene oxide	Micrococcus lysodeikticus	Lysozyme	affinity guided desorptive	0.2 to 40 mgmL^{-1}	0.05 mgmL^{-1}	(Vasilescu et al., 2017)
Nickel-doped graphene (NDG)	LSPR	3-nitro-l-tyrosine (3-NT)	Graphene mediated direct absorption mediated	0.5 pg/m-1 ng/mL	0.13 pg/mL	(Ng et al., 2017)
GO	Gold SPR chip	Alpha-Fetoprotein	GO based signal amplification	0 ng/mL to 100 pg/mL	–	(Hu et al., 2014a)
FnCOOH-GO	Au chips	cytokeratin-19	FnCOOH-GO-Au amplified field energy propagation intensity	0.001–100 pg/mL	1 fg/mL	(Chen et al., 2018a,b)
GO-AuNRs	Ab	Transferrin	The high loading capacity of GO & AuNR for high sensitivity.	0.0375–40 μg/mL	0.0375 μg/mL	(J. Zeng et al., 2013a)
valinomycin doped chitosan-graphene oxide thin film	Spin coated on gold chips	potassium ion (K+)	C–GO–V film acts as a high impending sensing substrate	0.00948–100 ppm	0.00948 ppm	(Zainudin et al., 2018)
Ag-GO	Au SPR chips	Dopamine	GO increased surface adsorption.	–	49 nM	(Zangeneh Kamali et al., 2015)
PDA-rGO	Carcinoembryonic antigen (CEA)-antibody	Carcinoembryonic antigen (CEA)	Sandwich immunocomplex for 1st & PDA on Au for 2nd amplification	0.5–50 ng/mL and above	500 pg/mL	(Hu et al., 2014b)
DNA Fn Graphene gold- coated fiber	tilted fiber Bragg grating (TFBG)	Dopamine	NC amplifies the surface refractive index modulation over the fiber surface	10^{-8} to 10^{-13}	10^{-13}	(Hu et al., 2018)
silver-GO-silver NPs	enzyme entrapped gel	Cholesterol	large surface area of GO nanosheets	0–10 mM	0 mM	(Semwal and Gupta, 2018)
Graphene/ZnO bilayer	Au SPR chips	ssDNA	No. of graphene layer affecting the sensitivity of sensogram	72-76 deg/RIU	–	(R. Kumar et al., 2018)
self-assembled PDDA-GO	S–Au chip	galectin-3	SAP-GO layer enhanced sensitivity	10.0–50.0 ng mL^{-1}	2.0 ng mL^{-1}	(Emiliano N. Primo et al., 2018)
Graphene	Au–MoS2	dsDNA	large surface to volume ratio of graphene	87.8 deg/RIU	–	(Rahman et al., 2017)
GO	nucleotides	double-stranded DNA (dsDNA)	GO based nucleotide-dsDNA interaction	–	–	(Rahman et al., 2017)
Phosphorus nanoparticles	Graphene coating	cDNA	desorption based cDNA detection	125°/RIU	–	(Pal et al., 2018)
Graphene	AuNS-ssDNA	ssDNA	Graphene/AuNS	Linear dynamic range up to 10^{-8}	500 aM	(Zagorodko et al., 2014)
rGO-AuNPs	–	GM food	hairpin catalysis	0.5–500 nM	12 pM	(Z. Chen et al., 2018)
Graphene coated chip	Gold	ssDNA	High signal to noise ratio	1 μM	–	(Song et al., 2010)
Graphene	–	miRNA-141	Graphene layer based	1 pM	1 fM	(Wang et al., 2016)
GO	AuNPs	miRNA, adenosine	GO surface for immobilization, AuNP for amplification	0-100 fM, 0.1 pM-2.0 nM	0.1 fM, 0 fM	(Yang et al., 2017)
GO/Au	AuNPs-ssDNA	ssDNA	indirect competitive inhibition assay (ICIA)	10^{-6} to 10^{-8}	1.6 μg/mL	(Xue et al., 2014a)
GO/p-Au	p-Au	Alpha- thrombin	Graphene aptamer non-covalent attachment	0-150 nM	0.05 nM	(Wang et al., 2011)
PDA-Ag@Fe_3O_4/rGO	Gold doped rGO	IgG	Improved Sandwich assay	0.019–40.00	0.76 nM	(Li et al., 2018)
graphene sheet- amine-modified Au film	Au bypyramid	IgM	gold bipyramid nanoparticles as sensitivity enhancement and selective wavelength-modulation	0.03–32 μg mL^{-1}	–	(Zeng et al., 2014)
COOH-Graphene	SAM layer of ODA	IgG in clenbuterol	SAM layer and COOH-graphene based sensitivity	0.01–10 ng mL^{-1}	6.57 pg mL^{-1}	(Yan et al., 2014)
GO-SPA	Au chips	IgG	SPA modified GO-Au sensor increase the sensitivity	0.1–50 μg/mL	0.1 μg/mL	(J. Zeng et al., 2013b)
GO/AuNPs	Ag coated polymer Si fiber	IgG	Enhancement of electric field due to GO	0.4985 nm/(μg/mL)	10 fM	(Wang and Wang , 2018)

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Table 1 (*continued*)

Nanocomposite	Fabrication	Analyte	Methods of sensitivity enhancement	Sensitivity	Limit of Detection	Refer.
GO	SPA co-modified TFBG	IgG	TTBG increased more light and analyte interaction	30–100 μg/mL	0.5 μg/mL	(Q. Wang et al., 2018)
GO	gold bypyramids	IgG	SPA enhanced the number of antibody scaffold for sensing	0.15–40 μgmL^{-1}	–	(Wu et al., 2015)
AuNRs-GO	EDC/NHS based anti-IgG	IgG	GO & Au for high sensitivity.	1.6–50 μg/mL	–	(H. Zeng et al., 2013)
PDDA-GO, GO-CHIT, rGO-CHIT	Covalent attachment	BSA	GO involved direct quantification of protein			(Emiliano N Primo et al., 2018)
AgNPs/GO NCs	EDC/NHS mediated fabrication	anti-BSA	C=O group of GO BSA protein conjugation	1.45 nM to 145 fM	145 fM	(Chen et al., 2018a,b)
Au/GO-COOH	(C=O)fn-GO NCs–Au chip	BSA	bio-molecular adsorption via hydrophillicity	–	–	(Chiu et al., 2017)
Graphene sheets	Graphene/Cr/Au	Bovine serum albumin	affinity-amplified immunoassay	–	10 ng/mL	(Chiu et al., 2015)
GO layer	FBG (Fiber Bragg Grating)	Ethanol	FBG-light interaction and graphene layer-sensing enhancement	2.5 fold than Au layer	–	(Arasu et al., 2016)
rGO-AuNPs	Au–S-ssDNA	GM food	hairpin catalysis	0.5–500 nM	12 pM	(Z. Chen et al., 2018)
graphene nanoplatelets (GNP)	tin oxide (SnO2)	hexachlorobenzene (HCB)	Nanocomposites used to enhance the chemical reactivity for HCB	0.0 g/L to 10^{-2} g/L	8.67×10^{-13} g/L	(Sharma et al., 2017)
graphene	–	Methylene blue (MB)	Electrostatic attraction between MB-Gr	0.01–10 μM	–	(Kaya et al., 2018)
Graphene layer	MoS_2 layers	Urea	Gr-MoS2 layer signal efficiency	5.24%	50 nM	(Jamil et al., 2019)
Graphene/Au	Gold-based SPR	Carbohydrate-lectin	Increased adsorption	–	1 $\mu g\,mL^{-1}$	(Penezic et al., 2014)
Au chip	Coated with rGO-PEI-glycan	E. Coli (discrimination between E. coli strain)	electrostatic interactions, charge variations in between close to the binding pockets	–	10^2 cfu/mL	(Subramanian et al., 2014)
GO & dextran	Capped AuNPs	concanavalin A	Sandwich assay	1.0–20.0 μg/mL	0.39 μg/mL	(Huang et al., 2013)
Go/Au	Lipase 4-ATP stabilized	Tributyrin	Gold-graphene induced sensitivity	20-350 mg/dL	20 mg	(Bhardwaj and Basu, 2018)
Au-graphene monolayer	Gold SPR chip	A gas sensor (NO_2,H_2,CO)	Reactivity of oxygen of graphene	1–10000 ppm	1 ppm	(Cittadini et al., 2013)
NPs/rGO	Gold SPR chip	Nitrogen gas	Nanocomposites enhanced sensitivity	250 ppm	–	(Khalili Fard et al., 2018)
GO-CS	Gold SPR chip	PB (II)	GO-CS covalent bonding	5 ppm	1.112 ppm	(Lokman et al., 2014)
ferric oxide-magnetite-reduced GO	Au/Fe2H2O4–Fe3O4-rGO	Arsenic	Nanocomposite based enhancement	2.196 °ppb−1 and 0.960 °ppb−1	0.1 ppb	(Al-rekabi et al., 2019)
PMMA/rGo	Gold SPR chip	NH_3 Gas	Nanocomposites enhanced sensitivity	10-100 ppm	–	(Mishra et al., 2014)
subuliform fiber optics	graphene-wrapping	NH_3 Gas	graphene based dielectric constant and effective index change in a waveguide manner	–	0.015 nm/pp	(Y. Zhao et al., 2018)
GCNT (Graphene Carbon Nanotubes)	Optical fiber cladding	Methane gas	GCNT based methane carbon adsorption based interaction	10-100 ppm	10 ppm	(Mishra et al., 2015)
cellulose/graphene oxide composite thin film	hexadecyltrimethylammonium bromide	Copper ion	Thin film nanocomposites enhanced the sensitivity	0.01–0.5 ppm	0.01 ppm	(Daniyal et al., 2018)
cellulose/graphene oxide composite thin film	hexadecyltrimethylammonium bromide	Nickel ion	Thin film nanocomposites enhanced the sensitivity	0.01–0.1 ppm	–	(Daniyal et al., 2019)
chitosan-graphene oxide composite	4-(2-pyridylazo) resorcinol (PAR)	Cobalt ion	Thin film nanocomposites based enhancement	10-100 ppm	0.00069° ppm^{-1}	(Saleviter et al., 2018)
graphene-molecule imprint polymer (Gr-MIP)	Au chips	L-Tryptophan	Hydrogen bonding based specific L-type detection	0.150–2.50 mmol/L	0.105 mmol/L	(Xu et al., 2018)
Graphene-gold spr	Cu2+ coordinated NTA	Cholera toxin	Ultrathin graphene	0.004 ng mL^{-1}- 4 ng mL^{-1}	4 pg mL^{-1}	(Singh et al., 2015)
GO	*S.typhi*antigen	S. typhi mab	GO on gold SPR increasing the adsorption of Antigen	1:25–1:1800 dilution of Ag:Ab	1:1800	(Singh et al., 2012)
aptamer-graphene oxide	SAM gold	prion disease-associated isoform (PrPSc)	specific affinity sandwich SPR	0.001^{-1} ng/mL	4.24×10^{-5} nM	(Gu et al., 2017)
AgNPs-rGO-Au	Magainin I	Escherichia coli O157:H7	Sandwich nanocomposites	1.0×10^3 to 5.0×10^7 cfu/mL	5.0×102 cfu/mL	(Zhao et al., 2018a,b,c)
GO-coated	Au-chip	Specific gravity based Food product detection	GO-coated Au-double sensitivity	10^5	–	(Garifullina et al., 2018)

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Table 1 (*continued*)

Nanocomposite	Fabrication	Analyte	Methods of sensitivity enhancement	Sensitivity	Limit of Detection	Refer.
Graphene	ssDNA	Mycobacterium tuberculosis complex	π-π stacking with hydrogen bond influences the hybridization	–	28 fM	(Prabowo et al., 2016)
Graphene	Au SPR chips	Pseudomonas	high adsorption efficiency of graphene	33.98 (Degree/RIU)	0.2987	(Verma et al., 2015)
Graphene/chalcogenide prism/ gold film	graphene-multilayer and gold		high adsorption efficiency of graphene	43.18°/RIU	–	(Maharana et al., 2014)
Graphene	Gold film	Antibody anti-cholera toxin	Ultrathin graphene layer controlled immobilization and amplification	–	4 pg mL-1	(Singh et al., 2015)
Graphene nanosheets	Au bipyramids	bovine IgM	Specific arrangement of graphene sheets and gold bipyramids	0.03–32 μgmL^{-1}	0.03 μgmL^{-1}	(Zeng et al., 2014)
graphene	Gold SPR chips	folic acid protein (FAP)	CVD grown graphene chips	5–500 fM	5 fM	(He et al., 2017)
rGO in chitosan modified silica sol-gel with fiber	Silver coated probe	Caffeine	Nanocomposites based modification of dielectric function	0–500 nM	1.994 nM	(Kant et al., 2017)
reduced graphene-polyaniline (rGO-PA)	Fiber optics	pH sensor (2.4–11.35)	Change PA, change in band gap of rGO with respect to pH	75.09 nm/pH	–	(Semwal and Gupta, 2019)

GO decorated with AuNR–antibody conjugates to 10 mg/mL solution of transferrin resulted in 10.2 nm shift in resonant wavelength. Also, GO/AuNR-AB had a good response over the concentration range of 0.0375–40.00 (saturation concentration)μg/mL compared to 1.25–40.00 μg/mL by Au-chipset. Zainuddin*et al*(Zainudin et al., 2018). prepared a novel system for potassium ion (K^+) detection. They used a combination of valinomycin doped chitosan-graphene oxide (C–GO–V) thin film. C–GO–V thin film was deposited on the gold surface using a spin coating technique. The comparative investigation concludes to monitor the SPR signals for K^+ detection in solution with and without C–GO–V thin film. The sensor produces a linear response for K^+ ion up to 100 ppm. The sensitivity and detection limit observed 0.00948 ppm and 0.001 ppm, respectively. These experimental findings showed that the C–GO–V film is high impending as a sensor element for potassium that had been proved by the SPR measurement.Kamali et al.(Zangeneh Kamali et al., 2015)reported chemical reduction based approach for synthesis of silver-GO nanocompositesfor detection of different biomolecules such as DA (dopamine), AA (ascorbic acid) and UA (uric acid). The density functional theory portrayed DA and Ag/GO sensor had more interaction which probably one of the reasons for having a LOD of 49 nM and 30 nM for intensity and position based SPR as obtained, which had sensitivity and selectivity as compared to other two biomarkers. Although, the same sensor would have detected AA and UA with LOD of 634 nM, 1.64 μM and 927 nM, 2.15 μM specific for intensity and position respectively. Detection of these biomolecule is essential for early prognosis of pathogenic conditions of the body.Hu et al. (2014b) mentioned PDA (Polydopamine)-graphene nanocomposite based sensing of carcinoembryonic antigen (CEA).Wherein, dopamine was oxidatively polymerized in the presence of GO in an alkaline medium followed by conjugation of antibody. The increase in sensitivity had been seen with LOD 500 pg/mL. This high level of sensitivity was obtained due to the dual signal amplification achieved via improved sandwich assay and secondly via PDA catalyzed reductive gold deposition on PDA-rGO. Detection of dopamine was further extended using aptamer-fiber optics based SPR by Hu et al. (2018) reported higher sensitivity detection of dopamine. They had explored a nanoscale metal-coated tilted fiber Bragg grating (TFBG) imprinted in a commercial single mode fiber core without further modifications. The TFBG sensor possesses a single layer graphene coating over the gold-coated fiber surface, functionalized with a selective DNA aptamer for highly sensitive detection of target molecules. Due to conformational changes in response to dopamine surface affinities resulting in aptamer it established the capture of dopamine molecules by the DNA aptamer. Overall process amplifies the surface RI modulation over the fiber surface to enable specific dopamine concentration measurement in real time via monitoring of the SPR signals. The sensor showed a linear response for dopamine concentration in the range from 10^{-13} M to 10^{-8} M with a lower limit of detection of 10^{-13} M. The detecting concentration was found to be lower than the concentration fluctuations of dopamine in the human brain; hence, it may be the futuristic application for the real-time measurement of the dopamine. Semwal et al.(Semwal and Gupta, 2018) claimed that the large surface area of GO nanosheets imparts good performance in SPR sensors, its involvement in the decomposition of hydrogen peroxide results in changed RI. A graphene based LSPR and SPR sensors were fabricated for cholesterol oxidase detection. Three sensors were prepared viz. first, an enzyme entrapped gel layer over silver coated unclad fiber; second, an enzyme immobilized over silver and GO coated unclad fiber; and third,an enzyme immobilized over silver-GO-silver NPs coated unclad fiber. The performance of all these probes has been compared in conditions of operating range, sensitivity and LOD. Fabrication of sensitive fiber-optic cholesterol sensor utilizing cholesterol oxidase (ChOx) has been carried out. The effect of pH, selectivity, repeatability and stability of the sensor had also been investigated. The sensor drawn result for pH 7 with good repeatability, stability. In addition, it can be operated in the range 0–10 mM and its SPR/LSPR spectra lie in the visible region.Primo

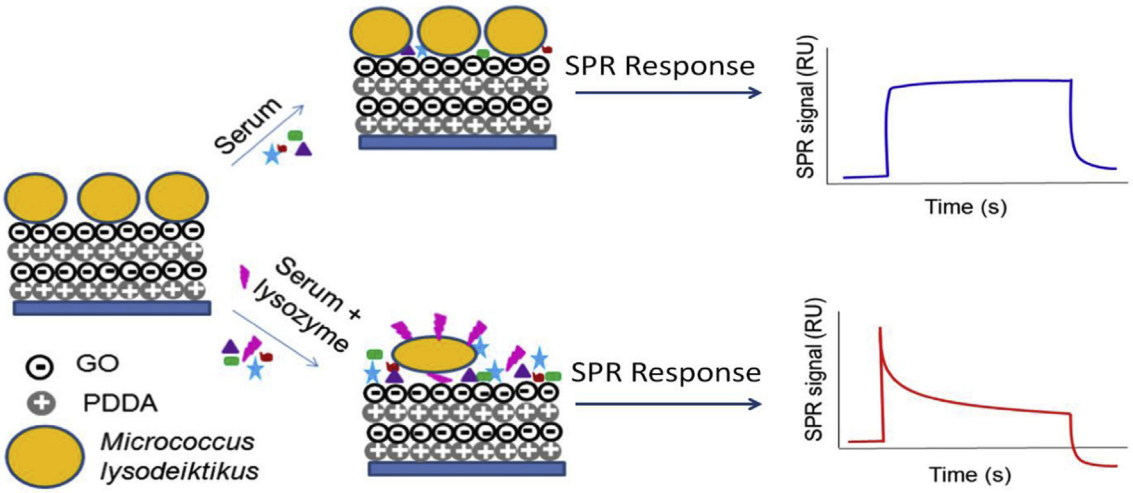

Fig. 6. SPR responses in absence and presence of lysozyme. Reprinted with permission from (Vasilescu et al., 2017), Copyright 2017 Elsevier.

et al. (Emiliano N. Primo et al., 2018a,b)reported the first of its type of novel optical sensor for cardiac biomarker galectin-3 by implying anti-Gal3 antibody, this bioaffinity guided interaction was enhanced by thiolated Au chips with four bilayer of SAM polymer named as poly-(diallyldimethylammonium chloride) (PDDA) and GO. To achieve this, Au-chips were thiolated with MPS solution, after which a layer of PDDA was added andfinallyreactedwith GO for specified time to get uniform layers.The procedure can be repeated depending upon the number of desired layers. The sensor surface had been evaluated by different spectroscopic techniques prior to immobilization sensing. Here, GO was advantageous in both ways as antibody adsorption and as amplification tag increasing the sensitivity up to 10.0 and 50.0 ng mL^{-1}. Researchers also used the 3-aminephenylboronic acid (3ABA) as bio-recognition element for antibody (anti-Gal3 antibody) immobilization, the results were obtained without further amplification and specificity obtained via 3ABA providing LOD 2.0 ng mL^{-1}. A good recovery ratio (1:3) of 30.0 ng mL^{-1} Gal3 with SPR signal 108% showed the ability of the proposed sensor in a clinical diagnostic setting for early detection of heart disease.

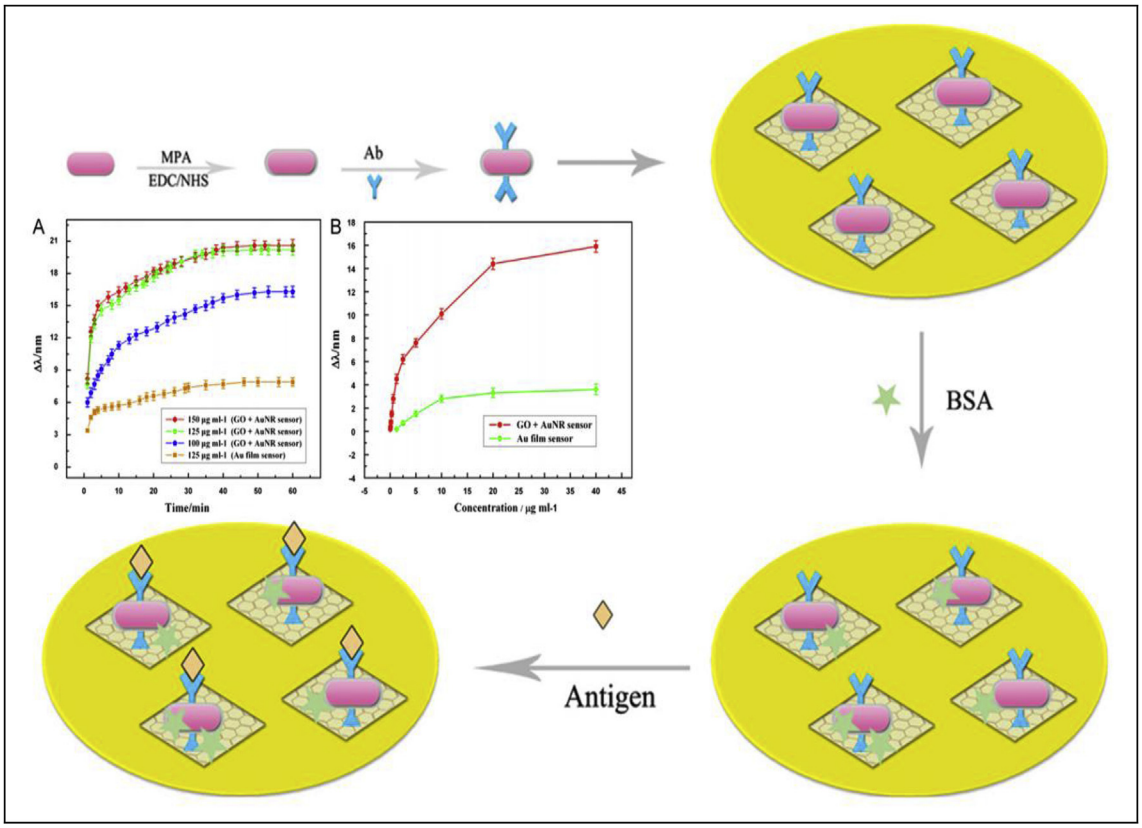

Fig. 7. Scheme of fabrication, functionalization and antigen immobilization. For clarity, all of the molecules were not drawn to scale. Reprinted with permission from (J. Zhang et al., 2013a), Copyright 2013 Elsevier.

3.2. Sensing of DNA, RNA and oligonucleotides

Detection of alteration in DNA and RNA sequence is required in the rapid clinical diagnosis of genetic diseases and disorders. Detection of mismatch occurring at a macromolecule level by sensing false DNA and RNA as well as pair mismatch or proofreading of the pair could give the probable outcomes of future genetic diseases.Thus, it is necessary to detect the base pair mismatch which is strongly enhanced by the use of nanomaterial as a signal amplification tags(Y. X. Chen et al., 2018). Many DNA binding assays have been carried out using conventional SPR sensors(Milkani et al., 2011) but they lacks in reliable SPR technique, which can detect minute and sequence mismatch.Likewise, quantification of DNA/RNA based SPR should have to be sensitive up tofemto (10^{-15}) to attomolar (10^{-8}) level pertaining to their expression in the body is quite low. Meanwhile, the short sequence mismatch, for single and double mismatch DNA, which is out of limits of detection of conventional SPR as a low RI change used happens. GO with other nanomaterials would serve the proposed purpose of ultrasensitive detection of nucleotides-double-stranded DNA (dsDNA) binding events (Rahman et al., 2017). The authors have investigated the state-of-the-art graphene coated SPR biosensor using tungsten disulfide for the detection of DNA hybridization had been added. In addition, they reported remarkable enhancement of the overall performance of the sensors with graphene with decrease in the other performance parameter also. Their numerical analysis showed that the variation of the SPR angle for unjust DNA strands is quite negligible while that for complementary DNA strands is significantly countable(Rahman et al., 2018).

GO being a DNA quencher especially for ssDNA(Robinson et al., 2008) is the first choice for DNA based biosensors(Gao et al., 2014). Also, the SPR signal is very sensitive to a change of oxide state of GO. After the first precise use of GO for aptamer sensing (Wang et al., 2011), extensive work had been taken out by the researcher for grapheneous material based DNA/RNA sensing(Akca et al., 2011). Choi et al. (2011b) numerically demonstrated the ability of graphene silver interfaces for detection of DNA hybridization phenomenon, particularly three times higher than the usual gold substrate. However, the selections of a number of graphene sheets certainly interfere with the sensitivity, which must consider this point, was the additional outlay of proposed study. Similarly, Pal et al. (2018)numerically studied the proposed phosphorous nanocomposite having graphene coating on it, the results drawn from studying the hybridization events between nanocomposites and cDNA, the proposed nanocomposite has shown the higher sensitivity influenced by desorption of the nanostructures leads to the detection of DNA hybridization events. In experimental background, Zagorodko et al. (2014)realizedDNA hybridization events in the ~500aM lower concentration. The method used was the non-covalently functionalization of gold nanostars carrying single-stranded DNA (ssDNA) on graphene coated SPR. Similarly, the ultrasensitive detection of short sequence mismatch involving with precision up to two base mismatch of HP1 and HP2 (Hairpin 1 and 2) and in a lower concentration of twelve pM (picomolar). This methodology gives hope for future outlook for an alternative solution for single base pair mismatch sensors (Z. Chen et al., 2018). Moreover, graphene's property as a reducing agent makes it a valuable candidate to fabricate MNPs onto themselves by means of reducing the size of MNPs. Also,the planar nature of graphene plays a role for direct immobilization of DNA for making sensing probe or directly to give a signal by adsorbing DNA through pi force(Akca et al., 2011; L Wu et al., 2010a,b)and leads to precise sensing of DNA binding events or hybridization by virtue of molecular recognition.Graphene-based SPR has shown a larger shift and 8 folds sensitivity of RI than conventional SPR sensors. The first successful demonstration of graphene-based gold sensor could efficiently sense a single strand DNA (ssDNA) bySong et al. (2010)found that graphene conjugated gold materials are precise in the sensing of ssDNA, a single pair mismatch could be easily identified owing to high signal to noise ratio provided by graphene.Wang*et al*(Wang et al., 2016).detected miRNA-141 (microsomal RNA) a part of a cell, with the help of GO/AuNPs SPR chip system. The four cancerous cell lines with closely related sequence and high homology had been chosen than the relative expression of mi-RNA 141 in target and 22Rv1 cell lines and determined using standardized qRT-PCR protocol and proposed chip system. Both of which were relative to each other and with reported literature. It was reported that the GO/Au system had three order low magnitude i.e. 1 fM, than gold SPR system i.e. 1 pM. It was reported that the sensitivity and the number of graphene layer has a direct co-relation.GO was combined with plasmon AuNPs to generate the LSPR for detection of the miRNA and adenosinewhere GO based high surface area provided the convenience for the immobilization of captured DNA molecules. On the other hand, AuNPs enabled the signal amplification to obtain the LOD and detection in the range of 0.1 fM and 0–100 fM, 0.1 pM - 2.0 nM respectively for both molecules (Yang et al., 2017). Particularly, sensitivity for ssDNA had increased by using ZnO bimetallic graphene nanoconjugates(R. Kumar et al., 2018). More specifically,Xue et al. (2014a) developed chip inclusive of indirect competitive inhibition assay (ICIA) for detection of ssDNA and dsDNA hybridization. The ssDNA and GO interacts by means of hydrogen bonding. While for dsDNA observed a weak binding with GO due to interstrand binding between dsDNA hindering the process.Compliment strand (csDNA) however, used to block the ssDNA adsorption, the dsDNA so form by this interaction. Thus, this observation of indirect competitive inhibition was further utilized to detect a particular sequence of ssDNA using GO SAM onto gold sensor chip to achieve the sensitivity in micro to femto range for csDNA with HIV-1 U5 long terminal repeat sequence and the femto molar for ssDNA. Wang et al. (2011) conducted an aptamer sensing study with the help of graphene/positively charged gold nanocomposites. The graphene was chemically reduced in the presence of hydrazine and assembled with positively charged gold nanocomposites. This electrostatic interaction provided a firm film of Gr/Au. Further on the aptamer i.e. alpha-thrombin was non-covalently attached with graphene. This fabricated assembly was very sensitive on aptamer and target interaction as this leads to ruptured graphene-aptamer bond. The proposed mechanism had practically been seen in the dynamic concentration range of 0–150 nM with LOD of 0.05 nM.

3.3. Application in immunosensing

Nowadays, SPR plays a centralized role for immunoassays(Mullett et al., 2000). Where a graphene-based nanocomposites had shown high sensitivity towards biological interaction, such as antigen-antibody binding events(Li et al., 2018), the sandwich assay supported IgG detection methodologies had been amplified by using Polydopamine-Ag@Fe_3O_4/reduced graphene oxide (PDA-Ag@Fe_3O_4/rGO). Nanocomposites provides the multifunctional effects because of different property oriented materials into one platform, giving them edge over single nano-material in terms of character based sensitivity enhancement. The Ag-rGO chemistry, the doping of gold particles onto free sites of rGo had amplified the response. Long chain polymers with functional groups surpasses the activation process for antibody immobilization. With such bulky arrangements, the sensitivity of proposed sensor was higher than the normal gold chips ranging in between 0.019-40.00 μg/mL. Zhang et al. (2014) developed gold bipyramid nanoparticles as sensitivity enhancement and selective wavelength-modulation for the determination of IgM. Graphene with larger surface area and numerous functionalities were prepared and assembled on nanosized sheet on amine-modified Au film. In comparison to the earlier literature, prepared AuBPs as sensitivity enhancer showed an acceptable response to bovine IgM in the concentration range of 0.03–32 $\mu g mL^{-1}$.In addition to that, Yan et al. (2014)approached to build platform for immunoassay of clenbuterol a livestock feed additive with sensitivity in the range of 0.01 $ng\,mL^{-1}$ to 10 $ng\,mL^{-1}$ and muchlower LOD i.e. 6.57 $pg\,mL^{-1}$. A

sensor platform was fabricated firstly with a SAM layer of Octadecanoic acid (ODA) followed by the carboxy-modified graphene (carboxy-GR), wherein, the carboxy group absorbed the goat anti-mouse IgG. GO-Au chips were first modified with staphylococcal protein A (SPA) owing to its specificity towards Fc portion of antibodies(J. Zhang et al., 2013b), This gold decorated GO and SPA modified sensor had shown sensitivity enhancement for IgG without chemical modification of substrate for antibody binding. The responses were in the range of 0.1–50 μg/mL. The SPA modified chip had shown not only the signal amplification but also bypassing the chemical modification of the antibody. Similarly, in a recent attempt, Wang et al. (Wang and Wang, 2018) carried out immunoassay of human IgG using goat anti-human IgG with LOD of 0.04 μg/mLand high sensitivity of 0.4985 nm/(μg/mL) when detected by using optical platform having silver coated polymer cladding silica fiberwith immobilized GO. For IgG functionalization, the platform was immersed in EDC/NHS (1:1 v/v) for 20 min at room temperature followed by washing with PBS the group used staphylococcal protein A (SPA) providing the proper orientation favoring the increase in the amount of goat anti-human IgG binding. Finally, the antibody (100 g/mL) soaked the fiber for 2 h, then further dipping in a BSA solution (1% w/v) to block unreacted carboxyl group.The chipset had given reusability up to six cycles when immersed in 10 mM NaOH for 20 min with relative standard deviations of 2.4% and 24.5% for seventh regeneration with specificity for human IgG(Fig. 8). The same effort has been taken by Wang et al.(Q. Wang et al., 2018) for human immunoglobulin G (IgG) detection based on GO and SPA co-modified tilted fiber Bragg gratings (TFBG), this group of researchersalso used SPA again owing to its specificity towards Fc portion of antibodies. The experiment was resulted in highly oriented antibody immobilization on the sensor surface and high antigen–antibody binding efficiency.The responses were in the range of 30–100 μg/mLwith LOD of 0.096 dB/(μg/mL) and 0.5 μg/mL. The SPA modified chip had shown not only the signal amplification but also bypassed the chemical modification of the antibody.The biosensor based on SPA showed response in a concentration range of 1.6–50 μg/mL. Likewise, GO with gold by the pyramids (GBP) was fabricated, where SPA was used to mediate the IgG antibody binding (Wu et al., 2015).The GBP was modified, followed by covalent binding with GO. SPA provided specificity towards the Fc region of different IgGs. Thus, SPA-Ab conceded the conformational changes in the Fc region making the Fab area with IgG binding scaffold protruding outward to react with IgG in the analyte.Other factors included the high level of sensing such as GO enabled large surface area for antibody adsorption by SPA also the fiber optics and TFBG also induced more light and analyte interaction leading to high sensitivity.

This proposed SPA modified GO sensor system could be a valuable tool for the future biomolecules binding assays.GO-AuNRs had been used for the immunoassay of human IgG. The AuNR synthesized by seed-mediated method(Brasil et al., 2015)was conjugated with human IgG antigen of different concentrations. The Au film was modified by GO and attached with goat anti-human IgG solution via carbodiimide-amine chemistry. For IgG-AuNRs, limit of quantification was 16 times lower than conventional MPA (unmodified human IgG) biosensor. The increase in sensitivity of SPR by using GO-AuNR conjugates was explained by Zhang et al.(H. Zhang et al., 2013).Similarly, Chung*et al* (Chung et al., 2015).reported the BSA sensing with the differential in a number of GO and rGOlayer onto the gold based plasmon sensing platform.Xiong et al. (2018)described spin-coated over a layer of GOS (graphene oxide sheet) on the gold SPR sensor chip and utilized for detection ofBSA in between 0 and 10 mg/mL with 14.98 nm shift and 39.35% sensitivity. Chiu et al. (2014)developed highly sensitive immunesensor for BSA. The high sensitivity was due to the formation of a strong covalent bond between GOS and BSA. The author also proposed that the GOS property could be further explored via chemical modification to achieve high sensitivity for protein interaction study.

3.4. Detection of harmful chemicals from food, environmental and pathogen

The molecules having the adverse effect on food and the environment, ultimately leading to the suffering of humans need to be detected at the earliest. Arasu et al. (2016) performed the ethanol sensing on GO layer. In order to generate the evanescent SPR wave, the team used FBG (Fiber Bragg Grating) increasing the light rays from the core region to

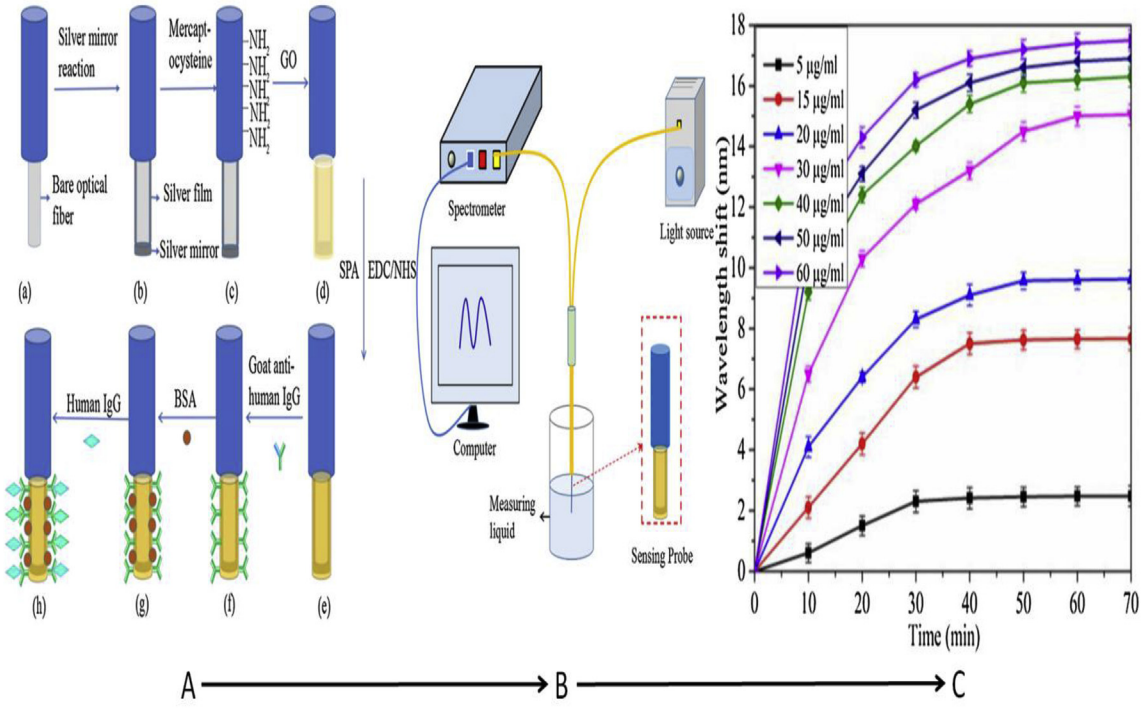

Fig. 8. A. Sensor fabrication process. B. Experimental setup for sensor platform, C. Response curve for different concentrations of human IgG a linear increase can be interpreted from the graph. Reprinted with permission from (Wang and Wang, 2018), Copyright 2018 Elsevier.

cladding region. Hence, light interacting with interfaces, resulted in sensitivity with 2.5 times compared to traditional gold coated SPR. Sensing enhancement was achieved by the addition of a nanostructured GO that proved an importance of graphene as the sensing element. Biotechnological advancement created genetically modified (GM) food that are resistant to the plant diseases providing additional nutrients. But, their side effects may have a significant health and environmental problems. Identifying this, would be helpful for ongoing biotechnological advancement for safety perspectives. Chen et al. (Z. Chen et al., 2018)addressed this with their work for standardization of genetically modified organism by using advance SPR techniques based on fiber optic. A fabrication process involved thiol bonding between the Au and thiol-modified ssDNA afterwards reduction of the disulfide bond of S-ssDNA afforded stable Au–S-ssDNA bond. A signal was amplified by use of rGO-AuNPs. The said nanocomposite was further reacted with HP1 (Hair Pin 1) to achieve the ultrasensitive detection up to two base mismatches with HP2 (Hair Pin 2) with a detection limit of 12 pM. Kushwaha et al. (2018) elaborated theoretical interpretation of sensing of pathogens like *pseudomonas*. Performance analysis of SPR biosensors for detection of pseudomonas-like bacteria with improved sensing ability was also attempted by incorporating graphene layer(Verma et al., 2015). A fiber optic nano-hybrid sensor was designed using graphene nanoplatelets (GNP) supported with tin oxide (SnO_2) nanoparticles for detection of hexachlorobenzene (HCB)(Sharma et al., 2017). Where, a nano-hybrid of SnO_2 nanoparticles decorated on GNP were assembled and combined effects of both were used to enhance the chemical reactivity for HCB detection. GNP/SnO_2 nano-hybrid owing to its synergistic properties. The proposed sensor had high sensing performance to detect HCB concentration in a wide range from 0.0 g/L to 10^{-2} g/L. The authors claimed a lower limit of detection of the fabricated sensor, about 8.67×10^{-13} g/L, compared to the reported in the literature. The sensor with robust nature can be used for online monitoring and remote sensing.

Besides this, Kaya et al. (2018) applied graphene monolayer assessed binding kinetics of small dye molecules Methylene blue (MB) using the SPR system. The binding kinetic study was further differentiated into three parameters mainly; angular dip (θ_{min}), intensity (I_0), and width (w). However, the same system was employed for the determination of the nature of interaction with more or less results using more or less similar kind of molecules (nile blue A, calmagite). The high adsorption efficiency of graphene and MB's optical property relative to the optical system wavelength had helped to get the desired sensitivity and detection (0.01–10 μM). The MB is a redox indicator useful for electrochemical sensor system being a redox mediator the proven electrostatics attraction with graphene is by fact is true. So far the multiparameter SPR and graphene based sensor could be an enormous source to observe the binding kinetics of small molecules on absorbing surfaces as well as for analyte molecules that absorb well under the measurement wavelength. Jamil et al. (2019) proposed the biosensor with Kretschmann configuration based on the graphene and MoS_2 layers based on the FDTD method, computer-based simulation studies the interaction of material layers with urea. The biosensor platform has shown good response in terms of the reflectance intensity as well as FWHM. The outcomes of the study promised the development of future experimental protocol.

During the disease or disease progression there are a number of interactions takes place at the cellular level, such as a virus-host cell, protein-protein, pathogen-cell to name a few. Usually, this involves a number of chemical signaling between cell components and foreign or simply studying this cell content like carbohydrate, amino acids, protein, has shown effective for insight and future development against the diseases. Graphene-accustomed optical sensors provide long-range RI detection and therefore a promising means in the biological cell detection field (Yang et al., 2018). A Carbohydrate-modified graphene/gold interface was implied to study the carbohydrate–lectin interactions as demonstrated by using a different lectin model (Penezic et al., 2014). Subramanian et al. (2014) performed the study for the cell surface and pathogen interaction taking place. Huang et al. (2013) observed probable interaction between carbohydrate (dextran) and concovalin A protein. The team observed the amplified signal 28.7 times with one pot synthesized dextran-gold nanoparticles (Dex-AuNPs) nanomaterial were fabricated on GO-Au SPR meticulously via π-π interaction and detected at a 1.0–20.0 μg/mL concentration range in PBS (phosphate buffer saline) at a 7.4 pH. Surprisingly the LOD reported was at 0.39 μg/mL. In a similar manner, Bhardwaj et al. (Bhardwaj and Basu, 2018) studied the lipase enzyme immobilized on 4-ATP (4-amino thio phenol) stabilized GO/Au for triglyceride detection using tributyrin as a model molecule with a linear range between 20-350 mg/dL. However, the system has detected the analyte with the binding of 3.4 mM owing to the presence of enzyme but it lacks the specificity which is a limitation in nowadays for commercialization of graphene-based biosensor. Gas sensors are crucial and play important role in the industrial and environmental investigation. In a pioneering work, B. Liedberg et al. (Bo et al., 1983)developed gas sensorspaving way for the SPR based sensor. While Maharana et al. (2015) numerically derive the high sensitivity with graphene-silver nanocomposite for gas sensing. In a similar manner, with an emphasis on both oxidant and reductive gases Cittandini*et al* (Cittadini et al., 2013). detected oxidants and reducing gases based on the fact that GO would change its electronic and optical properties, which shall further be raised by use of Au monolayer, favoring a well-resolved shift in the presence of both reducing and oxidizing gases. The underlying mechanism for which depends upon the surface absorbed oxygen of GO reacted with both the gases by subtracting and donating electron on the surface, for oxidant (NO_2) and reducing (H_2 and CO) gases respectively. The study was performed for both gases and in the mixture as well, in 1 ppm (parts per million) to 10,000 ppm concentration. Later,electro-plasmonic gas sensing highlighted the SPR sensitivity shift of 5 nm using NPs/rGO, which for AuNPs and SPR was found 1.4 nm, tested again the same concentration of nitrogen gas (250 ppm)(Khalili Fard et al., 2018). Monitoring heavy metals in food, drugs and in the environment is an essential and challenging task as it can lead to several diseases, also proper and real-time detection of them will be help in risk management. Lokman et al. (2014) detected lead (II) in water (5 ppm concentration) with enhancement of sensitivity up to 1.112 ppm which was attributed to property of GO that covalently bonded specifically with chitosan via oxygen-nitrogen bond formation. It is worth mentioning that the GO have superior sensing ability due to high surface-volume ratio and rough textured surface. The Arsenic (II &V) metal in parts per billion range was sensed with the help of sonicated rGO and hydrous ferric oxide-magnetite in a magnetic stirrer assisted method to get 7 nm thick nanofilm(Al-rekabi et al., 2019).As observed in AFM a nanocomposite layer with root means square (RMS) value of 5.2 nm. However, this after absorption of arsenic almost doubled and for AS(III) given 23.7 RMS observed, showing the highly selective absorption potential of prepared film which further decoded as 1° resonance angle shift for each ppb in linear range of 0.1–1 ppb in SPR with LOD of 0.1 ppb. More specifically, Mukhtar et al. (Mukhtar, 2018)detected the lead ion with variation based on nanocomposite thickness. Ammonia is one of the most toxic, gas which can be more selectively detected by using graphene owing to its high surface coverage and signal amplification properties (V. Kumar et al., 2018). Mishra et al. (2014) performed the ammonia gas sensing on fiber optic covered with polymer named polymethyl methacrylate (PMMA) reduced nanocomposite (rGO). The nanocomposites sheets were prepared by bulk polymerization of MMA with respect to the different concentrations of rGO (in 0–15%) by casting methods. The fabrication of optical fiber was done in such a way that PMMA/rGO film in intimate contact with a copper coated unclad section of fiber optics. Although the group studied sensing ability of proposed fiber optics based biosensor for a number of gases of which ammonia was detected with high sensitivity. However, other gases would also be detected by a wavelength interrogation by relative

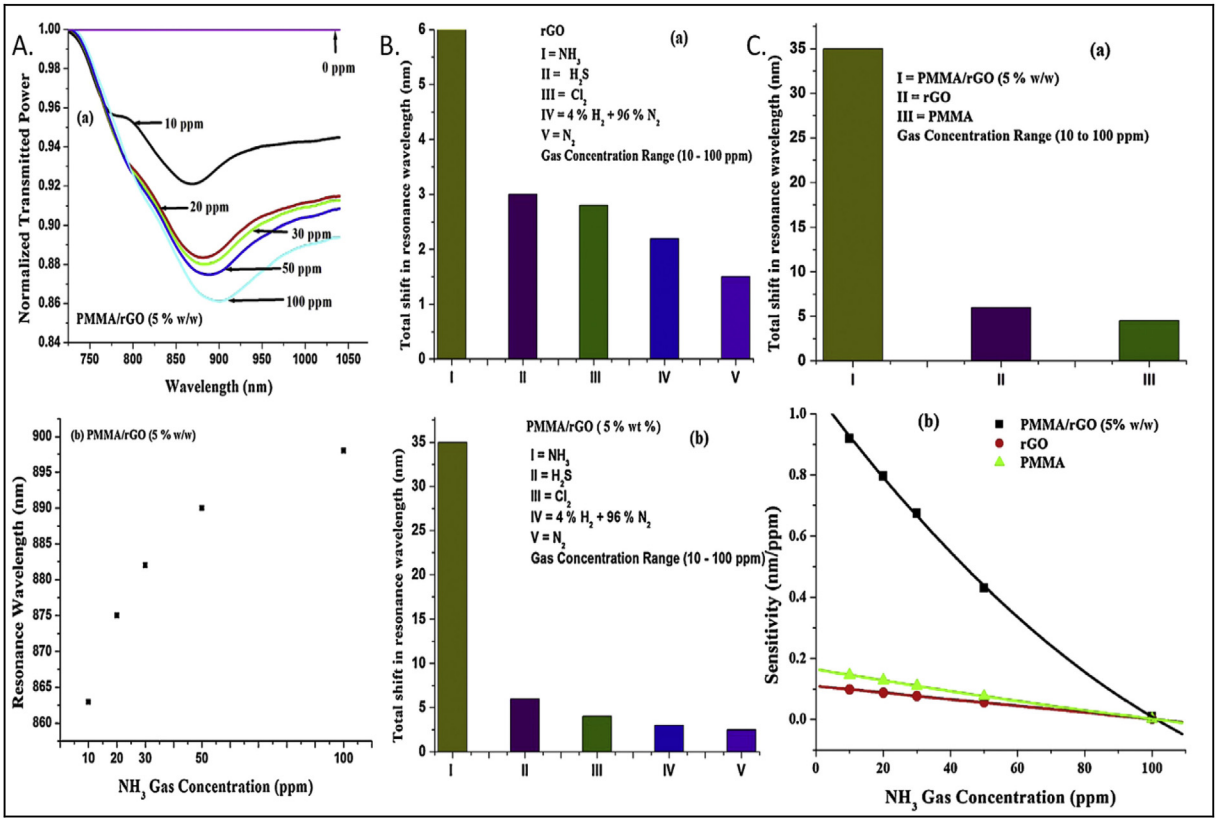

Fig. 9. A. SPR spectra for different concentrations of ammonia gas, with variation of resonance wavelength with the concentration of ammonia gas for copper/(PMMA/rGO) B. Total shift in the resonance wavelength for different gases with rGO and PMMA/rGO. C. The variation of sensitivity with the concentration of the NH_3 gas for three different sensing probes. Reprinted with permission from (Mishra et al., 2014),Copyright 2014 Elsevier.

wavelength spacing between the wavelengths of gases under a study as shown in Fig. 9. As discussed earlier, this fiber optics were capable of online monitoring, probes miniaturization, low cost, and resistant to electromagnetic field interference.

Zhao et al.(Y. Zhao et al., 2018) illustrated the sensitivity of the new graphene-wrapped on the subuliform fiber over layered between a down-taper and an up-taper. Itconsiderably increased the evanescent field and thereby high adsorption capacity of graphene changes the dielectric constant and the effective index of the entire mixed waveguide onto the surface which causes corresponding wavelength drift and attenuation to achieve the measurement of ammonia. A significant sensitization effect on the ammonia gas sensing, this platform provided efficient sensing of ammonia as low as 0.015 nm/ppm. Alternative nanocomposites were used for effective sensing of methane and other gases through optical fiber based on SPR module, the graphene involved GCNT (Graphene Carbon Nanotubes) confirmed via FTIR and TEM analysis and had high sensitivity for the different concentration with redshift for each gradual concentration ranging between 10-100 ppm. However, the probe was more reactive to methane in above the concentration most probably due to graphene carbon nanotubes based methane carbon adsorption based interaction, changing the dielectric constant and wavelength shift (Mishra et al., 2015). Hexadecyltrimethylammonium bromide modified nanocrystalline cellulose/graphene oxide composite thin film was fabricated using a spin coating technique and used for sensing copper ion(Daniyal et al., 2018). In addition, the SPR observation showed that copper ion can be detected up to 0.01 ppm. Detection of metal ions was further carried outusing nanocrystalline cellulose-graphene oxide nanocomposites based enhance sensitivity for nickel ion detection in a certain range between 0.01 and 0.1 ppm (Daniyal et al., 2019); study describes binding affinity constant, FWHM, data accuracy, and S/N ratio by implying different mathematical expressions on obtained SPR spectra. The sensor platform was excellent in sensitivity and selectivity aspects which could be helpful for harmful nickel ion detection in biological system and in atmosphere in extremely low ppm concentration. An immobilized 4-(2-pyridylazo) resorcinol (PAR) in chitosan-GO composite was assembled using spin coating technique for the effective detection of cobalt ions(Saleviter et al., 2018). The experimental findings of SPR curves were obtained for different concentration of the Co^{2+} metal ion solution to assess the potential of the composite for metal sensing. The fabricated composite sensor produces a linear response tothe higher concentration of Co^{2+} with a sensitivity of 0.00069 ppm^{-1}. The SPR is a very sensitive in terms of molecular interaction(Xu et al., 2018) of chiral selective detection of L-tryptophan by a new and advanced technique of molecular imprint, which involves the one-pot oxidative polymerization of dopamine with L-tryptophan as a template molecule and GO. During reaction this template molecule was trapped in the cross-linked polymeric network due to the strong hydrogen bonding as evident by DFT followed by chain branching and cross-linking of PDA which after removal of the template molecule leave behind a cavity specific to structure of template i.e. L-tryptophan, this generated graphene-molecule imprint polymer (Gr-MIP) composites was further immobilized on gold SPR chips. The proposed biosensor system was able to distinguish a chiral compound between L-tryptophan (selective due to MIP scaffold) and D-tryptophan based on the hydrogen bonding between composite and L-tryptophan. The ability of this chiral recognition was tested in a condition like different pH in 2.5 mmol/L. The LOD of sensor towards L-tryptophan determined as 0.105 mmol/L with a linear range in between 0.150 and 2.50 mmol/L. The MIP based activity was further stretched by Yao et al. (Qin et al., 2015). Authors synthesized the molecularly imprinted film by a molecular precipitation method in which 'grafting to' instead of 'grafting from' method had been used giving edge as a ultrathin and ready to use with repeated use of the film. In addition, gold nanoparticles (AuNPs)

and rGO provided signal amplification. The MIP scaffold containing ractopamine (0.1 mmol) as a sensing substrate was synthesized with functional monomer unit MAA in an acetonitrile after sonication a 2 mmol of cross linker and 120 mg of the initiator. The process was followed by degassing with nitrogen to remove oxygen, which could interfere otherwise in chain linking layer. The finalmixture was exposed to heat in a gradual manner for a period of 2 h and maintained constant up to 24 Hrs. This precipitated MIP then pour into the final fabrication medium containing AuNPs and rGO in THF medium and drop casted on to the gold chip followed by spinning at 7000 RPM for 15s to obtain the uniform covering. Prepared conjugate has detected the ractopamine with good RI sensitivity provided by AuNPs-rGO.

Salmonella typhi is a pathogenic bacteria which is a causative agent for human typhoid. It is an epidemic worldwide, burdening economical losses and threat to humanity as a potential concern for food safety (Rowe et al., 1997). The CVD grown ultrathin graphene for the controlled immobilization of cholera toxin antigen (biotinylated) on nitrilotriacetic acid (Cu^{2+} conjugated)has been studied (Singh et al., 2015). The ultrathin layer achieved the best-amplified sensitivity enhancement (up to 80%) as compared to a conventional setup with a single layer thickness of graphene and limit of detection as low as 4 pg mL^{-1}. Singh et al. (2012) has designed and developedlabel-free detection using SPR sensor. The detection usually involved the detection of antigen-antibody response between *S. typhi* and *S. typhi* monoclonal antibody (mab), respectively in the various dilutions. More interestingly, the graphene was synthesized by electrochemical exfoliation using a pencil as graphene source which could be the new way for efficient and economical large scale synthesis of GO. In particular range, this carbodiimide chemistry mediated detection methodologies provide future perspective for the low cost detection system. Gu*et al*(Gu et al., 2017). deployed the prion disease-associated isoform (PrP^{Sc}) on to a gold surface in a SAM, its responses was amplified due to highly specific affinity of aptamer-GO (AGO) towards PrP^{Sc} this sandwich SPR assay techniques had given the good sensitivity, LOD (0.001–1 ng/mL/ 4.24×10^{-5} nM) and specificity. However, the proposed sensor could be used for the other low weight molecules with intra-molecular thiol groups by changing the configuration of the sandwich assay format. Zhou et al. (2018a,b,c) detected the *Escherichia coli* O157:H7 a one of the food contaminants, specifically known for severe food poisoning, through Magainin I a well-known antimicrobial peptides that binds to *E. coli*. A communication optical fiber had been cut followed by hydroxylation with piranha solution further coated with AgNPs-rGO solution with final fabrication of gold. This probe is then finally immobilized with 0.4 mM Magainin I at 4 °C for 2 h to immobilize AMP by Au–S bond self-assembly. The prepared FOSPR is went through various characterization techniques and had shown sensitivity in 1.0×10^3 to 5.0×10^7 cfu/mL concentration range with LOD for the microbial agent was found to be 5.0×10^2 cfu/mL (S/N = 3) with the recoveries and relative standard deviations (RSDs) varied from 88% to 110% and 1.8%–7.59%, respectively suitable for practical applicability. A RI of solution changes in response to the amount of solute (analyte) in the solution (sample). This RI is correlated with the concentration of analyte in the study performed by Garifullina et al. (2018)by comparing the specific gravity of sample and standard. This GO-coated Au- sensor had given the double sensitivity over conventional Au sensor owing to high adsorption and optoelectronicity, the observed data was fallen into 95% confidence interval providing the accuracy of obtained results with reference to RI, which will leads to label-free, real-time standardization of food products. An alternative pH sensor having higher sensitivity had been proposed where reduced graphene-polyaniline (rGO-PA) played an active role. For which a SPR based optical fiber sensor coated with rGO-PA was dipped into the different standard pH solution. The band gap of rGO played an active role in accordance with the change in the pH. The change of RI of the sensor surface mainly indicated and correlated with the pH change (Semwal and Gupta, 2019). The SPR sensorsare advantageous in terms of miniaturization, stability, low cost, and more importantly low concentration detection when rightly tailored nanomaterials has been directed to the subsequent stage. The graphene materials is important in achieving the highest binding efficiency, as well as, for the conjugation based sensitivity enhancement (Ghasemi et al., 2017; Tong et al., 2018; Vahed and Nadri, 2019).

4. Conclusion and future perspectives

SPR biosensing has drawn huge attention to researchers as a real-time, *in-situ* and cost-effective tool to explore the molecular interaction without the need of any label. SPR sensor provides the essential insights for molecular binding events in kinematic perspective.The real-time detection of bio/chemomolecules having low molecular weight or low concentration can be done by SPR sensing both in sensitivity as well as kinematical calculations. Recently, nanomaterials based sensing enhancement provided the ultra depth functioning of SPR system to counter disadvantages of other techniques. Graphene-based nanomaterials have emerged as a new promising and pervasive technology due to its innate advantages over traditional materials.Graphene-based nanocomposites are sensor material of choice for SPR system to amplify the signals, promising it for simplistic and enhance sensitivity of SPR particularly useful for detection of small molecules and in low limit of the detection (i.e. fM to aM). On a more specific background, structural characteristics like high adsorption and optical properties are the driven forces for them as sensor material. More precisely, the presence of functional groups that can interact in an ionic, covalent or non-covalent manner makes it the material of choice to extract the biomolecules with the highest efficiency. Graphene is easy to functionalize with other nanomaterials; easy to fabricate and surface modification is another property of it. For instance, graphene and GO thin films are conjugated with several secondary antibodies, anti-immunoglobins and with various substrates such as enzymes, ssDNA, gas, etc. As a result, some of them are currently commercially available for use in different domains. So far graphene emerges as a potential alternative to the conventional SPR for amplification and sensitivity enhancing tags end results in ultrasensitive detection. Overall, graphene-based nanocomposites act as a sensing enhancement substrate and functional amplification tags. Graphene was utilized in a large span of shapes including film, ultrathin film, sphere, arrays, nano-rods, etc.particularly, with a metal substrate like gold and silver. These nanocomposites are able to enhance SPR signals by coupling of gold based LSPR on graphene-based PSPR wave, with an added advantage of the retardation of metal surface oxidation. Optimized thin layer graphene onto the metal surface via self-assembled monolayer in nanometer scale are able to generate a strong signal in response to target molecules. Graphene was fabricated with other nanomaterials for more precise sensing using a conventional SPR sensor to newly commenced optical fibers. It is worth to mention here that GO is highly ranked functional substrate owing to its oxygen, which sometimes also leads to taking a part in the surface modification interactions and sometimes indirect detection of reactive gases and DNA hybridization. The use of TFB gratings with an addition to graphene gold nanocomposites provided vicinity for more light and analyte interaction. Similarly, different simulation and experimental based analysis had proven the graphene with other materials like ITO, MoS_2, etc. as an enhancing substrate. As π- π stacking interaction is not specific and any organic molecule or biomolecules of the appropriate aromatic structure can integrate onto the graphene matrix.On the other hand, the key difference between electronic properties of graphene and GO film, low conductivity affect the sensitivity of GO based nanocomposites. This is currently a major limitation of these SPR interfaces. However, the same limitation could be improved for adsorption of biomolecule for which the binding specificity needs to be addressed properly by altering the surface chemistry of grapheneous derivative. For fabrication, the graphene substrate layer should be adjusted in the optimum number to the silver/gold substrate as an increase in thickness decreases the SPR sensitivity. Due to lack of reliable technique to get a

thin layer of graphene, new emerging trends in new fabrication technique show the promising effect to get one or two atoms thick graphene which could effectively be tuned for enhancing optical sensitivity for better performance with several trials and error modification. The electro-sputtering and CVD are the newer approach to look forward. So far graphene is emerged as valuable material for SPR sensor chip design and modification. The Ability of large-scale synthesis of graphene will definitely lead to a future of graphene-based nanocomposite as a SPR sensor not only in point of care diagnostics but also in online monitoring perspective in reference to fiber optics. In the light of such an extravagant and exceptional quality, the upwards and onwards journey of SPR is definitely having room for graphene-based nanomaterials as sensing enhancement probes, to lay down the future SPR based graphene nano-sensors.

Conflicts of interest

Authors declare no conflict of interest.

Declaration of interests

√ The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

CRediT authorship contribution statement

Pravin O. Patil: Conceptualization, Methodology, Writing - original draft, Supervision, Writing - review & editing, Visualization, Funding acquisition. **Gaurav R. Pandey:** Writing - original draft, Visualization, Writing - review & editing. **Ashwini G. Patil:** Writing - original draft, Visualization, Writing - review & editing. **Vivek B. Borse:** Writing - review & editing, Resources. **Prashant K. Deshmukh:** Writing - review & editing, Visualization. **Dilip R. Patil:** Writing - review & editing, Resources, Supervision. **Rahul S. Tade:** Writing - review & editing. **Sopan N. Nangare:** Writing - review & editing. **Zamir G. Khan:** Writing - review & editing. **Arun M. Patil:** Writing - review & editing, Resources. **Mahesh P. More:** Writing - review & editing. **Murugan Veerapandian:** Resources, Writing - review & editing. **Sanjay B. Bari:** Writing - review & editing, Resources.

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