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Rapid detection of SARS-CoV-2 using graphene-based IoT integrated advanced electrochemical biosensor

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ABSTRACT

Unique characteristics like large surface area, excellent conductivity, functionality, ease of fabrication, etc., of graphene and its derivatives, have been extensively studied as potential candidates in healthcare applications. They have been utilized as a potential nanomaterial in biosensor fabrication for commercialized point-of-care (POC) devices. This review concisely provided innovative graphene and its derivative-based-IoT (Internet-of-Things) integrated electrochemical biosensor for accurate and advanced high-throughput testing of SARS-CoV-2 in POC setting.

1. Introduction

Coronavirus disease (COVID-19) illness triggered by the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) has greatly affected mankind worldwide. Nowadays, it is not only the major cause of demises but also harms the healthcare sector as well as socio-economic conditions globally [1]. Commonly, the real-time polymerase chain reaction (RT-PCR), enzyme-linked immunosorbent assays (ELISA), computed tomography, and lateral flow immunoassay (LFA) are being utilized for COVID-19 diagnostics. Although these modalities offer highly sensitive results but also suffer from few drawbacks like they require a sophisticated laboratory setup, tedious processes, and inaccurate outcomes. Hence, advanced diagnostics modalities with ultra-sensitivity and specificity are required to manage the swiftly spreading COVID-19 [2,3]. Subsequently, electrochemical biosensors have been validated as potential candidates in healthcare applications. They have been utilized as a potential nanomaterial in biosensor fabrication for commercialized point-of-care (POC) devices. This review concisely provided innovative graphene and its derivative-based-IoT (Internet-of-Things) integrated electrochemical biosensor for accurate and advanced high-throughput testing of SARS-CoV-2 in POC setting.
enzymes by providing the active sites that make them suitable in an aqueous medium. Contrarily, graphene possesses hydrophobic nature and hence lacks such functionalities. Therefore, graphene is considered an inferior material in an aqueous medium [13,14]. However, GO can achieve graphene-like characteristics by conversion into rGO through chemical reduction [15].

Currently, there have been several studies reported regarding the graphene-based label or label-free electrochemical biosensors for the detection of viral infections such as influenza, dengue, and human immune deficiency virus (HIV) with high sensitivity and specificity. In contrast, recently, there are few reports suggested to analyse SARS-CoV-2 effectively [16,17]. This mini-review is aimed at the graphene-based electrochemical biosensors integrated with smart IoT for the monitoring and management of SARS-CoV-2 with high-throughput devices.

2. Electrode surface functionalization with nanomaterials

The properties of electrode materials greatly affect the performance of electrochemical biosensors. Subsequently, graphene, GO, rGO, AuNPs, carbon black, metal oxides/sulfides, etc. [18], have been utilized...
in the surface modification of electrodes. Nanomaterials govern the characteristics of the biosensor (sensitivity, specificity, and direct electron transfer processes), where immobilization of the biorecognition elements specific to target analytes takes place. A nano-enabled biosensing system has the ability of rapid detection of the disease-specific biomarker with the ultra-low detection limit up to fM [19]. Out of these nanomaterials, graphene, and its derivatives hold much attention for electrode fabrication. Since graphene has a planer surface and high π-electron cloud. Therefore, it has excellent electrochemical conductivity due to hetero electron transfer. In addition, the high surface area and presence of functional groups in abundance in GO make them perfect substrates for performing direct electron processes in electrochemical biosensors [20–22]. The graphical representation of sampling, detection and analysis of graphene-based IoT integrated electrochemical biosensor is shown in Fig. 1.

3. Graphene-based electrochemical immunosensor for SARS-CoV-2

Graphene-based nanomaterials hold many advantages over other materials. Some of the following studies show the potential of the work which attracts the scientific community to explore feasible, practical applicability of such material-based devices commercially. Very recently, Yakoh et al. [23] developed a label-free electrochemical paper-based analytical device (ePAD) for aiming at SARS-CoV-2 antibodies. Briefly, the solution of GO was implanted in a permeable framework at the test region of the graphene working ePAD and dried at room temperature. An embedded GO, 1-ethyl-3-(3-dimethyl aminopropyl) carbodiimide/N-hydroxysulfosuccinimide chemistry was employed via a reversed electrode architecture to immobilize spike protein at the hydrophilic paper substrate of the working ePAD, for selective binding of IgG and IgM SARS-CoV-2 antibodies. The square-wave voltammetry (SWV) technique was used to monitor the electrochemical response. In clinical sera samples, the reported sensing platform has shown satisfactory results. Fig. 2 (A and B) show the detection principle of ePAD for SARS-CoV-2.

In another study, a graphene-based electrochemical biosensor developed by Alafeef et al. [24] selectively detected the SARS-CoV-2 virus. The density of the ssDNA probe affected sensor sensitivity after its immobilization on the sensor surface. Significantly, thiol-modified antisense oligonucleotides (ssDNA)-capped AuNPs on the gold electrode surface showed better sensitivity in disparity to the lone ssDNA without AuNP conjugation. The proposed sensor is specific for the nucleocapsid protein of SARS-CoV-2. Alike four ssDNA probes were configured to trigger the two distinct zones to enhance the diagnostic performance of the assay simultaneously in the field of the similar viral N-gene. The output signal response time was less than 5 min having a sensitivity of 231.0 copies/μL. Consequently, SARS-CoV-2 RapidPlex was demonstrated by Torrente-Rodriguez and co-workers [25] as a multiplexed, hand-held, and wireless electrochemical system for quick diagnosis of SARS-CoV-2. The multiplexed biosensor is engineered based upon mass-producible laser engraved graphene (LEG) electrode that quantitatively detects the IgM and IgG antibodies (immune response), viral antigen nucleocapsid protein (viral infection), as well as C-reactive protein (disease severity) in both saliva and blood. Fig. 2 (C) shows the detection principle of the electrochemical RapidPlex platform for multiplex diagnosis.

Zhao et al. [26] described a sandwich-type recognition strategy-based electrochemical biosensor using p-sulfocalix(8)arene modified graphene to enhance toluidine blue (TB) signal for SARS-CoV-2 RNA detection. The detection limit is the lowest of 200.0 copies/mL amongst reported RNA measurements of SARS-CoV-2 for the clinical specimen. In another study, Ali et al. [27] reported an advanced nanomaterial-enabled biosensing system fabricated by nano printing of three-dimensional (3D) electrodes and further coated with rGO nanoflakes. They integrated the electrodes with the microfluidic device and detected the antibodies to SARS-CoV-2 spike S1-protein and receptor-binding-domain (RBD) with LOD of $2.8 \times 10^{-15}$ and $16.9 \times 10^{-15}$ mol/L, respectively. Additionally, they proposed a smartphone-assisted user interface for signal readout. The regeneration time of the sensor is < 60 seconds with no cross-reactivity for other antibodies and proteins. The modification of 3D-printed micropillar electrode and 3D-printed COVID-19 test chip (3DcC) sensor operation integrated with a smartphone is shown in Fig. 3 (a-g). Respective Graphene-based IoT integrated
electrochemical immunosensors for SARS-CoV-2 diagnostics are listed in Table 1.

Very recently, Beduk and group [28] revealed miniaturized laser-scribed graphene (LSG)-based electrochemical biosensor with 3D gold nanostructures for diagnosis of COVID-19. This electrochemical immunosensor quantitatively analysed S-protein from 5.0 to 500.0 ng/mL with LOD of 2.9 ng/mL. This smartphone-assisted setup also provides faster and accurate results than other commercial conventional detection techniques.

4. Significance of IoT for COVID-19 disease management

During this pandemic, screening, monitoring, and diagnosis of COVID-19 are the need of the hour. In this regard, telemedicine or mobile health is well considered to locate patients and prevent the swift spreading of the coronavirus [29]. The IoT integration with POC devices is a technological link between on-ground operation and online data collection and analysis. The IoT is a device that joins numerous platforms like electronics, smartphones, sensors, actuators, and networks at a local level. Hence, the medical data analysis can be done faster, remotely, along with high-throughput testing [19]. In the modern trend, smartphone-assisted POC devices take advantage of smartphone as a mediator for processing and transmitting the signal into digital and readable form. Such POC devices developed by integration of smartphones with biosensors make them inexpensive, hand-held, and wearable that modernizes the diagnosis [30]. Moreover, ease of sampling and quick detection makes the IoT-integrated electrochemical biosensor one of the most prominent diagnostic tools for COVID-19 management. Such advances help in efficient health management for the COVID pandemic on a global scale.

5. Conclusion and future outlook

The upsurge of the pandemic in the current scenario requires astonishing solutions for monitoring and management of this unanticipated problem faced by mankind. Accordingly, nanotechnology with smart electronics paved a way to combat the SARS-CoV-2 effectively. Herein we reviewed potential ultrasensitive graphene-based IoT integrated advanced electrochemical biosensors that will accomplish the diagnostic demand as well as screen SARS-CoV-2 transmission with high throughput potency. In addition, IoT integration provides better infrastructure for both patients and physicians to diagnose and manage SARS-CoV-2 cases at the preliminary stage. These aforementioned biosensors detect biomarkers associated with SARS-CoV-2 with high selectivity and sensitivity, making them effective allies against current and future pandemics.

Declaration of Competing Interest

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.

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References

[1] M. Asif, M. Ajmal, G. Ashraf, N. Muhammad, A. Aziz, T. Itikhar, J. Wang, H. Liu, J. Purr. Electrochem. 23 (2020) 174–184.
[2] P. Ranjan, A. Singhal, S. Yadav, N. Kumar, S. Murli, S.K. Sanghi, R. Khan, Int. Rev. Immunol. 40 (2021) 126–142.
[3] S. Yadav, M.A. Sadiq, P. Ranjan, N. Kumar, A. Singhal, A.K. Srivastava, R. Khan, A.C.S. Appl. Mater. 4 (2021) 2974–2995.
[4] A. Demeke Teklemariam, M. Samaddar, M.G. Alharbi, RR. Al-Hindi, A.K. Bhunia, Mol. Cell. Probes. 54 (2020) 101662.
[5] Ü. Anik, Y. Tepeli, M. Sayhi, J. Nsiri, M.F. Diouani, Analyst. 143 (2017) 150–156.
[6] M.F. Abd Muain, K.H. Cho, M.N. Omar, A.S. Amir Hamzah, H.N. Lim, A.B. Saleh, W.S. Tan, A. Ahmad Tajudin, Biosens. Bioelectron. 122 (2018) 199–205.
[7] M. Ahmadi, F. Ashour, Anal. Methods. 12 (2020) 4541–4550.
[8] S. Li, L. Ma, M. Zhou, Y. Li, X. Fan, C. Cheng, H. Luo, Curr. Opin. Biomed. Eng. 13 (2020) 32–41.
[9] C.I. Justino, A.R. Gomez, A.C. Freitas, A.C. Duarte, T.A. Rocha-Santos, TrAC Trends Anal. Chem. 91 (2017) 63–66.
[10] T.R. Fadel, D.F. Farrell, L.E. Friedersdorf, M.H. Grieb, M.D. Hoover, M.A. Meador, M. Meyyappan, ACS Sens. 1 (2016) 207–216.
[11] A.K. Srivastava, N. Dwivedi, C. Dhand, R. Khan, N. Satish, M.K. Gupta, R. Kumar, S. Kumar, Mater Today Chem. 18 (2020), 100385.
[12] G. Seo, G. Lee, M.J. Kim, S.H. Baek, M. Choi, K.B. Ku, C.S. Lee, S. Jun, D. Park, H. G. Kim, S.J. Kim, ACS nano. 14 (2020) 5135–5142.
[13] A. Nag, A. Mitra, S.C. Mukhopadhyay, Sens. Actuators A. 270 (2018) 177–94.
[14] J. Peña-Bahamonde, H.N. Nguyen, S.K. Fanoureas, D.F. Rodrigues, J. Nanobiotechnology. 16 (2018) 1–17.
[15] N.M.S. Hidayah, W.-W. Liu, C.-W. Lai, N.Z. Noriman, C.-S. Khe, U. Hashim, H. C. Lee, AIP Conf Proc. 1892 (2017), 150002.
[16] J. Huang, Z. Xie, Z. Xie, L. Xie, L. Huang, Q. Fan, Y. Zhang, S. Wang, T. Zeng, Anal. Chim. Acta. 913 (2016) 121–127.
[17] R. Singh, S. Hong, J. Jang, Sci. Rep. 7 (2017) 42771.
[18] L. Fabiani, M. Saroglia, G. Galata, R. De Santis, S. Fillo, V. Luca, G. Faggioni, N. D’Amore, E. Regalbuto, P. Salvadori, G. Terova, Biosens. Bioelectron. 171 (2021), 112686.
[19] M.A. Mujawar, H. Gohel, S.K. Bhardwaj, S. Srivinasan, N. Hickman, A. Kaushik, Mater Today Chem. 17 (2020), 100386.
[20] A. Chen, S. Chatterjee, Chem. Soc. Rev. 42 (2013) 5425–5438.
[21] M. Zhou, Y. Zhai, S. Dong, Anal. Chem. 81 (2009) 5603–5613.
[22] H. Gao, H. Duan, Biosens. Bioelectron. 65 (2015) 404–419.
[23] A. Yakob, V. Pimpikul, S. Fengguipat, N. Hiranyakorn, O. Chalilapakul, S. Chaiyo, Biosens. Bioelectron. 176 (2021), 112912.
[24] M. Alafaeef, K. Dighe, P. Moitra, D. Pan, ACS nano. 14 (2020) 17028–17045.
[25] R.M. Torrente-Rodriguez, H. Lukas, J. Tu, J. Min, Y. Yang, C. Xu, H.B. Rossiter, W. Gao, Mater. 3 (2020) 1981–1998.
[26] H. Zhao, F. Liu, W. Xie, T.C. Zhou, J. Ou-Yang, L. Jin, H. Li, C.Y. Zhao, L. Zhang, J. Wei, Y.P. Zhang, Sens. Actuators B Chem. 327 (2021), 128899.
[27] M.A. Ali, C. Hu, S. Jahan, B. Yuan, M.S. Salih, E. Ju, S.J. Gao, R. Punat, Adv. Mater. 33 (2021) 200647.
[28] T. Beduk, D. Beduk, J.L. de Oliveira Filho, F. Zlinioglu, C. Cicek, R. Sertoz, B. Arda, T. Goksel, K. Turhan, K.N. Salama, S. Timur, Anal. Chem. 93 (2021) 8585–8594.
[29] H. Lukas, C. Xu, Y. Yu, W. Gao, ACS nano. 14 (2020) 16180–16193.
[30] B. Purushot, A. Kumar, K. Mahato, P. Chandra, Curr. Opin. Biomed. Eng. 13 (2020) 42–50.