A mini review of electrochemical genosensor based biosensor diagnostic system for infectious diseases

N.A. Parmin1*, Uda Hashim1, Subash C.B. Gopinath1,2, Farrah Aini Dahalan3,4, C.H. Voon1, M.N.A. Uda1,2, M.N. Afnan Uda1, Zulida Rejali3, Amilia Afzan5, F Nadhirah Jaapar6, F Syakirah Halim1

1Institute of Nano Electronic Engineering, Universiti Malaysia Perlis, 01000 Kangar, Perlis, Malaysia.
2Faculty of Chemical Engineering Technology, Universiti Malaysia Perlis, 02600 Arau, Perlis, Malaysia.
3Faculty of Civil Engineering, Universiti Malaysia Perlis, 02600, Arau, Perlis, Malaysia.
4Centre of Excellence Water Research and Environmental Sustainability Growth, 02600, Arau, Perlis, Malaysia
5Department of Obstetrics and Gynaecology (OG), Faculty of Medicine, Health Sciences, Universiti Putra Malaysia, 43400 UPM Serdang, Selangor.

Abstract

The quest for alternative methods is driven by the need to provide expertise in real time in biological fields such as medicine, pathogenic bacteria and viruses identification, food protection, and quality control. Polymerase Chain Reaction (PCR) and Enzyme Linked Immunosorbent Assay (ELISA) are examples of traditional methods that have some limitations and lengthy procedures. Biosensors are the most appealing option because they provide easy, dependable, fast, and selective detection systems compared to conventional methods. This review provides an overview of electrochemical genosensor based biosensor diagnostic system for infectious diseases detection as well as their applications, demonstrating their utility as a fast and responsive tool for detecting pathogenic bacteria, viruses, GMOs, and human diseases.

Keywords:
Electrochemical sensor; infectious diseases; DNA probe; nanoparticles; nucleic acid complementation

1 Introduction

The development and application of electrochemical genosensors is progressing at a rapid rate, and the description and classification of electrochemical genosensors cannot definitively address all information and nuances. The signal transduction mode or the biological specificity conferring mechanism may also be used to classify biosensors. In electrochemical genosensors, the biosensor may be a probe with a short series of oligonucleotides, as in electrochemical DNA-based genosensors, or an aptamer, a synthetic oligonucleotides sequence, as in electrochemical Aptamer-based genosensors that are immobilized at the transducer surface (Nor Azizah Parmin, Hashim, and Gopinath 2018; Uda et al. 2018). Genosensors based on electrochemical DNA may be paired with nanoparticles or nanocomposites such as gold nanoparticles (AuNP) (Azizah et al. 2016) and quantum dots (QCD) (Ma, Li, and Zhang 2018) to enhance both oligonucleotide hybridization sensitivity and sequence immobilization on the transducer surface. Genosensor based on DNA specific detection become the most promising method for infectious disease detection.

DNA can be successfully isolated from different biological sources like blood (Madhad and Senthil 2014), saliva (Durdiakov et al. 2012; Lazarevic et al. 2013; Quinque et al. 2006) and swab tissue (Baay et al. 2011; Ghiottoni et al. 2010; Verdon, Mitchell, and van Oorschot 2014; Yang et al. 2014). Conventionally, DNA analysis by using human samples was a lengthy process consumes several days or weeks. DNA extraction from biological samples, quantification of DNA extract, amplification by using Polymerase Chain Reaction (PCR), separation of amplified products by using gel electrophoresis, analysis of data for confirmation basically using DNA sequencing and result reporting are the routine analysis for DNA (Yang et al. 2014). All these analyses require at least 8 to 11 hours or more depends on skilled personnel like molecular biologist to complete the process under laboratory conditions.

Some DNA extraction methods become the most important thing in molecular biology and for the analysis on DNA biosensor. Purification of viral DNA with minimal elution volumes is required for higher sensitive detection for viruses. In the present study, the virus Human Papillomavirus (HPV) is targeted and analysed on the Interdigitated Electrodes (IDE) sensor (T. LakshimiPriya et al. 2016; S. Nadzirah et al. 2020; N.A. Parmin et al. 2019). Preparation of HPV DNA from the free viral particles is needed, especially for Specimen Transport Medium (STM) swab, typically used for sample collection system in Pap smear by Obstetrics and Gynaecology doctors. Direct swab method has used in the clinical laboratories and forensic laboratories (Gangano et al. 2013; Holland and Wendt 2015) and also for commercial sample processing equipment. Swab lysis in liquid lysate form is highly suitable for the microfluidic system (Brun et al. 2012; Kim et al. 2009; Yang et al. 2014).
2 Electrochemical biosensor

Biosensors are a significant analytical instrument that alternate to conventional cellular and biological assays. It has arisen for viral detection that use organs, tissues, cells, and invasive methods. For many years, electrochemical biosensors have been used in a number of fields including medical and environmental. This type of biosensor analyzes any changes in dielectric properties and charge distribution on the electrode surface caused by the interaction of analyte and biorecognition factor. Electrochemical biosensors are graded as amperometric, potentiometric, voltammetric, and impedimetric based on the method of transduction. In molecular biology, the transduction was defined as the mechanism by which a virus or viral vector introduces foreign DNA into a cell.

An electrode transducer converts a chemical reaction into an electrical signal, which is used in electrochemical biosensors. Electrochemical biosensor was said to be highly sensitive transducers because they have ultra-low sensitivity (parts per trillion or sub-pico or femto molar ranges), linear efficiency, low power consumption, and good resolution. These sensors have the potential to be used as point-of-care (POC) and point-of-need detection systems. When evaluating a patient’s care reaction, such testing helps medical staff to make fast decisions depending on the patient’s condition. It is important to provide a wide understanding of the most recent novel methods and the problems that come with infectious disease electrochemical diagnostics. This kind of technology assist the researchers in developing the best possible solution of biorecognition elements that effectively overcome the difficulties that electrochemical transducers were facing.

3 Genosensor

A new type of affinity biosensor is created by combining nucleic acid layers with electrochemical or optical transducers as a DNA biosensor or well known as genosensor for low-molecular-weight molecules. Biological substance in various combinations in conjunction with various forms of transducers are a fascinating research subject to explore nowadays. A genosensor, also known as a gene-based or DNA biosensor, measures specific binding processes at the sensor surface, such as the formation of DNA–DNA and DNA–RNA hybrids. It also measures the interactions between proteins or ligand molecules and DNA. The following measures are involved in the design of a genosensor: i. The sensor surface is changed to provide an active layer for the DNA probe to bind to. ii. The probe molecule immobilization on the surface, preferably with a balanced density and packing orientation. iii. DNA hybridization at the sensor–liquid (aqueous form) interface to detect target gene sequences, preferably with controlled packing density and orientation of molecules on the surface.

In DNA biosensors based on electrochemical hybridization transduction, the high specificity of hybridization reactions is combined with the excellent sensitivity and portability of electrochemical transducers. The ultimate aim of all research studies involving genosensor is to build DNA biosensors in order to lay the groundwork for a potential DNA microarray device. Electrochemical genosensors based on various materials and transducers, in terms of chip-based technology, have recently been developed in response to clinical demand for promising results. Biosensors based on nucleic acids (genosensors) are being designed to involve the groundwork for a DNA microarray device. Electrochemical genosensors have attracted a lot of attention among genosensors because of their fast response, sensitivity, and cost-effectiveness, as well as their ideal for point-of-care (POC) research due to their compatibility with microfabrication technology and easy operating mode.

4 Infectious disease cause by pathogenic virus

Pathogens were classified as microorganisms that cause infectious disease. Pathogenic virus can damage the cells by entering the body and cause problems. During vaccination, pathogens are introduced into the body in weakened form, allowing the body to produce enough white blood cells to protect against the pathogens and prevent disease. Antibiotics are useful in the fight against bacteria that are not resistant to antibiotics, but not against viruses. Early, fast, and accurate identification of infectious virus is directly linked to the efficient management of the spread of disease and the enhancement of patient outcomes are two issues that need to be discussed. The identification of unique nucleic acid sequences of the target virus is the most common method for diagnosing and assessing viral infections (Kelley 2017). Infectious diseases spread directly from one person to another in some cases, but not all. Several viruses, including HIV, Human Papillomavirus (HPV) (N.A. Parmin et al. 2019), dengue virus (Nuzaihan et al. 2016), and hepatitis virus, have been detected using electrochemical genosensors. Dengue viral RNA can be identified at levels of 103 to 106 copies per milliliter in biological samples (blood, saliva, or urine) (Hue et al. 2011), a special DNA capture probe is modified and sandwiched with a digoxigenin-labeled detector probe (HPV-16 and HPV-18) to detect DNA sequences from high-risk human papillomavirus (hrHPV) strains (Bartosik et al. 2016). HPV also can be detected by using electrochemical genosensor due to easy to handle compared to molecular methods (Azizah et al. 2015; N.A. Parmin et al. 2019).

5 Infectious disease cause by pathogenic bacteria

Infectious diseases spread directly from one person to another in some cases, but not all. Bacterial detection and identification are primarily based on microbiological and biochemical recognition of various microorganisms techniques, which can take anywhere and the results will take anywhere from 3 to 7 days to appear (Bifulco, Ingianni, and Pompei 2013). Electrochemical genosensor was developed based on an oligonucleotide probe derived from the Escherichia coli O157:H7 genome’s 16s rRNA coding region (Sh. Nadzirah et al. 2015; Rajapaksha et al. 2017). A life-threatening infection can be caused by as little in a milliliter of blood as 1 to 10 colony-forming units (cfus) of bacteria [28]. When used as a target, ribosomal RNA should have a copy number of 103 or 104, which corresponds to detecting a subfemtomolar RNA concentration in a sample with an overwhelming abundance of non-target RNA. Direct detection methods for bacterial infection face a significant challenge as a result, and molecular-level analysis is normally performed after bacterial culture enrichment (Kelley 2017). The pathogens that cause urinary tract infections (UTIs) such as Staphylococcus saprophyticus, Escherichia coli, Proteus mirabilis, Klebsiella pneumoniae, and Pseudomonas aeruginosa. Urine that contains more than 105 cfu mL-1 is considered positive for UTIs (Karaba et al. 2021).

6 DNA structures at electrodes

The electrochemical behavior of deoxyribonucleic acid (DNA) and its reactions with different types of ligands and complemen-
tary or partially complementary nucleic acid sequences are both influenced by the surface state of DNA immobilized at electrodes. Since they directly associate with the presence of an active infection, DNA and RNA sequences are ideal candidates for infectious disease surveillance (Kelley 2017). Gold-standard techniques include microscopy, plating and culturing methods, nucleic acid-based approaches, and immunological assays were used for diagnosing bacterial and viral infections. The most popular approach for determining virus-specific antigens is to use Enzyme Linked Immunosorbent Assay (ELISA), which have been granted regulatory approval and are commercially available. However, they are time-consuming and multi-stage, have low sensitivity, can produce false negative results, and highly dependent on operator skills (Karaba et al. 2021; Uda et al. 2018).

The identification of nucleic acids can be more precise and sensitive than immunological methods (Nor Azizah Parmin, Hashim, and Gopinath 2017). Since they associate with the existence of an asymptomatic microorganism, these special DNA and RNA sequences are ideal targets for infectious disease study. Electrochemical biosensors combined with low-cost field-portable and programmable battery-operated instrumentation open up exciting new possibilities for non-experts to perform easy and cost-effective analytical strategies (Thangavel Lakshmipriya et al. 2016).

7 Conclusion and future perspectives

The rapid development and the use of biosensors can never be limited. The use of nanoparticles, nanocomplexes, and nanostructures has increased, allowing progress in electrochemical genosensors for pathogenic bacteria, virus detection, plant breeding, food safety, and quality control. Electrochemical genosensors are still the most appealing choice for developing a functional and a portable device is used to provide easy, accurate, fast, and selective detection systems.

Declaration of competing interest

The authors declare no known competing interests that could have influenced the work reported in this paper.

Acknowledgments

This research was supported by the Ministry of Higher Education (MOHE) under Grant Nos. RACER/1/2019/TK04/UNIMAP/2 and the author would like to thank all staff members of the Institute of Nanoelectronic Engineering in Universiti Malaysia Perlis for their technical advice.

References

Azizah, N., U. Hashim, S.C.B. Gopinath, and S. Nadzirah. 2016. “Gold Nanoparticle Mediated Method for Spatially Resolved Deposition of DNA on Nano-Gapped Interdigitated Electrodes, and Its Application to the Detection of the Human Papillomavirus.” Microchimica Acta 183(12).

Azizah, N., U. Hashim, S. Nadzirah, and A.R. Ruslinda. 2015. “Rapid and Sensitive Strategy for Human Papillomavirus (HPV) Detection Using a Gene-Based DNA Nanobiosensor.” In IECBES 2014, Conference Proceedings - 2014 IEEE Conference on Biomedical Engineering and Sciences: “Miri, Where Engineering in Medicine and Biology and Humanity Meet,”

Baay, Marc F D et al. 2011. “The Presence of Y Chromosomal Deoxyribonucleic Acid in the Female Vaginal Swab: Possible Implications for Human Papillomavirus Testing.” Cancer Epidemiology 35(1): 101–3.

Bartosik, Martin et al. 2016. "Electrochemical Chip-Based Genomagnetic Assay for Detection of High-Risk Human Papillomavirus DNA." Biosensors and Bioelectronics 83.

Bifulco, Laura, Angela Ingiammi, and Raffaello Pompei. 2013. "An Internalin a Probe-Based Genosensor for Listeria Monocytogenes Detection and Differentiation." BioMed Research International.

Brun, M. et al. 2012. "A New Microfluidic Device for Electric Lysis and Separation of Cells." In Proceedings of the Annual International Conference of the IEEE Engineering in Medicine and Biology Society, EMBS., 6281–84.

Connors, Emily et al. 2019. "Identification and Validation of Reliable Aeromonas Salmonicida Subspecies Salmonicida Reference Genes for Differential Gene Expression Analyses." Infection, Genetics and Evolution 73.

Durdiaak, Jaroslava et al. 2012. "Comparison of Different Collection Procedures and Two Methods for DNA Isolation from Saliva." Clinical Chemistry and Laboratory Medicine 50(4): 643–47.

Gangano, Stefanie et al. 2013. "DNA Investigative Lead Development from Blood and Saliva Samples in Less than Two Hours Using the RapidHITTM Human DNA Identification System." Forensic Science International: Genetics Supplement Series 4(1).

Ghittoni, Raffaella et al. 2010. "The Biological Properties of E6 and E7 Oncoproteins from Human Papillomaviruses." Virus Genes 40(1): 1–13.

Holland, Mitchell, and Frank Wendt. 2015. “Evaluation of the RapidHIIT??, 200, an Automated Human Identification System for STR Analysis of Single Source Samples.” Forensic Science International: Genetics 14: 76–85.

Hue, Kien Duong Thi et al. 2011. “Validation of an Internally Controlled One-Step Real-Time Multiplex RT-PCR Assay for the Detection and Quantitation of Dengue Virus RNA in Plasma.” Journal of Virological Methods 177(2).

Karaba, Sara M. et al. 2021. “Prevalence of Co-Infection at the Time of Hospital Admission in COVID-19 Patients, A Multicenter Study.” Open Forum Infectious Diseases 8(1).

Kelley, Shana O. 2017. "What Are Clinically Relevant Levels of Cellular and Biomolecular Analytes?" ACS Sensors 2(2): 193–97.

Kim, Jungkyu, Michael Johnson, Parker Hill, and Bruce K Gale. 2009. “Microfluidic Sample Preparation: Cell Lysis and Nucleic Acid Purification.” Integrative biology: quantitative biosciences from nano to macro 1(10): 574–86.

Lakshmipriya, T., U. Hashim, S.C.B. Gopinath, and N. Azizah. 2016. “Microfluidic-Based Biosensor: Signal Enhancement by Gold Nanoparticle.” Microsystem Technologies 22(10).

Lakshmipriya, Thangavel, Uda Hashim, Subash C. B. Gopinath, and N. Azizah. 2016. “Microfluidic-Based Biosensor: Signal Enhancement by Gold Nanoparticle.” Microsystem Technologies. http://link.springer.com/10.1007/s00542-016-3074-1.

Lazarevic, Vladimir et al. 2013. “Comparison of DNA Extraction Methods in Analysis of Salivary Bacterial Communities.” PLoS ONE 8(7).

Ma, Fei, Chen chen Li, and Chun yang Zhang. 2018. “Development of Quantum Dot-Based Biosensors: Principles and Applications.” Journal of Materials Chemistry B 6(39).

Madhad, Vaibhavi J, and K P Sentheil. 2014. “The Rapid Non-Enzymatic Isolation of DNA from the Human Peripheral Blood Suitable for Genotyping.” European Journal of Biotechnology and Bioscience 1(3): 1–16.

Nadzirah, S. et al. 2020. “State-of-the-Art on Functional Titanium Dioxide-Integrated Nano-Hybrids in Electrical Biosensors.” Critical Reviews in Analytical Chemistry.

Nadzirah, Sh. et al. 2015. “Titanium Dioxide Nanoparticle-Based
Interdigitated Electrodes: A Novel Current to Voltage DNA Biosensor Recognizes E. Coli O157:H7. Plos One 10(10): e0139766. http://dx.plos.org/10.1371/journal.pone.0139766.

Nuzaihan, M M N et al. 2016. “Biosensors and Bioelectronics Electrical Detection of Dengue Virus (DENV) DNA Oligomer Using Silicon Nanowire Biosensor with Novel Molecular Gate Control.” Biosensors and Bioelectronic 83: 106–14. http://dx.doi.org/10.1016/j.bios.2016.04.033.

Parmin, N.A. et al. 2019. “Voltammetric Determination of Human Papillomavirus 16 DNA by Using Interdigitated Electrodes Modified with Titanium Dioxide Nanoparticles.” Microchimica Acta 186(6).

Parmin, Nor Azizah, Uda Hashim, and Subash C.B. Gopinath. 2018. “Designing Probe from E6 Genome Region of Human Papillomavirus 16 for Sensing Applications.” International Journal of Biological Macromolecules.

Parmin, Nor Azizah, Uda Hashim, and Subash C B Gopinath. 2017. “Designing Probe from E6 Genome Region of Human Papillomavirus 16 for Sensing Applications.” International Journal of Biological Macromolecules: 1–9. http://dx.doi.org/10.1016/j.ijbiomac.2017.10.051.

Quinque, Dominique et al. 2006. “Evaluation of Saliva as a Source of Human DNA for Population and Association Studies.” Analytical Biochemistry 353(2): 272–77.

Rajapaksha, R. D.A.A. et al. 2017. “Target SsDNA Detection of E.Coli O157:H7 through Electrical Based DNA Biosensor.” Microsystem Technologies 23(12): 5771–80.

Uda, M. N.A. et al. 2018. Nanobiosensors for Biomolecular Targeting A Disposable Biosensor Based on Antibody-Antigen Interaction for Tungro Disease Detection. Elsevier Inc. http://dx.doi.org/10.1016/B978-0-12-813900-4.00006-3.

Verdon, Timothy J., Robert J. Mitchell, and Roland A H van Oorschot. 2014. “Swabs as DNA Collection Devices for Sampling Different Biological Materials from Different Substrates.” Journal of Forensic Sciences 59(4): 1080–89.

Yang, Jianing et al. 2014. “An Integratable Microfluidic Cartridge for Forensic Swab Samples Lysis.” Forensic Science International: Genetics 8(1): 147–58. http://dx.doi.org/10.1016/j.fsigen.2013.08.012.