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Advances in Biochemical Techniques for Monitoring Emerging Infectious Diseases in Humans

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ABSTRACT

Major global health challenges like infectious diseases are causing significant morbidity and mortality with approximately 25% of the 60 million annual deaths attributed to these diseases. However, microbial culture and immunological assays along with the face, speed, limitations, and specificity are the historic diagnostic methods used to hinder effective disease management. Molecular diagnostic techniques like Polymerase chain Reaction and high-throughput sequencing have revolutionized pathogen detection and offered rapid and precise identification. Diagnostic capabilities are enhanced through the innovation in point-of-care (POC) devices by offering rapid, on-site testing. The GeneXpert for tuberculosis and Human Immuno deficiency Virus are notable examples of Point of care. However, often lack full integration and automation indicating a need for further innovation in Point of care devices to achieve beneficial solutions. Additionally, electrochemical biosensors offer rapid, selective, and cost-effective virus detection. These biosensors identify the various viral biomarkers such as nucleic proteins, nucleic acids, and antibodies essential for monitoring infections and guiding treatment. Infectious disease occurrences like Ebola, zika, and coronaviruses (e.g., SARS-CoV-2) highlight the necessity for advanced diagnostic tools

Keywords: molecular techniques, GeneXpert, biosensors, corona viruses, electrochemical

Introduction

Infectious diseases can occur through bacteria, viruses, parasites, and fungi that can produce various diseases that are referred to as infectious and lead to causes morbidity and mortality. Recent statistics analyzed that approximately 60 million deaths occur globally each year with at least 25% associated with infectious diseases. The significant challenges are influencing public health and economic stability due to the occurrence of these diseases. However, vaccines, antimicrobial, and antiviral drugs can reduce infections to some extent and their effectiveness decreases with the increase of new infectious organisms and antibiotic-resistant microbes. Moreover, the innovation in Immunizations and pharmaceuticals like long-term clinical testing, hinders rapid and effective disease control. The fast and precise diagnosis of pathogens is crucial to selecting appropriate detection techniques when no availability of proper vaccines and drugs. This approach enabled a swift counteraction to major public health threats enhanced treatment efficiency and reduced disease spread [1].

Some traditional diagnostic techniques are microbe culture, hemagglutination inhibition testing, and enzyme-linked immunoassay) techniques. Culturing harmful microorganisms is the most time-consuming and primarily dependent on indistinct morphological features and sensitivity. Immune-based techniques like hemagglutination inhibition assays and ELISAs serve to detect microbes-specific antibodies or antigens that are straightforward to perform. Although, suffer from high false-positive rates, high costs, and poor thermal stability. The molecular diagnostic techniques focus on nucleic acid detection due to advancements in genomic and genetic research that have revolutionized the diagnosis of infectious diseases to offer short turnaround times and high sensitivity. Furthermore, such techniques can detect multiple pathogens by analyzing drug resistance genes and conduct pathogen homology analysis which has become an essential tool for early diagnosis. Nowadays, molecular identification techniques for infectious diseases are PCR, isothermal amplification, gene chip techniques, and high-capacity sequencing are being used [2].

Point-of-care devices for identifying infectious diseases

The first point-of-care (POC) apparatus was underscored by the World Health Organization by a GeneXpert during the first identification of tuberculosis linked with Human Immunodeficiency virus in 2010. A consolidated sample preparation occurs by combining nucleic acid amplification and detection in a single apparatus capable of completing nucleic acid detection within 2 hours and has been a proliferation of POC devices designed for Infectious ailments diagnosis. The automated nucleic acid test instrument analyzer for Human Immuno deficiency virus identification, Truelab Uno® for detecting malaria, and GenePoC for Influenza A and B virus detection are notable examples [6].

Mostly, existing Point of care devices are not integrated or automated and are essentially simple adaptations of conventional diagnostic steps. Furthermore, these are expensive and bulky indicating a significant opportunity for developing Point of care devices that are seamlessly integrated, swift, delicate, economical, and completely automated technology. The POC diagnostic industry is still in its early stages, with many challenges remaining. The most critical challenge is to deliver rapid, sensitive, and sample-to-result detection that is possible through seamlessly combining all identification processes into a completely automated device [10].

However, the POC devices can be affected by the rapid progress made in the fields of microfluidics, nanotechnology, and Micro-electro chemical systems technologies. Consequently, to detect infectious diseases a variety of POC devices have been designed like lab-on-a-chip (LOC) technology, lab-on-a-disc (LOAD) systems, microfluidic paper-based analytical devices (μPADs), lateral flow devices, and miniaturized Polymerase chain reactions devices are utilized., and integrated genetic material amplification analysis apparatus [6].

Optical technologies for detecting viruses

Virus Sub-micron-sized often termed "organisms at the edge of life" have been linked to some of the most severe human pandemics in history like the influenza pandemic from1918. Furthermore, the smallpox outbreak in 1978 and the current COVID-19 pandemic. As of September 17, 2020, COVID-19 spread through the SARS-CoV-2 virus affecting nearly 29 million people worldwide and resulting in 936,521 deaths. The full-length genome sequences are analyzed through genomic studies of SARS-CoV-2-infected individuals have revealed a genome similarity of approximately 80% with SARS-CoV, 50% with MERS-CoV, and 96% with the bat coronavirus RaTG13 **[6].**

The aerosolized droplets, direct contact, and oral-fecal transmission are the various pathways that have been implicated in the spread of the pandemic with a viral incubation period of up to 24 days COVID-19 detection explored many clinical samples, with conventional methods primarily utilizing respiratory specimens through the upper and lower respiratory tract. The acute infection stage enabled the collection of nasal swabs often within 7 days of

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viral exposure and has shown high viral loads compared to throat specimens [7].

The blood serum is used in Immunological assays and also for diagnostic purposes and the detection of viral samples from saliva and stool specimens that require fewer sampling procedures than respiratory samples. However, improving sensitivity and reducing false negatives demonstrated in clinical studies through testing samples from multiple sites can [8].

Electrochemical Diagnostics for Infectious Viral Diseases: Emerging Trends and Challenges

Viruses are the smallest transmissible Messengers that cause several diseases like Chikungunya, Chickenpox, Dengue, Ebola, Flu, Hepatitis, Influenza, and Middle East Respiratory Syndrome. Normally, a viral particle has nucleic acids at its core and proteins in its outer shell. Various viruses use RNA or DNA to encode proteins. However, the viruses can spread rapidly and can cause significant threats to global public health. They enter through several mechanisms in host cells and their replication occurs in host metabolism. Consequently, the spread of infectious diseases is launching new challenges and severe consequences. So, precise and high-throughput virus detection and analysis are crucial for effective disease control. Recently, the COVID-19 pandemic is the best example of a viral spread that affected millions worldwide within a few months [6].

Viral isolation, immunofluorescence microscopy, and enzyme-linked Immunoglobulin assays are historic viral diagnostic techniques that are becoming outdated for routine clinical testing. The mentioned techniques often have long turnaround times that range from 2 to 14 days which is insufficient for rapidly spreading viruses. Additionally, the given diagnostic tests are expensive but are rapid and reliable, reproducible analytical methods that are urgently needed to identify viral pathogens in various samples. Biosensors occurred like significant analytical tools to offer a substitute for traditional cellular and biological assays for viral detection by using tissues, cells, and invasive organ approaches. In addition, Electrochemical biosensors have been utilized for many years across different fields among the many types of biosensors. However, it is crucial to understand the newest creative methods and hurdles in electrochemical diagnostics for viral infectious diseases [4].

The importance of infectious virus detection is crucial to improve clinical diagnostics

Various biomarkers including RNA, DNA, glycoproteins, peptides, antibodies, antigens, etc. are utilized to detect many viral infections and are classified into two main categories, antigens and antibodies. As mentioned above; to encode proteins the most viruses have either RNA or DNA. The genetic material (DNA or RNA), nucleocapsid protein, and capsid proteins are the three structural components of viruses. The genetic material is encapsulated Firstly, nucleocapsid proteins cover genetic material and then cover by capsid proteins [4].

The virus enters the body through attachment to cell-surface receptors and ends with the delivery of the viral genome to the cell cytoplasm. This occurrence takes place by endocytic (clathrin-mediated endocytosis and penetration) or non-endocytic (fusion at the cell surface) routes.

viral infection monitoring is important for treating affected individuals which is achieved by detecting other biomarkers including C-reactive protein, interleukins, glutamate, breath pH, tumor necrosis factor (TNF-α), interferons, hematological biomarkers, and D-dimer. The electrochemical methods with suitable biorecognition elements are used in these biomarker detections [6].

Developing Biosensing Technologies for Diagnosing Viral Infectious Diseases

As discussed above, numerous infectious diseases are caused by several viruses. Although, their occurrence in different regions can cause a significant threat to global health and economies. Coronavirus stains including SARS-CoV, MERS-CoV, and particularly SARS-CoV-2 are responsible for COVID-19 that caused widespread illness and ongoing pandemic concerns. While aerosols and fomites spread these viruses [9].

DNA sequencing of bronchoalveolar lavage fluid (BALF) samples and RT-PCR are used for clinical diagnosis to detect viral nucleic acid and offer high sensitivity but high costs, complex equipment, and the need for specialized personnel remain limitations. Additionally, RT-PCR can only work within a limited time to identify active infections, and during outbreaks its demand increases. The importance of monitoring humoral immunity through immunology or serology assays is shown by evolving SARS-CoV=2 and emerging mutations that detect antibodies like IgM, IgG, and IgA. While rapid and low-cost testing is achieved through serological assays such as ELISA and lateral flow immunoassays (LFA) contains have lower accuracy and specificity as compared to RT-PCR [5].

Conclusion

The monitoring of emerging infectious diseases is enhanced significantly by advancement in biochemical techniques through addressing critical public health challenges. The microbial cultures and ELISAs are traditional methods used to identify essential pathogens that cause diseases and used to find often suffer from limitations such as long turnaround times and high false-positive rates. In contrast, molecular diagnostic techniques like PCR and high-throughput sequencing, offer rapid, sensitive, and specific pathogen detection are crucial for early diagnosis and effective disease management. The progress offers benefits, but some challenges remain in integrating and automating these technologies to make them more accessible and affordable. Overcoming from these diseases is possible due to innovations in microfluidics, nanotechnology, and electrochemical diagnostics hold promise and ultimately enhancing our ability to control and prevent infectious diseases.

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