Chapter 16
Nanotechnology-A New Frontier in Medical Microbiology

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Abstract Nanotechnology relates to microbiology at a number of levels as the microbial entities are nano-machines. In the second half of this decade, nanotechnology expanding its applications in the field of medical microbiology. Nanotechnology is clinically appropriate and retains the potential to be valuable in the diagnosis of general and microbial infections. The rapid detection of pathogenic microbes at the point of care is extremely critical. The application of nanoparticles permits for the detection of infectious pathogens in small sample volumes directly in a sensitive, specific, and rapid format at lower costs than current in-use technologies. A bio-conjugated nanoparticle-based bioassay for in situ pathogen quantification can detect a single microbe. The waveguide technology is an emergent area in the medical microbiology for the fast and successful diagnosis of infectious diseases. Nanotechnology is demonstrated for the detection of Avian influenza virus H5N1, Respiratory Syncytial Virus (RSV), HIV, and Severe acute respiratory syndrome (SARS) Coronavirus in clinical samples with a great degree of sensitivity. Nanoparticle-based bio-barcode amplification (BCA) assay is being applied for early detection of HIV-1 capsid antigen. The gold nanoparticle interferometer sensor has been validated for detection of Herpes simplex virus (HSV) and silver nanorod array substrates can detect spectral differences between the viral strains. A nanoparticle label technology with highly fluorescent chelated nanoparticle label has been developed for Adenovirus and Human papillomavirus (HPV). The nano-gold labelled amplification is a novel technique for the detection of Hepatitis B virus, Hepatitis C virus, and Hepatitis E virus in patient’s samples. Norovirus is a leading cause of gastroenteritis and nanospray mass spectrometry is evaluated for norovirus detection. With the manifestation and intensification of microbes resistant to antibiotics, silver nanoparticle antiseptics have been evaluated for the antimicrobial
activity against Gram-positive and Gram-negative bacteria. All these technologies would have to be assessed in clinical settings prior to their complete admission is highly recommended.

**Keywords** Nanotechnology · Microorganisms · Virus · Bacteria · Infectious diseases

### 16.1 Introduction

Microbiology is the science that deals with microscopic analysis of microorganisms, which are not visible to the naked human eye. Microscopic invention by Antony van Leeuwenhoek in the year of 1676 marked the beginning of microbiology.

Advent of light microscopy by Ernst Abbe and Carl Zeiss (1880s) and electron microscopy by Ernst Ruska (1931) produced high resolution images for visual characterization and released a new avenue in the study of biological structures. Microorganisms were the primitive life recorded on earth. Microbes occupy a place as significant component of biota with peculiar cell structure and ability to survive under extreme environmental conditions. They exist as unicellular, multicellular, and acellular organisms. Microorganisms are beneficial as well as harmful to human beings (Minocheherhomji 2016). They can be divided into six major types: bacteria, archaea, fungi, protozoa, algae, and viruses.

Nanotechnology is a highly promising field of research in the modern era of scientific research. Eric Drexler popularized the word ‘nanotechnology’ in 1986 in his book ‘Engines of creation: The Coming Era of Nanotechnology (Shinde 2012). It is an interdisciplinary science, whose potential has been widely touted in various domains of lifecycle with its development from submissive nanomaterial to active nanotechnology (e.g., drug delivery system) and nanosystems (e.g., robotics). Nanotechnology, in other forms, means building things from the bottom up, with atomic precision. This theoretical capability was envisioned as early as 1959 by the renowned physicist Richard Feynman in his talk ‘There’s Plenty of Room at the Bottom’ at Caltech, USA (Feynman 1960) and promoted by Eric Drexler by his book *Engines of Creation: The Coming Era of Nanotechnology* in the 1980s.

Nanotechnology is the study and application of nanoparticles. Nanoparticles are particles that exist on a nanometre scale (i.e., below 100 nm in at least one dimension). They possess physical properties such as uniformity, conductance or special optical properties and are of pronounced technical interest (Prasad et al. 2016). They form a bridge between bulk materials and atomic or molecular structures. Nanoparticles were used by artisans as far back as the ninth century in Mesopotamia for generating a glittering effect on the surface of pots. In the fourth and fifth centuries BC it was used by traditional medical practitioners in the world with the name ‘Swarna Bhasma’ for therapeutic purposes in treating cognitive disorders and syphilis (Paul and Chugh 2011; Balzani 2005; Drexler and Peterson 1989; Dykman and Khlebtsov 2011). The worldwide market of nanomaterials used
in the biomedical, pharmaceutical, and cosmetic industries showed an elevation from $170.17 million in 2006 to $684.4 million in 2012.

16.2 Role of Nanotechnology in Medical Microbiology

The boom of Nanotechnology revolutionized microbiology. Nanotechnology proved significant in medical microbiology in the detection of pathogens. Microbial nanotechnology has gained prominence with the advancement of chip-based DNA control, quantum dots, and carbon nanotubes (CNTs). Secure and cost viable novel materials are designed utilizing the viral like protein frames for biomedical imaging, Bioengineering, Drug Design, Enzyme Technology, etc. Additionally, synthesis of nanomaterials utilizing the energy of microorganisms also opens a new-fangled opportunity for ‘green’ synthesis, which is eco-friendly cost-effective process (Ahmad et al. 2016; Prasad et al. 2016, 2018; Prasad 2019a, b; Srivastava et al. 2021). Multidrug resistance is the present day concern (Andersson and Hughes 2010; Magiorakos et al. 2012). Biofilms offer resistance to antimicrobial agents while present in the human body and oblige as a reservoir of bacteria that can cause continuous and chronic infections (Watnick and Kolter 2000; Inamuddin et al. 2021). A 80% of infections in the hospitals are reported due to biofilms (Costerton 1999; Beyth et al. 2015). There is an immense need for substitute to antibacterial and anti-biofilm agents (Prasad et al. 2020). The nanotechnology can provide novel diagnostics and treatments for the bacterial infections and encouraged extensive investigation. Nanotechnology marks an innovative tool to address this challenge. Antibacterial nanodrugs are active against MDR bacteria and biofilms. A nanoporous polymer matrix composed of sodium dodecyl sulfate proved to have excellent anti-biofilm activity against \textit{E. coli}. Vitamin E-conjugated cationic polymer cross-linked hydrogels were found to have good bactericidal and antifungal effects (Lee et al. 2013).

16.2.1 Broad Spectrum of Nanoparticles

Liposomes are the nanoparticles comprised of lipid bilayer membranes, which is surrounded by an aqueous interior. Liposomes can be widely applied for delivery of antibiotics to the target sites (Druulis-Kawa et al. 2009). Liposomes exhibited biofilm inhibition and minimum sensing distraction on clinical variants of \textit{E. coli}, \textit{Acinetobacter lwofii}, \textit{A. baumannii}, \textit{Bordetella bronchiseptica}, \textit{Klebsiella pneumoniae}, and \textit{P. aeruginosa} (Druulis-Kawa et al. 2009). Dendrimers with lower molecular weight peptides are proven to be efficient antimicrobials against \textit{E.coli} and \textit{S. aureus} without any further antibiotics (Johansson et al. 2008). Fucose-specific lectins (LecB) from fucose peptide dendrimers prevented the formation of biofilm in \textit{P. aeruginosa}. A lipid dendrimer hybrid nanoparticle (LDHN) delivered vancomycin against methicillin resistant \textit{Staphylococcus aureus} (MRSA) infections.
Nano-emulsions with antimicrobial activity against bacteria such as *E. coli*, *Salmonella* sps, *S. aureus*, enveloped viruses (Human immunodeficiency virus and Herpes simplex), fungi (Candida), and spore forms of *B. anthracis* have been reported.

Bio-responsive smart nanoparticles, which are comprised with peripheral energy and energy absorbing NPs having the therapeutic properties towards respective antimicrobial infections (Ramasamy et al. 2016). Quantum dots (Qdots) are nanoscale semiconducting nanoparticles that can transport electrons. Qdots emit various colors (wavelength) when the light is excited in different wavelengths according to their size. Qdots have been extensively used in the Fluorescent Resonance Energy Transfer (FRET) based immunoassays for fast and accurate detection of *Aspergillus* sps. Qdot barcodes have been widely applied in the detection of HIV (Kaittanis et al. 2010; Kattke et al. 2011).

### 16.2.2 Nanoparticles with Intrinsic Antibacterial Properties

Silver is a renowned potent antimicrobial agent since prehistoric times (Rai et al. 2009; Reidy et al. 2013; Duran et al. 2015; Franci et al. 2015; Aziz et al. 2014, 2015, 2016). Silver based nanoparticles (AgNPs) exhibit antimicrobial activity against *Pseudomonas aeruginosa*, one of the important opportunistic pathogens triggering nosocomial infections, *Mycobacterium tuberculosis* (Mapara et al. 2015; Singh et al. 2015; El-Zahry et al. 2015; Pal et al. 2015) and *Staphylococcus aureus* (Actis et al. 2015). Silver nanoparticles are also inhibited the formation of biofilm by means of *P. aeruginosa* and *S. epidermidis* by more than 95% (Sinha et al. 2011).

Gold nanoparticles (AuNPs) adapted with various surfaces exhibit various enzyme-like accomplishments including peroxidase, glucose oxidase, superoxide dismutase, and catalase mimetics (He et al. 2013). MSN-AuNPs can inhibits the biofilm formation in *Bacillus subtilis* and even be able to collision existing biofilm (Tao et al. 2015). They proved effective against enteropathogenic *Escherichia coli* (EPEC), *Enterococcus faecium*, *Enterococcus faecalis* (including vancomycin-resistant strains) (Kaittanis et al. 2010). Gold and silver nanoparticles have been applied in resonance scattering confocal microscopy or two-photon luminescence confocal microscopy and as a transporters for drugs (Dykman and Khlebtsov 2011). ZnO NPs have better antibacterial activities and low toxicities in mammalian cells and is effective against *E. faecalis*, *S. aureus*, *S. epidermidis*, *Bacillus subtilis*, and *E. coli* (Lee et al. 2014).

Gold nanoparticle interferometric approach is a sensitive and quantitative detection method for sensing single particle, which is calibration-free allowing molecular counting and capable of directly processing with complex biological samples. Gold nanoparticles are used for sensing due to localized surface plasmon resonance (LSPR). The detection is typically based on the shifting of wavelength of the Localized surface plasmon resonance by scattering of light through a single gold nanoparticle. The experiment utilizes a nonlinear confocal microscope and interferometer arm similar to a Michelson interferometer. The modifications to the local RI
(refractive index) signal or absorption of molecules on the nanoparticle surface are captured. Gollmer et al. (2014) described the Gold nanotriangle (GNT) arrays synthesized by e-beam lithography. A biosensor designed from electrochemically produced with Gold nanoparticles-modified screen-printed carbon electrode coupled with the thiolated aptamer AG3 might achieve a limit of detection of around 180 virus particles of MNV (Giamberardino et al. 2013). Gold nanoparticles that have enzyme-like catalytic activity (i.e., NanoZyme activity) are immobilized with the AG3 aptamer (Kd of 18.5 nM) for Mouse norovirus (MNV) recognition.

16.2.3 A Bio-Conjugated Nanoparticle-based Bioassay

Cancer is a deadly disease caused by the abnormal cell growth, which affects any part of the human body. Early diagnosis of cancer (prior to metastasis) is a critical aspect in its treatment. Nanomaterials demonstrated as excellent materials for the early stage diagnose of many cancers. Gold nanoparticles (AuNps) comprised with biomolecules corresponding to target molecule are used for early detection of cancer by means of colorimetric detection. Kang et al. (2010) established a colorimetric procedure using anionic citrate coated AuNps for the diagnosis of cancer. Zhang et al. (2016) proposed Gold nanoparticles with telomerase primer on its surface and treated with human cell lines of leukaemia (HL-60, K562), hepatocellular carcinoma (HepG2), embryonic kidney (293 T), and normal skin fibroblasts (HSF). Guo et al. (2014) conjugated folic acid to Au nanorods (FGNRs) and produced optical detecting sensor with fluorescence and Localized surface plasmon resonance absorption feature, which facilitated in the cancer diagnosis.

The biomolecule-conjugated AgNPs shows exciting applications by challenging the clinical complexities such as multidrug resistance, designing biocompatible nanopharmaceutics, cancer therapy superior drug delivery transporters, fluorescence biosensors, and the next-generation antibiotics. The bio-conjugated Gold nanoparticles clenched unique optical and plasmonic features, which are exploited for developing diagnostic sensors and for the detection of biomarkers using immunoassays. Liu et al. (2013) reported a highly sensitive technique for the quantitative detection of HIV nucleic acids. Abbaspour et al. (2015) reported an assay for the detection of Gram-positive bacteria *Staphylococcus aureus* by dual-aptamers bio-conjugated AgNPs sandwich immunosensor. Kurdekar et al. (2017) explored the application of fluorescent AgNPs bio-conjugated with streptavidin for early diagnosis of HIV infection.

16.2.4 Nanotechnology for Viral Detection

Development of a variety of novel isolates of pathogenic viruses presently marked universal fatality challenging human health and demanding urge to develop suitable
detection methods, nanovaccines, and therapeutic nano-based options. Most respiratory tract infections are caused by frequently Influenza virus, Respiratory syncytial virus (RSV), Rhinovirus (RV), and Severe/acute respiratory syndrome (SARS) Coronavirus. Respiratory viruses affect infants, children, elderly people, and immune compromised patients. Nanomaterials are considered to be suitable aspirants against many viral infections, especially Coronavirus due to their capability to move into cells effortlessly and interact with viruses and interfere with viral genome replication. Gold NPs and quantum dots (QDs) form new nanotechnology-based detection procedures for several respiratory viruses. Au NPs comprised with silver staining and employed for the recognition of HPV (Human papillomavirus) in a cancer type called cervical carcinoma cell lines (Zehbe et al. 1997). Nanostructures like metal NPs, graphene oxide (GO), Quantum Dots, carbon nanotubes are used for virus testing and detection (Wang et al. 2009). Gold or silica based nanoparticles synthesized on thin silicon membranes form nanochips, which are used to screen clinical samples. This method enabled the diagnosis of HIV 1 virus in the plasma samples (Lee et al. 2004).

Mass spectrometry is employed in the detection of nuclear NoV(CCN3) protein and this was first evidenced by Colquhoun et al. (2006) using matrix-assisted laser desorption ionization coupled with a time-of-flight(MALDI-TOF) and nanoelectrospray ionization mass spectrophotometry (ESI-MS) in the detection of NoV virus like particles (VLPs) in clinically significant mediums. Khoris et al. (2019) demonstrated a silver-enhanced nanozyme-based immunoassay for the analysis of NoV with unaided-eye. Moreover, a LSPR-based fluorescence nanobiosensor reported by Takemura et al. (2017), which is capable to accomplish a limit of detection of 0.4 pg/mL of NoV VLPs, through incorporating antibody-mediated AuNPs and quantum dots.

Bio-barcode amplification assay (BCA) with nanoparticles have been reported for the detection of nucleic acid (DNA or RNA) and proteins at ultra-low level with short oligonucleotides as surrogate targets, which can be measured with light scattering, calorimetry, fluorescence or gel based assay (Singh et al. 2018; Draz and Shaﬁee 2018). NP-I-PCR is a modiﬁcation of BCA, where real-time PCR is used for detection. It helps in identiﬁcation of infectious microorganisms and can be used for biomarker discovery for different infectious diseases. In general, gold nanoparticles are being used in the BCA. Kim et al (Kim et al. 2008) developed 235 NP-based BCA for the detection of HIV-1 Gag p24 protein in blood samples, and after the conjugation of anti-HIV 1 p24 Gag pAbs (coated on MMPs) to p24 Gag present in samples, the biotinylated anti-HIV p24 Gag mAbs were added followed by the addition of streptavidin coated GNPs and the biotinylated oligonucleotides. Perez et al. (2011) developed Nanoparticle-ampliﬁed I-PCR technique for the rapid detection of RSV surface protein with the help of synthesized GNPs.
16.2.5 **Nano-based Antimicrobial and Anti-Biofilm Coatings**

Catheters are coated with copper (Cu) and silver (Ag) nanoparticles to accelerate killing of *E. coli* (Rtimi et al. 2016). Silicon coated urinary devices killed different bacterial strains that commonly cause urinary tract infections (UTI) such as *E. coli*, *Staphylococcus aureus*, *Staphylococcus epidermidis*, and *Pseudomonas aeruginosa*. ZnO and MgF2 nanoparticles proved to prevent biofilm formation of *Staphylococcus aureus* and *Streptococcus pneumoniae*. Catheters coated iron NPs inhibited biofilm in *S. aureus* and *P. aeruginosa* (Anghel et al. 2012) iron NPs eliminated *S. epidermis* infection on orthopaedic implants.

Nanocantilevers consist of tiny pieces of silicon-based materials that can recognize proteins and detect pathogenic bacteria and viruses (Kumar 2006). A technique was developed using 16S rRNA gold nanoprobe-nucleic acid sequence-based amplification (NASBA), for the detection of *Salmonella* (Mollasalehi and Yazdanparast 2013). Surface Enhanced Raman Scattering (SERS) nanoparticles help in sensitive detection of pathogens in complex samples (Weidemaier et al. 2015).

16.2.6 **Nanosystems for Food Borne Pathogen Detection and Biofilm Inhibition**

Surface enhanced Raman spectroscopy (SERS) is applied as a nano-biosensing technique for the detection of highly sensitive infectious microorganisms directly in a rapid and accurate way (Chandra et al. 2011). Different types of nano-biosensors have been employed to identify foodborne pathogens (bacteria and virus) (Thakur and Ragavan 2013; Li et al. 2004). Silver nanocolloids are commonly used in SERS (Baranwal et al. 2016) to detect microbes as silver nanocolloids increase Raman signals. In addition to silver nanocolloids, graphene oxide (rGO), magnetic beads, carbon nanotubes, and plasmonic gold are most commonly used nanomaterials for the detection of foodborne pathogens. Furthermore, synthetic DNA molecular beacon probes labelled with colour-codes are used as nanobarcodes to detect food pathogens (Li et al. 2004). Therefore, surfaces of refrigerators and storage containers are coated with silver nanoparticles to prevent growth of foodborne pathogens and food spoilage bacteria.

Nanofibers are efficiently employed in the prevention of biofilm-associated infections (Zhang et al. 2011). Furthermore, nickel oxide nanoparticles (NiO-NPs) are proposed as potential antibacterial and antitumor agents. Over and done with a green approach and using *Eucalyptus globulus* leaf extract, researchers (Sallem et al. 2017) synthesized NiO-NPs, which are in the size of 10–20 nm and evaluated their anti-biofilm activity.
16.2.7 Nanotechnology for Designing Vaccines

Nanoparticles/nanocarriers act as vaccine adjuvants and nanovaccine delivery platforms as they retain physicochemical characteristics that enhance their immunogenicity. This offers novel opportunities of novel nano-therapeutics and diagnostics which is the urge of present drug resistance age. These vaccines are more effective, safe, and convenient over conventional vaccines. Silver Nanoparticles inhibited the viral entry into host cells, for HIV-1 virus by interacting with the cell receptors (Kerry et al. 2019; Zhu et al. 2019). Gold Nanoparticles stabilized by biocompatible polymers showed antiviral activity against HIV-1 and influenza virus (e.g., H1N1, H3N2, H5N1) (Seo et al. 2020; Rauch et al. 2018). Multidrug nanoparticles (with great biocompatibility and drug loading) for the reduction of uncontrolled inflammation is very promising, particularly in the case of COVID-19 (Callaway 2020; Le et al. 2020; Martinez 2020).

16.3 SARS-COVID

Severe acute respiratory syndrome (SARS) is a respiratory disease caused by novel coronavirus called SARS-associated corona virus (SARS-CoV) that caused the major pandemic of this decade (Drosten et al. 2003). For the first time, SARS has been reported in Asia in 2003 and then the infection was transmitted to more than 12 countries including America, Europe, and other Asian countries before the SARS global outbreak of 2003 is contained.

Recently, the global outbreak has outstretched by the novel pathogenic viral transmission caused by the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) which is identified on January 2020 by Chinese scientists and originated in Wuhan, China and spread around the world (Pandemic). The World Health Organization (WHO) officially declared the COVID-19 epidemic as a public health emergency of global concern. Infection with SARS-CoV-2 virus can cause sickness, ranging from common cold to more severely, respiratory diseases, such as SARS and MERS. The virus is highly transmitted human-to-human with rapid rate. The beginning of SARS-CoV-2, since the SARS-CoV in 2002 and Middle East respiratory syndrome coronavirus (MERS-CoV) in 2012, manifests the third introduction of a highly pathogenic and large-scale epidemic coronavirus into the human population in the twenty-first century. Coronavirus is a betacoronavirus, enveloped, non-segmented, single-stranded, positive-sense RNA virus. Coronavirus resembles a crown and the electron microscopic image of Coronavirus is illustrated in Fig. 16.1.

Progressive resources are the principal basis of various technologies that can reduce the effect of Coronavirus. Nanovaccines are the influential proxies that could inhibit viral contaminations and nanosystems with antiviral activity might diminish the virus for its inactivation. Nanotechnology is a versatile technology that can serve
the solution to the pandemic situations such as Coronavirus outbreaks. Nanodiagnostics resemble nanoparticles, which encounter molecules of interest to generate a signal by permitting the recognition of infective viruses (Jackson et al. 2017). Nanotechnology provides significant benefits including rapid analysis, sensitive, highly accurate, and reproducible results. Nanotechnology-based biosensors can greatly reduce the use of chemical reagents needed for the analysis. A biosensor is a device that assembles with a profound biological detection module and a physical transducer to recognize viruses in the body fluids.

The characteristics of nanomaterial based biosensors include stability in fluids, suitable surface chemical properties, more surface energy and high amplification effect to generate measurable signals. Biosensors designed with the Metallic nanoparticles (MNPs), carbon-graphene based nanotubes, and photonic crystals (PCs) are widely used in the field of viral diagnostics. Figure 16.2 illustrates the various types of nanoparticle-based biosensors for SARS-CoV-1 and MERS-CoV detection. The principle involved in the technology is specific adsorption. The developed biosensors are the prominent modules for the development of SARS-CoV-2 biosensors. Table 16.1 presents the Biosensors developed for the detection of closely related COVID19 viruses.

16.4 SARS-CoV-1, MERS-CoV and SARS-CoV-2

16.4.1 Nucleic Acid-based Biosensors

Researchers designed a biosensor based on the surface plasmon resonance (SPR) on a chip with nucleotides for targeting the general respiratory viruses such as SARS-CoV. The test RNA samples were collected and subjected to the RT-PCR and hybridization has been carried out on the nanochip, which intensifies the signal and enhances the biosensor accuracy. The multi-target biosensor for the identification of target genetic materials from different infectious pathogens such as MERS-CoV was evaluated. The arch-shaped biosensors are applied to detect the other
Fig. 16.2 Different types of Biosensors and their principle involved in the detection of virus infections
| Biosensor                        | Virus            | Detection Probe          | Detection mechanism | Major findings                                                                 | Reference     |
|--------------------------------|------------------|--------------------------|---------------------|--------------------------------------------------------------------------------|---------------|
| SPR chip with immobilized      | SARS-CoV-1       | Oligonucleotide (DNA)    | RI signal           | Simultaneous identification of SARS-CoV-1, influenza A and B, H1N1, PIV-1, PIV-2, PIV-3, RSV, Adenovirus | Shi et al.    |
| specific oligonucleotides      |                  |                          |                     |                                                                                 |               |
| Photo crystals based biosensor | SARS-CoV-1       | Surface envelope protein | RI signal           | Detection of protein-protein, DNA-DNA and protein-metal interactions          | Park et al.   |
| chip                           |                  |                          |                     |                                                                                 |               |
| Carbon nanotube field-effect    | SARS-CoV-1       | Engineered antibody mimic protein (AMP) | Conductance | Efficient detection of SARS nucleocapsid protein                              | Ishikawa et al.|
| transistor (FET)-based biosensor|                 |                          |                     |                                                                                 |               |
| Arch-shaped multi-target        | MERS-CoV         | Oligonucleotide (DNA > 50 bp) | Resonance wavelength | Rapid detection of MERS-CoV                                                   | Koo et al.    |
| sensor                         |                  |                          |                     |                                                                                 |               |
| RCA microfluidic device        | MERS-CoV         | Oligonucleotide (ssDNA)  | Naked eye           | Multiple detection of pathogens                                              | Na et al.     |
| ESPS microfibers (electrospun  | MERS-CoV         | His-MERS NP antigen protein | Fluorescence       | MERS NP detection                                                             | Hoy et al.    |
| polystyrene)                   |                  |                          |                     |                                                                                 |               |
| Electrochemical immunoSENSOR   | MERS-CoV         | S1 protein               | Square wave         | Detection in spiked nasal samples                                             | Layqah et al. |
| based on an array of carbon     |                  |                          | voltammetry         |                                                                                 |               |
| electrodes (DEP) modified with  |                  |                          |                     |                                                                                 |               |
| gold nanoparticles              |                  |                          |                     |                                                                                 |               |
| S. aureus nanobioparticles      | MERS-CoV         | MER-CoV nucleoprotein    | Agglutination test  | MERS NP detection                                                             | Qiao et al.   |
| Dual-functional plasmonic      | SARS-CoV-2       | Oligonucleotide (DNA)    | Plasmonic resonance | High sensitivity towards selected SARS-CoV-2 sequences                         | Qiu et al.    |
| biosensor                      |                  |                          | wavelength          |                                                                                 |               |
| FET-based biosensor (COVID-19  | SARS-CoV-2       | Specific antibody against | Electrical          | SARS-CoV-2 in medium culture and in nasopharyngeal swab samples from COVID-19  | Seo et al.    |
| FET)                           |                  | SARS-CoV-2Â S protein    | response             | patients                                                                      |               |

Table 16.1 Detection of COVID19 viruses (SARS-CoV-1 and MERS-CoV) by Biosensors
infectious pathogens including Zika virus, Ebola virus, and SARS-CoV-1. Recently, the plasmonic photo-thermal (PPT) and localized surface plasmon resonance (LSPR) sensing transduction based biosensors are developed to detect the RNA from SARS-CoV-2. The sensor is assimilated on a chip by two-dimensional gold nanoislands (AuNIs). The developed device was also validated for the detection of various genome sequences from both the SARS-CoV-2 and CoV-1 and the significant outcomes are exhibited.

16.4.2 Antigen-based Biosensors

A photosensitive based biosensor is designed for the detection of SARS-CoV-1 S antigen. The biosensor comprised of photonic crystals (PCs) synthesized with chemical carboxyl groups (aldehyde or ketones) for the attachment of S protein, which endorsed identifying SARS-CoV-1 S antibodies. Researchers proposed biosynthesized nanoparticles inside the Staphylococcus aureus cells, where the MERS and Ebola virus nucleoproteins (MERS NP and EBOV NP) are coupled with the help of a cell wall binding domain (CBD) from a bacteriophage lysin PlyV12. The nanoparticles are designed using responding the monotetrazolium redox dye at room temperature for 15 min to produce insoluble formazan crystals inside the cells followed by inactivation at high temperature (65°C). S. aureus nanoparticles are used to execute an agglutination test to detect the IgG antibodies of Ebola virus and MERS nanoparticles.

16.4.3 Antibody-based Biosensors

Ishikawa et al., invented biosensors with carbon nanotubes which work based on the field-effect transistor. The designed nanotubes are capable to detect the nucleocapsid (N) protein of SARS-CoV. A novel biosensor based on antibody for the detection of S protein from SARS-CoV-2 is developed. The sensor containing grapheme bundles of the field-effect transistor (FET), which are smeared with CoV-2 antibodies. Furthermore, the performance of biosensor was evaluated for the detection of nasopharyngeal swab samples from COVID-19 patients.

16.4.4 Virus Inhibition Using Nanosystems

Virus inhibition can be done with Nanosystems against SARS-CoV-2 and worked based on the detection of inactivate enveloped viruses. The hydrophobic regions present on the nanosystems interact with the fatty acids on the surface portion of the virus endorsing its denaturation, which causes the inhibition of virus. Researchers
developed various types of nanoparticle-based nanosystems to detect the viruses by following the inhibition mechanism. The graphene oxide and their derivatives act as antiviral agents against Porcine epidemic diarrhea virus (PEDV) and Pseudorabies virus. Graphene oxide-silver nanosystems (GO-Ag) are verified against enveloped and non-enveloped virus including feline Coronavirus and infectious bursal disease virus. The magnetic hybrid colloids (MHC) comprised with Ag nanoparticles have a vital role in inactivation of viral pathogens. Functionalized carbon quantum dots (CQD) are the most effective therapy against human coronavirus (HCoV-229E). The carbon quantum dots are produced using an aqueous carbonization process. Ag\textsubscript{2}S nanoclusters (NC) having quantum dots with glutathione are the good inhibition properties against coronavirus proliferation. The curcumin based carbon dots are developed with the antiviral activity against intestinal Coronavirus.

Nanosystems can be effectively applied to destroy the virus as well. The electromagnetic irradiation is engaged by producing reactive species (RS) or photo-thermal heating. Hence, the viruses might be destroyed by the redox or denaturation processes. The carbon nanohorns (CNH) synthesized with a polyethylene glycol (PEG) derivative on one side and a T7 promoter tag antibody on the other side PEG is effective approach to kill the virus.

### 16.4.5 Nanovaccine Models Against COVID-19

Nanotechnology proved effective in combating COVID-19 with the production of nanovaccines. Nanovaccines consist of nanoparticles functioning as delivery vectors for antigens that activate defensive immunity. Various nanoparticles have the inherent immune-stimulatory characteristics that favour vaccine activity. Hence, nanosystems can be effectively used for the designing of vaccines. Virus like Particles (VLPs) mark significant in nanovaccine production. Most of the recombinant vaccines available in the healthcare industry are, indeed, based on VLPs (HBsAg and HPV). Additionally, other nanoparticles based vaccines are being explored including liposomes and particles comprised of gold and chitosan. Additionally, gold nanoparticles (AuNPs) based nanosystems have been demonstrated in the development of nanovaccines against Coronaviruses.

### 16.5 Conclusion

Microbial nanotechnology is the present day promising field. Nanotechnology-based advancements are commonly denoted to as systematically extracted information and open new avenues of research and applications that will progressively affect all portions of the world. Hence, there is an immense requirement from the scientific community for a complete and comprehensive understanding of the toxicity of the particle if any, their interactions within the ecosystem and ultimately the fate of the
biosynthesized nanoparticles so that this powerful technology could be adapted for the welfare of the human and animal welfare as well in the environmental conservation. The lack of exhaustive and complete toxicological data is also due to the actual difficulty to characterize, detect, and measure nanoparticles alone and in complex matrices like food/feed and biological samples. Concerns are elevated as no administration has established a monitoring structure that reveals the nanoparticles. Therefore, extensive research is prerequisite to reach strong and dependable risk assessment procedures for nanomaterials to be applied in medical the field of microbiology and disease diagnosis.

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