Interdisciplinary Biotechnological Advances

P. M. Visakh
Oguz Bayraktar *Editors*

Recent Progress in Nanobiotechnology

Modern Techniques in Biomedical Applications

Springer

Interdisciplinary Biotechnological Advances

This series is an authoritative resource that explains recent advances and emerging directions in biotechnology, reflecting the forefront of research clearly and reliably, without excessive hype. Each volume is written by authors with excellent reputations and acknowledged expertise in the topic under discussion. The volumes span the entire field from an interdisciplinary perspective, covering everything from biotechnology principles and methods to applications in areas including genetic engineering, transgenic plants and animals, environmental problems, genomics, proteomics, diagnosis of disease, gene therapy, and biomedicine. The significance of these applications for the achievement of UN Sustainable Development Goals is highlighted. The series will be highly relevant for Master's and PhD students in Biotechnology, Nanochemistry, Biochemical Engineering, and Microbiology, medical students, academic and industrial researchers, agricultural scientists, farmers, clinicians, industry personnel, and entrepreneurs.

P. M. Visakh • Oguz Bayraktar
Editors

Recent Progress in Nanobiotechnology

Modern Techniques in Biomedical Applications

Springer

Editors
P. M. Visakh
Department of Chemical Oceanography
Cochin University of Science and Technology
Cochin, Kerala, India

Oguz Bayraktar
Chemical Engineering
Ege University
İzmir, Bornova, Türkiye

ISSN 2730-7069 ISSN 2730-7077 (electronic)
Interdisciplinary Biotechnological Advances
ISBN 978-981-97-2842-8 ISBN 978-981-97-2843-5 (eBook)
https://doi.org/10.1007/978-981-97-2843-5

This Springer imprint is published by the registered company Springer Nature Singapore Pte Ltd.
The registered company address is: 152 Beach Road, #21-01/04 Gateway East, Singapore 189721, Singapore

Preface

Recent progress in nanobiotechnology: Modern techniques in biomedical applications—State of the art and new challenges are given in the first chapter. In this chapter, I have reported short explanation of all chapters in this book, with different titles such as nanobiotechnology for brain tumor-targeted therapies, cell culture models and nanobiotechnology for advanced drug delivery research, bionanocomposites in medical and food packaging applications, cell culture models and nanobiotechnology for advanced drug delivery research, biomolecules organize nanomaterials for medical applications, polymer-based nanotechnology to combat the emergence of drug resistance in bacteria, microbial compartments and their biomedical applications, and bio- and medical applications of carrageenan-based bionanocomposites.

The second chapter offers a review of nanobiotechnology for brain tumor-targeted therapies.

This chapter covers the brain tumor treatment that needs more research and technology development based on the nanobiotech to improve brain cancer patient's life. Authors of this chapter proposed the future research that should focus on enhancing the therapeutic potential of nanobiotechnology-based medicines and undertaking in-depth pharmacological studies to combine existing scientific knowledge into effective brain tumor treatment with minimal side effects. The conventional cancer diagnosis, clinical symptoms and physical examination, positron emission topography, nanobiotechnology for cancer diagnosis are described in Chap. 3. Finally, authors discuss the quantum dots, gold nanoparticles, and carbon nanotubes in this chapter.

Chapter 4 deals with bionanocomposites in medical and food packaging applications. In this chapter, biopolymers in medical application, introduction to food packaging, bionanocomposites preparation for food processing applications, and incorporation techniques are discussed. The applications of piezoelectric materials in tissue engineering and bionanocomposites characterization are also reported. The fifth chapter discusses the cell culture models and nanobiotechnology for advanced drug delivery research. In addition, the cell culture models, cell culture models and nanobiotechnology, cell culture models and nanobiotechnology for advanced drug delivery are also discussed.

Biomolecules organize nanomaterials for medical applications are covered in Chap. 6. Introduction to biomolecules organize nanomaterials and biomolecules organize nanomaterials for medical applications are discussed in this chapter. In Chap. 7, polymer-based nanotechnology to combat the emergence of drug resistance in bacteria is discussed. Also, cationic polymers, polymeric nanocarriers, and applications of polymeric nanocarriers in eradicating bacteria and their biofilms are also discussed. Microbial compartments and their biomedical applications are discussed in Chap. 8; preparation and characterization of biodegradable nanocomposites are also reported. The final chapter of this book discusses biomedical applications of carrageenan-based bionanocomposites. In this book, chapter authors have reviewed different aspects of recent progress in nanobiotechnology—modern techniques in biomedical applications. This book is a valuable reference source for faculties, professionals, research fellows, senior graduate students, and researchers working in the field of nanobiotechnology—modern techniques in biomedical applications. The use of modern techniques in biomedical applications are considered as a promising area of research, a lot of research activities are going in the area of biomedical applications. Finally, we would like to express our sincere gratitude to all the contributors of this book, who made excellent support to the successful completion of this venture. We also thank the publisher Springer for recognizing the demand and importance of modern nanobiotechnology in biomedical applications.

Cochin, Kerala, India — P. M. Visakh
İzmir, Bornova, Turkey — Oguz Bayraktar

Contents

About the Editors

P. M. Visakh (MSc, MPhil, PhD) is a prolific editor with more than 40 books already published. Now he is working in the Department of Chemical Oceanography, School of Marine Sciences, Cochin University of Science and Technology, Cochin, Kerala, India. He was an Associate Professor in TUSUR University, Tomsk, Russia since 2017 till July 2023. He did his post doc research in Tomsk Polytechnic University, Tomsk, Russia (2014–2017). He obtained his PhD, MPhil, and MSc degrees from the School of Chemical Sciences, Mahatma Gandhi University, Kottayam, Kerala, India. He edited 40 books from Scrivener (Wiley), Springer; Royal Society of Chemistry, Elsevier and more than 25 books in press (from Wiley, Springer, Royal Society of Chemistry and Elsevier). He has been invited as a visiting researcher in Russia (2014–2023), Portugal (2013, 2014), Czech Republic (2012, 2013), Italy (2009, 2012), Argentina (2010), Sweden (2010, 2011, 2012), Switzerland (2010), Spain (2011, 2012), Slovenia (2011), France (2011), Belgium (2012), and Austria (2012) for his research work. He visited 12 countries; he visited 15 universities in Europe. He published 20 publications, 4 reviews, and more than 45 book chapters. He has attended and presented more than 28 conferences; he has 2723 citations and his h-index is 24. He acts as guest editor for four ınternational journals.

Oguz Bayraktar received his Bachelor's degree from Ege University, Chemical Engineering Department in 1993. He received Master's and Doctorate degrees from West Virginia University in the United States. In 2001, he started to work as an Assistant Professor at the Izmir Institute of Technology, Department of Chemical Engineering. In 2006, he received the title of Associate Professor in the Department of Chemical Engineering. He received the title of Professor in the Department of Chemical Engineering in 2012 and worked as a faculty member at the Izmir Institute of Technology, Department of Chemical Engineering. He worked as a Professor at the Department of Chemical Engineering at Ege University between 2014 and 2020. In 2022, he established the NATCO Ltd. start-up company. He is currently working as a Professor in the Department of Bioengineering at Ege University. His fields of study are

biotechnology and bioengineering, and he actively continues his research in the interdisciplinary field. Determination of biological activities of natural compounds obtained from plants, isolation, purification, production, bioconversion of natural compounds, cosmetics, and functional food applications are among his main interests.

Recent Progress in Nanobiotechnology: Modern Techniques in Biomedical Applications: State-of-the-Art and New Challenges

P. M. Visakh

Abstract In this chapter providing a short version of all chapters, here I am writing about the different chapter topics such as nanobiotechnology for brain tumor-targeted therapies, cell culture models, nanobiotechnology for advanced drug delivery research, bionanocomposites in medical and food packaging applications, cell culture models and nanobiotechnology for advanced drug delivery research, biomolecules-organized nanomaterials for medical applications, polymer-based nanotechnology to combat the emergence of drug resistance in bacteria, microbial compartments and their biomedical applications, and bio and medical applications of carrageenan-based bionanocomposites.

Keywords Nanobiotechnology · Brain tumor · Nanomedicine · Food packaging · Drug delivery · Carrageenan · Bionanocomposites

1 Nanobiotechnology for Brain Tumor-Targeted Therapies

Brain tumors are terrible diseases that grab a lot of attention because of their poor prognosis and high mortality. The current brain cancer treatment hasn't advanced far enough to allow them to be cured fully. The key factors are the heterogeneity of tumor, lack of specialized targeted treatments, tumor aggressiveness, the difficulty of getting medications through the blood–brain barrier, and the extensive spread of brain tumors. Over the last couple of decades, several nanobiotechnology-based therapies have been investigated for a variety of biological applications, including the treatment of brain tumors. The main applications of these drugs include targeted treatment, gene and pharmaceutical delivery, and improving the immune system for cancer treatment (Mangraviti et al. 2016; Qamar et al. 2019).

P. M. Visakh (✉)
Department of Chemical Oceanography, School of Marine Sciences, Cochin University of Science and Technology, Cochin, Kerala, India

P. M. Visakh, O. Bayraktar (eds.), *Recent Progress in Nanobiotechnology*, Interdisciplinary Biotechnological Advances,
https://doi.org/10.1007/978-981-97-2843-5_1

innovative drug designs (also known as nanomedicines) with fewer side effects.

Nanobiotechnology integrates many techniques to improve the delivery of biotech medications, whose properties can be altered using various chemical approaches. The main challenge in many disease therapies is getting therapeutic drugs to the target region. For example, in brain disorders, delivering drugs to cross the blood–brain barrier is a crucial task that nanobiotechnology can help with. In traditional uses of a few drugs, drug delivery control is characterized by limited efficacy, poor biodistribution, and a lacking of selectivity. There are some of the nonspecific symptoms for brain tumor patients. The other most common symptom in people with brain tumors is seizures. Among seizures, acidosis, hypoxia, metabolic, immunological, and inflammatory problems can also be seen in some patients. Nausea and vomiting are also indicators of a brain tumor when the chemo trigger zone in the postrema area, on the floor of the fourth ventricle, is activated (Pickering et al. 2021; Slegers and Blumcke 2020).

The research and implementation of nanobiotechnology in medical science developments have shown considerable promise in improving human living standards. Nanobiotechnology has a substantial impact on human health care for the prevention, early detection, diagnosis, treatment, and follow-up of a variety of disorders across time. Its numerous applications have attracted the attention of academics, manufacturers, clinicians, legislators, and the general public over the last three decades. Nanobiotechnology has the potential to enhance medication encapsulation, preserve pharmaceuticals from degradation, increase solubility, and target specific diseases (Jewett and Patolsky 2013; Gulati et al. 2022; Adam et al. 2019).

Exosomes can penetrate the blood–brain and blood–brain tumor barriers, and their properties have been employed in animal models to mediate cell-based therapy for brain tumor, Parkinson's disease, intracranial inflammation, Alzheimer's disease, and stroke. Exosomes have the potential to overcome the disadvantages of low bioavailability while also lowering cytotoxicity and immunogenicity. Transmembrane and membrane-anchoring proteins are also found in exosomes, which help in endocytosis (Gao et al. 2021; Dai et al. 2020; Aqil and Gupta 2022).

Zhu et al., designed a multifunctional ginsenoside Rg3-based liposomal system. The safety of the liposomes was determined by histological changes in the mice's organs following drug administration, and no pathological abnormalities were found in any of the liposome groups, suggesting that the liposome delivery system is safe and biocompatible. In glioblastoma, liposomes improve the chemotherapeutic efficacy of paclitaxel by having a synergistic tumor-killing effect, modulating the brain-tumor microenvironment, and activating the immune microenvironment, resulting in paclitaxel having a greater antitumor effect (Zhu et al. 2021).

Polyphenol nanoparticles, developed by Liu et al., have dual roles, limiting the creation of new arteries while also causing targeted disruption of the existing tumor tissues. The nanoparticles are made using a combination of iron coordination and polymer stabilization techniques, resulting in substantial drug loading and intrinsic tumor vascular targeting. After intravenous delivery, nanoparticles bind to VEGFR2, which is overexpressed in tumor vasculature. Nanoparticles have the ability to use as promising antiangiogenic medicines for the treatment of brain cancer due to their

amazing effects on tumor growth and medication transport (Liu et al. 2022). Arduino et al., developed polyethylene glycol-stabilized solid lipid nanoparticles containing Pt(IV)-prodrugs developed from kiteplatin for the treatment of glioblastoma. An in vitro study on the human glioma cell line (U87) showed that the Pt(IV)-prodrugs were taken up more readily by the cells, indicating that the solid lipid nanoparticles have better anticancer activity. The results showed that drugs encapsulated in solid lipid nanoparticles were able to cause a greater loss in cell viability than their free counterpart. In the measured lipid concentration range, polyethylene glycol-loaded solid lipid nanoparticles without drug were determined to be noncytotoxic (Arduino et al. 2020). Monoclonal antibodies have evolved from scientific tools to potent therapies for disorders of numerous systems, including cardiovascular, respiratory, hematological, immunology, and oncology, over the previous three decades. However, because of the brain–blood barrier (BBB) restriction, delivering mAb therapies into the central nervous system has been extremely difficult (CNS) (Sousa et al. 2018; Viola et al. 2018).

2 Cell Culture Models and Nanobiotechnology for Advanced Drug Delivery Research

Cancer can generally be classified into two categories, i.e., benign or malignant. Benign tumors are confined to the initial focal point without spreading to surrounding cells or tissues whereas malignant tumors are actively invading and spreading to surrounding cells and tissues. Cancer is characterized as uncontrolled cell proliferation that has the capability of spreading and invading various parts of the body from the initial point of occurrence and possibly causing death (Jin et al. 2020). Nanotechnology has been used for the diagnosis of cancer focusing at the molecular level through the development of gold nanoparticles, quantum dots, biomarkers, and nanoshells, among others (Rai et al. 2021). Thence, recent advancements in the field of nanobiotechnology provide great potential in the early detection and diagnosis of cancer as it can produce sensitive, specific, and rapid detection of abnormal or cancer-related molecules, enabling the detection of molecular changes even in a considerably small percentage of cells. Pain is a common symptom of cancer and is mostly experienced when the cancer has metastasized. Pain is a common early-stage sign of bone cancer, colorectal cancer, testicular cancer, pancreatic cancer, and ovarian cancer; in addition, a consistent painful headache is also commonly reported for brain tumors (Stark et al. 2012). Those with lung cancer may cough up blood whereas abnormal bloody vaginal discharge or bleeding could be caused by endometrial or cervical cancer. Blood in urine could be a sign of kidney or bladder cancer and on the other hand, bloody stools may indicate colon or rectal cancer (Pakish et al. 2016). In brief, some of the other signs or symptoms commonly associated with cancer include unexplained and sudden weight loss, extreme tiredness, visible

changes in skin complexion, changes in bladder or bowel movements, anemia, and hoarseness.

The X-ray emission tube launches a narrow and thin x-rays beam through the patient's body while rotating, simultaneously the electronic x-ray detectors found opposite to the x-ray source detect and measure the amount of radiation that is being absorbed by the body which is transmitted to a computer. The CT computer used specialized and sophisticated computer programs and mathematical techniques to interpret the data collected into two-dimensional cross-sectional image slice of the body (Cole and Hespel 2020). Ultrasound also known as ultrasonography or sonography, uses sound waves of high frequency typically between the range of 3 to 10 megahertz (MHz) for medical diagnostics which is approximately a hundred times greater than the human hearing limit. The high-frequency acoustic energy emitted from the transducer goes through the human body and is reflected by the organs and tissues creating echoes. These echoes generated from the reflection and scattering of acoustic waves are captured and converted into real-time images also known as a sonogram.

There are numerous benefits associated with the use of MRI for diagnostics. It can produce multiplanar and 3D images where the acquisition of sagittal, direct, oblique, and coronal image is possible. MRI is effective in portraying, staging the tumors, and also monitoring the efficacy of treatments for tumors as it has superior soft tissue resolution. Through MRI, physiological, biological, and histopathological information can be obtained (Lu et al. 2013). Increased accumulation of FDG is often observed in cancerous cells because of overexpression of glucose transporters on malignant tumor cells and spike in hexokinase concentration or action. However, there are some limitations with the use of PET where inflammatory conditions may show high-activity areas mimicking cancer (Shukla and Kumar 2006).

The antibodies used for IHC are modified in the laboratory to specifically recognize immune proteins linked to cancer. In IHC stains, the labeling antibody is altered with colored dyes or fluorescent dyes to enable easier tracing of microscopic or computerized analysis. IHC can be used to identify the origin and aggressiveness of the cancerous cells, as well as determine if the cancer has metastasized (Duraiyan et al. 2012). The magnifying power of an electron microscope is approximately 1000 times greater than that of a conventional light microscope. This exceptional resolving power of the electron microscope produces beneficial morphologic data which aids in establishing tumor histogenesis (Eyden 2002). The customary techniques used in molecular pathology and cytogenetics include fluorescence in situ hybridization (FISH). In FISH technique, a fluorescent tag is attached to a specific DNA segment which acts as a probe. The probe is later denatured into single strands that are exposed to denatured target DNA where the probe identifies and binds to the complementary gene sequence of the denatured target DNA under hybridization conditions. Subsequently, the sample is visualized by immunocytochemical or autoradiographic method where the probe bound to the gene is represented by bright spots. FISH technique is ideal to be applied in cancer diagnosis and management as chromosome dysregulations are often associated with malignant cells such as

duplicated genes, amplified genes, gene deletion, or translocated genes (Varella-Garcia 2003).

Advancements in nanobiotechnology have demonstrated promise in molecular level detection for cancer diagnosis. It is proven through extensive research that nanoparticles are well taken up and accumulated in cells because of its enhanced permeation and retention effect, even in cancerous cells. Thence, opens great potential in detection of molecular changes related to cancer even in a small percentage of cells (Vishwakarma et al. 2008). A new range of multifunctional Quantum Dots (QD) probes were reported for tumor targeting and imaging simultaneously in live animals. These new QD conjugates consist of targeting ligands, amphiphilic triblock copolymer and multiple PEG molecules for in vivo protection, recognition of tumor antigen, and improved circulation and biocompatibility respectively (Gao et al. 2005). The unique surface chemistry and affinity of AuNPs to thiol and amine groups offer potential for surface functionalization with molecules such as DNA, siRNA, antibodies, peptides, nucleic acid, and receptors. AuNPs are nontoxic, biocompatible, and the shape as well as size of the particles can be controlled (Ali et al. 2020). Sun et al. in 2019 demonstrated the application of glycol-chitosan-coated gold nanoparticles (GC-AuNPs) as a photoacoustic contrast agent for the imaging of cancer cell. The cellular uptake of GC-AuNPs in breast cancer cells was successful through the synergistic effect of glycol, chitosan, and gold nanoparticles. Strong photoacoustic signals were also produced after cellular uptake in cancer cells due to the plasmon coupling effect of GC-AuNPs establishing GC-AuNPs performance as contrast agent which were confirmed with dark-field microscopy (Sun et al. 2019).

A peptide nucleic acid (PNA) immobilized SiNW-FET was used to detect miRNAs for early diagnosis of cancer as miRNA plays a vital role in cell development related to certain cancers. The PNA functionalized SiNW-FET detected PNA-miRNA hybridization through base pairing and was capable of sensing specific miRNA in total RNA that was extracted from HeLa cells (Chen et al. 2011). Silicon nanowires (SiNWs) have a large surface-area-to-volume ratio which permits alterations in surface charge because of interaction with biological molecules or ions which consequently leads to physicochemical properties change of the SiNWs which are measurable. SiNWs can also be functionalized with capture ligands for excellent selectivity, specificity, and sensitivity in the detection of analyte of interest (Smith et al. 2020).

3 Bionanocomposites in Medical and Food Packaging Applications

Sultana et al. (Sultana et al. 2017) have prepared PLLA nanofibers piezoelectric bio-based e-skin. This can be used in energy harvesting and piezotronics applications. They have performed this e-skin as a function of different deformation frequencies investigated. Among the studied biopolymers used for biomedical

purposes, polyhydroxyalkanoates (PHA) have a high ability to bioincorporate. All these studies were used for biomedical applications, polyhydroxyalkanoates (PHA) have a high ability to bioactive ability than other biomaterials. Poly(3-hydroxybutyrate-co-3-hydroxyvalerate) (PHBHV) can be used as a bone filler. PHBHV is a copolymer of the PHA family. The PHBHV is biocompatible with different cells such as osteoblasts and chondrocytes, and the by-products from these materials are nontoxic. It can be seen in human blood and case for tissue regeneration. In addition, there is no change in the pH of the environment when degradation of PHA Polylactic acid (PLA) when degraded into lactic acid (Pouton and Akhtar 1996; Voinova et al. 2019; Guo et al. 2022), also PLA widely used biopolymers in medical fields especially as biomedical devices. Because of its biodegradability and biocompatibility, poly(3-hydroxybutyrate) (PHB) is one of the most widely used synthetic polymers in tissue engineering, as well as noncytotoxicity of its metabolic products (Suwantong et al. 2007). PHB has an asymmetric crystal cell structure, which defines its piezoelectric properties (Chernozem et al. 2018). PHB-based piezoelectric scaffolds are successfully used as a material for engineering bones, skin, and nerve tissues (Nagiah et al. 2013; Cao et al. 2009).

There are some package applications of these materials these packaging as a product container. These container protecting from compressions, shocks and plays a very important role in the preservation of food, these packaging product acting as a barrier to biological, chemical, and physical contamination and deterioration elements (Souza and Fernando 2016).

Polyunsaturated lipids, essential oils, carotenoids, probiotics, phenolic compounds, peptides, and vitamins are an asset when added to food. And they confer a broad variety of functional properties, such as antimicrobial, anti-inflammatory, and antioxidant (Souza et al. 2019; Pascoal et al. 2015; Santos et al. 2018).

The method is dependent on the characteristics and type of the coating material and the core, even though, no other proposed systems can be officially recognized as a standard (Shishir et al. 2018). The nanoemulsion-based encapsulation consists of three phases, a dispersed/discontinuous internal phase, an interphase called an emulsifier, and a continuous external phase. The nano-ultrafine emulsions have been obtained by processes with high energies such as microfluidizers inversion high-pressure valve homogenizers and spontaneous emulsification.

Biopolymers are obtained from renewable sources and the incorporation of active compounds and also some functional properties such as antifungal and antimicrobial properties, pigments, and other nutrients (Mohanty and Swain 2017; Souza et al. 2020; Souza et al. 2018). In the bionanocomposites preparation, inorganic nanoparticles have been used as reinforcement, and in most cases metal or metal oxide nanoparticles were used. Some metal nanoparticles such as gold, zinc, and silver are commonly used for food packaging applications. Nanocellulose, added into a polymeric matrix allows the improvement of the mechanical properties as well as oxygen barrier properties. They are well-renewable and recycling materials, it can be used as a nanofiller in packaging applications (Pires et al. 2019).

Sahraee et al. (2020) studied the effect of ZnO NPs incorporated in bilayer films, and these films were used for the sponge cake packaging. A film was prepared by

layering method, first layer of ZnO NPs, N-chitin, and gelatin and a second layer with gelatin emulsion. The ZnO NPs show the improvement of the barrier properties of the film.

Piezoelectric materials are used for many applications such as sensors, actuators, energy harvesting electronics, and bioimplants (Sirohi and Chopra 2000). The direct piezoelectric effect is the charge separation in response to external mechanical stress, this effect is useful for applications in energy harvesting and sensors applications. An applied external electric field induces mechanical deformation in the reverse piezoelectric effect, this gives a potential application in mechanical actuators. The reverse or direct piezoelectric energy conversions have found their way into different biological applications such as drug delivery platforms, biosensors, and bioactuators (Jariwala et al. 2021; Chen et al. 2017).

There are many bioelectrical activities and piezoelectricity happening in the various tissues of the body such as nerves, cartilage, ligaments, dentin, and bones. The body tissues are attempts being made to use piezoelectric materials as a self-powered platform for electrical stimulation of cells/tissues in tissues, this can be used in engineering applications and regenerative medicine (Lang 1966; Yin et al. 2010; Zhang 2015; Bhang et al. 2017; Jacob et al. 2018).

4 Cell Culture Models and Nanobiotechnology for Advanced Drug Delivery Research

The ability to modify genes and molecular pathways is one of the advantages of employing cell culture methods. Moreover, well-defined culture systems, or the homogeneity of specific cell types as well as clonal cell populations, eradicate both environmental factors and meddling genetic variables, enabling data generation with consistency and high reproducibility, which is difficult to guarantee while studying complete organ systems (Segeritz and Vallier 2017). In medicine and biotherapeutics, nanobiotechnology provides some novel possibilities (Bawa 2010). Nanobiotechnology is an interdisciplinary field that deals with the use of nanomaterials in biotechnology. This discipline has major implications in health, pharmaceutical application, medicine, tissue engineering, imaging, cosmetics, agriculture, immunoproteomics, and pharmacy. Many pharmaceutical researchers are studying drug metabolism and transport at explicit biological barriers in cell culture in order to develop new delivery mechanisms for therapeutic candidates developed through rational design of drug recombinant technologies of DNA (Wilson et al. 2012).

In addition, a cell culture study can accurately predict toxicity at low cost or cost-effectively, biotransformation, or permeability; thus, reducing the number of animal tests needed to narrow down or reduce a list of potential new drugs with the best preclinical potential. Another advantage of cell culture models is the identification of particular molecular pathways at the cell level (Karunaratne et al. 2005; Kalashnikova et al. 2016). After a certain number of passes, many primary cultured

cells are no longer stable, and some will not multiply (Freshney 2006). Immortalized cellular lines could be created through viral /chemical means, and some of the cells, most notably malignant cells, are naturally continuous. Primary cells have several advantages over immortalized cell lines, despite the fact that corresponding immortalized cellular lines are frequently more appropriate. To ensure that the system exhibits biological barrier properties, the corresponding cell system's barrier properties, along with its biochemical or morphological attributes, must be assessed after choosing a suitable cell culture model (Borchardt et al. 1991).

For example, consider the Caco-2 system (an in vitro system for determining pharmaceutical drug absorption), which has characteristics that suggest it could be a model of the intestinal epithelium that is polarized. The principal drug absorption site, the epithelium of small intestinal, is made up of monolayer of cells with high morphological and functional heterogeneity (Trier 1981). In order to anticipate the pharmacodynamic (PD) and pharmacokinetic (PK) behavior of many distinct kinds of medications, several blood–brain barrier (BBB) models have been developed, ranging from in silico to in vivo (Zhang et al. 2017; Vastag, and Keseru. 2009; Banerjee et al. 2016). The isolation of bone marrow microvascular endothelial cells (BMEC) from various species is used to create BBB primary cell culture model. To simulate or mimic the in vivo settings, astrocytes, pericytes, and neurons can all be cocultured with primary cells. Calu-3 is a cell line that is extensively used to study airway epithelium. Calu-3, an adenocarcinoma cellular line with tight connections, forms a confluent monolayer and has the ability to produce mucus. Transport experiments can be conducted in submerged culture after 6–8 days and in air interface culture after 10–14 days (Kreft et al. 2015; Harcourt et al. 2013; Sousa and Castro 2016).

The cell culture model is an essential tool in a wide range of clinical and biological research (Amelian et al. 2017). Fundamental information regarding cytotoxicity, drug penetration, and drug accumulation in cells is provided by 2D cell culture models. They're critical while looking for new compounds with appropriate biopharmaceutical properties to be candidates for new drugs (Vunjak-Novakovic and Freshney 2006). In comparison to 2D growing cells, 3D tissue models more precisely imitate in vivo settings and characteristics of real tissue. These cell culture models lower drug development cost, improve medical compound screening, and eliminate the use of animals in clinical trials (Geraghty et al. 2014). Some cell culture-based conclusions depending upon nanoparticle research cannot be transferred to animals or people due to the absence of the endocrine system, immunological impact, blood proteins, and a general lack of complicated animal interaction in most in vitro systems (Kura et al. 2014; Blaauboer 2008).

Current diagnostic approaches for a wide range of diseases depend on observable signs/symptoms before healthcare professionals can determine whether or not a patient has a specific sort of illness. In the agricultural and biomedical industries, several enzymatic and electrochemical biosensors have been developed for disease diagnosis (Verma 2017a, b). Thus, the earlier disease detection, improved the possibility of cure. The most advantageous method is diagnosing and treating the patient before the symptom manifests.

5 Biomolecules-Organized Nanomaterials for Medical Applications

The biofunctionalized nanomaterials exhibit conspicuous properties such as unique structure, ultra-small sizes, large surface area, physicochemical reactivities, and high compatibility for surface modifications (Veerapandian and Yun 2011). Biomolecules with nanoscale structures, functions, and processes are used to develop innovative functional nanostructured biological materials known as bionanomaterials, which assist to broaden the research area in nanotechnology and biotechnology-related fields (Nagamune 2017). The distinct properties of bionanomaterials enable them to play numerous roles in the biomedical field, with multiple applications, including therapeutic and diagnosis, drug and gene delivery, tissue engineering, cancer treatment, antibacterial activity, etc. Bionanomaterials could also be used to self-assemble complex devices in environmentally friendly mild conditions (Singh et al. 2021).

Nanobiotechnology is used similarly to nanotechnology where it is used to create materials, devices, and systems for studying biological systems and developing new biological assays, diagnostic, therapeutic, information, storage, and computing systems, among other. Undoubtedly, tremendous progress has been made in the development of biomolecule-based nanomaterials and nanostructures, as well as their applications in sensing, nanocircuitry, nanoscale machinery, logic operations, and fabrication of nanodevices over the last few decades (Willner and Willner 2010). Biomolecules can be used as both stabilizing and reducing agents in the growth of MNPs. Furthermore, biomolecules were used as the templates for the functionalization and multidimensionality of MNPs (Zhang et al. 2020). Biomolecule-conjugated MNPs are not only more structurally stable than pure MNPs, but they can also retain the functions of the biomolecules themselves, achieving therapeutic and diagnostic functions (Mabrouk et al. 2021).

Electrostatic interactions, van der Waals forces, hydrogen bond forces, and combinations of two or more of these forces contribute to stabilization through capping agents. However, the function of capping agents is not limited to protecting the NPs but can also control the interaction between the NPs and their environment. Proteins are major biomolecules that are present in the body of all living organisms. Peptides are compounds that consist of three or more amino acids linked together by peptide (amide) bonds, which are also intermediates of protein hydrolysis. There are several outstanding advantages of using peptides to synthesize metal nanoclusters, including easy access to their resources, renewable resources, biocompatibility and biodegradability, the presence of multiple functional groups that can carry large amounts of drugs, and the ability to connect targeting groups to them (Seeman and Sleiman 2017; Kianfar 2021).

Some of the biomolecular engineering, such as protein engineering, genetic engineering, DNA and RNA engineering, self-assembly technology, site-specific chemical, enzymatic conjugation technologies, and massive high-throughput screening (HTS) methods, will be enabled to improve the functions and properties

of biological materials (Tam and Lo 2015; Wang and Ding 2013). Due to the compatibility of nanomaterials with biomolecules, nanomedicine includes the development of NPs (among other nanocomponents and devices) for molecular diagnostics, treatment, and prevention of human diseases. The adsorption of dozens of proteins with different identities and quantities on NPs changes their physicochemical identity and influences physiological responses such as cellular uptake, targeting, blood circulation lifetime, and toxicity. For DNA, RNA, and proteins, the sensing techniques for biological analyses help more efficient detection and sensitive of these analytes at low concentrations.

There are many applications of using NPs for sensors such as detecting and capturing cells, detecting analytes at very low concentrations, detecting molecular and cellular functions detecting and separating pathogens. NPs have composition-dependent properties, high surface-area-to-volume ratio, and a small size that allows the use of surface ligands to increase the faster detection or detection threshold (Doria et al. 2012). This is an advantage in early disease diagnosis and therefore treatment at the early stages of a particular condition. Polysaccharides, such as hyaluronic acids (HA), have been used in biomedicine for therapeutic purposes due to their inherent targeting and therapeutic properties. HA is a nonimmunogenic and more specifically a mucopolysaccharide component of the extracellular matrix (Han et al. 2015).

HA-based nanostructures are the specific receptor for CD44 that is overexpressed in many cancers. Hence, HA-based nanostructures could be used for CD44-mediated targeting and cell uptake (Choi et al. 2014; Buschmann et al. 2021). Tissue engineering is primarily concerned with skin tissue engineering, cartilage and bone construction, corneal tissue engineering, vascular tissue engineering, oral tissue engineering, nerve tissue engineering, tendon and ligament tissue engineering. Hydrophobic drug administration via conventional routes has several drawbacks, including poor water solubility, rapid biodegradation, nonspecific delivery, and serious side effects. To get around this, drug delivery systems could encapsulate these therapeutic agents. Due to their biocompatibility, noncytotoxicity, and biodegradability, self-assembled nanomaterials are an excellent solution for drug delivery applications. Nanocarriers should encapsulate molecules at high concentrations, protect them from dilution and degradation, and release them in a controlled and prolonged manner for drug loading and release (Olshefsky et al. 2022) producing enhanced effects with less wastage.

6 Polymer-Based Nanotechnology to Combat the Emergence of Drug Resistance in Bacteria

One of the powerful strategy to overcome the emergence of drug resistance is using nanocarriers to encapsulate the conventional antibiotics and release the cargoes in a controlled manner. In the past decades, various polymeric nanocarriers for

antibiotics have been developed. With the assistance of nanocarriers, conventional antibiotics can be temporarily protected and prevented from their interactions with normal tissues and cells, minimizing the side effects of antibiotics. Of note, the drug-loaded nanocarriers can pass by the bloodstream and accumulate at an infection site via the enhanced permeability and retention (EPR) effect (Ding et al. 2020). To prepare the polymeric micelles, various methodologies have been developed (Cagel et al. 2017) for example, direct dispersion, nanoprecipitation, emulsification, solvent volatilization, dialysis, thin film, etc. Direct dispersion is to directly place the amphiphilic polymer in an aqueous solution to form polymeric micelles. This method is a convenient and quick way to prepare polymeric micelles. Besides, organic solvents are not involved in the whole procedure, therefore, the potential cytotoxicity of residual organic solvents could be prevented. As aforementioned, polymersomes or polymeric vesicles are usually self-assembled by block or grafted amphiphilic copolymers with a packing parameter $p > 1/2$. Compared with ordinary liposomes, polymersomes have similar structures yet better stability. A typical polymersome is a hollow sphere with a hydrophilic cavity surrounded by a double-layer membrane. Generally, polymersomes are composed of high molecular weight amphiphilic copolymers, and their membranes are about 3–4 nm thick and relatively strong.

To increase the stability of polymeric micelles, commonly used strategies include core cross-linking or shell cross-linking via chemical covalent bonds, hydrogen bonding, and π-π stacking. Of note, the cross-linked polymeric micelles will no longer be affected by the CMC and can resist the disintegration/dissociation of micelles caused by infinite dilution. More importantly, stimuli-responsive domains could be constructed into the polymeric micelles or cross-linkers, to achieve the triggered disintegration of micelles and on-demand drug release in response to either internal or external stimuli. Besides, charged polymers can attract oppositely charged polymers through electrostatic interactions to form the polyion complex (PIC) micelles (Harada and Kataoka 2018). PIC micelles are usually prepared from completely hydrophilic polymers, and their size can generally be well controlled below 100 nm with a narrow size distribution. This is mainly because the preparation is carried out in an aqueous solution, unlike other methods that involve solvent exchange.

The antimicrobial drug loaded in polymeric nanocarriers needs to be released into the body to treat bacterial infection. So, understanding the drug release behavior is important for designing and application of polymeric nanocarriers. In general, the drug release behavior was affected by multiple factors coming from the polymeric nanocarriers (e.g., the stimuli responsiveness, structure, and geometry), the release environment (e.g., temperature, pH, ionic strength, and enzyme), and the properties of the drug (e.g., solubility and interaction with nanocarriers) (Liu et al. 2021).

Unlike polymeric micelles, polymersomes possess a hydrophilic cavity and a hydrophobic shell. Therefore, both hydrophilic and hydrophobic therapeutics can be encapsulated in polymersomes. For example, ciprofloxacin can be encapsulated in the hydrophilic cavity of the polymersomes composed of

poly(ε-caprolactone)-*block*-poly-(glutamic acid), and the poly-(glutamic acid) shell further chelates Ce^{3+}. The yielded drug-loaded polymersomes are efficient to eradicate the bacteria and quench the reactive oxygen species caused by staphylococcal infections in open wounds, as well as accelerate the wound closure (Wang et al. 2021).

7 Microbial Compartments and Their Biomedical Applications

In nature, various intricate and delicate micro−/nanostructures are constructed in the living things on Earth. Of particular, eukaryotic cells possess membrane-bound compartments such as mitochondria, nucleus, and Golgi apparatus (Cohen et al. 2018). On the contrary, prokaryotes have neither a distinct nucleus with a membrane nor other specialized organelles. Instead, prokaryotic cells complete many of their biological processes in the multimeric protein-derived compartments (Ding et al. 2021) called bacterial compartments (BCs) or prokaryotic microcompartments, which were found in the 1950s with 80–400 nm pseudo-icosahedral structures. The ideal size that facilitates the systemic administration ranges from 20 nm to 200 nm (Liu et al. 2019). If the size of particles is smaller than 7 nm, they are easily effluxed from blood circulation via renal clearance. Particles with a size bigger than 500 nm are readily opsonized and cleared by the reticuloendothelial system (RES) in vivo. Therefore, the size of particles that are suitable for biomedical applications is suggested to be 20–200 nm. These particles often possess applaudable blood circulation time and can accumulate at a lesion via the enhanced penetration and retention (EPR) effect (Ding et al. 2020). Epy ferritin is a soluble protein that exists widely in organisms and is highly conserved. It plays an important role in the process of iron ion homeostasis, embryonic development regulation, cell proliferation, and cell apoptosis. Excessive iron ions can produce reactive oxygen species through the Fenton reaction. Meanwhile, excessive reactive oxygen species can cause oxidative stress and directly damage DNA, lipids, and proteins, and ultimately lead to cell apoptosis. Ferritin can chelate iron ions, thereby protecting cells from apoptosis induced by oxidative stress (Watt et al. 2010).

A vaccine could be injected directly or be accompanied by a carrier. Recently, protein cages have been widely used as antigen delivery systems to dendritic cells, subsequently inducing antigen-specific T cell proliferation. For example, Ovalbumin (OVA) peptides OT-1 and OT-2 could be genetically inserted into protein cages such as encapsulins (Choi et al. 2016) and lumazine synthase (Ra et al. 2014). Hydrogels are commonly used scaffolds in tissue engineering due to their structural similarities to the natural extracellular matrix. However, the drug release inside of hydrogels is a major challenge that limits the biomedical application of hydrogels in tissue engineering. Protein-based scaffolds have been used as matrices and delivery systems in tissue engineering (Wozney and Seeherman 2004). In this regard, protein

cages are an ideal drug delivery system since protein cages can provide a considerable stable environment to prevent the denaturation and degradation of drugs. With rational modification, protein cages serve as building blocks to construct the hydrogels for tissue engineering.

8 Bio and Medical Applications of Carrageenan-Based Bionanocomposites

Carrageenan is a term that denotes a family of water-soluble and sulfated polysaccharides, which are extracted from certain species of red seaweeds (macroalgae). The species that commercially used for the production of carrageenan include *Eucheuma*, *Chondrus*, *Kappaphycus*, *Hypnea*, *Gigartina, Solieria, Agardhiella, and Sarconema (*Hernandez-Carmona et al. 2013*;* Kalsoom Khan et al. 2017*)*. The use of carrageenan is including a broad range from food industry, such as gelling, stabilizing, emulsifying, and thickening agents, to pharmaceuticals and cosmetics (Zia et al. 2017). The carrageenans are classified into three types of kappa (κ), iota (τ), and lambda (λ) according to the presence or not of anhydrogalactose unit and the sulfation pattern. Minor types are mu (μ-) and nu (υ-), which correspond to κ- and τ-precursors, respectively (Freile-Pelegrin et al. 2011). In most cases, a plasticizer is added to the film to improve the dispensability, extensibility, and flexibility of the developed film while these changes in molecular organization reduce the cohesion and rigidity of the film at the same time (Espitia et al. 2014). Glycerol is the most important plasticizer used in film making, because of their stability and compatibility with hydrophilic biopolymeric packaging chain (Chillo et al. 2008). Meindrawan, Suyatma (Meindrawan et al. 2018) prepared a bionanocomposite coating based on carrageenan and zinc oxide (ZnO) nanoparticles to extend the shelf life of mango. They first prepared a solution containing κ-carrageenan and ZnO, and also glycerol as plasticizer. After formulating, the mango was dipped in the coating solution for desired time and drained. In another study, Salgueiro, Daniel-da-Silva (Salgueiro et al. 2013) synthesized τ-carrageenan-coated magnetic iron oxide nanoparticles. The prepared nanoparticles are dispersed into the carrageenan solution, and the final bionanocomposite is usually synthesized by cross-linking with an iron cation. The nanoparticles are prepared using a suitable methods developed by researchers such as coprecipitation, chemical reactions in emulsions or plant extract, hydrothermal. They first synthesized the Au nanospheres and nanorods by reduction of a gold (III) complex and seed-mediated growth method, respectively. Afterwards, the nanocomposites were prepared by blending the nanoparticles with the polymer matrix at distinct aspect ratios. Azizi, Mohamad (Azizi et al. 2017) also fabricated bionanocomposite hydrogel beads based on κ-carrageenan and biosynthesized silver nanoparticles (Ag-NPs).

To improve the barrier properties of carrageenan coatings, the carrageenan/chitosan nanolayered coating was prepared by dipping method and then characterized