American Journal of Bioscience and Clinical Integrity
https://biojournals.us/index.php/AJBCI
ISSN: 2997-7347

The Therapeutic Effect of Zno-Loaded Alginate Nanoparticles on Pancreatic Cancer

Hasan Mustafa Hamdi, Ghada Abbas Raoof
Republic of Iraq, Ministry of Higher Education and Scientific Research, University of Technology, Department of Biomedical engineering, Medical Devices Branch

Asst. Lec. Mustafa Hatem, Asst. Lec. Israa Fawzi

Aya Abduljalel Lazim
Republic of Iraq, Ministry of Higher Education and Scientific Research, University of Technology, Department of Biomedical engineering, Medical Devices Branch

Received: 2025, 15, Mar
Accepted: 2025, 21, Apr
Published: 2025, 19, May

Annotation: Nanoparticles have garnered significant attention in the field of cancer therapy due to their unique properties and potential applications. In this study, Zinc Oxide/Alginate Nanoparticles (ZnO/Alg NPs) were synthesized and characterized for their morphology, chemical composition, and cytotoxic effects on pancreatic cancer cell lines. The preparation of ZnO/Alginate involved the interaction of zinc ions with sodium Alginate, resulting in the formation of stable nanoparticles embedded within the alginate matrix. Atomic Force Microscopy (AFM) revealed the nanometer-scale morphology of the nanoparticles, with a size estimated at 58.01 nm. Fourier Transform Infrared (FTIR) spectroscopy confirmed the presence of characteristic functional groups associated with ZnO nanocomposite, including Zn-O bond stretching and –OH vibrations. Field Emission Scanning Electron Microscopy (FESEM) showed spherical-shaped nanoparticles with diameters ranging from 30 to 40 nm, indicating potential reactivity due to their high surface area-to-volume ratio. Cytotoxicity evaluation of ZnO/Alginate Nanoparticles on pancreatic cancer cell lines demonstrated a

significant inhibitory effect at higher concentrations (200, 400, and 800 µg/ml), with an IC50 value of 200 µg/ml. Lower concentrations showed minimal impact on cell viability, while the highest dose exhibited a pronounced decrease in cell proliferation.

Overall, this study highlights the potential of ZnO/Alginate Nanoparticles as promising candidates for cancer therapy, offering insights into their synthesis, characterization, and cytotoxicity profile against pancreatic cancer cells. Further investigations are warranted to explore their therapeutic efficacy and biocompatibility in preclinical and clinical settings.

Chapter One

Introduction & Literature review

1. Introduction & Literature review

1.1 Introduction

The integration of nanotechnology into medical therapeutics has revolutionized the approach to cancer treatment. Zinc oxide (ZnO) nanoparticles, in particular, have emerged as a promising tool in the nanomedicine landscape due to their unique physicochemical properties. When conjugated with alginate, a naturally derived biopolymer, ZnO nanoparticles exhibit enhanced biocompatibility and targeted drug delivery capabilities, making them a potent agent against pancreatic cancer, one of the most challenging malignancies to treat (Patra *et al*., 2018).Nanoparticles, by virtue of their size, offer a high surface area for drug loading and the ability to interact with biological membranes, facilitating targeted delivery to the tumor site while minimizing systemic toxicity (Barui *et al*., 2020). The biocompatibility of alginate further augments this delivery system, providing a safe passage for the nanoparticles through the body's immune defenses (Spoială *et al*., 2022). Recent advancements have seen the development of ZnO-loaded Alginate nanoparticles that can be engineered for controlled drug release, responding to specific stimuli within the tumor microenvironment. This targeted approach ensures a higher concentration of the therapeutic agent at the site of the tumor, thereby enhancing the efficacy of the treatment (Barui *et al*., 2020).

Aim of the study

The primary aim of this study is to investigate the therapeutic effect of zinc oxide (ZnO) nanoparticles loaded into alginate matrices for the treatment of pancreatic cancer following procedures:

1. Preparation of Zinc oxide nanoparticles using laser ablation method.
2. Preparation of nanocomposite of Zinc Oxide/Alginate.
3. Characterization of synthesized nanoparticles by using AFM, FTIR and FESEM.
4. Testing the cytotoxic effect of nanoparticles on the cell line (Pancreatic cancer).

1.2 Literature Review

1.2.1 Nanotechnology

In the realm of nanoscale technology, the creation of various materials has been attributed to nanotechnology. Notably, nanoparticles (NPs), a broad category encompassing particulate substances, are distinguished by possessing a minimal diameter of 100 nanometers. (Laurent *et al*., 2010).

These materials may assume forms ranging from 0D to 3D, the overall shape serves as a contingency factor for them. (Tiwari *et al*., 2012). Nanotechnology focuses on manipulating individual atoms, molecules, or compounds, facilitating the transformation of large structures into minuscule ones, specifically in the size range of 0.1-100 nm (Patra *et al*., 2018). polymeric nanoparticles have been recognized for their potential in the sustained release of bioactive agents. Their small size significantly enhances their efficiency in targeted drug delivery due to increased surface area (Anwer *et al*., 2016).

A notable shift in properties, attributed to the augmented surface area and the predominance of quantum effects, is observed at diminutive sizes, leading to a substantial surface area-to-volume ratio. These changes position nanotechnology and nanoparticulate carriers as promising tools in various biomedical applications, including disease prevention, diagnosis, and controlled drug delivery (Biswajit *et al*., 2014). Nanoparticles, complex entities, they are comprised of three distinct layers: (a) a modifiable surface layer containing various small molecules, metal ions, surfactants, and polymers, (b) a chemically unique shell layer, and (c) a core which represents the central portion of the nanoparticle. (Shin *et al*., 2016).

The extension of the utility of nanoparticles is observed in the realm of drug delivery. (Lee *et al*., 2011), Biological and chemical sensing is facilitated by their utilization. (Barrak *et al*., 2016), CO2 capture (Ganesh *et al*., 2017), gas sensing (Ullah *et al*., 2017), among other applications (Shaalan *et al*., 2016).

Nanomaterial manufacturing employs two primary approaches: top-down and bottom-up. The former involves reducing bulk material into smaller entities through mechanical or chemical energy. At the same time, the latter starts with atomic or molecular species, facilitating growth in particle size via chemical reactions (Boverhof *et al*., 2015). Nanoscience and nanotechnology are poised for significant advancements, with anticipated applications across various fields, including agriculture, electronics, energy, and medicine (Mansoori *et al*., 2017).

1.2.2 Nanobiotechnology

The ongoing revolution in nanotechnology, characterized by its provision of advantageous tools, has enabled the generation of individual molecules at the nanoscale for various applications. Nanobiotechnology, A fusion of nanotechnology and biology, characterized by a focus on scientific applications involving nanosized devices, is observed., Novel biological applications and innovative medical treatments are encompassed by them. (Garalleh, 2013).

A broad and diverse range of technologies is encompassed within this multidisciplinary field., and this amalgamation of different fields has given rise to new materials and techniques for their fabrication (Shoseyov & Levy, 2008). Consequently, the integration of nanotechnology and biology, termed "nanobiotechnology," is recognized for its critical role in the advancement and application of numerous tools in life sciences (Fakruddin *et al*., 2012).

Within this multidisciplinary field, a broad and diverse range of technologies are included. The properties and characteristics of nanoscale individual molecules are the focus of concern in nanobiotechnology. The applications involving the interaction of biological nanostructures are included, and the interfacing between biological entities is also addressed in this field, In this field, the focus is also placed on the interfacing among biological, chemical, and physical systems. (Garalleh, 2013).

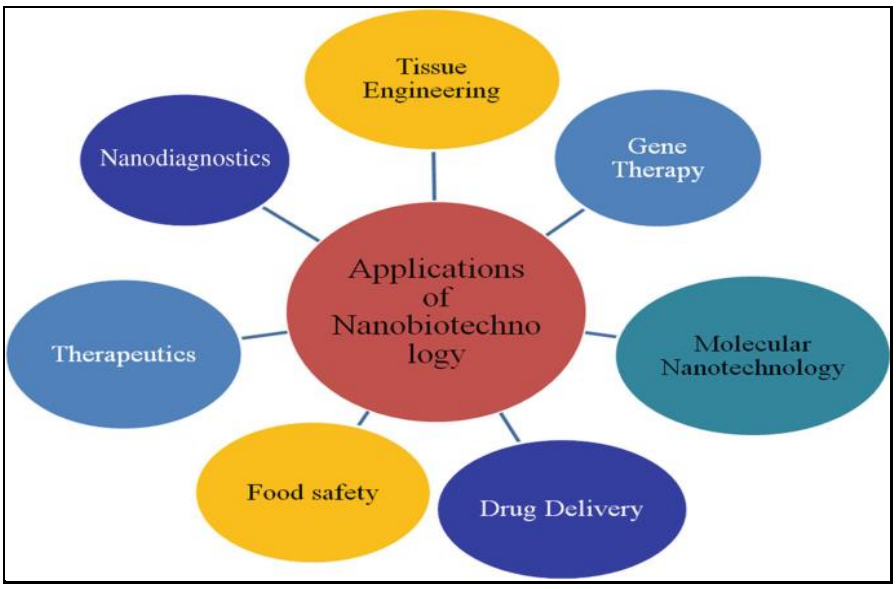


Figure (1.1): Applications of Nanobiotechnology

1.2.3 Advantages of Nanobiotechnology

The enhancement of drug delivery and transport through the use of nanoparticles is recognized as a pivotal advantage in the field of nanobiotechnology. (Andrieux *et al*., 2005). It has been observed that various Nano-products tend to accumulate at higher concentrations as compared to conventional drugs. (Vasir *et al*., 2005). Additionally, it is noted that Nano-system have the capability to selectively localize in inflamed tissues. (Allen & Cullis, 2004).

Enhanced vascular permeability and impaired lymphatic drainage in tumors are factors that facilitate the improved transmission and retention of Nano-systems in tumors or inflamed tissues. (Maeda *et al*., 2000). Furthermore, it is observed that the modification of cell and tissue distribution, achieved through the loading of drugs onto nanoparticles, results in more selective delivery of bioactive compounds. This process enhances drug efficacy and reduces toxicity. (Feng *et al*., 2004; Villarroya *et al*., 2004).

The utilization of nanotechnology in medication delivery systems has been noted to introduce new possibilities for sustained and targeted release of drugs. (Dinauer *et al*., 2005). It has been found that specially designed nanoparticles are capable of reaching less accessible body sites, evading phagocytosis, and penetrating tiny capillaries. Controlled drug release from Nano-formulations has been observed to maintain more constant drug levels in the bloodstream over extended periods. The encapsulation of the active ingredient in a polymer matrix facilitates the navigation of the drug through the restrictive cavities of the matrix., Potentially, this leads to a reduction in the dosage and frequency of administration. (Das *et al*., 2005; Patil *et al*., 2011).

1.2.4 Nanoparticles

Nanoparticles, which are characterized as microscopic matter particles, are typically measured within the nanoscale range of 1–100 nanometers. (Divya *et al*., 2015). The production of nanoparticles in various sizes and shapes, including spheres, cubes, cylinders, rings, triangles, or disks of differing dimensions, has been reported. (Omrani *et al*., 2016; Husain *et al*., 2017).

By comparison, it has been estimated that the diameter of human hair is approximately 80,000-100,000 nm, which may be about ten times the diameter of the hydrogen atom. The DNA double helix, a diminutive form of cell life, measures two nanometers, while the length of Mycoplasma bacteria is 200 nanometers, and viruses are about 30-50 nanometers in size (Anajwala *et al*., 2010).

The exceptional characteristic of nanoparticles lies in their large surface area and energy, rendering them highly reactive and ideal for use as catalysts. Nanoparticles have been noted for

their unusual optical characteristics necessary to confine electrons and facilitate quantum-size production (Koçak *et al*., 2018). Their classification commonly depends on their nature, being either organic, inorganic, or carbon-based (Elias *et al*., 2017).

Carbon-based nanoparticles, mainly consisting of carbon, are categorized into fullerenes, graphene, and carbon nanotubes (Teleanu *et al*., 2018). In contrast, inorganic nanoparticles are primarily metal-based, including metals like silver, iron, gold, aluminum, copper, cobalt, cadmium, and zinc, and their metal oxide-based counterparts, such as iron oxide, titanium oxide, silicon dioxide, magnetite, zinc oxide, and cerium oxide, which possess improved properties over their metal equivalents (Higa *et al*., 2016).

The organic nanoparticles, often recognized as polymeric nanoparticles, are commonly biodegradable and non-toxic, encompassing dendrimers, ferritin nanoparticles, and hollow spheres like micelles and liposomes (Faisal *et al*., 2017; Teleanu *et al*., 2018). Different types of nanoparticles have been synthesized using various physical, chemical, biological, and hybrid methods. (Diallo *et al*., 2017; Sheoran *et al*., 2018). Despite the widespread use of chemical and physical technologies, The use of toxic chemicals in the application of nanoparticles has been significantly limited, particularly in the field of biomedical applications and clinical areas (Mafuné *et al*., (2001).

Biodegradable polymers, derived from eco-friendly materials such as alginate, chitosan, starch, dextran, and cellulose, can degrade into water, carbon dioxide, and small inorganic molecules (Han *et al*., 2018). These polymers undergo various degradation processes, including photo, thermal, mechanical, and chemical. Molecular chains within these polymers can be actively split through enzyme-catalyzed hydrolysis or passively through hydrolysis, and oxidation may also occur (Woodard *et al*., 2016; Mohammadikhah *et al*., 2016; Woodard *et al*., 2018).

It is typically observed that the type of degradation depends on the specific bonds within the polymer's backbone. (Glaser *et al*., 2019). It has been found that enzymatically degradable polymers contain hydrolytic-labile bonds, which are too stable under physiological conditions and require enzymatic catalysts for degradation. (Zhu *et al*., 2019).

In the medical field, nanoparticles are used for diagnosing and treating various diseases. The harmful effects of nanoparticles on normal cells and living organs have been observed to significantly limit their application in medical fields, including in the treatment of conditions such as cancer and autoimmune diseases. The diversity of nanoparticles and their chemical attributes, including surface area, are considered tangible factors in this context, the safety or toxicity of nanoparticles is influenced by crucial elements such as particle size, shape, dispersity, and the effects of the protein corona, as has been observed (Ajdary *et al*., 2018).

The toxicity of nanoparticles is a principal factor limiting their use in disease detection and treatment in living organisms. Choosing a suitable model for in vivo assessment of toxicity (experimental animals) and in vitro (cell lines) is paramount in accurately determining the toxicity of nanoparticles on living cells and organisms. The induced damage to cells and tissues depends on the characteristics of nanoparticles, both chemical and physical; the route of entry into the body can be through skin, inhalation, digestion, or blood flow (Cheng *et al*., 2011; Moulton *et al*., 2010).

1.2.5 Types of Nanoparticles

Nanoparticles, characterized by their diminutive size ranging from 1 to 100 nm diameter, are acknowledged for their distinct physical and chemical attributes. These include optical, magnetic, catalytic, thermodynamic, and electrochemical properties. It has been observed that nanoparticles comprise either organic polymers or inorganic elements (Sanvicens & Marco, 2008). The diversity in the structure and shape of nanoparticles is notable, encompassing both organic and inorganic types. Various forms have been identified, such as spherical, tubular, and

irregular shapes. Furthermore, a distinction is made based on whether nanoparticles exist in fused aggregates or agglomerated forms (Bouwmeester *et al*., 2009).

Concerning organic nanoparticles, examples include liposomes and carbon nanomaterials. Carbon nanotubes are particularly notable, comprising coaxially arranged graphite sheets rolled into cylindrical shapes. These nanotubes find utility in applications such as biosensors and drug carriers. In the realm of inorganic nanoparticles, types such as quantum dots, magnetic, and metallic nanoparticles are recognized. Spherical nanocrystals with an iron core are employed as agents for labelling biomolecules in bioassays and for active targeting in both in vivo and in vitro diagnostics. Additionally, it is noted that metallic nanoparticles are utilized, encompassing gold, silver, nickel, and titanium dioxide, are prepared in various geometries like nanospheres, Nano-shells, nanorods, or nanocages.

1.2.5.1 Nanocapsules

In the described structure, it is noted that the drug is encapsulated within a reservoir or cavity, which is then enveloped by a polymer membrane or coating, A core-shell configuration is formed as a result. (Letchford and Burt, 2007; Anton *et al*., 2008).The active substance, which may be in the form of a liquid, solid, or molecular dispersion, has been described, It can be contained within the cavity.(Radtchenko *et al*., 2002).

The reservoir's composition, either lipophilic or hydrophobic, is determined by the methods employed in preparation and the raw materials used. In recent years, it has been observed that drug-loaded polymeric nanocapsules have demonstrated promising applications within the realm of drug delivery systems. (Rong *et al*., 2011). Polymeric nanocapsules, a distinct category of polymeric nanoparticles, are utilized to enhance biological effects. (Frank *et al*., 2015).

High drug encapsulation efficiency, attributed to the optimal solubility of the drug in the core and a lower polymer content relative to other nanoparticulate systems like nanospheres, are advantages nanocapsule systems offer when used as carriers of active substances.(Pinto *et al*., 2006; Anton *et al*., 2008).The utilization of nanocapsules offers various advantages, such as the enhancement of drug efficiency and the reduction in toxicity and tissue irritation. (Fessi *et al*., 2010).

1.2.5.2 Nanomicelle

Colloidal particles of nanoscale dimensions, commonly recognized as Nano micelles, are typically characterized by a size range of 10 to 100 nm, characterized by their self-assembling nature, these entities possess a hydrophobic core which is enveloped by a hydrophilic shell (Trivedi and Kompella, 2010).

Hydrophobic drugs are solubilized through entrapment within the hydrophobic core of mixed micelles, which is complemented by a corona of hydrophilic chains extending outward. (Velagaleti *et al*., 2010). Nanomicelles are currently being employed successfully as carriers for water-insoluble drugs, Various appealing characteristics have been exhibited by nanomicelles, including biocompatibility, longevity, and high stability both in vitro and in vivo. Additionally, it has been observed that various poorly soluble drugs can be solubilized by them, effectively altering the release profile of the incorporated drugs.

The accumulation of pharmaceutical agents in the target zone is leveraged by the enhanced permeability and retention effect. (Movassaghian *et al*., 2015). In the field of drug delivery, the applications of these polymeric micellar nanocarriers are primarily observed in areas such as anticancer therapy, treatment of neurodegenerative diseases in the brain, administration of antifungal agents, and ocular drug delivery. (Kshirsagar *et al*., 2011).

1.2.5.3 Nanotherapeutics

In the medical field, the emergence of nanotherapeutics as a vital discipline, particularly the rapidly advancing nanoparticle-based drug delivery for cancer therapy, has been noted. This

development addresses the constraints of traditional drug delivery systems, for their high stability and carrying capacity, drug carriers of nanometric scale, optimized in size and surface properties, are recognised. Additionally, it is facilitated by the capability to incorporate both hydrophilic and hydrophobic substances and the adaptability of various administration routes, leading to controlled drug release from the matrix. Thereby, drug bioavailability is enhanced (Gelperina *et al*., 2005).

1.2.6 Classification of Nanoparticles

Nanoparticles (NPs) are categorically divided into groups based on their morphology, size, and chemical properties. The classification of these nanoparticles is commonly determined by their physical and chemical characteristics, as outlined below. (Khan *et al*., 2019).

1.2.6.1 Carbon-based Nanoparticles

Two principal categories of carbon-based nanoparticles (NPs) are identified Carbon Nanotubes (CNTs) and fullerenes, with the latter being characterized by their globular hollow cages, Considerable commercial interest has been generated in such nanomaterials, including allotropic forms of carbon, particularly in the context of nanocomposites for various applications, including fillers.(Saeed and Khan, 2016).High efficiency gas adsorbents are utilized in environmental cleaning applications.(Ngoy et 2014).They are also employed as a support medium for organic and inorganic catalysts.(Mabena *et al*., 2011).

1.2.6.2 Metal Nanoparticles

The unique optoelectrical features of these nanoparticles (NPs), which originate entirely from metal, are attributed to their well-recognized characteristics of localized surface plasmon resonance. (Zada *et al*., 2020). In contemporary advanced materials, the significance is attributed to metal nanoparticles (NPs) characterized by controlled facets, size, and shape. (Dreaden *et al*., 2012).

The enhanced optical properties of metal nanoparticles (NPs) lead to their application in various research fields. Scanning electron microscope (SEM) It is expected that the sampling will employ a coating of gold nanoparticles (NPs) to augment the electronic stream, which facilitates the acquisition of high-quality Scanning Electron Microscopy (SEM) images. (Dreaden *et al*., 2012).

1.2.6.3 Ceramics Nanoparticles

Ceramic nanoparticles, characterized as nonmetallic inorganic solids, are synthesized through heating and cooling processes. The formation of these nanoparticles results in diverse structures, encompassing amorphous, polycrystalline, dense, porous, and hollow configurations. (Sigmund *et al*., 2006). Increased interest in these types of nanoparticles has been observed among scientists, attributable to their applications in photodegradation, photocatalysis, dye processes, catalysis, and imaging applications. (Thomas *et al*., 2015).

1.2.6.4 Semiconductor Nanoparticles

Properties intermediate between metals and nonmetals are exhibited by nanoparticles, and they are utilized across a broad spectrum of applications. (Ali *et al*., 2017; Khan *et al*., 2017a). Significant band gaps are possessed by semiconductor nanoparticles, and dramatic alterations in their characteristics are observed when these band gaps are subject to tuning. (Sun, 2000), It has been established that high efficiency in water-splitting applications is exhibited by several semiconductor nanoparticles, attributed to their optimal band gap and band edge placements. (Hisatomi *et al*., 2014).

zinc sulfide (ZnS) it’s a highly necessary semiconductor material, having been commonly utilized in solar energy conversion, light emitting diode, photocatalysis, emission, fuel cells and so on (Ghawade *et al*.,2022). Lead sulfide (PbS) is a significant compound that belongs to the

IV–VI group of binary semiconductors. PbS is a significant direct band gap semiconductor material that has a narrow band gap of)0.41 eV(. It is commonly used in near-IR communications. (Mamiyev et al.,2015).

AgS (Silver Sulfide) semiconductor nanoparticles have been studied for their structural, optical, and electronic properties. These nanoparticles exhibit a direct band gap of approximately 0.96 eV (Zamiri et al.,2015).

1.2.6.5 Polymeric Nanoparticles

Nanoparticles are particles that fall within the size range of 1 to 1000 nm. They have the ability to carry active chemicals, which can either be confined within the polymeric core or attached to their surface through adsorption. The term "nanoparticle" encompasses both nanocapsules and nanospheres, which are differentiated based on their morphological structure. Polymeric nanoparticles have demonstrated significant promise in the precise administration of medications for the therapeutic intervention of many ailments. (Zielińska *et al*.,2020).

Nanoparticles primarily composed of organic materials are called polymer nanoparticles (PNPs). The most prevalent forms of these nanoparticles are nanospheres and nanocapsules. (Mansha *et al*., 2017). (Abouelmagd *et al*.,2016). Polymeric nanoparticles (NPs) provide several advantages as drug carriers. These include their potential for controlled release of drugs, their capacity to protect drugs and other biologically active molecules from the environment, and their ability to enhance bioavailability and therapeutic index. (Zielińska *et al*.,2020).

1.2.6.6 Lipid-based Nanoparticles

In various biomedical applications, the utilization of nanoparticles possessing lipid moieties is observed, with lipid nanoparticles, it is typically observed that they exhibit a spherical shape and a diameter ranging from 10 to 1000 nm. Like polymeric nanoparticles, it is observed that lipid nanoparticles consist of a solid lipid core and a matrix including soluble lipophilic compounds. (Rawat *et al*., 2011).

The external core of these nanoparticles is stabilized by surfactants or emulsifiers. Lipid nanotechnology (Mashaghi *et al*., 2013) is identified as a subfield of nanotechnology that is dedicated to the design and production of lipid nanoparticles, which are employed in various applications, including serving as carriers and facilitating drug delivery. (Puri *et al*., 2009).

1.2.7 Synthesis of Nanomaterials

The fabrication of nanomaterials, which are defined as materials with at least one dimension at the nanoscale level (1-100 nm), is noted, Moreover, the creation of nanostructured surfaces, which fall under the category of nanomaterials, is acknowledged, it is recognized that methods involved in the production of nanoparticles can be generally categorized into two groups.

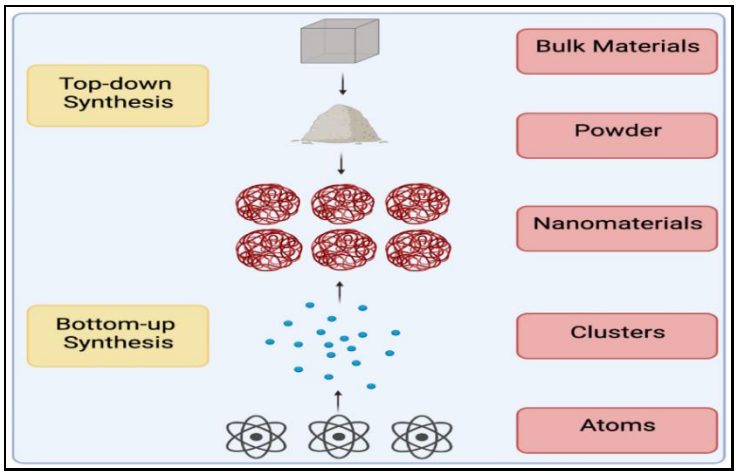


Figure (1.2): Synthesis of nanomaterials (top-down synthesis and bottom-up synthesis)

1.2.7.1 Bottom-up Method

In the bottom-up approach, it is observed that molecular components autonomously assemble into more complex structures, progressing from atom to molecule to cluster, exemplified by crystal growth. This method employs molecular self-assembly, a concept long recognized by chemists, to arrange molecular components into functional conformations. It is pivotal in synthesizing tiny nanomaterials and typically results in nanostructures with fewer defects than those created by the top-down approach. (Wang and Xia, Chapter Two Literature Review 11 2004).

1.2.7.2 Top-down Method

The top-down approach involves the creation of nanoscale devices through the guidance of more significant, externally controlled devices for their assembly. This method often utilizes conventional workshop or microfabrication techniques, wherein materials are shaped, cut, and milled into the desired form and arrangement using external tools. Characterized by a destructive process, it is characterized by the process where larger molecules are broken down into smaller units, which are then converted into suitable nanoparticles. Techniques such as grinding/milling, chemical vapor deposition (CVD), physical vapor deposition (PVD) are employed, and various decomposition processes exemplify this technology. (Iravani, 2011).

1.2.8 Application of Nanoparticles

Nanotechnology's contributions are significantly enhancing, and in some cases revolutionizing, numerous sectors, Including areas such as information technology, homeland security, medicine, transportation, energy, food safety, and the environment. The Science. Because of their distinct chemical and physical characteristics, nanoparticles are becoming increasingly vital in developing new nanodevices for a wide array of applications in physical, biological, biomedical, and medicinal fields. The optical characteristics of nanomaterials find use in the creation of optical detectors, sensors, lasers, displays, and solar cells, as well as in biomedicine and photo electrochemistry. Carbon nanotubes constitute the electrodes in microbial fuel cells. Nanocrystalline zinc selenide is utilized in display screens to enhance zinc selenide is utilized in display screens to enhance pixel microelectronics industry focuses on the miniaturization of circuits, including transistors, diodes, resistors, and capacitors. (Loureiro *et al*., 2016).

Nanoparticles (NPs) and nanostructured materials are being increasingly utilized for drug delivery to specific target zones due to their advantageous characteristics, such as optimal size and efficient drug loading and release capabilities. These materials are distinguished by their controllable shapes and high surface-to-volume ratios, rendering them exceptional candidates for drug delivery applications. The efficacy of these Nano-delivery systems is further enhanced by their ability to deliver pharmaceuticals in precise dosage ranges, often leading to increased therapeutic efficiency and improved patient compliance while minimizing side effects. Additionally, the potential of these systems to traverse the blood-brain barrier is anticipated to pave new pathways for transporting drugs into the brain. Furthermore, the conjugation of drugs with nanocarriers is known to augment the biological distribution of medications and extend their circulation time within the bloodstream. (Alexis *et al*., 2008; Majumder *et al*., 2019). Superparamagnetic iron oxide nanoparticles, when appropriately surface-engineered, have been identified as versatile in their in vivo applications.

These applications encompass MRI contrast enhancement, tissue repair, immunoassays, medication delivery, cell division, biological fluid detoxification, and extreme heat. However, fulfilling these biomedical applications necessitates nanoparticles possessing high magnetization values, diminutive sizes (less than 100 nm), and narrow widths less than 100 nm. (Laurent *et al*., 2010).

Semiconductor and metallic nanoparticles, predominantly gold and silver, are widely recognized for their potential in cancer detection and treatment. Gold nanoparticles, in particular, their

ability to effectively convert absorbed light into localized heat has been identified, A property that can be harnessed for targeted laser photothermal therapy in cancer treatments is recognized. Beyond these uses, nanoparticles have also demonstrated significant antineoplastic effects, efficiently inhibiting tumor growth with minimal toxicities. For instance, multi-hydroxylated nanoparticles have shown high efficiency in their antineoplastic actions. Additionally, the antibacterial properties of silver nanoparticles have led to their increasing incorporation in various applications, including wound dressings, catheters, and numerous household products. (AshaRani *et al*., 2009).

1.2.9 Toxicity of Nanoparticles

The increasing utilization of nanoparticles (NPs) and nanostructured materials in drug delivery to targeted areas is attributable to their beneficial properties, including optimal sizing and effective drug loading and release functions. The notable features of these materials, such as their controllable shapes and substantial surface-to-volume ratios, position them as outstanding choices for drug delivery. The effectiveness of these Nano-delivery systems is enhanced owing to their capability to administer pharmaceuticals in Accurate dosage ranges often result in heightened therapeutic effectiveness, better patient adherence, and reduced adverse effects. The ability of these systems to cross the blood-brain barrier is also projected to create novel opportunities for drug transportation into the brain. Moreover, the integration of drugs with nanocarriers is recognized for biologically improving medication distribution and prolonging their presence in the bloodstream (Brohi *et al*., 2017).

When surface-engineered appropriately, superparamagnetic iron oxide nanoparticles are recognized for their versatility in various in vivo applications. These diverse applications include but are not limited to, enhancing MRI contrast, repairing tissue, conducting immunoassays, detoxifying biological fluids, inducing hyperthermia, delivering drugs, and separating cells. To effectively fulfil these biomedical applications, it is necessary for the nanoparticles to exhibit high magnetization values, maintain small dimensions (less than 100 nm), and have narrow widths (also under 100 nm).In the cancer detection and treatment field, semiconductor and metallic nanoparticles, especially those composed of gold and silver, have gained wide acknowledgment for their potential. Gold nanoparticles are particularly noted for their efficiency in transforming absorbed light into localized heat, a characteristic that can be utilized in targeted laser photothermal therapy for cancer. Moreover, nanoparticles have been found to exhibit significant antineoplastic effects, effectively inhibiting tumor growth while maintaining low toxicity levels. For example, multi-hydroxylated nanoparticles have been efficient in antineoplastic activity. In addition, the antibacterial qualities of silver nanoparticles have prompted their increased use in various contexts, such as wound dressings, catheters, and an array of household items. (Vance *et al*., 2015) worries about the possible negative impacts of nanoparticles Extensive research into the toxicity of nanoparticles (NPs) has been conducted using mice and rats, species that share genetic similarities with humans. However, the use of these mammalian models is limited due to the duration of their developmental cycle and the ethical concerns associated with their use. (Zieliska *et al*., 2020).

In the treatment of human diseases, various methodologies are available for the administration of nanoparticles, with oral administration being one of them, Transdermal distribution, intravenous injection, and surgical implantation are encompassed as methods within the spectrum of available approaches in an academic context. (Wennerberg *et al*., 2011). Exposure to nanoparticles (NPs) that is unintentional in nature. can occur through inhalation, cutaneous contact, or ingestion. Once entering the human circulation, these nanoparticles are subject to metabolic processes and elimination or retention in various bodily compartments. The characteristics of nanoparticles' size, shape, and surface, which vary due to their distinct physicochemical characteristics, profoundly affect their distribution, metabolism, excretion, and absorption in the human body and within cells, all determined by their specific physicochemical

properties.(Oberdörster *et al*., 2005).The metabolism, elimination, distribution, and absorption mechanisms are collectively known as the ADME Processes. (Borel & Sabliov, 2014).

1.2.10 Zinc Oxide Nanoparticles

Zinc oxide nanoparticles, referred to as zinc oxide nanoparticles, are recognized when their diameters are below 100 nanometers in an academic context. A substantial surface area in relation to their size is exhibited by them, accompanied by pronounced catalytic activity in an academic context. The synthesis methods used determine the precise physical and chemical properties of zinc oxide nanoparticles. Academic literature has different methods for producing ZnO nanoparticles, such as laser ablation. Academic researchers use hydrothermal processes, electrochemical depositions, and the sol-gel method to synthesize zinc oxide nanoparticles. Chemical vapor anodization, co-precipitation, electrophoretic deposition, deposition, thermal breakdown, combustion techniques, ultrasound, and the microwave-assisted combustion method are encompassed as methods utilized in academic research for the synthesis of zinc oxide nanoparticles, Two-step mechanochemical–thermal synthesis and precipitation processes that rely on solution concentration are included as methods employed in academic studies for the synthesis of zinc oxide nanoparticles, At room temperature, ZnO exhibits a 3.37 eV energy gap, and its characteristics are influenced by factors such as pH and the washing medium. It is regarded as a broad-band gap semiconductor in academic contexts. (kumar *et al*., 2013).

ZnO nanoparticles are widely recognized as one of the three most often manufactured nanomaterials in academic discussions, along with titanium dioxide and silicon dioxide nanoparticles. (Zhang *et al*.,2015). The academic community has worries about the possible hazards associated with ZnO nanoparticles, as it is a relatively novel material. Their minute size allows for their traversal of various bodily barriers, as demonstrated in animal studies, including the placenta, blood-brain barrier, individual cells, and even cell nuclei, as observed in academic research. Detection becomes challenging due to the facilitation of tissue absorption by their small size. However, when it comes to applications like sunscreen, it has been observed academically that human skin successfully blocks ZnO nanoparticles. With the exception of skin abrasions, accidental ingestion of small quantities of ZnO nanoparticles may occur, as observed in academic contexts, In certain instances, such as during the application of sunscreen, a potential route of entry into the body is presented. The possible release of ZnO nanoparticles into runoff water, which can enter the food chain, is a concern when sunscreen is washed off. Until 2011, there were no documented cases in academic literature of human ailments caused by exposure to manufactured nanoparticles. (Kessler *et al*., 2011).

1.2.11 Application of Zinc Oxide Nanoparticles

The synthesis of metal oxide semiconducting nanoparticles is necessitated by the sensitivity of biomolecules to solution pH and temperature. This synthesis is undertaken for their potential application in biological sensing, labelling, drug and gene delivery, and nanomedicines within the academic context. In various biological applications, ZnO nanoparticles are regarded as a preferred choice due to their ease of fabrication, environmentally friendly attributes, and synthesis routes that are non-toxic, as acknowledged within academic discourse. Nevertheless, the essential prerequisites for the application of ZnO nanoparticles in biological contexts include their water solubility and biocompatibility, as acknowledged in academic discussions. (Griffin *et al*., 2006).

Nanomedicine, a rapidly evolving branch of nanotechnology, is changing the environment for illness diagnosis and treatment At both the cellular and molecular levels, significance has been attributed to metal oxide nanoparticles in biology and medicine over recent decades, owing to their distinct physicochemical properties, as noted in academic literature. Particularly, zinc oxide nanoparticles (ZnO-NPs) have attracted considerable interest from researchers for therapeutic and diagnostic applications in academic circles. Their appeal stems from their reduced toxicity, biodegradable characteristics, and affordability. In various biomedical applications, these ZnO-

NPs find employment, encompassing the delivery of biomolecules (such as drugs and genes), cancer therapy, angiogenic therapy, antibacterial applications, tissue engineering, bioimaging, biosensing, and more, as observed within the academic discourse. (Jiang *et al*., 2018).

1.2.12 Polymers

Polymers, comprising both natural and synthetic substances, are comprised of giant macromolecules formed from multiples of simpler chemical units known as monomers, as described within academic discourse. These substances constitute an integral component of the materials present in living organisms, including proteins, cellulose, and nucleic acids, as recognized in academic contexts. Furthermore, polymers play a fundamental role in the composition of various minerals, in addition to their presence in artificial materials such as concrete, as acknowledged within academic discussions, Glass, paper, plastics, minerals like feldspar and quartz, as well as rubbers, are comprised of polymers. The term 'polymer' denotes an unspecified quantity of monomer units within the academic context. Compounds with an exceptionally high number of monomers are frequently referred to as high polymers within the academic discourse. The diversity of polymers is noteworthy, as they are not constrained to monomers of identical chemical composition or molecular weight and structure. Although a single type of monomer constitutes some natural polymers, the majority of both natural and synthetic polymers are formed from two or more distinct monomers classified as copolymers within academic discussions (Britannica, 2023).

1.2.13 Alginate Polymer

Alginate, a polysaccharide, is found naturally in the cell walls of brown algae (also known as brown seaweeds) and in the bacterial capsules of Azotobacter sp. and Pseudomonas sp., as recorded in academic literature. Academic study has proven that alginate, found in brown algae, greatly improves the structural integrity and flexibility of cell walls. This imparts a protective buffer to the algae against potential damage caused by the force of solid sea waves, as elucidated in academic discourse. (Venkatesan,2014). In bacteria, a protective capsule is formed by alginate, which also assists in the formation of biofilms. (ID. Hay,2013) Alginate assists in bacterial adherence and colonization. (H.W. Liu,2010).

Alginate is highly recognized in academic circles for its capacity to create a hydrogel, making it a versatile stabilizer, thickener, gelling agent, and emulsifier. The extraction of alginate from brown algae encompasses several processes, including the initial treatment with mineral acid to Transform the salts of alginic acid (Alginates) found in the algae into unbound alginic acid, although biocompatibility is a requirement for all biomaterials to prevent post-acute or chronic adverse effects, as emphasized in academic discourse. The use of biomaterials that can undergo degradation will obviate the necessity for a subsequent surgical procedure to remove the material once the tissues have healed and regenerated, as highlighted in academic contexts. Another crucial factor is the capacity to regulate the degradation rate of biomaterials, as emphasized in academic discussions. The criterion for ensuring proper tissue healing and regeneration involves the requirement that the degraded biomaterials must not be toxic and should be readily eliminated from the body.

Alginate, a biological substance, is employed in the healing and regeneration of human tissue. Particularly for wound dressing purposes, alginate, which is employed in food preparation for human consumption, is regarded as safe for biomedical applications. Alginate is additionally characterized by its biodegradability, as it undergoes slow dissolution within the body due to the release and exchange reaction of cross-linking agents in the alginate with monovalent cations present in bodily fluids. The extraction of alginate from brown algae encompasses several processes, including the initial treatment with mineral acid to convert the salts of alginic acid (Alginates) in the algae into free alginic acid, Although biocompatibility is a requirement for all biomaterials to prevent post-acute or chronic adverse effects .The use of biomaterials that can undergo degradation will obviate the necessity for a subsequent surgical procedure to remove the

material once the tissues have healed and regenerated. Another crucial factor is the capacity to regulate the degradation rate of biomaterials. The criterion for ensuring proper tissue healing and regeneration involves the requirement that the degraded biomaterials must not be toxic and should be readily eliminated from the body. Alginate, a biological substance, is employed in the healing and regeneration of human tissue. Particularly for wound dressing purposes, alginate, which is employed in food preparation for human consumption, is regarded as safe for biomedical applications. Alginate is additionally characterized by its biodegradability, as it undergoes slow dissolution within the body due to the release and exchange reaction of cross-linking agents in the alginate with monovalent cations present in bodily fluids.

1.2.14 Characterization of Nanoparticles

Nanoparticles are small particles with dimensions ranging from 1 to 100 nm, displaying various physical and chemical characteristics. A comprehension of nanoparticles would unveil remarkable characteristics and potential uses that would assist in the expansion of thin film technology. The choice of synthesis methods, such as top-down, bottom-up, chemical, biological, and mechanical processes, significantly affects the properties displayed by nanomaterials. Nanoparticles are also analyzed to determine their morphological, structural, optical, elemental, size, and physiochemical properties. Nanoparticles are minute particles with sizes ranging from 1 to 100 nm, and they possess various physical and chemical characteristics. An in-depth knowledge of nanoparticles would unveil their remarkable properties and possible uses, which would contribute to the expansion of thin film technology. The choice of synthesis methods, such as top-down, bottom-up, chemical, biological, and mechanical processes, significantly affects the properties displayed by nanomaterials. Nanoparticles are also assessed to determine their morphological, structural, optical, elemental, size, and physiochemical properties (Rawle A. F et al., 2017).

1.2.14.1 Atomic Force Microscopy (AFM)

Also recognized as scanning force microscopy, is widely known in the scientific community. A method recognized for its capacity to generate surface images via a scanning probe, Atomic Force Microscopy (AFM), is particularly well-suited for the quantitative assessment of surface roughness at the nanometer scale. It excels in the visualization of nanotexture on a variety of material surfaces, including polymer nanocomposites. The analysis of mechanical characteristics at the Nano level for individual particles and molecules is facilitated by AFM, leading to enhanced considerations of physiological aspects. (Vozza *et al*., 2018)

1.2.14.2 Fourier Transform Infrared (FTIR) Spectroscopy

FTIR spectroscopy, recognized as an efficient method for chemical identification, depends on interactions among functional groups or their existence. This technique can measure inorganic and organic compounds in solid, liquid, and gaseous forms within an unknown mixture. The infrared study involves the vibrational motion of atoms or molecules. The wide usage of FTIR spectroscopy in studying the nature of surface adsorbents in nanoparticles is attributed to the large surface area of these particles. Modification of the nanoparticle surface with suitable adsorbates can result in varied properties. The principle of FTIR lies in the absorption of light by some molecules in the infrared field of the electromagnetic spectrum. It measures the frequency in the 4000–400 cm-1 range as wave numbers and records data as interference patterns, subsequently converting this data into a spectrum. (Choudhary *et al*. 2017).

1.2.14.3 Filed Emission Scanning Electron Microscopy (FESEM)

The field emission scanning electron microscope (FE-SEM) is an electron microscope that uses a high-energy beam of electrons to scan the surface of a sample in a raster scan pattern in order to create a picture. The field emission gun was utilized to generate electron emitters. These electron emitters have the capability to generate emissions that are up to 1000 times greater than those

produced by a tungsten filament. Nevertheless, they need significantly more stringent vacuum conditions.

Once the electrons beam emerges from the electron gun, they are further restricted and concentrated into a narrow, uniform beam by means of metal apertures and magnetic lenses. Ultimately, electron microscopes are equipped with detectors specific to each sort of electrons, which capture signals in order to generate an image of the material. (Lemine O. M. *et al.*, 2012)

1.2.15 Pancreatic Cancer

One of the poorest prognoses among all cancers is characterized by pancreatic cancer, as evidenced by a mere 11% five-year survival rate. A significant factor contributing to this dire prognosis is the frequent diagnosis of pancreatic cancers (90%) at an advanced disease stage, where the tumor becomes inoperable, primarily due to delayed clinical manifestation and systemic metastases in over 50% of patients. Despite its relatively low overall incidence rate (5.7 per 100,000 person-years), the latest data from the Global Burden of Diseases highlights the severity of this cancer type. The Injuries and Risk Factors Study observed a more than twofold increase globally in the total number of fatalities and incident cases are to be considered in this context. From 1990 to 2017, an adjustment in years of life for handicaps due to pancreatic cancer has been observed, along with the total number of fatalities and incident cases.

In recent years, improvements have been made to traditional chemotherapy and radiotherapy for pancreatic cancer. The development of both first-line and second-line palliative treatments has marked the improvements made in recent years to traditional chemotherapy and radiotherapy for pancreatic cancer. Additionally, adjuvant treatments have been employed in clinical trials to enhance their efficacy in alleviating symptoms and improving disease outcomes. However, Given the low five-year survival rate, investigation into new treatment methods, such as targeted therapy and immunotherapy, is needed. The direct costs of pancreatic cancer are observed to be higher compared to other cancer types, influenced by factors such as tumor type and cancer stage. In contrast, substantial indirect costs are primarily attributed to high mortality rates. These high costs place a significant economic burden on society and adversely affect patients' health-related quality of life (HRQOL).

In response to the impact of cancer on global health, public health policies are aimed at enhancing global survival through three key pillars. Health promotion, timely diagnosis, and comprehensive treatment and supportive care are the three key pillars through which public health policies aim to enhance global survival. Consequently, an assessment of the global public's... (please continue the sentence for further rephrasing). Interest in this disease is deemed essential, and it has been observed that patients who have access to cancer information tend to engage more actively in disease management. Access to this information prepares patients for treatment, aids in coping with adverse effects, and diminishes anxiety and depression. Treatment satisfaction is heightened, communication with family is enhanced, and an improved quality of life is contributed to by this access. Over recent decades, technological advancements, such as patient-centered care coordination through mobile applications, have been developed. Cancer illness awareness among the population has been enhanced by technological advancements such as patient-centered care coordination through mobile applications.

The Internet has become a resource for patients seeking information on disorders. Medications, and treatments have become more accessible through the Internet. Consequently, this type of search traffic data could serve as a crucial source for analyzing health-related trends and gauging public interest in various health topics. Although still emerging, the potential of "infodemiology" for extensive data analysis is promising, as evidenced by its applications in infectious diseases and Internet-based surveillance systems. (Gianfredi *et al*., 2023).

Chapter Two

Materials and Methods

2. Materials and Methods

2.1 Materials

2.1.1 Apparatus and Equipment

Apparatus and equipment utilized in this study are listed in Table (2.1)

Table (2.1) : Apparatus and Equipment utilized in This Study.

Apparatus and equipment	Company /origin
1064nm AN DYG LASER	Homemade (UOT)
Magnetic stirrer device	labTech/ China
Magnetic stirrer bar	China
Beaker 250 ml	simax/ czech
Laboratory balance	OHAUS/Malaysia
Filtration funnel	---------
Grinder	---------

2.1.2 Chemicals and Biological Materials

Chemical and Biological Materials utilized in this study are listed in Table (2.2).

Table (2.2): Chemical and Biological Materials utilized in This Study.

Chemicals and Biological Materials	Company /origin
Sodium Hydroxide flakes 97% (NaOH)	Loba chemie/ India
Sodium Alginate	Loba chemie/ India
Zinc target 99.99%	British Drug House (BDH) / England
Calcium Chloride Dihydrate ($CaCl_2.2H_2O$)	AVONCHEM/ UK
Demineralized Water (D.W)	Pioneer
Aloe Vera plant	----------

2.2 Methods

2.2.1 Preparation of Zinc Oxide Nanoparticles Using Laser Ablation Method

Laser ablation is done through the steps below: -

1. A sample of the zinc target was taken, its purity (99.99%), size (1*1 mm), and it was placed in a beaker of size (250 ml).
2. 5 gram of sodium hydroxide was diluted in (100ml) of deionized water inside a closed container, then mixed well and waited for a few minutes until the reaction took place, and this is known by little increase the temperature of the container.
3. After the reaction is complete, the mixture of sodium hydroxide (NaOH) and deionized water is added to the target sample, and then, the sample is ready for laser ablation.
4. The laser ablation method is carried out by ablate the sample with a wavelength 1065 nm and frequency 8HZ using three different laser powers in sequence (280mj, 360mj, 460mj), and with a number of pulses (347,500,1000) in sequence. As the laser power and the number of pulses increase, the corrosion of the target sample increases and occurs Slight turbidity occurs in the medium in which the sample is immersed, indicating the formation of nanoparticles.

This method was carried out at the Nanoscale Research Center at the University of Technology in Baghdad, Iraq

Figure (2.1): AN DYG LASER that was used has a wavelength of 1064nm and a frequency of 8Hz.

Figure (2.2): Zinc target sample into baker (250ml).

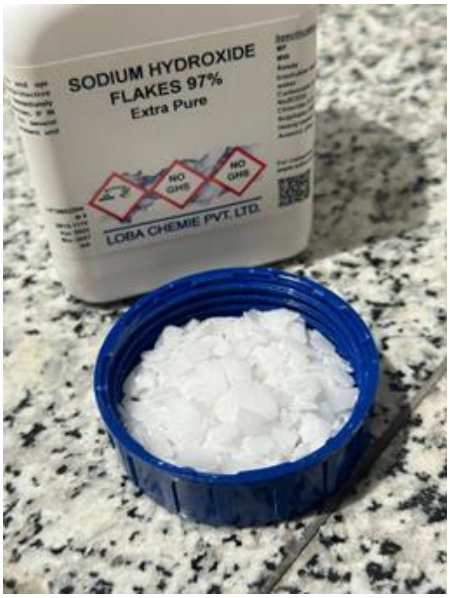


Figure (2.3): Pure sodium hydroxide.

Figure (2.4): The addition of sodium hydroxide solution to the zinc target To make it ready for laser ablation prosess.

Figure (2.5): Laser targeting of the sample with different powers and pulses, nanoparticles are then formed.

2.2.2 Bio synthesis of Zinc oxide nanoparticles

1. Aqueous plant extracts of aloe vera leaves were prepared in the traditional way by washing fresh leaves with well running water to remove surface contaminants, soaking them for 30 minutes after removing moisture and drying them at room temperature.
2. 10 g of dried plant leaves were placed in a 250 mL glass beaker containing 160 mL of deionized water and then heated in a hot plate at 90 °C for approximately 1 hour.
3. The sample solution was then filtered using filter papers and thus the sample extract was obtained.
4. Then we prepare zinc oxide particles from aloe vera extract in the traditional way by heating 30 ml of the plant extract using a hot plate at a temperature of 40 - 45 degrees Celsius.
5. At 45°C, add 5 g of zinc acetate to the solution and continue heating until the viscosity of the sample increases. Thus, the sample is ready for examination.

This method was carried out in the laboratories of the Biomedical Engineering Department at the University of Technology in Baghdad, Iraq

Figure (2.6): 10g of Aloe Vera powder into 250ml glass baker.

Figure (2.7): The picture shows heating the aloe vera solution at a temperature of 90 degrees

Figure (2.8): Filter the solution using filter papers.

Figure (2.9): Zinc acetate

Due to the similarity of the method and preparation using the plant, reliance was placed on the laser and its connection with alginate. During any production process, by-products are generated, which are mainly harmful, and if not appropriately utilized, they cause environmental pollution. In addition, wastewater generated from nanoparticle synthesis can contain harmful chemicals. The low yield is another disadvantage: only a small percentage of the starting materials is converted into nanoparticles, generating raw material waste.

2.2.3 Preparation of Zinc Oxide/Alginate

Preparation of nanocomposite accordingly with modified to (Abdul Bakil *et al*., 2020)

1. Firstly, we dissolved 1g of Sodium Alginate into 100 ml of distilled water, the solution was stirred for 30 min at room temperature until completely dissolved and became thick liquid.
2. Then, we added 10 ml of Zinc oxide that prepared by laser ablation to the sodium alginate solution.
3. After that we dissolved 0.215g of Calcium chloride dihydrate **($CaCl_2.2H_2O$)** into 10 ml of distilled water
4. When the Calcium chloride dihydrate (**$CaCl_2.2H_2O$**) solution was ready, it was gradually added to the Zinc oxide/Alginate solution to prevent the solution from clumping, and it was stirred at room temperature for an hour in order to produce Zinc oxide/Alginate solution.

This method was carried out in the laboratories of the Biomedical Engineering Department at the University of Technology in Baghdad, Iraq

Figure (2.10): Addition of Sodium Alginate into distilled water.

Figure (2.11): Sodium alginate powder.

Figure (2.12) The addition of prepared zinc oxide to the sodium alginate solution

2.3.1 Cytotoxicity of Cell Line

After the cells dispensed in the monolayer were, the following steps were done:

1. The cells were washed with a solution of 2 mg/ml PBS after removing the growth media.
2. A volume of 2 to 3 milliliters of Trypsin/Versene solution was added to the cell. The cells were inverted to ensure thorough coverage of the monolayer with mild agitation. The cells were incubated at a temperature of 37 °C for a duration of 1 to 2 minutes, during which time the cells became detached from the vessel.
3. A fresh, full volume of RPMI medium (15-20 ml) was added, and the cells were detached from the surface using a pipette and transferred into a growth medium.
4. The cells were rearranged to get the desired concentration and placed in culture vessels, flasks, or plates as necessary. They were then kept at a temperature of 37 °C in an incubator with 5% CO2.
5. Cell concentration was determined by enumerating the cells using a hemocytometer and applying the following formula:
6. Total Cell Count/ml: cell count × dilution factor (or sample volume) ×104

7. Assay (Freshney, 2010)

- Tumor cells (1x104 cells/ml) were cultivated in 96 flat well micro-titer plates, with a final amount of 200 µL of complete culture media per well. The microplate was sealed with sterile parafilm and agitated gently.
- Plates were incubated at 37 °C, 5 % CO2 for 24 hrs.
- The medium was taken out, and the necessary concentrations of synthetic NPs (50, 100, 200, 400, and 800 µg/mL) were introduced to the wells after being diluted twice.
- Three replicates were utilized for each concentration, as well as for the controls (cells treated with media lacking serum). The plates were placed in an incubator set at a temperature of 37 °C and with a carbon dioxide concentration of 5% for a specific duration of 24 hours.
- Each well received 10 µl of the MTT solution. Subsequently, the plates were incubated at a temperature of 37°C and a carbon dioxide concentration of 5% for a duration of 4 hours.
- Following incubation, the media were carefully extracted, and 100 µL of solubilization solution was introduced into each well for a duration of 5 minutes.
- Utilizing an ELISA reader to measure at a wavelength of 575 nm. The optical density data was analyzed statistically to assess the concentration of chemicals.

2.3.2 Statistical Analysis

SPSS for Windows version 22 was used to analyze data (SPSS Inc. Chicago, Illinois, United States). Data have been represented in proportions and frequencies. (Forthofer and Lee, Glover and Mitchell, 2008).

Chapter Three

Result and Discussion

3. Results and Discussion

3.1 Characterization of Zinc Oxide/Alginate Nanoparticles (ZnO/Alg NPs)

3.1.1 Atomic force Microscope (AFM)

The surface morphology and topography were assessed using an Atomic Force Microscope (AFM). Furthermore, it was selected as an imaging technique that offers nanometer-level precision and the ability to capture three-dimensional surface images with minimal sample preparation. It also permits imaging in both ambient and liquid environments. The Atomic Force Microscope (AFM) provides a high-resolution, two and three-dimensional representation of the surface of nanoparticles at the atomic scale, as depicted in Figure (3.3).

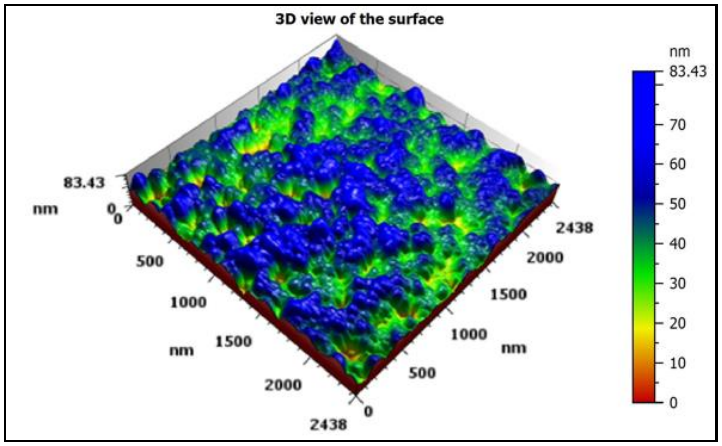


Figure (3.1): Atomic force microscope for ZnO/Alg NPs

The size of ZnO/Alginate Nanoparticles was estimated by using AFM as shown in table (3.1). The result showed that ZnO/Alginate size was 58.01 nm.

Individual results				
Parameters	**Projected Area**	Area	Mean diameter	Z-maxim...
Unit		nm²	nm	nm
Particle #1	Small	8041	96.90	514.5
Particle #2	Small	3986	65.90	496.6
Particle #3	Large	72581	239.8	601.0
Particle #4	Small	9159	108.0	500.7
Particle #5	Small	91.37	5.439	458.6
Particle #6	Small	2334	48.34	518.5
Particle #7	Small	4716	64.82	495.0
Particle #8	Medium	20499	146.4	540.4
Global statistics				
Mean	*****	**7491**	**58.01**	**500.0**
Min	*****	25.70	**3.542**	457.3
Max	*****	181781	**475.5**	762.5

Table (3.2): The size of ZnO/Alg NPs by AFM.

3.1.2 Fourier transforms infrared (FTIR) characterization

The FTIR spectra, depicted in Figure 3.4, provide insight into the specific functional group linked to the ZnO nanocomposite. The FTIR spectra of ZnO nanoparticles shown in Fig. (3.4) exhibit prominent peaks at 424, 520, and 600 cm−1, which correspond to Zn–O bond stretching. Additionally, there are peaks at 1630 and 3416 cm−1, which indicate –OH bending and stretching vibrations. The vibrations of the –OH bond in alginate are caused by its interaction with ZnO, resulting in slight variations in the peak about 3400 cm−1. The band observed at 1120 cm−1 is associated with the symmetric stretching of the C–O bond in CO2. This molecule is found in the air and is absorbed on the surface. The small shift in the vibrational frequency of carboxylic acid suggests that Zn2+ has undergone reduction and has been stabilized by the formation of O-Zn type lattices through the interaction with oxygen electrons (lone pairs).55 The intense peaks observed at 600–400 cm−1 in the ZA1, ZA2, and ZA3 bio nanocomposites are caused by the presence of ZnO nanoparticles.

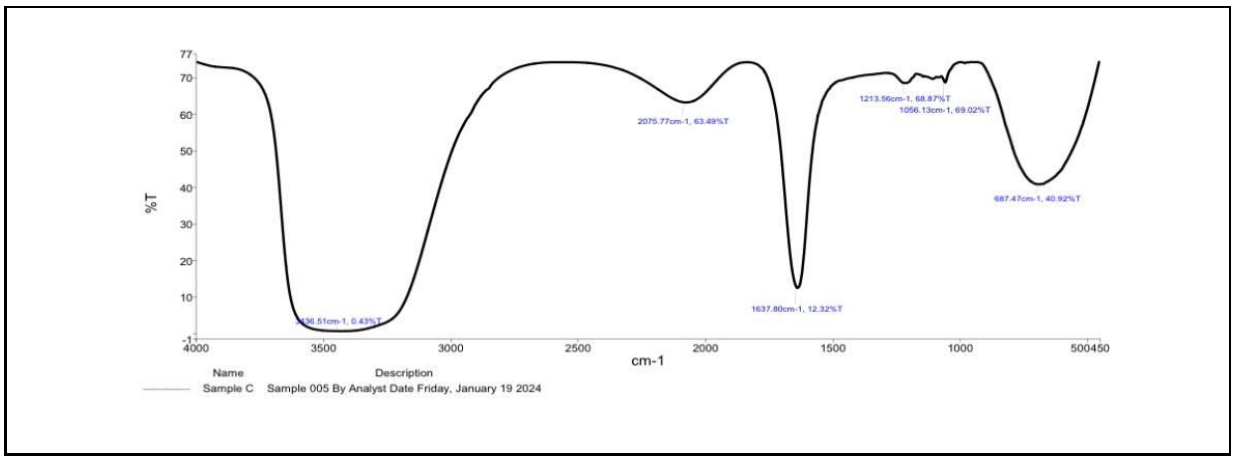


Figure (3.3): The Fourier Transforms Infrared (FTIR) Spectroscopy Measurement

3.1.3 Filed Emission Scanning Electron Microscopy (FESEM)

The SEM was used to examine the morphological characteristics of the ZnO/Alginate nanostructures that were created. Figure (3.4) illustrates the results. The findings indicate the

presence of spherical structures with diameters ranging from 30 to 40 nm. In addition, due to the thermal energy generated during the annealing procedure, a few of the particles experienced a minor agglomeration. Nevertheless, the diminutive particles exhibited a significant level of reactivity due to their jagged edges, which had a substantial volume-to-surface ratio, resulting in a greater proportion of exterior atoms and lower energy compared to the bulk materials.

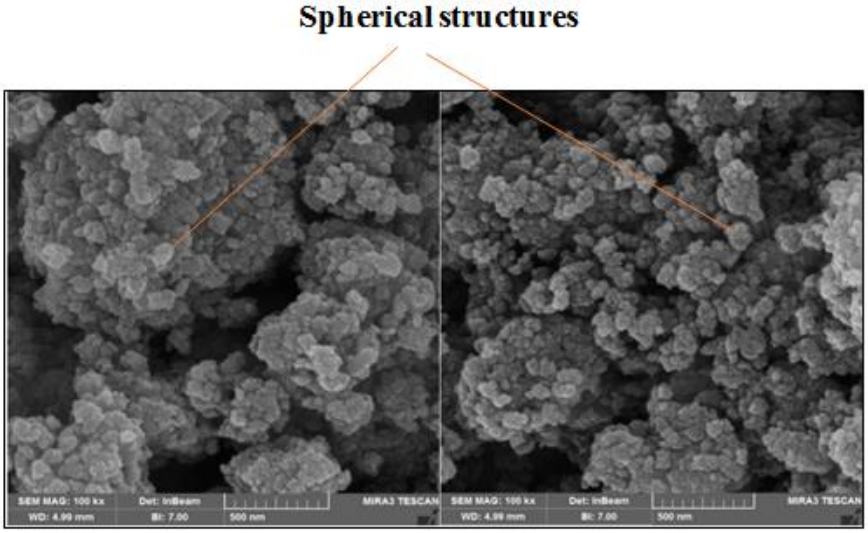
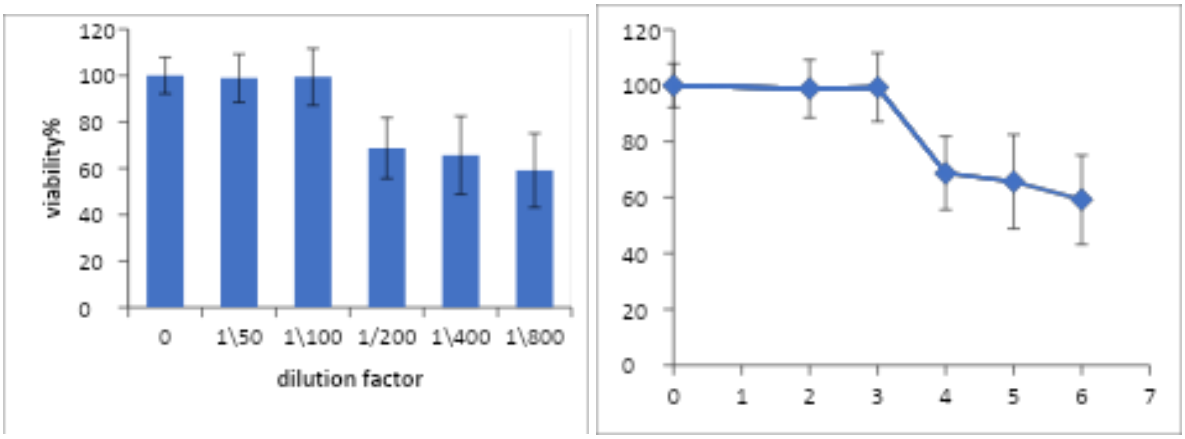


Figure (3.4): morphological characteristics of the ZnO/Alginate nanostructures, the results of FESEM indicate presence of spherical structures with diameters ranging from 30 to 40 nm

3.2 Cytotoxicity of Nanocomposite

Cytotoxic effect of ZnO/Alginate NPs on cell lines in vitro. Results showed a highly significant ($P < 0.001$) effect of Zinc Oxide/Alginate nanoparticles on pancreatic cancer cell lines at different concentrations. The percentage of cell viability varied as cells were treated with different concentrations of zinc oxide nanoparticles. After treatment with different concentrations (50, 100, 200, 400, and 800 µg/ml) and 5 different readings for each concentration, the high dose was greatly effective in decreasing the proliferation of cells, as shown in Figure (3.6). So, the highest dose (800 µg/ml) slowed the cell line's growth to a mean optical density (M.O.D.) of 0.165, which was lower than the control M.O.D. of 0.5431. As the concentration of treatment had increased gradually and respectively (0.5, 1, 5, 10) µg/ml in low doses, there were no significant effects of treatment on cancer cells. The M.O.D. of the cell line was (0.4834, 0.4342, 0.4228, and 0.362), and the viability was similar with a slight inhibition in cells. After decreasing the dose of treatment to 50 or 100) µg/ ml, the viability with M.O.D. decreased to (0.3456, 0.1752), which was highly significant and reduced the growth of cancer cells. The dose of treatment with Zinc Oxide/Alginate nanoparticles increased to 200 and 400 µg/ml, the nanoparticle decreased the viability of the cell line, and the M.O.D. was (0.1514 and 0.155. Zinc Oxide/Alginate nanoparticles were highly effective as cells were treated with 200, 400, and 800 µg/Ml . We did not use a concentration of more than 800 because that leads to a toxic and harmful effect on the surrounding healthy cells and organs. A curve that showed inhibition was used to figure out the IC50 value for the amount of ZnO/Alginate NP that was needed to stop cell growth by 50%. The result showed that the IC50 for the cell line was 200 µg/ml (Figure3.6)



(a) (b)

Figure (3.5): Cytotoxicity of Pancreatic cancer exposed to ZnO-Alginate NPs. (a) The figure shows the 50% inhibitory concentrations (IC50). (b) The graph illustrates the dose-dependent response of Pancreatic cancer to varying concentrations of ZnO-Alginate NPs, indicating a decrease in cell viability with increasing doses.

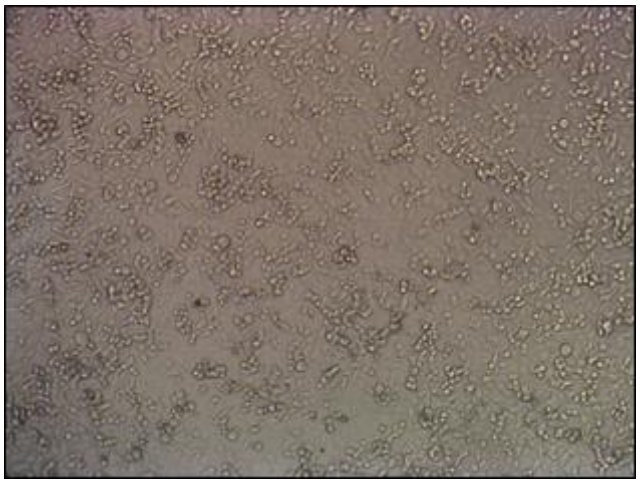


Figure (3.6a): Microscopic view of in vitro cells, it appears that there is no effect on pancreatic cancer cell lines when the concentration of ZnO-alginate nanoparticles is zero in cytotoxicity of cell line test

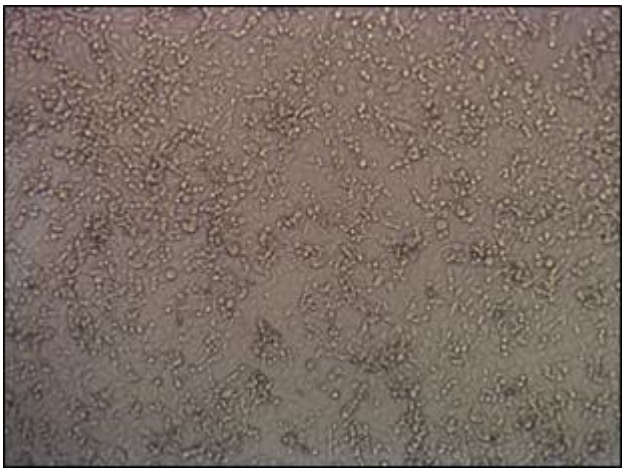


Figure (3.6b): Microscopic view of in vitro cells, it appears that there is mild inhibition effect of pancreatic cancer cell lines when the concentration of ZnO-alginate nanoparticles is 50 in cytotoxicity of cell line test

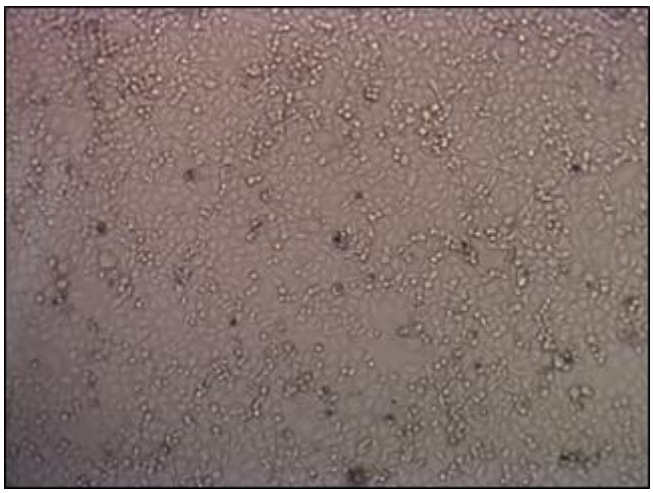


Figure (3.6c): Microscopic view of in vitro cells, it appears that there is moderate reduction in pancreatic cancer cell lines when the concentration of ZnO-alginate nanoparticles is 100 in cytotoxicity of cell line test

Figure (3.6d): Microscopic view of in vitro cells, it appears that there is significant decrease in pancreatic cancer cell viability when the concentration of ZnO-alginate nanoparticles is 200 in cytotoxicity of cell line test

Figure (3.6e): Microscopic view of in vitro cells, it appears that there is potent anti-cancer effect demonstrated in pancreatic cancer cell when the concentration of ZnO-alginate nanoparticles is 400 in cytotoxicity of cell line test

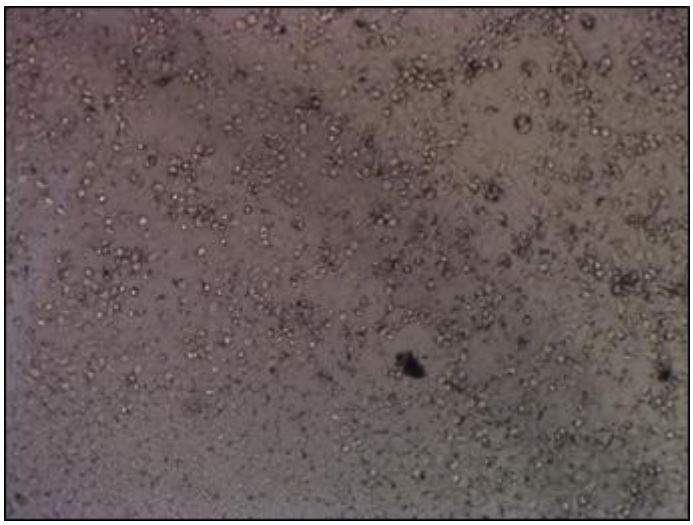


Figure (3.6f): Microscopic view of in vitro cells, it appears that there is highly effective suppression of pancreatic cancer cells when the concentration of ZnO-alginate nanoparticles is 800 in cytotoxicity of cell line test

Chapter Four

Conclusion &

Recommendation

4. Conclusion & Recommendation

4.1 Conclusion

1. Zinc Oxide/Alginate Nanoparticle was synthesized by the chemical ionotropic gelation method.
2. The Zinc Oxide/Alginate prepared was characterized by AFM, and the results indicated the synthesize Zinc Oxide/Alginate NPs was in the range of nanometer with average diameter size 58.01nm.
3. ZnO/Alginate Nanocomposite could have toxic effect dependent on the concentration.
4. ZnO/Alginate Nanocomposite had cytotoxic effect on the cell lines that used the pancreatic cell line.

4.2 Recommendation

1. Complete the characterization of polymeric nanoparticles such as zeta potential and X-ray diffraction (XRD).
2. studying the antimicrobial activity.
3. Use of EPO, EREF, CYP2E1, Hcy, GSH, MDA and Caspase-3 biomarkers as indicators of Nanoparticles toxicity of animal organs tissues instead of sera.

References

[A]

1. Abdul Bakil, S. N., Kamal, H., Abdullah, H. Z., & Idris, M. I. (2020). Sodium Alginate-Zinc Oxide Nanocomposite Film for Antibacterial Wound Healing Applications. Biointerface Research in Applied Chemistry, 10(5), 6245-6252.
2. Abouelmagd, S.A., Meng, F., Kim, B.-K., Hyun, H., and Yeo, Y., (2016). Tannic acid-mediated surface functionalisation of polymeric nanoparticles.ACS Biomater. Sci. Eng., 6b00497

3. Ajdary, M., Moosavi, M. A., Rahmati, M., Falahati, M., Mahboubi, M., Mandegary, A., and Varma, R. S. (2018). Health concerns of various nanoparticles: a review of their in vitro and in vivo toxicity. Nanomaterials, 8(9), 634.

4. Al Garalleh, H. (2013). Modelling molecular interactions arising in nanobiotechnology. Thesis (Ph.D). Wollongong University.

5. Al Garalleh, H. (2013). Modelling molecular interactions arising in nanobiotechnology. Thesis (Ph.D). Wollongong University.

6. Ali, S., Khan, I., Khan, S.A., Sohail, M., Ahmed, R., Rehman, A., Ur Ansari, M.S., and Morsy, M.A., (2017). Electrocatalytic performance of NiOPt core–shell nanoparticles supported on carbon nanotubes for methanol oxidation reaction. J. Electroanal. Chem. 795, 17–25.

7. Ali, S., Khan, I., Khan, S.A., Sohail, M., Ahmed, R., Rehman, A., Ur Ansari, M.S., and Morsy, M.A., (2017). Electrocatalytic performance of NiOPt core–shell nanoparticles supported on carbon nanotubes for methanol oxidation reaction. J. Electroanal. Chem. 795, 17–25.

8. Allen, T. M. and Cullis P.R. (2004). Drug delivery systems: entering the mainstream. Science 303:1818-1822.

9. Alyamani, A. M. O. L., & Lemine, O. M. (2012). FE-SEM characterisation of some nanomaterial. In Scanning electron microscopy. IntechOpen.

10. Anajwala, C. C., Jani, G. K., & Swamy, S. V. (2010). Current trends of nanotechnology for cancer therapy. International journal of pharmaceutical sciences and nanotechnology, 3(3), 1043-1056.

11. Anton, N.; Benoit, J. P. and Saulnier, P. (2008). Design and production of nanoparticles formulated from nano-emulsion templats–a review. J. Control. Release 128, 185-199.

12. Anton, N.; Benoit, J. P. and Saulnier, P. (2008). Design and production of nanoparticles formulated from nano-emulsion templats–a review. J. Control. Release 128, 185-199.

13. Anwer, M. K., Al-Mansoor, M. A., Jamil, S., Al-Shdefat, R., Ansari, M. N., and Shakeel, F. (2016). Development and evaluation of PLGA polymer based nanoparticles of quercetin. International Journal of Biological Macromolecules, 92: 213-219.

[B]

14. Barrak, H., Saied, T., Chevallier, P., Laroche, G., M'nif, A., and Hamzaoui, A.H., (2016). Synthesis, characterisation, and functionalisation of ZnO nanoparticles by N-(trimethoxysilylpropyl) ethylenediamine triacetic acid (TMSEDTA): investigation of the interactions between phloroglucinol and ZnO@TMSEDTA. Arab. J. Chem.

15. Bartneck,M., Ritz, T., Keul, H. A.,Wambach,M., Bornemann, J.,Gbureck, U., and et al. (2012). Peptide-functionalised gold nanorods increase liver injury in hepatitis. ACS Nano 6, 8767–8777.

16. Barui, S., Percivalle, N. M., Conte, M., Dumontel, B., & others. (2020). Development of doped ZnO-based biomimicking and tumor targeted nanotheranostics to improve pancreatic cancer treatment. Research Square.

17. Biswajit, M., Niladri, S. D., Ruma, M., Priyanka, B., Pranab, J. D., and Paramita, P. (2014).Current Status and Future Scope for Nanomaterials in Drug Delivery. Application of Nanotechnology in Drug Delivery. InTech

18. Borel, T., and Sabliov, C. M. (2014). Nanodelivery of bioactive components for food applications: types of delivery systems, properties, and their effect on ADME profiles and toxicity of nanoparticles. Annual review of food science and technology, 5, 197-213.

19. Borel, T., and Sabliov, C. M. (2014). Nanodelivery of bioactive components for food applications: types of delivery systems, properties, and their effect on ADME profiles and toxicity of nanoparticles. Annual review of food science and technology, 5, 197-213.

20. Bouwmeester, H.; Dekkers, S.; Noordam, M. Y.; Hagens, W. I.; Bulder, A. S.; De Heer, C.; and Sips, A. J. (2009). Review of health safety aspects of nanotechnologies in food production. Regulatory toxicology and pharmacology, 53(1), pp :52-62.

21. Boverhof, D. R., Bramante, C. M., Butala, J. H., Clancy, S. F., Lafranconi, M., West, J., & Gordon, S. C. (2015). Comparative assessment of nanomaterial definitions and safety evaluation considerations. Regulatory Toxicology and Pharmacology, 73(1), 137-150

22. Britannica, T. Editors of Encyclopaedia (2023, September 4). polymer. Encyclopedia Britannica.

23. Brohi, R. D., Wang, L., Talpur, H. S., Wu, D., Khan, F. A., Bhattarai, D., and Huo, L. J. (2017). Toxicity of nanoparticles on the reproductive system in animal models: a review. Frontiers in pharmacology, 8, 606.

24. Brohi, R. D., Wang, L., Talpur, H. S., Wu, D., Khan, F. A., Bhattarai, D., and Huo, L. J. (2017). Toxicity of nanoparticles on the reproductive system in animal models: a review. Frontiers in pharmacology, 8, 60

25. Brohi, R. D., Wang, L., Talpur, H. S., Wu, D., Khan, F. A., Bhattarai, D., and Huo, L. J. (2017). Toxicity of nanoparticles on the reproductive system in animal models: a review. Frontiers in pharmacology, 8, 606.

[C]

26. Cheng, R., Feng, F., Meng, F., Deng, C., Feijen, J., and Zhong, Z. (2011). Glutathione-responsive nano-vehicles as a promising platform for targeted intracellular drug and gene delivery. Journal of controlled release, 152(1), 2-12.

27. Choudhary, R. C., Kumaraswamy, R. V., Kumari, S., Pal, A., Raliya, R., Biswas, P., & Saharan, V. (2017). Synthesis, characterisation, and application of chitosan nanomaterials loaded with zinc and copper for plant growth and protection. In Nanotechnology (pp. 227-247). Springer, Singapore.

28. Choudhary, R. C., Kumari, S., Kumaraswamy, R. V., Pal, A., Raliya, R., Biswas, P., & Saharan, V. (2019). Characterisation Methods for Chitosan-Based Nanomaterials. In Plant Nanobionics (pp. 103-116). Springer, Cham.

[D]

29. Das, S.; Banerjee, R. and Bellare, J. (2005). Aspirin loaded albumin nanoparticles by coacervation: Implications in drug delivery. Trends Biomater. Artif. vol.18, pp. 203-212.

30. Diallo, A., Doyle, T. B., Mothudi, B. M., Manikandan, E., Rajendran, V., & Maaza, M. (2017). Magnetic behavior of biosynthesised Co3O4 nanoparticles. Journal of Magnetism and Magnetic Materials, 424, 251-255.

31. Dinauer, N.; Balthasar, S.; Weber, C.; Kreuter, J.; Langer, K. and Brisen, H.V. (2005). “Selective targeting of antibody-conjugated nanoparticles to leukemic cells and primary T-lymphocytes”. Biomaterials. vol.26, pp. 5898-5900.

32. Divya, C., & Muthuvinothini, A. (2015). Nanoscience and Nanotechnology. Edorium Journal of Nanotechnology, (2), 1-3.

[E]

33. Ealias, A. M., & Saravanakumar, M. P. (2017, November). A review on the classification, characterisation, synthesis of nanoparticles and their application. In IOP Conf. Ser. Mater. Sci. Eng (Vol. 263, p. 032019).

[F]

34. Faisal, N., & Kumar, K. (2017). Polymer and metal nanocomposites in biomedical applications. BIOINTERFACE RESEARCH IN APPLIED CHEMISTRY, 7(6), 2286-2294.
35. Feng, S.S.; Mu, L. and Win, K.Y. (2004). Nanoparticles of biodegradable polymers for clinical admenstration of paclitaxel. CurrMed Chem.11: 413-424.
36. Ferreira, A.J., Cemlyn-Jones, J., and Robalo Cordeiro, C. (2013). Nanoparticles, nanotechnology and pulmonary nanotoxicology. Rev. Port. Pneumol. 19, 28–37.
37. Ferreira, A.J., Cemlyn-Jones, J., and Robalo Cordeiro, C. (2013). Nanoparticles, nanotechnology and pulmonary nanotoxicology. Rev. Port. Pneumol. 19, 28–37.
38. Frank, L. A.; Contri, R. V.; Beck, R.C.R.; Pohlmann, A. R. and Guterres, S. S. (2015). Improving drug biological effects by nanocapsulation into polymeric nanocapsules. Nanobiotechnol 7(5):623-639.

[G]

39. Ganesh, M., Hemalatha, P., Peng, M.M., and Jang, H.T., (2017). One pot synthesised Li, Zr doped porous silica nanoparticlefor low temperature CO2 adsorption. Arab. J. Chem. 10, S1501–S1505.
40. Garcés, M., Cáceres, L., Chiappetta, D., Magnani, N., & Evelson, P. (2021). Current understanding of nanoparticle toxicity mechanisms and interactions with biological systems. New Journal of Chemistry, 45(32), 14328-14344.
41. Garcia-Garcia, E.; Gill, S.; Andrieux, K.; Desmaele, D.; Nicolas, V.; Taran, F.; Georgin, D.; Andreux, J.P.; Roux, F. and Couvreur, P. (2005). A relevant in vitro rat model for the evaluation of bloodbrain barrier translocation of nanoparticles. Cell Mol Life Sci, 62(12):1400-1408.
42. Gelperina, S.; Kisich, K.; Iseman,M. D. & Heifets, L. (2005). The potential advantages of nanoparticle drug delivery systems in chemotherapy of tuberculosis. American Journal of Respiratory and Critical Care Medicine, 172, 1487–1490.
43. Ghawade, S. P., Pande, K. N., Dhoble, S. J., & Deshmukh, A. D. (2022). Tuning the properties of ZnS semiconductor by the addition of graphene. In V. B. Pawade, S. J. Dhoble, & H. C. Swart (Eds.), *Nanoscale Compound Semiconductors and their Optoelectronics Applications* (pp. 351-381). Woodhead Publishing.
44. Gianfredi, V., Nucci, D., Nardi, M., Santangelo, O. E., & Provenzano, S. (2023). Using Google Trends and Wikipedia to Investigate the Global Public's Interest in the Pancreatic Cancer Diagnosis of a Celebrity. International Journal of Environmental Research and Public Health, 20(3), 2106.
45. Glaser, J. A. (2019). Biological Degradation of Polymers in the Environment. In Plastics in the Environment. IntechOpen.

[H]

46. Han, J., Zhao, D., Li, D., Wang, X., Jin, Z., & Zhao, K. (2018). Polymer-based nanomaterials and applications for vaccines and drugs. Polymers, 10(1), 31.

47. Higa, A. M., Mambrini, G. P., Hausen, M., Strixino, F. T., & de Lima Leite, F. (2016). Ag-nanoparticle-based nano-immunosensor for antiglutathione S-transferase detection. Biointerface Research in Applied Chemistry, 6(1).

48. Hisatomi, T., Kubota, J., and Domen, K., (2014). Recent advances in semiconductors for photocatalytic and photoelectrochemical water splitting. Chem. Soc. Rev. 43, 7520–7535.

49. Hisatomi, T., Kubota, J., and Domen, K., (2014). Recent advances in semiconductors for photocatalytic and photoelectrochemical water splitting. Chem. Soc. Rev. 43, 7520–7535.

50. Hossain, M. S., & Iqbal, A. (2014). Production and characterisation of chitosan from shrimp waste. Journal of the Bangladesh Agricultural University, 12(1), 153-160

51. Husain, Q. (2017). Nanosupport bound lipases their stability and applications. Biointerface Research in Applied Chemistry, 7(6), 2194- 2216.

[I]

52. Iravani, S., (2011). Green synthesis of metal nanoparticles using plants. Green Chem. 13, 2638.

[K]

53. Kessler, Rebecca (2011-03-01). "Engineered Nanoparticles in Consumer Products: Understanding a New Ingredient"

54. Khan, I., Abdalla, A., and Qurashi, A. (2017a). Synthesis of hierarchical WO3 and Bi2O3/WO3 nanocomposite for solar-driven water splitting applications. Int. J. Hydrogen Energy 42, 3431–3439.

55. Khan, I., Abdalla, A., and Qurashi, A. (2017a). Synthesis of hierarchical WO3 and Bi2O3/WO3 nanocomposite for solar-driven water splitting applications. Int. J. Hydrogen Energy 42, 3431–3439.

56. Khan, I., Saeed, K., and Khan, I. (2019b). Nanoparticles: Properties, applications and toxicities. Arabian journal of chemistry, 12(7), 908- 931.

57. Koçak, A., & Karasu, B. General Evaluations of Nanoparticles. ElCezerî Journal of Science and Engineering, 5(1), 191-236.

58. Kshirsagar, R.V.; Dhembre, G.N. and Moon, R.S. (2011). A review on polymeric micellar nanocarriers. International Journal of Pharma and Bio Sciences 2(2):109-116.

59. Kumar, Surabhi Siva; Venkateswarlu, Putcha; Rao, Vanka Ranga; Rao, Gollapalli Nageswara (2013-05-07). "Synthesis, characterisation and optical properties of zinc oxide nanoparticles"

[L]

60. Laurent, S., Forge, D., Port, M., Roch, A., Robic, C., Vander Elst, L., and Muller, R.N., (2010). Magnetic iron oxide nanoparticles: synthesis, stabilisation, vectorisation, physicochemical characterisations, and biological applications. Chem. Rev. 110.

61. Lee, J.E., Lee, N., Kim, T., Kim, J., and Hyeon, T., (2011). Multifunctional mesoporous silica nanocomposite nanoparticles for theranostic applications. Acc. Chem. Res. 44, 893–902.

62. Letchford, K. and Burt, H. (2007).A review of the formation and classification of amphiphilic block copolymer nanoparticulate structures: micelles, nanospheres, nanocapsules and polymersome. Eur. J. Pharm. Biopharm. 65, 259-269.

63. Levy [eds.]. Nanobiotechnology Bioinspired Devices and Materials for the Future. Humana Press Inc., New Jersey, USA.

64. Loureiro, A., Azoia, N.G., Gomes, A.C., Cavaco-Paulo, A., (2016). Albumin-based nanodevices as drug carriers. Curr. Pharm. Des. 22, 1371–1390.

[M]

65. Mabena, L.F., Sinha Ray, S., Mhlanga, S.D., and Coville, N.J., (2011). Nitrogen-doped carbon nanotubes as a metal catalyst support. Appl. Nanosci. 1, 67–77.

66. Maeda, H.; Wu, J.; Sawa, T.; Matsumura, Y. and Hori, K. (2000). Tumor vascular permability and the EPR in macromolecular therapeutics: a review. J Control Release. 65:271-284.

67. Mafuné, F., Kohno, J. Y., Takeda, Y., Kondow, T., & Sawabe, H. (2001). Formation of gold nanoparticles by laser ablation in aqueous solution of surfactant. The Journal of Physical Chemistry B, 105(22), 5114-5120.

68. Mamiyev, Z. Q., & Balayeva, N. O. (2015). Preparation and optical studies of PbS nanoparticles. *Optical Materials, 46*, 522-525. https://doi.org/10.1016/j.optmat.2015.05.017

69. Mansha, M., Khan, I., Ullah, N., and Qurashi, A., (2017). Synthesis, characterisation and visible-light-driven photoelectrochemical hydrogen evolution reaction of carbazole-containing conjugated polymers. Int. J. Hydrogen Energy.

70. Mansoori, G. A. (2017). An introduction to nanoscience and nanotechnology. In Nanoscience and Plant–Soil Systems (pp. 3-20). Springer, Cham.

71. Mashaghi, S., Jadidi, T., Koenderink, G., and Mashaghi, A., (2013). Lipid nanotechnology. Int. J. Mol. Sci. 14, 4242–4282.

72. Mohammadikhah, R., & Asadi-Malekshah, R. (2016). ThermalOxidative Degradation of PGA, PLLA, and Random Binary PLLA-PGA Copolymers. Journal of Petroleum Science and Technology, 6(2), 66-83

73. Mora-Huertas, C.E.; Fessi, H. and Elaissari, A. (2010). Polymerbased nanocapsules for drug delivery. International journal of pharmaceutics. 385(1–2):113–42.

74. Moulton, M. C., Braydich-Stolle, L. K., Nadagouda, M. N., Kunzelman, S., Hussain, S. M., and Varma, R. S. (2010). Synthesis, characterisation and biocompatibility of “green” synthesised silver nanoparticles using tea polyphenols. Nanoscale, 2(5), 763-770.

75. Movassaghian, S.; Merkel, O. M. and Torchilin, T. (2015). Applications of Polymer Micelles for Imaging and Drug Delivery, WIREs Nanomedicine & Nanobiotechnology.7:691–707.

[N]

76. Nanosized carrier for drug delivery. Pp. 329-349.In: O. Shoseyov and I.

77. Ngoy, J.M., Wagner, N., Riboldi, L., and Bolland, O., (2014). A CO2capture technology using multi-walled carbon nanotubes with polyaspartamide surfactant. Energy Procedia 63, 2230–2248.

[O]

78. Oberdörster, G., Oberdörster, E., and Oberdörster, J. (2005). Nanotoxicology: an emerging discipline evolving fromstudies of ultrafine particles. Environ. Health Perspect. 823–83

79. Oberdörster, G., Oberdörster, E., and Oberdörster, J. (2005). Nanotoxicology: an emerging discipline evolving fromstudies of ultrafine particles. Environ. Health Perspect. 823–839.

80. Omrani, M. M., Ansari, M., & Kiaie, N. (2016). Therapeutic effect of stem cells and nano-biomaterials on Alzheimer's disease. Biointerface Research in Applied Chemistry, 6(6).

[P]

81. Patra, J. K., Das, G., Fraceto, L. F., Campos, E. V. R., del Pilar Rodriguez-Torres, M., Acosta-Torres, L. S., and Habtemariam, S. (2018). Nano based drug delivery systems: recent developments and future prospects. Journal of Nanobiotechnology, 16(1): 71.
82. Patra, J. K., Das, G., Fraceto, L. F., Campos, E. V. R., Rodriguez-Torres, M. D. P., Acosta-Torres, L. S., … & Shin, H.-S. (2018). Nano based drug delivery systems: recent developments and future prospects. Journal of Nanobiotechnology, 16(1), 71.
83. Pinto, C.; Neufeld, R.; Ribeiro, A. and Veiga, F. (2006). Nanoencapsulation II. Biomedical applications and current status of peptide and protein nanoparticulate delivery systems. Nanomedicin: NBM 2, 53-65.
84. Puri, A., Loomis, K., Smith, B., Lee, J.-H., Yavlovich, A., Heldman, E., Blumenthal, R., (2009). Lipid-based nanoparticles as pharmaceutical drug carriers: from concepts to clinic. Crit. Rev. Ther. Drug Carrier Syst. 26, 523–580.

[R]

85. Radtchenko, I. L.; Sukhorukov, G. B. and Möhwald, H. (2002). A novel method for encapsulation of poorly water- soluble drugs: precipitation in polyelectrolyte multilayers shells. Int. J. Pharm. 242, 219-223.
86. Raus, R. A., Nawawi, W. M. F. W., & Nasaruddin, R. R. (2021). Alginate and alginate composites for biomedical applications. Asian Journal of Pharmaceutical Sciences, 16(3), 280-306.
87. Rawat, M.K., Jain, A., Singh, S., Mehnert, W., Thunemann, A.F., Souto, E.B., Mehta, A., and Vyas, S.P., (2011). Studies on binary lipid matrix based solid lipid nanoparticles of repaglinide: in vitro and in vivo evaluation. J. Pharm. Sci. 100, 2366–2378.
88. Rawle, A. F. (2017). Characterisation of nanomaterials. Metrology and Standardization of Nanotechnology: Protocols and Industrial Innovations, 129-150.
89. Rong, X.; Xie, Y.; Hao, X.; Chen, T.; Wang, Y. and Liu, Y. (2011). Application of polymeric nanocapsules in field of drug delivery systems. Curr Drug Discov Technol. 8(3) 173-187.

[S]

90. Saeed, K., and Khan, I., (2016). Preparation and characterisation of single walled carbon nanotube/nylon 6,6 nanocomposites. Instrum Sci. Technol. 44, 435–444.
91. Sahoo, S.K.; T.K. Jain, M.K.; Reddy, and Labhasetwar, V. (2008).
92. Shaalan, M., Saleh, M., El-Mahdy, M., and El-Matbouli, M., (2016). Recent progress in applications of nanoparticles in fish medicine: a review. Nanomed. Nanotechnol. Biol. Med. 12, 701–710.
93. Sheoran, N., & Kaur, P. (2018). Biosynthesis of nanoparticles using eco-friendly factories and their role in plant pathogenicity: a review. Biotechnology Research and Innovation.
94. Shin, W.-K., Cho, J., Kannan, A.G., Lee, Y.-S., and Kim, D.-W., (2016). Cross-linked composite gel polymer electrolyte using mesoporous methacrylate-functionalised SiO2 nanoparticles for lithium-ion polymer batteries. Sci. Rep. 6, 26332.
95. Sigmund, W., Yuh, J., Park, H., Maneeratana, V., Pyrgiotakis, G., Daga, A., Taylor, J., and Nino, J.C., (2006). Processing and structure relationships in electrospinning of ceramic fiber systems. J. Am. Ceram. Soc. 89, 395–407.

96. Spoială, A., Ilie, C.-I., Ficai, D., Ficai, A., & Andronescu, E. (2022). From Biomedical Applications of Alginate towards CVD Implications Linked to COVID-19. Pharmaceuticals, 15(3), 318.

97. Sun, S., (2000). Monodisperse FePt nanoparticles and ferromagnetic FePt nanocrystal superlattices. Science 80 (287), 1989–1992.

[T]

98. Teleanu, D., Chircov, C., Grumezescu, A., Volceanov, A., & Teleanu, R. (2018). Impact of nanoparticles on brain health: An up to date overview. Journal of clinical medicine, 7(12), 490.

99. Thomas, S., Harshita, B.S.P., Mishra, P., and Talegaonkar, S., (2015). Ceramic nanoparticles: fabrication methods and applications in drug delivery. Curr. Pharm. Des. 21, 6165–6188.

100. Tiwari, J.N., Tiwari, R.N., and Kim, K.S., (2012). Zero-dimensional, one dimensional, two-dimensional and three-dimensional nanostructured materials for advanced electrochemical energy devices. Prog. Mater Sci. 57, 724–803.

101. Trivedi, R. and Kompella, U.B. (2010). Nanomicellar formulations for sustained drug delivery: strategies and underlying principles. Nanomedicine (Lond.) 5(3), 485–505.

[U]

102. Ullah, H., Khan, I., Yamani, Z.H., and Qurashi, A., (2017). Sono chemical driven ultrafast facile synthesis of SnO2 nanoparticles: growth mechanism structural electrical and hydrogen gas sensing properties. Ultrason. Sonochem. 34, 484–490.

[V]

103. Vance, M. E., Kuiken, T., Vejerano, E. P., McGinnis, S. P., Hochella, M. F. Jr., Rejeski, D., and et al., (2015). Nanotechnology in the real world: redeveloping the nanomaterial consumer products inventory. Beilstein J. Nanotechnol. 6,1769–1780.

104. Vance, M. E., Kuiken, T., Vejerano, E. P., McGinnis, S. P., Hochella, M. F. Jr., Rejeski, D., and et al., (2015). Nanotechnology in the real world: redeveloping the nanomaterial consumer products inventory. Beilstein J. Nanotechnol. 6,1769–1780.

105. Vasir, J.K.; Reddy, M.K. and Labhasetwar, V. (2005). Nanosystems in drug targeting: opportunities and challenges. Curr Nanosci. 1:47-64.

106. Velagaleti, P.R.; Anglade, E.; Khan, I.J.; Gilger, B.C. and Mitra, A.K. (2010). Topical delivery of hydrophobic drugs using a novel mixed nanomicellar technology to treat diseases of the anterior and posterior segments of the eye. Drug Deliv. Technol. 10(4), 42–47.

107. Vozza, G. (2018). Formulation and in Vitro Characterisation of Fungal Chitosan Nanoparticles Coated With Zein for Improved Oral Delivery of Selenoamino Acids.

[W]

108. Wang, Y., and Xia, Y., (2004). Bottom-up and top-down approaches to the synthesis of monodispersed spherical colloids of low melting-point metals. NanoLett. 4, 2047–2050.

109. Woodard, L. N., Page, V. M., Kmetz, K. T., & Grunlan, M. A. (2016). PCL–PLLA Semi-IPN Shape Memory Polymers (SMPs): Degradation and Mechanical Properties. Macromolecular rapid communications, 37(23), 1972-1977.

[Z]

110. Zada, A., Muhammad, P., Ahmad, W., Hussain, Z., Ali, S., Khan, M., and Maqbool, M. (2020). Surface Plasmonic-Assisted Photocatalysis and Optoelectronic Devices with Noble Metal Nanocrystals: Design, Synthesis, and Applications. Advanced Functional Materials, 30(7), 1906744.

111. Zamiri, R., Abbastabar Ahangar, H., Zakaria, A. et al. The structural and optical constants of Ag2S semiconductor nanostructure in the Far-Infrared. Chemistry Central Journal 9, 28 (2015). https://doi.org/10.1186/s13065-015-0099-y

112. Zhang, Yuanyuan; Leu, Yu-Rui; Aitken, Robert J.; Riediker, Michael (2015-07-24). "Inventory of Engineered Nanoparticle-Containing Consumer Products Available in the Singapore Retail Market and Likelihood of Release into the Aquatic Environment"

113. Zhu, L., Pelaz, B., Chakraborty, I., & Parak, W. J. (2019). Investigating Possible Enzymatic Degradation on Polymer Shells around Inorganic Nanoparticles. International journal of molecular sciences, 20(4), 935.

114. Zielińska, A., Carreiró, F., Oliveira, A. M., Neves, A., Pires, B., Venkatesh, D. N., Durazzo, A., Lucarini, M., Eder, P., Silva, A. M., Santini, A., & Souto, E. B. (2020). Polymeric Nanoparticles: Production, Characterisation, Toxicology and Ecotoxicology. Molecules (Basel, Switzerland), 25(16), 3731.