

## REVIEW

# Recent advancements in enzyme-incorporated nanomaterials: Synthesis, mechanistic formation, and applications

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## Abstract

Over the past decade, nanotechnology has been developed and employed across various entities. Among the numerous nanostructured material types, enzyme-incorporated nanomaterials have shown great potential in various fields, as an alternative to biologically derived as well as synthetically developed hybrid structures. The mechanism of incorporating enzyme onto a nanostructure depends on several factors including the method of immobilization, type of nanomaterial, as well as operational and environmental conditions. The prospects of enzyme-incorporated nanomaterials have shown promising results across various applications, such as biocatalysts, biosensors, drug therapy, and wastewater treatment. This is due to their excellent ability to exhibit chemical and physical properties such as high surface-to-volume ratio, recovery and/or reusability rates, sensitivity, response scale, and stable catalytic activity across wide operating conditions. In this review, the evolution of enzyme-incorporated nanomaterials along with their impact on our society due to its state-of-the-art properties, and its significance across different industrial applications are discussed. In addition, the weakness and future prospects of enzyme-incorporated nanomaterials were also discussed to guide scientists for futuristic research and development in this field.

## KEYWORDS

agro-food, biocatalysts, enzymes, immobilization, nanomaterials

## 1 | INTRODUCTION

The use of nanotechnology has been continuously gaining attention in several applications, especially in the pharmaceutical and medical (Bayda et al., 2019), wastewater treatment (H. Li et al., 2019), food and beverage, packaging (Gomez et al., 2021; Mousavi & Rezaei, 2011), electrical as well as optical industries (Y. Zhang et al., 2019). Some benefits of nanoparticles (NPs) include a larger surface-to-volume ratio which increases their

adsorption efficiency for elevating kinetics for reactions, better charge transfer, and carrier immobility (Shende et al., 2018). Furthermore, the addition of nanomaterials increases the stability, particularly when they are fabricated via physical or chemical approaches (Ye et al., 2016). However, physical and chemical synthesis of nanomaterials utilizes costly equipment and toxic reducing agents, which restricts them to be used for biomedical applications. Thus, greener or environmentally friendly approaches are currently being widely utilized for the synthesis of

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less/nontoxic NPs (X. Wang et al., 2014). Figure 1 shows common nanostructures that are currently under extensive research or utilized in commercial applications which are further explained in Section 2.

Proteins are essentially macromolecules, polymers made up of amino acids. Hundreds and thousands of these amino acids link together to form long chains of proteins (Blanco & Blanco, 2017). Enzymes, on the other hand, are proteins that act on substrate complexes to reduce the activation energy of a chemical reaction (Lewis & Stone, 2022). Recently, hybrid nanomaterials are gaining significant attention among scientists, which are synthesized through the fusion of compositions at a molecular level to produce a conjugate that can reduce the disadvantages of singular compounds. Organic-inorganic hybrid nanoflowers (NFs) are three-dimensional (3D) objects that resemble flowers under the microscope. Their synthesis, physical characteristics, protein activity, stability, and repeatability are all now being studied (Cui & Jia, 2017). Inorganic-organic hybrid NFs have shown promising results in stabilizing enzyme activity, drug delivery, biosensor, cell imaging, and other biomedical applications. Hybrid NFs have a unique shape and structure, where it has a larger surface-to-volume ratio, compared to spherical-shaped NPs (J. Li et al., 2016). Further, these hybrid NFs possess essential qualities, such as the ability to have increased stability, and physical and reproducibility properties, compared to conventional materials (S. Lee et al., 2015).

Organic-inorganic hybrid nanostructures are the fusion of organic (enzymes/proteins) and inorganic (metallic components), which exhibits

promising characteristics in various industries (Abd-Elsalam, 2020). The incorporation of enzymes onto nanomaterials has the natural ability to accommodate the enzymatic activity, structures as well as functions (Al-Maqdi et al., 2021). Further, enzymes and proteins are highly distinct from each other because enzymes are catalysts that exist naturally within living organisms and play a vital role in the regulation of both intracellular and extracellular systems (M. Chen et al., 2017). The designing of nanomaterial-enzyme conjugates can improve the overall enzymatic performance by imparting their novel properties onto the system, compared to conventional free enzymes, which often experience limitations in terms of deactivation and/or denaturing and ease of extraction (M. Cao et al., 2021; Tao et al., 2014). Thus, this review is an overview of various enzyme-incorporated nanomaterials, their history, mechanism, bond formation between enzyme and nanostructures (combinatorial technology), their impact, and applications across various industries.

## 2 | TYPES OF NPS

### 2.1 | Carbon-based NPs

In general, carbon-based NPs are divided into two major groups, such as fullerenes and carbon nanotubes (CNTs). Fullerenes are made up of allotropic carbon forms usually taking the shape of globular hollow cages. Some unique properties of fullerenes include their high

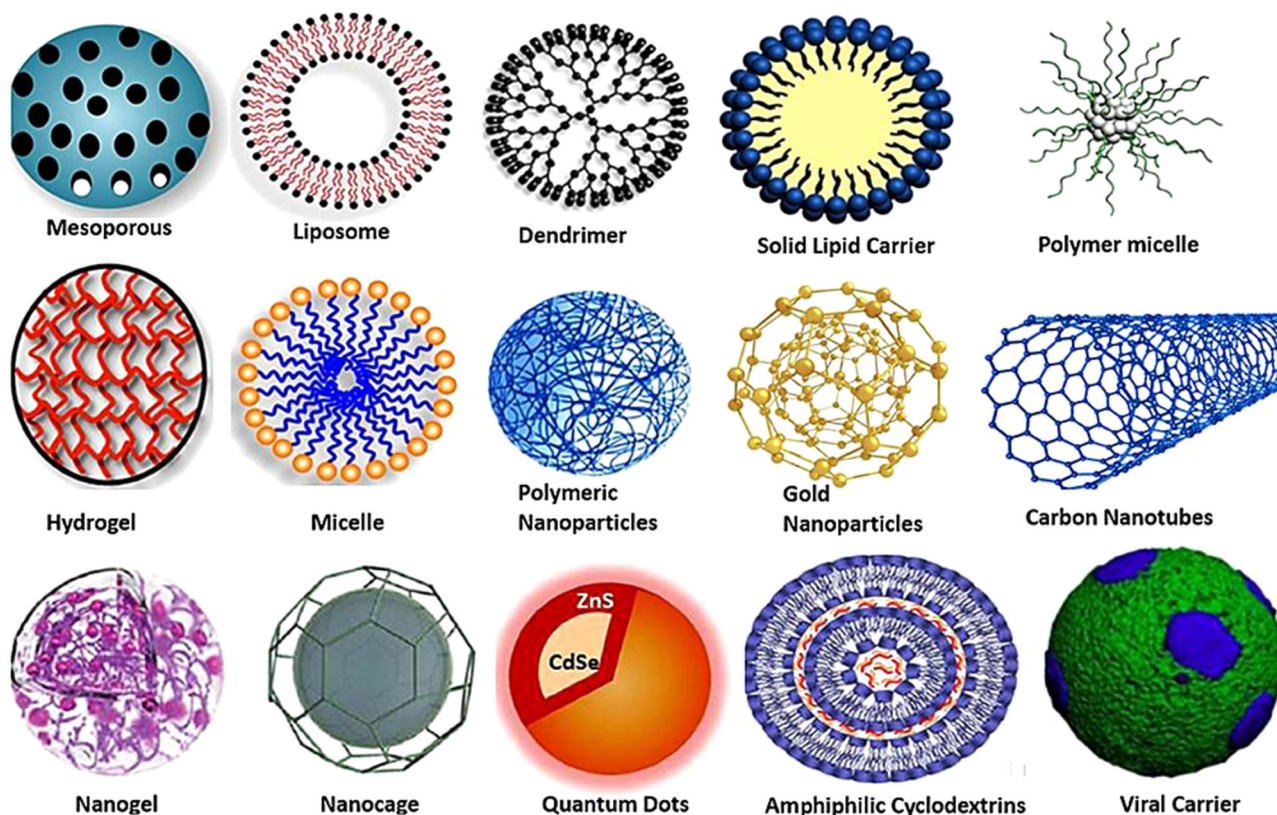


FIGURE 1 Representation of different nanosystems (Yaqoob et al., 2020)

strength and structure, electrical conductivity, electron affinity, and versatility (Astefanei et al., 2015). CNTs, on the other hand, are more elongated and tubular in structure with dimensions between 1 and 2 nm (Ibrahim, 2013). They typically roll into sheet-like forms similar to graphite sheets whereby, these sheets vary between single (SWCNTs), double, or multiwalled (MWCNTs). Vaporized atomic carbons from graphites and/or deposition of carbon precursors are widely used in the synthesis of CNTs. Some techniques involved during synthesis are, laser, electric arc onto metal particles as well as chemical vapor deposition (CVD) (Elliott et al., 2013).

## 2.2 | Metal NPs

Metal-based NPs are made from metal precursors. Metal-based NPs are well known for localized surface plasmon resonance (LSPR) which enhances their optoelectrical properties (Khan et al., 2019). Some other properties include their small size whereby metal NPs have a high surface-to-volume ratio. The difference in size contributes to the difference in electric, magnetic, and chemical properties (Liu, 2006). Metal NPs are also stable, highly functional (Bhatia, 2016), able to withstand high temperatures, good welding, superparamagnetic, catalytic, and selective properties (Chavali & Nikolova, 2019). They are used in multiple applications including drug and gene delivery, thermal ablation, radiotherapy enhancement as well as agriculture (Bansal et al., 2006).

## 2.3 | Polymer-based NPs

Polymer-based NPs are nanospherical and/or nano capsule-shaped particles that can embed and/or encapsulate agents or additives within the particle due to its complex polymeric matrix. It can also be adsorbed or attached to the surface of the particle as well (Din et al., 2017; Labhasetwar et al., 1997). Polymeric particles are mainly used in biomedical applications such as drug delivery due to their exceptional characteristics including biodegradability and biocompatibility to be able to be injected into a body, the ability to release drugs controllably (Kumar et al., 2012) and the ability to entrap drugs by different mechanisms (e.g., cross-linking, ionic cross-linking and ionic complexation) (Arnaldi et al., 2020; Prabaharan & Mano, 2004). Some disadvantages of polymeric NPs include high cost, higher toxicity levels, low purity due to the presence of solvent residue during synthesis and purification method, and the inability to produce in large-scale, degradability as well as specific requirements for biodegradable polymers (e.g., high purity and quality) (C.-C. Chen et al., 2010; Kahraman et al., 2017; Müller et al., 2007).

Polymeric NPs can be divided into natural and synthetic materials, whereby, the natural and synthetic materials can be further divided into biodegradable and nonbiodegradable polymeric NPs. Some examples of natural, biodegradable polymeric NPs are chitosan, cellulose, gelatine, alginate, and so forth, on the other hand, synthetic biodegradable polymers such as polylactic acid (PLA), poly-(lactide-co-glycolide) (PLGA), and polyanhydrides are well-known. Furthermore, nonbiodegradable,

synthetic polymers including polymethacrylate and polymethylmethacrylate (PMMA) have also been used in drug delivery and diagnostic imaging applications (Kumar et al., 2012).

## 2.4 | Lipid-based NPs

Lipid-based NPs are nano-spherical particles that are made of a lipidic core. Lipid NPs are synthesized by combining an oil phase with emulsifiers such as phospholipids. The oily core is how fat-soluble drugs such as paclitaxel (Mo & Lim, 2004), hematoporphyrin (P. Stevens et al., 2004), and lipid-conjugates (P. J. Stevens et al., 2004) are carried. There are various lipid-based carriers, solid-lipid NPs (SLN), lipid-drug conjugates (LDC) nanostructured lipid carriers (NLC) (Kumar et al., 2012). Lipid-based NPs thus far have been used in medicinal applications of DNA/RNA due to their low cytotoxic levels (Puri et al., 2009; Xiao et al., 2018).

## 2.5 | Lipid-polymer hybrid nanoparticles (LPN)

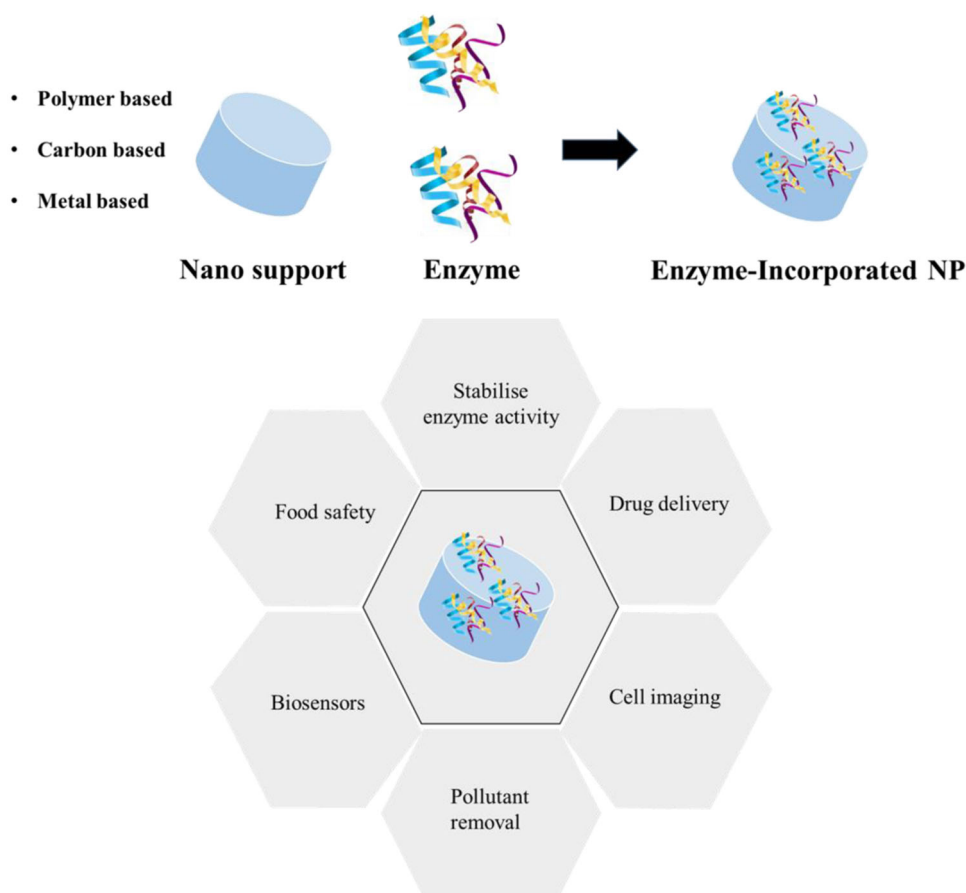
The idea of lipid-polymer-based NPs is the combination between polymeric and lipid-based NPs (Chan et al., 2010; Thevenot et al., 2007; L. Zhang et al., 2008). LPNs consist of a hydrophobic polymeric core, hydrophilic shell, and lipid shell in between the core and shell. The hydrophobic core acts to entrap poorly water-soluble drugs, whereas the hydrophilic shell enhances stability and half-life due to their antibiofouling properties, and the interface lipid shell acts as a protective wall to ensure the drug encapsulation efficiency, high loading yield, controlled release of drugs (Kamaly et al., 2016; L. Zhang et al., 2008), high immunocompatibility properties (e.g., complement and coagulation system activation and the ability to bind onto human plasma protein) (Salvador-Morales et al., 2009).

## 2.6 | Inorganic-organic hybrid NPs

The hybrid NFs are composed of binding an inorganic compound to an organic compound resulting in hybrid nanomaterial. Some of the best qualities of hybrid NPs include the ability to immobilize enzymatic activities, significantly better stability, physical and reproducibility properties compared to conventional materials. Some applications of hybrid NFs include stabilizing enzyme activity, drug delivery, biosensor, cell imaging, and other biomedical applications (S. Lee et al., 2015). Figure 2 shows an illustration of the combined nanotechnology between enzyme and NPs and their potential applications.

## 3 | ENZYME-INCORPORATED NANOMATERIALS IN SOCIETY

The enzymes that are incorporated in nanomaterials are utilized in various fields of society, such as food from agriculture, human health, and the environment.



**FIGURE 2** Illustration of enzyme-incorporated nanoparticle and its potential uses

### 3.1 | Agri-food

Global food production is an increasingly challenging task due to current risks including food safety, malnutrition, food wastage, and wastewater production (Henchion et al., 2019). Recent studies showed that nanoscale technologies can overcome the current challenges in agriculture and food-based industrial sector through sustainable growth (Handford et al., 2014; Parisi et al., 2015). The application of nanotechnology in agriculture can be classified into nano-inside, where the material is ingested by target species, and nano-outside, where it is used in/as the packaging material (Siegrist et al., 2008; Yang & Hobbs, 2020).

Nanotechnology-based products are beneficial in this sector by producing nutritious, high-quality, and safe foods (Gomez et al., 2021; Mousavi & Rezaei, 2011). The application of nanotechnology in the agri-food industry can be divided into five main sectors, such as primary production, food processing, nutrition, food safety, health, and packaging applications. The use of the nanomaterial in primary production, includes agrochemical delivery, pesticide detection, antimicrobial activity, genetic engineering, field sensing, diagnosis, and prevention of animal diseases (Gomez et al., 2021; Henchion et al., 2019). In food processing, nanotechnology is applied in improving the texture, aroma, and consistency of food, as an anticaking and gelation agent, and viscosity-increasing agents, whereas it is used to aid nutrient delivery, fortify

minerals and vitamins, senses characteristics of supplements and also as nutraceuticals in the nutrition sector. Further, nanotechnology in food safety and health improves shelf life, sensor diagnostics for contaminants, security devices, and disinfection products for surfaces and equipment (Handford et al., 2014; Henchion et al., 2019).

Diverse conjugations of organic-inorganic hybrid nanomaterials have been produced to increase plant protection through antimicrobials, nano-pesticide sensors, degradation, water treatment, as well as food safety. Furthermore, enzyme-incorporated hybrid nanomaterials have also been promising in food analysis and safety due to their excellent single-molecule and multiplex mycotoxin detection properties (Abd-Elsalam, 2020).

### 3.2 | Human health

Over the last six decades, there has been massive progress in the production and applications of biosensors. Biosensors are generally composed of three main components such as biological recognition compound, transducer, and processor (Nguyen et al., 2019). The progress of nanotechnology in human health, and ways to diagnose and prevent diseases have been excellent over the past years. Enzyme-incorporated hybrid nano-bio-sensors are analytical devices (derived biologically), that

are used to detect and sense various diseases and/or disorders, when combined with transducers to recognize and convert signals (Clark & Lyons, 1962; Hiratsuka et al., 2008; A. P. F. Turner, 2000; Updike & Hicks, 1967; Yoo & Lee, 2010).

The enzyme-incorporated hybrid nanomaterials in the production of biosensors and/or biomarkers are considered an alternative cutting-edge technology, compared to conventional methods, such as colorimetric sensors and protein biomarkers. Some advantages of the hybrid nanostructures include ultrasensitivity, improved viability, stability, and shelf-life of the enzymatic proteins (Shende et al., 2018). The use of enzyme-incorporated biosensors has shown promising results in the detection of common diseases such as Parkinson's, Alzheimer's, type 2 diabetes, and acquired immunodeficiency syndrome (AIDS) (El Harrad et al., 2018).

### 3.3 | Environment

Nanotechnology-based materials have also been studied and applied to control air pollution through sensing, removal, and/or adsorption of toxic gases present in the ambient air. Examples of toxic pollutants in the air include carbon dioxide (CO<sub>2</sub>), nitrous oxide (NO<sub>x</sub>), ammonia, dioxins, and volatile organic compounds (VOCs), which affect the composition of the breathable air depending on the threshold quantity. For instance, large quantities of dioxins present in the air affect the immune and endocrine system as well as are carcinogenic to human health. Therefore, enzyme-incorporated hybrid nanomaterials have been used as toxic gas sensors (e.g., phenolic compounds and hydrogen peroxide) to detect and eliminate them (Bollella & Katz, 2020).

Furthermore, the dye-processing industry is a major industry that contributes to more than 200,000 tons of discharge as aqueous effluent (Chequer et al., 2013) of which 10%–50% is lost in textile coloration (Mansour et al., 2012). However, the effect of dye (e.g., red 1, red 9, yellow 14, crystal violet, methylene blue, basic yellow) on human health is hazardous, as they can cause carcinogenic activity and allergic reactions (Lellis et al., 2019). NPs and composites have already been in use, however, it has a low adsorption efficiency (Sadegh et al., 2016). Thus, enzyme-incorporated hybrid nanotechnology implementation showed greater efficiency to remove dyes from wastewater effluents for overcoming the limitations of NPs (Azghandi et al., 2017).

## 4 | ENZYME-INCORPORATED NANOMATERIALS

### 4.1 | Mechanism of enzyme-incorporated nanomaterials

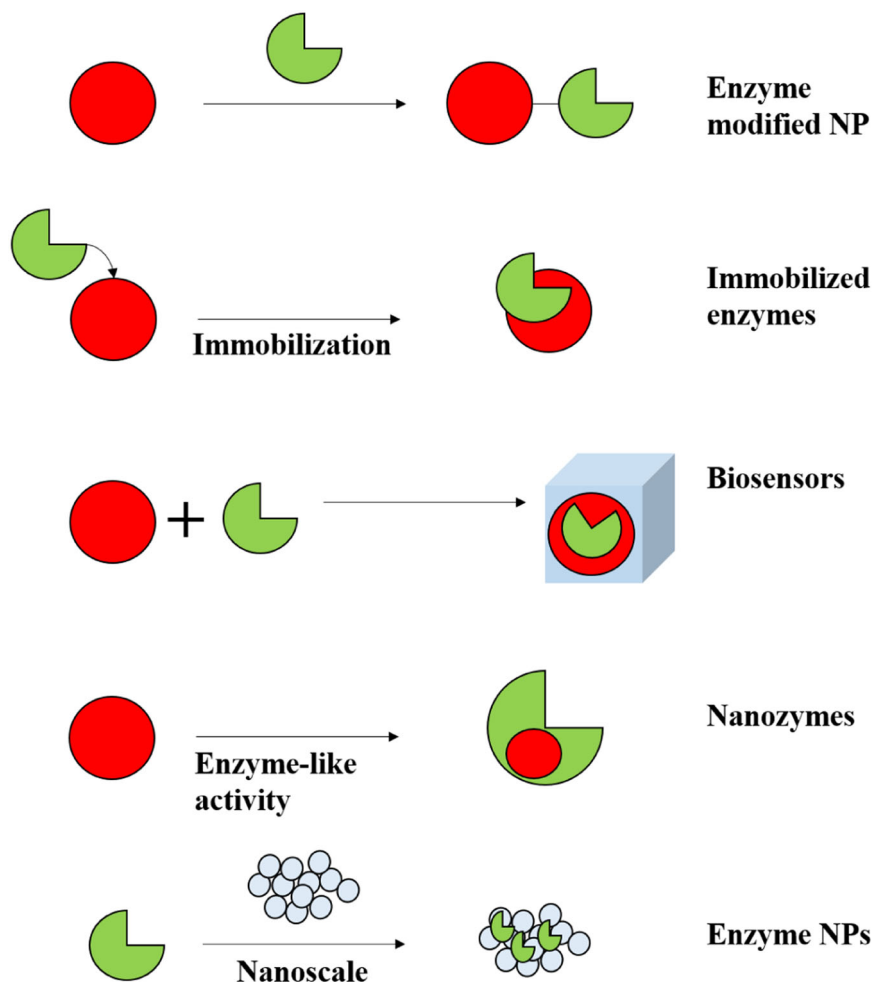
The interactions between enzymes and nanomaterials may have a greater impact on the chemical and physical properties of the enzymes or the nanomaterial. This is due to the binding nature of

enzymes onto the surface of nanomaterials which may potentially alter the structure, function, and properties of the nanomaterials. However, these potential changes may be different depending on the type of enzyme immobilization methods (e.g., physical or chemical). For example, an adsorption is a form of physical bonding, that generally causes conformational transitions, which would affect the catalytic activity of the enzymes. Some factors which may contribute to physical bond formation are the type of enzymes (e.g., amino acid composition, 3D structures, and orientation) and/or nanomaterials (e.g., size, structure, chemical properties) as well as environmental conditions (Castagnola et al., 2017; M. Chen et al., 2017; Johnson et al., 2014; X. Zhao et al., 2015). Further, enzyme orientations, typically affected by factors, such as physicochemical properties, pH, and temperature, are crucial, as improper orientations of the enzymes can impede and/or obstruct the active sites for enzymatic reactions (Johnson et al., 2014). Figure 3 shows probable pathways of the interaction between enzymes and NPs. There are various potential pathways of interaction between enzymes and NPs depending on the functionality, application, and uses. For instance, free enzymes can be encapsulated or combined with NPs to immobilize them for various functions. Nanozymes are NP-encapsulated materials that possess enzyme-like activity, whereas biodegradation is the use of enzymes to degrade NPs or nanomaterials. It is noteworthy that the interactional patterns between enzymes and nanomaterials depend on the physicochemical properties of enzymes (e.g., structure) and NPs (e.g., shape, size) (M. Chen et al., 2017).

The effect of varying conditions on the immobilization and the enzymatic reactivity is influential, where the distinct type of bond formations (e.g., physical, covalent, hybrid linking) shows significant changes in the overall enzymatic efficacy. One study by Asuri and coworkers showed that enzymes immobilized onto NPs of smaller diameters (resulting in higher curvatures) can retain better enzymatic activity. Higher curvatures (the radial distance between adjacent enzymes) can decrease or deter enzymes from each other preventing unnecessary interactions. A less curved surface could also potentially increase the enzyme denaturing process due to the interaction between enzyme and surface area of NPs (Asuri, Bale, et al., 2006; Asuri, Karajanagi, et al., 2006). Moreover, distinct bond formations affect the conjugates, whereby, past studies showed that physical binding alone has a lower enzymatic activity, compared to covalent as well as covalent binding with a linker (Garcia-Galan et al., 2011; L. Wang et al., 2010).

### 4.2 | Methods of incorporation of enzymes onto nanomaterials

There are different methods to synthesize enzyme-incorporated nanomaterials and the yield of the particles with unique structure, morphology, and size depends on the operating conditions (e.g., temperature, the concentration of components, duration of synthesis), presence of solvents, surfactants, additives, and/or polymers (Shende et al., 2018). The main techniques of enzyme immobilization



**FIGURE 3** Typical significant pathways of enzyme and nanoparticles interaction, Inspired by (M. Chen et al., 2017)

can be divided into physical and chemical bond formation as described in detail in this section.

### 4.3 | Physical bond formation

Physical bond formation can further be divided into adsorption and entrapment method. The adsorption method is relatively simple with high loading efficiency and feasible method, which depends on the physical mechanism based on the dipole attraction, hydrophobic attraction, Van der Waals forces, and/or hydrogen-based bond formation (Eş et al., 2015; Hwang & Gu, 2013; K. H. Kim et al., 2018). There are different methods of adsorption, such as (a) static process (b) dynamic batch process (c) reactor loading, and (d) electrodeposition. In the static process, the enzyme in a solution is allowed to be in contact with the carrier naturally, where the contact between enzyme and carrier is established through stirring when the carrier is placed inside a reactor. The electrodeposition process involves the use of current and it attracts the enzymes onto the carriers, that are placed close to the electrodes (K. H. Kim et al., 2018).

In addition, affinity binding uses the principle of selectivity between complementary biomolecules. The key advantages of this method are that it has high selectivity of interaction, the controlled

orientation of enzymes as well as minimal conformational changes (Brena & Batista-Viera, 2006; Reis & San Román, 2004). Affinity binding can be achieved in two ways (a) precoupling of support to ligand affinity and (b) an entity conjugated to an enzyme, which develops an affinity eventually (Datta et al., 2013). Coulombic, van der Waals and hydrogen bonding through bio-affinity layering can increase the affinity binding efficiency and reusability (Datta et al., 2013; Haider & Husain, 2008).

Adsorption is more commonly reversible as immobilized enzymes can be removed if/when enzyme activity decays over time. The process can be maneuvered under gentle conditions to regenerate and reload carrier/support with fresh enzymes (Brena & Batista-Viera, 2006; End & Schöning, 2004). Moreover, there are different types of carriers such as mineral, organic, or ion exchange resins. Some examples of carriers used are cotton fibers, gelatine, porous glass, and cellulose. However, adsorption bonding is not very stable due to weak bondings between enzyme molecules and the nanomaterial. Therefore, coatings would aid in stronger bondings and overall efficiency (K. H. Kim et al., 2018; Tang et al., 2004).

The entrapment method involves the entrapment of enzyme molecules through porous gel and/or fibers (Ahmad & Sardar, 2015b; Hwang & Gu, 2013). This method's advantages include providing a safe and confined environment that protects the enzymes from gas

bubbles, solvents, mechanical shear, high stability, simple mechanism, and continuous operations (Hwang & Gu, 2013). Entrapment can be conducted in three ways: inclusion in (a) gels (b) fibers, and (c) microcapsules. Some examples of carriers include polyacrylamide gels, gelatine, cellulose triacetate, agar, carrageenan, and alginate. Some key advantages of this method are that it reduces enzyme leaching and improves mechanical stability (Shen et al., 2011) ultimately reducing the potential denaturing of enzymes due to interaction with polymer (Datta et al., 2013). However, the disadvantage of this method are that it can be rigorous and has a mass transfer limitation (J. Kim et al., 2008; D.-H. Zhang et al., 2013). Other disadvantages include enzyme leaching typically due to irregular ratio of pores on support, enzyme deactivation while immobilization and reduced loading capacity, and support matrix abrasions (Górecka & Jastrzębska, 2011).

#### 4.4 | Chemical bonding

The covalent technique is the attachment of the enzyme onto the nanomaterial through covalent bonds (Ahmad & Sardar, 2015a). This technique has a strong bond formation between the enzyme and nanomaterial and therefore increases stability to be beneficial in various applications and industries (Eş et al., 2015; Hwang & Gu, 2013). The orientation of enzyme binding is a critical component that controls the stability of the covalent bond between enzyme and support. A study showed that when the active center amino acids are not implicated in the support binding, the highest level of enzyme activity is reached. Depending on the active groups present in the molecule to be immobilized, the coupling with the support can be accomplished in two ways. The reactive functional groups can be added to the support without any changes, or the nanomaterial can be changed to produce activated groups. The electrophilic groups formed on the support are expected to react with strong nucleophiles on the protein in both circumstances (Mohamad et al., 2015). When combined with other supports such as mesoporous silica, chitosan, and others, the approach boosts enzyme half-life and heat stability (Ispas et al., 2009). Because there is no barrier between the enzyme and the substrate, there is infinite covalent binding between them, which confers stability. The enzyme's location on the support's surface improves enzyme adhesion and the binding mechanism used to load the enzyme (Zhai et al., 2013). Covalent bonds can be formed via distinct methods including (a) diazotization (b) peptide bond formation, and (c) using polyfunctional reagents. The diazotization method involves creating bonds between amino groups and tyrosyl and/or histidyl groups from enzymes, whereas peptide bond formation is between amino group and enzyme and polyfunctional reagents create the bonds between amino groups of enzymes and carriers, such as cellulose and titanium oxide. Moreover, the disadvantage of this technique includes a decrease in enzyme mobility, reactivity as well as conformation restrictions (K. H. Kim et al., 2018).

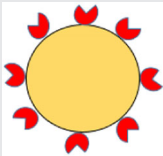
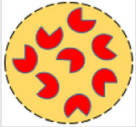
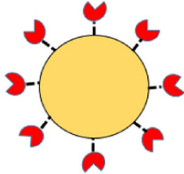
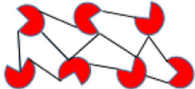
The cross-linking method involves the attachment of enzymes to each other using a reagent (Ahmad & Sardar, 2015a). Technically, cross-linking is accomplished by using bi- or multifunctional chemicals to produce intermolecular cross-linkages between enzyme molecules. Cross-linked enzyme aggregates (CLEAs) are made by aggregating enzymes in precipitants like acetone, ammonium sulfate, and ethanol, then adding a cross-linker (Hanefeld et al., 2009). CLEA was developed to improve the efficiency of enzyme immobilization whereby, enzyme precipitation from an aqueous solution and the combination of nonionic polymers, organic solvents, and/or salts (K. H. Kim et al., 2018; Sheldon, 2007). The reactions for enzyme immobilization can be carried out in three ways: mixing the prepolymers with a photosensitizer (e.g., benzoin ethyl ether), melting, mixing with an enzyme solution, and gelling by exposure to near ultraviolet (UV) radiation, or freezing the enzyme-containing monomer solution in the form of small beads. Then, the enzymes are either mixed in a buffered aqueous solution containing acrylamide monomer and a cross-linking agent before performing a chemically initiated polymerization, or the enzymes are mixed in a buffered aqueous solution containing acrylamide monomer and a cross-linking agent before performing a chemically initiated polymerization (Obzturk, 2001). Examples of reagents include glutaraldehyde and diazonium salts. The binding between glutaraldehyde and enzyme occurs between the aldehyde functional groups and amino group of enzymes. Glutaraldehyde is the most often used cross-linking reagent since it is inexpensive and readily available in large amounts (Górecka & Jastrzębska, 2011; Hanefeld et al., 2009; Sheldon, 2007).

The increased surface area and porosity, cross-linking enzymes to electrospun nanofibres has resulted in enhanced residual activity. Continuous antibacterial applications have also been described using lysozyme-immobilized electrospun chitosan nanofibres via CLEAs (Park et al., 2013). Advantages of this method include strong binding, preventing any leakage and desorption as well as good recovery and reusable values, whereas the disadvantage of this technique is that it reduces enzymatic activity due to the reagents used that may easily denature or modify (D.-H. Zhang et al., 2013). Table 1 shows the difference between all the four different enzyme immobilization techniques.

#### 4.5 | Self-assembly of enzymes and NPs

Noncovalent bonds, such as hydrogen bonds between a hydrogen atom and atoms of electronegative surface of the nanostructure can enhance the self-assembly of hybrid nanomaterials (L. Wang et al., 2019). Hydrogen bonds aid in the growth of biological components in one direction to form 1D structures, however, the hydrogen bonds provide cylindrical morphology for self-assembly in cyclic peptides (Rho et al., 2019). One study also revealed that hydrogen bonds formed between -NH group on peptide chains and carboxyl groups present on polymers, resulting in strong interaction without the need for external agents or binders (Agostinelli

TABLE 1 Difference between enzyme immobilization techniques

Enzyme immobilization technique		Advantage	Disadvantage	References
Physical bonding	Adsorption	<ul style="list-style-type: none"> <li>• Simple</li> </ul>	<ul style="list-style-type: none"> <li>• Unstable</li> </ul>	K. H. Kim et al. (2018); Zhang et al. (2013)
		<ul style="list-style-type: none"> <li>• High loading efficiency</li> </ul>	<ul style="list-style-type: none"> <li>• Weak bond formation</li> </ul>	
Chemical bonding	Entrapment	<ul style="list-style-type: none"> <li>• Simple, continuous operation</li> </ul>	<ul style="list-style-type: none"> <li>• Rigorous</li> </ul>	
		<ul style="list-style-type: none"> <li>• Protect enzyme activity</li> </ul>	<ul style="list-style-type: none"> <li>• Mass transfer limitation</li> </ul>	
Chemical bonding	Covalent	<ul style="list-style-type: none"> <li>• Strong bonding</li> <li>• High stability</li> </ul>	<ul style="list-style-type: none"> <li>• Decrease enzyme mobility and reactivity</li> </ul>	
		<ul style="list-style-type: none"> <li>• High stability</li> </ul>		
Chemical bonding	Cross-linking	<ul style="list-style-type: none"> <li>• Strong binding</li> <li>• Prevent leakage and/or desorption</li> </ul>	<ul style="list-style-type: none"> <li>• Decreased enzymatic activities</li> </ul>	
		<ul style="list-style-type: none"> <li>• Good recovery and reusability</li> </ul>		

et al., 2007). Van der Waals forces and dipole-dipole interactions are much weaker than hydrogen bonds and covalent bonds, however, they function to control self-assembly and any possible phase transitions (J. Cao et al., 2019; H.-Y. Gao et al., 2015).

Figure 4a shows hypothetical electrostatic interaction between NPs and enzymes. Electrostatic interactions are formed between negative and positively charged particles whereby, it has been identified to enhance self-assembly while maintaining the stability of the resultant hybrid NPs (Mauri et al., 2017). The strength of the interaction can be increased by varying the pH and/or charge screening of the ionic medium. Likewise, hydrophobic interaction-induced self-assembly of enzymatic-NPs has also been identified as a promising bond formation agent due to its excellent property to enhance secondary biomolecule structures into a specific cylindrical structure (Fu et al., 2014).

Covalent bonds, on the other hand, are required to form a more stable and stronger attachment between functional groups on enzymes and a particular surface. For instance, amine groups can react with the ester group to form amide bonds, thiol group, and unsaturated carbonyl groups form thioether bonds (Homaei

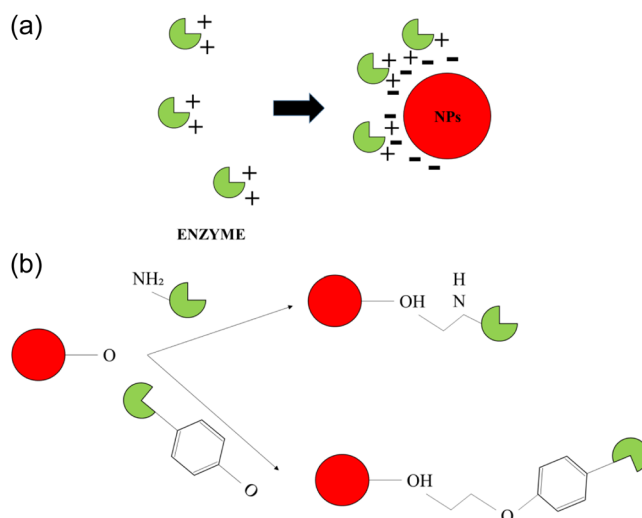


FIGURE 4 Hypothetical internal interactions between enzymes and nanoparticles (NPs) (a) electrostatic interaction (b) covalent linking through amino groups or phenolic groups of enzyme and NPs.



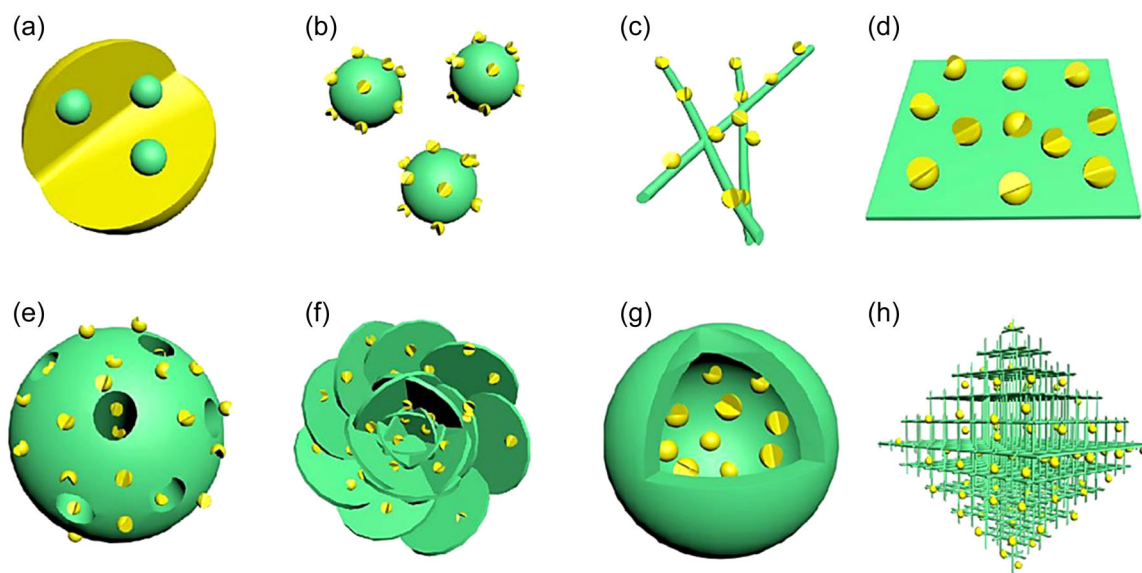
et al., 2013). Figure 4b shows hypothetical interaction between functional groups on enzymes and NPs' surfaces. Furthermore,  $\pi$ - $\pi$  bonds have been identified to possess major influence in enhancing the overall stability of structures, that are bound by noncovalent bonds (e.g., hydrogen, van der Waals). Also,  $\pi$ - $\pi$  stacking has been identified to cause fibrils that can reassemble a higher-order hierarchical system (J. Chen & Zou, 2019). In typical biomolecule self-assembly, the noncovalent interactions (charged or uncharged) form either electrostatic or hydrogen bonds, whereas the nonpolar amino acids form and stabilize through  $\pi$ - $\pi$  stacking or hydrophobic interactions (X. Ding & Wang, 2017). For example, 3D hybrid nanostructures, that are composed of pyrene-modified glucose oxidase/catalase-loaded graphene nanodots/gold electrodes, are stacked through  $\pi$ - $\pi$  bonds to exhibit better catalytic activity (up to 70%), compared to free standalone enzymes in the detection of glucose and  $H_2O_2$  in biological sample. It is noteworthy that the graphene-pyrene  $\pi$ - $\pi$  interactions can help to protect the pyrene conjugate structure while maintaining the electrical properties of graphene (J. Wang et al., 2015).

#### 4.6 | Factorial variation on the reactivity of enzymatic-NPs

The mechanism of the enzymatic-NPs' enhancement has improved the interaction between enzyme and nanomaterial, which eventually influences their rate of reactivity, as shown in Figure 5. Some factors which contribute to the enhancement of enzyme-NPs interaction include the morphology and structure of the nanomaterial, temperature, electron transfer efficiency, metal ion activation, conformational changes, and the possibilities of multienzyme systems.

Initially, different morphologies have distinct stability of the enzyme-incorporated nanomaterial, especially due to the difference in surface area and diffusion resistance (An et al., 2020). J. Li et al. (2016) developed spindle-like nanocrystals and DNA was encapsulated onto the structure, which showed a 143-fold increase in the enzymatic activity due to its larger surface area for a greater accumulation of compounds on their surface (Z. Li et al., 2016). Encapsulated enzymes also have shown increased catalytic activity due to protected enzymes against harsh external environmental conditions (M. Wang et al., 2015). Another study involved the synthesis of 3D nano shapes (flowers, plates, and parallel hexahedrons), where the NF structures showed the highest catalytic activity, due to the large surface-to-volume ratio, which reduces mass transfer limitations (L-B. Wang et al., 2013). In another study, nanowire meso-crystals resulted in one of the highest specific nano biocatalyst activity due to its octahedral structure which is developed via anisotropic nanowires, which has a large surface-to-volume ratio, interpenetrating channels, and decreases the overall mass transfer limitations (G. Li et al., 2018).

The use of metal salts with enzymes or proteins has been proven to be successful as metal ions can bind effectively with the enzyme/protein (which acts as precursors). Similarly, the incorporation of metal ions onto enzymes can enhance the electron transfer efficiency (Schwizer et al., 2018), where enzymatic redox reactions are often related to their electron transfer, particularly in oxidoreductase catalytic activities/processes (Wu et al., 2017). Conductive nanosized supports, such as gold (Au), cadmium sulfide (CdS), carbon dots, and/or tubes have shown promising results to support and improve the electron transfer of redox enzymes (An et al., 2020). Moreover, temperature plays a crucial role in the catalysis process as an increased temperature can enhance enzymatic activity. However, certain enzymes may denature at extreme temperatures, therefore, it is vital to learn the difference between enzymes and



**FIGURE 5** Hypothetical views on different morphologies between nanoparticles (NPs). (a) NPs on enzymes. (b–d) Enzymes are adsorbed and immobilized onto the surface of NPs. (e) Encapsulation of enzymes on/into porous nanomaterial. (f–h) Embedded enzymes on/into three-dimensional support matrices (An et al., 2020).

apply temperature accordingly (Rodrigues et al., 2013). Recently, it was identified that certain nanostructures can absorb light and/or electromagnetic waves and convert them into heat to enhance catalytic activity. Another study developed temperature-responsive enzyme-incorporated polymer as nano-catalysts, which showed excellent results at 40°C (J. Zhu et al., 2013). Photosensitive nanostructures, such as metal NPs and semiconductor nanomaterials also help to control the activation of light into heat strategy (C. Wang et al., 2017). Furthermore, magnetic NPs were also identified to possess the ability to convert electromagnetic energy into thermal energy. The use of magnetic nanomaterials further allows easier recovery of the nanostructures upon frequent usage (Alarcón-Correa et al., 2019). Later, Xiong et al. (2019) demonstrated that the ferromagnetic nano-ring-hybrid enzyme-incorporated structure can stimulate magnetic fields into localized heating, which improves reactivity by 1.8 fold (Xiong et al., 2019).

In addition, nanomaterial, or allosteric effectors can have desirable conformational changes in enzymes, which can improve catalytic activity (Darby et al., 2017). Certain metal ions such as calcium (Ca) and magnesium (Mg) have created major impacts as allosteric effectors (L.-B. Wang et al., 2013; M. Wang et al., 2015). Conformational changes within proteins or enzymes, unfortunately, may also lead to fibril formations from misfolded amyloid proteins from the works of Colvin & Kulinowski (2007) and thermodynamic instability making them prone to chemical denaturing (Chatterjee et al., 2010). This ultimately reduces the protein-protein interaction signals, which ultimately decreased the enzymatic activity of  $\alpha$ -chymotrypsin (CT) and soybean peroxidase (SBP) based on the works of Karajanagi et al. (2004). In addition, conformational epitopes are the alterations that can result in the specific folding of polypeptide chains. Nanostructures can either induce these novel epitopes or even unfold concealed epitopes originating from primary proteins (Saptarshi et al., 2013).

Multienzymatic systems have the involvement of more than one enzyme can also enhance the catalytic activity in enzymatic NPs (Vranish et al., 2017). This system has gained attention in cascade reactions to obtain desired substrates (Hwang & Lee, 2019). However, there are certain challenges in designing a multienzyme system, hence, it should be designed to reduce the movement of intermediate products between enzymes to improve overall activity, good spatial arrangement, number, and types of enzymes (H. Wang et al., 2018). Some examples of nanomaterials in designing multi-enzymatic systems for overall enhancement of enzymatic activity are metal-organic framework, a product of one enzyme that acts as a substrate for another (W.-H. Chen et al., 2018), and aggregation-induced NPs (S. Cao et al., 2021). A metal-organic framework is the addition of several enzymes onto NPs which acts as the nanoreactors (W.-H. Chen et al., 2018) whereby, the aggregation-induced-NPs multienzymatic framework can be described as the (a) formation of structural formation of NPs followed by (b) functionalization through enzymatic machinery layer-by-layer self-assembly process (S. Cao et al., 2021). Recent studies on the development of nanostructured hybrid particles and their increased enzymatic activity have been summarized in Table 2.

## 5 | ENZYME-INCORPORATED NANOMATERIALS IN VARIOUS APPLICATIONS

Recently, enzyme-incorporated nanomaterials are under extensive research to be beneficial in various applications, such as biocatalysts, biosensors, environment monitoring, drug delivery, and bioremediation.

### 5.1 | Biocatalysts

Nanosystems have been recently utilized as platforms to promote enzyme immobilization for ensuring efficient enzyme usage as the free enzymes often lose their activity easily involving high costs and low reusability value (Kuhad & Singh, 2013; R. N. Patel, 2008; N. J. Turner, 2009; Yu et al., 2005). Enzyme immobilization is the process of attaching enzymes over support to completely immobilize the attached enzyme (Chagas et al., 2015) and is highly beneficial as biocatalysts (Jeevanandam et al., 2022). It is often accomplished by four methods, namely, adsorption (K. H. Kim et al., 2018), entrapment, covalent bond formation (Ahmad & Sardar, 2015a), and/or cross-linking approach (Wong et al., 2019).

Some factors which affect the efficiency and performance of the enzyme immobilization process are the method of immobilization, type of immobilizer carriers (structure and property), and loading of enzymes (Talbert & Goddard, 2012). The type of carriers determines the mass transfer rate between the substrate and the enzyme, the interaction between molecules as well as the surface area for enzyme loading, and the electron transfer to the active site on the enzyme (H. Li et al., 2017). Immobilization allows the protection of enzymes from harsh conditions, such as pH, temperature, and type of solvent (D.-H. Zhang et al., 2013). Further, dye removal tests showed greater removal performance due to good enzyme adsorptive abilities (Pang et al., 2013; Pham et al., 2015), improved facilitation of direct electron transfer, good reusable property, and enzyme stability (Li et al., 2017). Table 3 shows the list of various types of enzymes and their support matrices as biocatalysts to immobilize enzymes.

### 5.2 | Biosensors

Enzyme-incorporated nanomaterials were widely utilized to fabricate biosensors for the effective monitoring of glucose in diabetic patients, biomarkers of cardiovascular diseases, cancer, other diseases, disorders, and ailments.

#### 5.2.1 | Monitoring of glucose in diabetic patients

Diabetic patients are required to control and monitor their daily sugar (glucose) intake to ensure a normal range of glucose levels in the blood to prevent severe complications, such as heart diseases, kidney failures, and loss of vision (Martín-Timón et al., 2014). Figure 6 shows

TABLE 2 Effect of various favorable interaction on the enhanced enzymatic activity

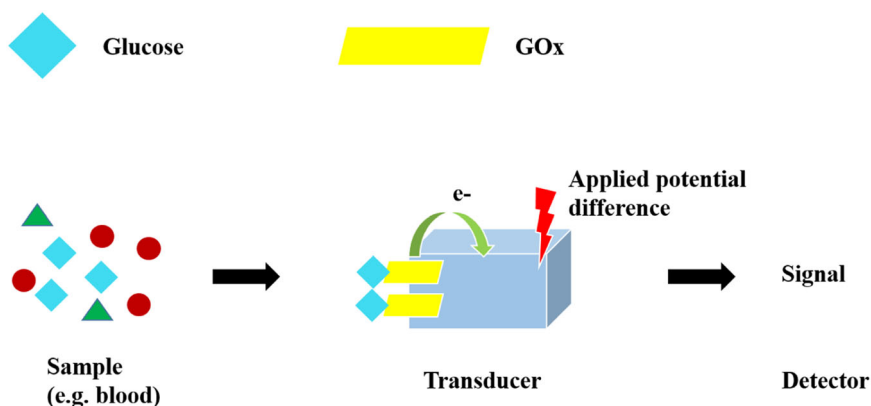
Enzyme	Support	Effect	Temperature (°C)	Increased activity (folds)	References
Laccase	Cu <sub>3</sub> (PO <sub>4</sub> ) <sub>2</sub> nanoflower	Cu <sup>2+</sup>	25	6.50	Ge et al. (2012)
Horse radish peroxidase	Cu <sub>3</sub> (PO <sub>4</sub> ) <sub>2</sub> nanoflower	Cu <sup>2+</sup>	Room temperature	5.06	Lin et al. (2014)
Laccase	Cu <sub>3</sub> (PO <sub>4</sub> ) <sub>2</sub> nanoflower	Cu <sup>2+</sup>	30	1.50	Batule et al. (2015)
Laccase	Au nanoparticle	Local plasma resonance effect	4	1.91	Guo et al. (2015)
Laccase	Carbon dots		4	1.92	H. Li et al. (2015)
Laccase	Cu <sup>2+</sup> /polyacrylic acid/poly(polyethylene glycol) acrylate	Cu <sup>2+</sup>	Room temperature	4.47	T. Chen, Xu, et al. (2017)
Laccase	Single-walled carbon nanotube		Room temperature	6.00	Wu et al. (2017)
Laccase	Cu <sub>3</sub> (PO <sub>4</sub> ) <sub>2</sub> hybrid microsphere	Cu <sup>2+</sup>	Room temperature	3.60	Rong et al. (2017)
$\beta$ -galactosidase	Mg-Al layered double hydroxide	Mg <sup>2+</sup> (allosteric effect)	4	30.00	Wang, Huang, et al. (2015)
$\alpha$ -chymotrypsin	Ca <sub>3</sub> (PO <sub>4</sub> ) <sub>2</sub> nanoflower	Ca <sup>2+</sup>	Room temperature	2.66	Yin et al. (2015)
Lipase	Cu <sub>3</sub> (PO <sub>4</sub> ) <sub>2</sub> nanoflower	Cu <sup>2+</sup>	4	4.60	Cui et al. (2016)
Organophosphorus hydrolase	Co <sub>3</sub> (PO <sub>4</sub> ) <sub>2</sub> ·8H <sub>2</sub> O nanocrystal	Co <sup>2+</sup>	25	3.00	L. Han & Liu (2017)
Carbonic anhydrase	Cu <sub>3</sub> (PO <sub>4</sub> ) <sub>2</sub> nanoflower, Ca <sub>8</sub> H <sub>2</sub> (PO <sub>4</sub> ) <sub>6</sub> nanoflower	Cu <sup>2+</sup> , Ca <sup>2+</sup>	4	2.86, 1.49	Duan et al. (2018)
D-psicose 3-epimerase	Co <sub>3</sub> (PO <sub>4</sub> ) <sub>2</sub> nanoflower	Co <sup>2+</sup>	4	7.20	L. Zheng et al. (2018)
Lipase	Carbon nanotube, Cu <sub>3</sub> (PO <sub>4</sub> ) <sub>2</sub> nanoflower	Cu <sup>2+</sup>	37, 25	68.00, 51.00	K. Li et al. (2018)
Laccase	Cu <sub>2</sub> O nanowire mesocrystal	Cu <sup>+</sup> , Cu <sup>2+</sup>	Room temperature	10.00	G. Li et al. (2018)
Laccase	Cu(OH) <sub>2</sub> nanocage	Cu <sup>2+</sup>	Room temperature	18.00	Silva-Torres et al. (2019)
Laccase	Fe <sub>3</sub> O <sub>4</sub> -NH <sub>2</sub> -PEI Fe <sub>3</sub> O <sub>4</sub> -NH <sub>2</sub>	Fe <sup>3+</sup>	25	101.33 74.45	Xia et al. (2016)
Glucose oxidase	Anodic alumina nanochannel		4	80.00	Mi et al. (2017)
Lipase	Zn <sub>3</sub> (PO <sub>4</sub> ) <sub>2</sub> hybrid nanoflower	Zn <sup>2+</sup>	30	1.47	B. Zhang et al. (2016)
Urease	Cu <sub>3</sub> (PO <sub>4</sub> ) <sub>2</sub> ·3H <sub>2</sub> O nanoflower	Cu <sup>2+</sup>	4	40.00	Somturk et al. (2016)
L-arabinitol 4-dehydrogenase, nicotinamide adenine dinucleotide oxidase	Cu <sub>3</sub> (PO <sub>4</sub> ) <sub>2</sub> ·3H <sub>2</sub> O nanoflower	Cu <sup>2+</sup>	4	2.46, 1.44	S. K. S. Patel et al. (2017)
Laccase	Copper alginate	Cu <sup>2+</sup>	4	3.00	S. Zhang, Wu, et al. (2018)
Hydroxylase	Cu <sub>3</sub> (PO <sub>4</sub> ) <sub>2</sub> ·3H <sub>2</sub> O nanoflower	Cu <sup>2+</sup>	Room temperature	1.62	Fang et al. (2018)

TABLE 3 List of various enzymes and support matrices as enzyme immobilizers

Enzyme	Nanomaterial	Type of immobilization	Application	Recyclability	References
Cellulase	Styrene/maleic anhydride copolymer NPs	Covalent bond	Catalytic hydrolysis of carboxymethylcellulose	80% after 10 cycles	Y. Wang et al. (2018)
Laccase	Polyurethane/amidoxime/polyacrylonitrile/ $\beta$ -cyclodextrin nanofiber membrane	Covalent bond	Carrier support for catalytic activity	47% after 10 cycles	Wu et al. (2018)
Lipase	Terpolymer poly (glycidyl methacrylate-co-methylacrylate)-g-polyethylene oxide nanofibrous membrane	Covalent bond	Hydrolysis of olive oil	45% after 5 cycles	X. Liu et al. (2018b)
Lipase	Poly (glycidyl methacrylate-co-methylacrylate)/feather polypeptide nanofibrous membrane	Covalent bond	Additive for catalytic activity	62% after 7 cycles	X. Liu et al. (2018a)
Lipase	Copper phosphate/carbon nanotube		Resolution reaction between 1-phenylethanol and vinyl acetate	97% after 8 cycles	K. Li et al. (2018)
Lipase	Chitosan-mesoporous silica/SBA-15 hybrid nanomaterial	Covalent/cross-linking	Hydrolysis of triacetin	85% after 10 cycles	Xiang, Ding, et al. (2018)
Lipase	Chitosan-mesoporous silica SBA-15 nanomaterial	Adsorption/cross-linking	Hydrolysis of triacetin	82% after 10 cycles	Xiang, Suo, et al. (2018)
Lipase	Zinc doped magnetic NPs	Adsorption	Hydrolysis of fish oil	>50% after 20 cycles	Verma et al. (2019)
$\alpha$ -chymotrypsin	Magnetic chitin nanofiber composite	Cross-linking	Support matrix for catalytic activity	84.9% after 20 days	Huang et al. (2018)
$\beta$ -galactosidase	Chitosan/polyvinyl alcohol blend nanofibers	Covalent bond	Catalytic activity	42% after 28 days	Haghju et al. (2018)

Abbreviation: NP, nanoparticle.

**FIGURE 6** Illustration of the working principle of an amperometric glucose biosensor



an illustration of the mechanism of amperometric glucose biosensors. Current enzymatic biosensors applied in the detection of blood glucose level includes the incorporation of glucose oxidase (GOx) due to high selectivity, ability to withstand higher pH and temperatures, and its cost-effectiveness (Shandilya et al., 2019).

For instance, GOx immobilized onto carbon nanotubes/Nafion matrix has been identified to be favorable due to the presence of carbon nanotubes, which act as the “electron pathways” with an electron transfer rate of  $1.47 \text{ s}^{-1}$  with better transportability and higher stability. The immobilization of enzymes via cross-linking approach also improved the overall sensitivity ( $16.26 \times 10^{-3} \text{ A}\cdot\text{M}^{-1}\text{cm}^{-2}$ ) of the biosensors (J. H. Kim et al., 2015). H. Lee and coworkers (2017) adapted a wearable sweat-based biosensor to detect glucose levels to better point-of-care in diabetic patients. The study showed good potential of the painless sweat-collecting combined with a transdermal drug delivery system. The electrodes are made from gold/GOx/Nafion NPs cross-linked with glutaraldehyde whereby, the limit of detection (LOD) was found to be  $1 \mu\text{l}$  (H. Lee et al., 2017). Another novel study developed a polyethylene terephthalate (PET) combined with gold NPs (GNPs) immobilized with GOx enzyme to detect glucose. The study showed an overall sensitivity of  $22.05 \mu\text{A}\cdot\text{mM}^{-1}\text{cm}^{-2}$  with a LOD of  $2.7 \mu\text{M}$  (Y. Wang et al., 2019).  $\text{MnO}_2$  NPs decorated on graphene-coated with GOx and Nafion showed good potential as disposable glucose sensors whereby, the linear range was from 0.1 to 1.4 mmol/L with a sensitivity of  $56.32 \mu\text{A}/\text{mmol}\text{cm}^2$  and LOD of 0.05 mmol/L (Vukojević et al., 2018). A novel approach using paper-based electrodes consisting of GNPs/graphene/GOx and conducting polymer, poly(9,9-di-(2-ethylhexyl)-fluorenyl-2,7-diyl)-end capped with 2,5-diphenyl-1,2,4-oxadiazole (PFLO) showed excellent potential as glucose biosensor whereby, the sensitivity was  $7.357 \mu\text{A}/\text{mM}\text{cm}^2$  with a LOD of 0.081 mM (Gokoglan et al., 2017). Another study involved the immobilization of GOx on graphene/multiwalled carbon nanotubes (MWCNTs)/GNP electrodes to improve the sensing of glucose, where the electron transfer rate was  $3.36 \text{ s}^{-1}$  and sensitivity of  $10 \mu\text{M}^{-2}\text{mM}$  with LOD of  $4.1 \mu\text{M}$  (Devasenathipathy et al., 2015). Furthermore, a novel study incorporated GOx onto tobacco mosaic virus nanotubes as carriers to improve the detection of glucose in biological samples. These biosensors exhibited 50%–60% of their initial enzymatic activity after 3 weeks (Bäcker et al., 2017).

## 5.2.2 | Biosensors in cardiovascular diseases

Cardiovascular diseases have been on the rise over the past decade and one way to curb or diagnose them at an early stage is through the detection of high levels of cholesterol in the blood. Recently, there are several novel enzymatic biosensors to detect cholesterol by using cholesterol oxidase on the surface of electrodes, which catalyses cholesterol into hydrogen peroxide ( $\text{H}_2\text{O}_2$ ). Cinti et al. (2015) immobilized cholesterol oxidase on the surface of screen-printed electrodes containing Prussian blue NPs and revealed that the sensitivity of the biosensor was  $2.1 \mu\text{A}/\text{mM}\text{cm}^2$  (Cinti et al., 2015). Another study involved the use of cholesterol oxidase and cholesterol esterase onto GNPs/screen-printed electrodes, where the detection limit was identified to be  $3.0 \mu\text{g}/\text{ml}$  (Huang et al., 2017). Other innovative biosensors include the incorporation of screen-printed electrodes with graphene oxide and iridium oxide NPs and tyrosinase to detect angiotensin-converting enzymes for the treatment of heart failures by inhibiting the tyrosinase and thioquinone enzymatic activity (Kurbanoglu et al., 2017).

## 5.2.3 | Other medical applications

The use of nanosized biosensors in the detection and monitoring of antidepressants by immobilizing monoamine oxidase on MWCNT-modified screen-printed electrodes to detect chemicals present in common antidepressant drugs (detection limit of  $6 \times 10^{-10} \text{ M}$ ,  $8 \times 10^{-10} \text{ M}$  and  $9 \times 10^{-10} \text{ M}$  for phenazepam, imipramine and afobazole respectively), such as imipramine, afobazole, and phenazepam (Medyantseva et al., 2015). The same team also developed silver NPs incorporated biosensors to detect imipramine and amitriptyline with a detection limit of  $4 \times 10^{-9} \text{ M}$ . The use of silver NPs aided in wider concentration limits as well as improved the overall sensitivity of the sensor (Medyantseva et al., 2015). Another study involved the synthesis of screen-printed electrodes, that are incorporated with carbon nanotubes and graphene oxide in the monitoring of moclobemide and amitriptyline concentrations in pharmaceutical drugs (Brusnitsyn et al., 2016). Similarly, Medyantseva and team (2017) successfully synthesized gold and

silver NPs modified biosensors, which were able to detect moclobemide, tianeptine, amitriptyline in urine and drugs with a detection limit of  $8 \times 10^{-10}$  M (Medyantseva et al., 2017). Another study involved the use of magnetic NPs and tyrosinase on screen-printed electrodes to detect methimazole, where the detection limit was identified to be 0.006  $\mu$ M (Kurbanoglu et al., 2015). Table 4 is a summary of the enzyme-based biosensors in the medical and pharmaceutical industries.

### 5.2.4 | Nano-based biosensors for environmental monitoring applications

In recent times, environmental protection is highly essential due to the presence of several toxins and pesticides in water bodies and soil as a result of the agriculture and industrialization process to meet the global demand of consumers (Justino et al., 2017). For instance, the detection of various pesticides through the incorporation of acetylcholinesterase enzyme has been identified to be a promising environmental monitoring agent.

Lang and team (2016) demonstrated the detection of paraoxon pesticides by synthesizing biosensors from glassy carbon electrodes/gold nanorods/acetylcholinesterase with a detection limit of 0.7 nM and an average recovery of 97% (Lang et al., 2016). Another study included the detection of methyl parathion from carbon paste electrode/chitosan/GNPs/Nafion with a detection limit of 5 fg/ml (Y. Deng et al., 2016). In addition, Wei and their team (2015) produced ionic liquids/GNPs/carbon composite/acetylcholinesterase to detect dichlorvos with a detection limit of 0.3 pM and recovery of 80.8%–93.1% (Wei & Wang, 2015). Furthermore, Zhang and the team (2021) synthesized reduced graphene oxide (GO)/titanium oxide nanorods and acetylcholinesterase to detect organophosphorus pesticides (dichlorvos [DDVP]) in waste samples. The sensor showed a response between 226 and 565 nM with a detection limit of 2.23 nM where it showed 78% of the initial state on Day 30 (J. Zhang et al., 2021). Another study successfully developed acetylcholinesterase on concanavalin A (Con A)/polydopamine (PDA)-reduced graphene oxide (RGO)-GNP nanostructures to detect carbofuran. The sensor was identified to have a detection range of 5–40  $\mu$ g/kg with a detection limit of 0.012  $\mu$ g/kg (Y. Li et al., 2019). Moreover, carbofuran has been successfully detected through the formation of glassy carbon electrodes/graphene oxide/MWCNTs (Zhuang et al., 2017). Later, Q. Liu and their team (2015) synthesized MWCNTs/graphene oxide/nanoribbons structure to detect carbaryl pesticides (Q. Liu et al., 2015).

Similarly, other innovative enzyme-incorporated biosensors include the addition of butyrylcholinesterase enzyme on screen-printed electrodes/carbon black NPs to detect paraoxon with a detection limit of 5  $\mu$ g/L and recovery of 96% (Arduini et al., 2015). Another study also revealed the usage of poly(3,4-ethylenedioxythiophene), graphene oxide nanosheets, and laccase on glassy carbon electrodes to detect catechol in contaminated water samples, whereby the study showed a 0.032  $\mu$ M of detection limit (Maleki

et al., 2017). Table 5 shows the summary of different nanostructured biosensors that are successfully fabricated to aid in environmental monitoring and protection.

### 5.2.5 | Nano-based biosensors for food safety

Food safety was introduced to preserve food by suppressing spoilage, contamination, as well as enhancing tenderness (of meat and muscle products) (Kerry et al., 2006). Nanosensors can detect toxins, pesticides, microbes, and infections through the change in color, taste, odor, and flavor (He et al., 2019). Besides, the incorporation of nanosensors in food packaging allows tracing of physical, chemical, and biological modifications during processing (Berekaa, 2015) as well as provides information on the quality of food during transportation and storage of product (Augustin & Sanguansri, 2009). Custom-made nanosensors are also able to detect pathogens and toxins (Berekaa, 2015). The advantages of enzyme-based nano-sized biosensors in food safety are because of their high level of sensitivity, selectivity, rapid response, and nanoscale compact size (Cavalcante et al., 2021). For instance, Navarro and coworkers (2020) developed immobilized tyramine oxidase on GNPs to detect tyramine in emmental cheese whereby the overall sensitivity of the biosensor lay between 0.033 and 0.25  $\mu$ M with a LOD of 2.9  $\mu$ M (Navarro et al., 2020).

Scombroid poisoning due to the presence of histamine; typically synthesized from the decarboxylation of histidine, is a well-known toxicant (Shkodra et al., 2020). Therefore, some recent studies have shown how biosensors can detect high levels of histamine as preventative care. Another study revealed that indium tin oxide NPs decorated with diamine oxidase showed good potential to detect histamine in kashar cheese. The biosensor showed a linear range of sensitivity of 0.069–6  $\mu$ M and LOD of 2.7  $\mu$ M (Kaçar et al., 2020). Shkodra and coworkers (2020) developed CNTs coated with horseradish peroxidase to detect histamine in fish whereby, the biosensor's linear range of detection lay between 0.005 and 50 ng/ml<sup>-1</sup> with a LOD of 2.48 pg/ml<sup>-1</sup> (Shkodra et al., 2020). Y. Zheng and their team (2015) synthesized enzyme-incorporated nanocomposite; acetylcholinesterase/glassy electrodes/ionic liquids, and gelatine to detect levels of carbaryl and monocrotophos found in tomato juice samples. The percentage of detection of carbaryl and monocrotophos in the sample was found to be between 92.5%–105% and 91.2%–110%, respectively (Y. Zheng et al., 2015). Another study using acetylcholinesterase incorporated onto Pt NPs/UiO66-NH<sub>2</sub> support matrix showed high sensitivity and detection of malathion in cabbage and apples with a detection limit of  $4.9 \times 10^{-15}$  M with an average recovery rate of 95% (L. Ma et al., 2019).

Furthermore, ethanol detection is vital during the fermentation process for the production of fermented beverages and/or products (Cavalcante et al., 2021). There have been several studies on how enzyme-incorporated nano-biosensors have been found promising. For example, alcohol dehydrogenase/diamond NPs on phenothiazine support matrix (Revenga-Parra et al., 2020), screen-printed electrodes

TABLE 4 Summary of enzyme-incorporated biosensors in medical and pharmaceutical applications

Application	Organic component	Biosensor materials	Detection	References
Antidepressant drugs	Monoamine oxidase	MWCNTs/screen-printed electrodes	• Imipramine	Medyantseva, Brusnitsyn, Varlamova, Beshevets, et al. (2015)
			• Afobazole	
			• Phenazepam	
Monoamine oxidase	MWCNTs/silver NPs screen-printed electrodes	• Imipramine	Medyantseva, Brusnitsyn, Varlamova, Maksimov, et al. (2015)	
		• Amitryptiline		
		• Moclobemide		
Monoamine oxidase	CNTs/graphene oxide/screen-printed electrodes	• Amitryptiline	Brusnitsyn et al. (2016)	
		• Moclobemide		
Monoamine oxidase	CNTs/graphene oxide/gold and silver NPs/screen-printed electrodes	• Moclobemide	Medyantseva et al. (2017)	
		• Amitryptiline		
		• Tianeptine,		
Congestive heart failure treatment	Tyrosinase	Magnetic NPs/screen-printed electrodes	• Amitryptiline	Kurbanoglu et al. (2015)
			• Methimazole	
			• Cholesterol	
Diabetes	Glucose oxidase	Prussian blue screen-printed electrodes	• Cholesterol	Cinti et al. (2015)
			• Angiotensin converting enzymes	
			• Glucose	
Diabetes	Glucose oxidase	Gold NPs/screen-printed electrodes	• Angiotensin converting enzymes	Huang et al. (2017)
			• Glucose	
			• Angiotensin converting enzymes	
Diabetes	Glucose oxidase	Graphene oxide/iridium oxide NPs/screen-printed electrodes	• Angiotensin converting enzymes	Kurbanoglu et al. (2017)
			• Glucose	
			• Angiotensin converting enzymes	
Diabetes	Glucose oxidase	CNTs/nafion	• Glucose	J. H. Kim et al. (2015)
			• Glucose	
			• Angiotensin converting enzymes	
Diabetes	Glucose oxidase	Graphene/MWCNTs/gold NPs	• Glucose	Devasenathipathy et al. (2015)
			• Glucose	
			• Angiotensin converting enzymes	
Diabetes	Glucose oxidase	Tobacco mosaicic nanotubes	• Glucose	Bäcker et al. (2017)
			• Glucose	
			• Angiotensin converting enzymes	

Abbreviations: MWCNT, multiwalled CNT; NP, nanoparticle.

**TABLE 5** Summary of enzyme-incorporated biosensors in environmental protection

Organic component	Biosensor materials	Detection	References
Acetylcholinesterase	Glassy carbon electrodes/gold nanorods	Paraoxon	Lang et al. (2016)
	Carbon paste electrode/chitosan/gold NPs/Nafion	Methyl parathion	Y. Deng et al. (2016)
	Ionic liquids/gold NPs/carbon composite	Dichlorvos	Wei and Wang (2015)
	Glassy carbon electrodes/graphene oxide/MWCNTs	Carbofuran	Zhuang et al. (2017)
	MWCNTs/graphene oxide/nanoribbons structure	Carbaryl	Q. Liu et al. (2015)
Butyrylcholinesterase	Screen-printed electrodes/carbon black NPs	Paraoxon	Arduini et al. (2015)
Laccase	Poly (3,4-ethylenedioxy-thiophene)/graphene oxide nanosheets/glassy carbon electrodes	Catechol	Maleki et al. (2017)

Abbreviations: MWCNT, multiwalled CNT; NP, nanoparticle.

**TABLE 6** Summary of enzyme-incorporated biosensors in food safety

Organic component	Biosensor materials	Detection	References
Acetylcholinesterase	Glassy electrodes/ionic liquids/gelatine	<ul style="list-style-type: none"> <li>• Carbaryl</li> <li>• Monocrotophos</li> </ul>	Zheng et al. (2015)
	Pt NPs/UiO66-NH <sub>2</sub> support matrix	<ul style="list-style-type: none"> <li>• Malathion</li> </ul>	L. Ma et al. (2019)
Alcohol dehydrogenase	Diamond NPs on phenothiazine support matrix	<ul style="list-style-type: none"> <li>• Ethanol</li> </ul>	Revenga-Parra et al. (2020)
	Screen-printed electrode/gold NPs/MWCNTs/polynuclear red film		Bilgi and Ayranci (2016)
	Fe <sub>3</sub> O <sub>4</sub> @Au NPs		Samphao et al. (2015)
Alcohol oxidase	Polyfluorene-g-polyethylene glycol/MWCNTs		Bekmezci et al. (2020)
Diamine oxidase	Indium tin oxide NPs	<ul style="list-style-type: none"> <li>• Histamine</li> </ul>	Kaçar et al. (2020)
Horseradish peroxidase	CNTs		Shkodra et al. (2020)
Tyramine oxidase	Gold NPs	<ul style="list-style-type: none"> <li>• Tyramine</li> </ul>	Navarro et al. (2020)

Abbreviations: MWCNT, multiwalled CNT; NP, nanoparticle.

modified with GNPs and MWCNTs/polynuclear red film/alcohol dehydrogenase (Bilgi & Ayranci, 2016), polyfluorene-g-polyethylene glycol/MWCNTs/alcohol oxidase (Bekmezci et al., 2020) and the immobilization of alcohol dehydrogenase on Fe<sub>3</sub>O<sub>4</sub>@Au NPs (Samphao et al., 2015). Table 6 shows the summary of the enzyme-incorporated biosensors using nanomaterials.

## 5.3 | Drug delivery

### 5.3.1 | Controlled site-specific drug delivery

Certain types of drugs have poor water solubility, biocompatibility, and controlled release of the agent, and thus, require a good carrier to ensure the drug is delivered to the specific site. Conventional drug delivery systems are not recommended for large-scale biomedical applications as it has toxic side effects. Therefore, NP-based drug delivery systems were employed recently, due to the specificity, dosage quantity, and its damage control efficiency. Uricase and HRP combined with calcium hydrogen phosphate (CaHPO<sub>4</sub>) nanofibers

(NFs) have shown promising results in the transdermal delivery of drugs in hyperuricemia-diagnosed patients (Hao et al., 2019).

In recent times, protease-responsive nanomaterials have been identified to be promising in targeted and controlled drug delivery applications. For example, a recent study developed a novel system consisting of enzyme-responsive activatable protein NPs coated with polyethylene glycol to deliver therapeutic drugs. The self-assembled NPs consist of therapeutic drugs (Melittin; anticancerous agent), that are embedded with peptides and would be activated in the presence of protease in an environment of diseased cells and/or tissues. The advantages of this system are its prolonged in vivo circulation, less toxicity, targeted, and controlled delivery of Melittin (Yu et al., 2018). In another study, peptides embedded with therapeutic drugs (cisplatin, adjuvin, and WKYMVm hexapeptide) were synthesized for triple-negative breast cancer drug therapy via transformable spherical NPs, that are transformed into nanorod structures upon addition of metalloproteinase-2 (MMP-2) sequence. This novel system showed a prolonged circulation time, targeted and controlled drug delivery, and deeper tumor penetration capability. The WKYMVm was also identified to increase the immune system by forming stable peptide interaction with



cancerous cells (C. Xu, Yu, et al., 2019). Furthermore, Ding and team (2017) developed a drug delivery system for breast cancer immunotherapy by synthesizing acrylamide polyethylene glycol copolymer-phenyl vinyl ethylene carbonate (PEGA-pVEC) peptides, hyaluronic acid, silicon dioxide (SiO<sub>2</sub>) NPs to deliver small interfering ribonucleic acid (siRNA) and anticancer drugs for the treatment of breast cancer cells *in vivo*. The system delivered targeted and controlled release of drugs in the presence of hyaluronidase due to degradation from the entry of lysosomes proving the efficiency in drug-resistant cancers (J. Ding et al., 2017). Another novel study showed that cadmium selenide-zinc selenide (CdSe/ZnS) quantum dots successfully delivered anti pancreatic cancer drug (gemcitabine) through polyethylene glycol decorated quantum dots from links of matrix metalloproteinase-9 (MMP-9) and cathepsin-B peptides. The two-linkage system showed a higher accumulation of gemcitabine, lower toxicity levels, and increased tumor penetration capability (H. Han et al., 2017). Other innovative drug delivery systems, include the development of transformable core-shell nanocarriers into micro-sized extracellular depots to suppress tumor sites with the incorporation of transglutaminase and human serum albumin (HSA) loaded on the support matrix. In the case of hyaluronidase overexpression, the anticancer therapeutics would be unloaded through the degradation of the cross-links between hyaluronic acid and micro depot systems (Hu et al., 2016).

### 5.3.2 | DNA functionalizing and programming for biomedical application

DNA is highly biocompatible and water-soluble, where it is used as building blocks to synthesize DNA nanostructures to be used in different medical applications. Conventional DNA strands have weak structures and may suffer structural integrity at low concentrations, which makes it difficult for therapeutic and imaging applications. The main concern regarding the DNA structures is mutations within the structure, whereby, mutations can take place through the addition or deletion of base pairs and/or DNA segments within chromosome structure (Mahdih & Rabbani, 2013). Moreover, the synthesis of conventional bulky DNA strands is more expensive and time-consuming than NFs. Therefore, DNA NFs have been developed through liquid crystallization (an anisotropic process that aligns concentrated polymers in order) and dense packaging of DNA building blocks (Lv et al., 2015). The noncovalent interactions in nanostructures resulting from vertices aid in the overall stability of the 3D DNA segments (Greschner et al., 2013). This synthesis process helps to improve the drug delivery system due to its two-fold strand, high biocompatibility, ability to program DNA, high drug tracing, loading, and controlled release to be beneficial as an efficient drug delivery system (Shende et al., 2018). One study showed the sequence binding of functionalized ligand onto HSA showed increased stability from the nucleases (otherwise known as, DNA hydrolyzing enzyme) and circulatory time (Jiang et al., 2012). Liu and coworkers (2011) found the coating of DNA nanostructures with HSA showed an increase of 13%–17% uptake of cells while maintaining low immunogenicity, degradation, and dissociation (X. Liu et al., 2011).

There have been several advancements over the years in the functionalization and preprogramming of enzyme-incorporated DNA nanostructures. For instance, Yang and coworkers (2018) developed DNA-oligonucleotide immobilized trypsin on polydopamine-modified magnetic NPs whereby, the DNA linkers aided in the overall catalytic enhancement and reusability values. The study showed the immobilized trypsin maintained 55% of its initial activity after 70 cycles and specificity constant which was 4.7-fold greater than that of free trypsin (Yang et al., 2018). Ngo and coworkers (2016) developed xylose reductase (XR)/xylitol dehydrogenase (XDR) cascaded reaction on a DNA origami structure. The novel system uses DNA binding adaptors (e.g., zinc finger protein and basic leucine zipper protein) to attach enzymes onto the origami resulting in a cascaded reaction. The metabolic pathway begins when XR converts xylose into xylitol in the presence of nicotinamide adenine dinucleotide cofactor. The second enzyme, XDR, then converts xylitol into xylulose in the presence of NAD<sup>+</sup> (Ngo et al., 2016). Another study by Liu and coworkers (2016) successfully synthesized a triple enzyme cascade system using malate dehydrogenase/oxaloacetate decarboxylase/lactate dehydrogenase to convert malic acid into lactic acid across three cycles. The study showed that the triple cascade system depended highly on the geometric arrangements of enzymes at short distances (10–30 nm) whereby, upon optimization, the overall catalytic activity showed a fivefold enhancement in comparison to free enzymes (M. Liu et al., 2016).

Besides, it is worthy to note the protection mechanisms of enzyme delivery to ensure high efficiency and good cellular delivery. A novel approach to coat  $\beta$ -galactosidase with DNA strands (oligonucleotides) showed that coating enzymes with DNA strands increased their catalytic functions significantly after the process of transfection (Brodin et al., 2015). Another study developed self-assembled DNA cages which are responsible for the trapping and releasing of horseradish peroxidase through conformational change, which can be controlled by temperature. The enzyme could enter and exit at 37°C but remained trapped at 4°C (Juul et al., 2013). Kohman and coworkers (2016) used the light-dependent (pulses of <60 s of light) approach to release bovine serum albumin-bound within the DNA origami structures (Kohman et al., 2016). Another approach of developing half-cages that can close into a shield-like box to store GOx and horseradish peroxidase showed good potential to enhance the catalytic activity of the enzymes and stability against proteases. The walls of the cages allowed for the entry and exit of substrates and products, however, blocks proteases (Z. Zhao et al., 2016).

### 5.4 | Pollutant removal and bioremediation

Conventional and environmental endangering methods of toxin removal and/or disposal are mainly through burying into the ground or dumping into the ocean. Newer methods such as incineration, dechlorination, and UV oxidation have been implemented, however, it is complex and has a larger cost (Karigar & Rao, 2011). These methods are highly toxic to the environment and can be a high potential health hazard to the ecosystem. Therefore, scientists have developed novel systems consisting of

biological components (e.g., microbial enzymes, fungi, bacteria, plants, algae) to convert contaminants into non or less hazardous wastes, over the past decade (Vidali, 2001). However, there are research gaps in the field of enzymatic-nanosystem-based water treatment methods as studies are dispersed and experimental in nature to exhibit bio-remediation in lab-scale conditions only.

#### 5.4.1 | Wastewater detoxification

All industrial manufacturers would contribute to the formation of toxic wastewater, which contains hazardous chemical components, including heavy metals, such as zinc, lead, chromium, and copper (J. Ma et al., 2017; J.-N. Zheng et al., 2014). Recently, scientists have initiated incorporating biological components into an existing nanosystem to enzymatically degrade or decompose wastes before disposing of the waste (Karigar & Rao, 2011). Figure 7 shows an illustration of the wastewater treatment mechanism using enzyme-incorporated NPs.

For instance, phenols can be produced using wastewater effluents from various industries, such as textiles and dyes, petroleum and coal, plastic as well as organic chemical industries. Conventional methods of phenol removal are less efficient and incomplete, therefore, researchers have utilized alternative approaches based on polyphenol oxidase for enzymatic degradation of phenols (Agarwal et al., 2016). A recent study showed highly efficient removal of phenolic compounds using tyrosinase immobilized iron oxide NPs, where the phenol degradation was identified to be >70% with a reusability value of 58% after seven cycles. The synthesized nano-bio-catalyst also was tested in a real water sample containing phenol and was identified to be able to degrade phenol up to 78% after 60 min of the incubation period (Abdollahi et al., 2018).

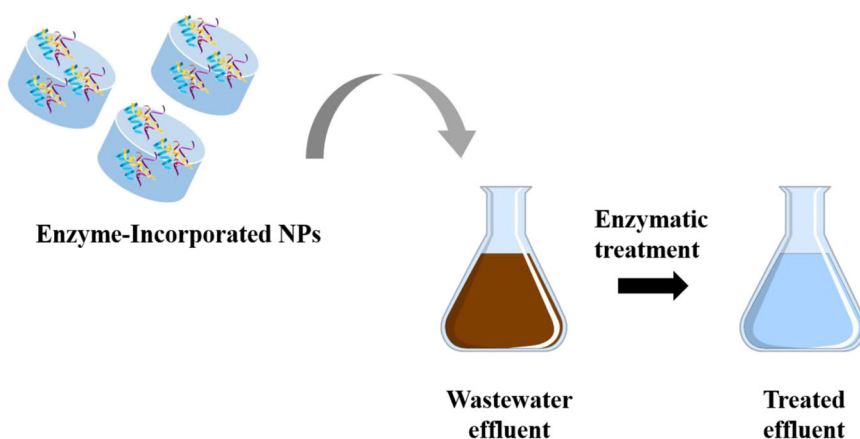
Zhang and team (2020) successfully developed a laccase immobilized chitosan/magnetic iron oxide ( $\text{Fe}_3\text{O}_4$ ) NPs for the removal of 2,4-dichloro phenol, and 4-chlorophenol, where the degradation was identified to be 91.4% and 75.5% after 12 h. The study also showed effective removal up to 10 cycles (K. Zhang et al., 2020). Another study by Qiu and coworkers showed good degradation efficiency of phenol (86.1%), 2,4-dichloro phenol (100%),

and 4-chlorophenol (93.6%) pollutants using laccase cross-linked on iron magnetic NPs using dialdehyde starch (X. Qiu et al., 2020, 2021).

Das and coworkers (2020) developed covalently bonded laccase on iron oxide NPs which showed good potential to degrade chlorpyrifos. The study showed a high sorption coefficient, 112.3 L/kg, and nitrogen content of 0.21%–0.22%. The high affinity of pesticide to soil and low nitrogen content within the column also describes the degradation, which ultimately showed the excellent potential of the novel system (Das et al., 2020). Another study using laccase immobilized on iron magnetic NPs also showed good potential for degradation of chlorpyrifos whereby, the high-performance liquid chromatography (HPLC) results showed four major peaks at retention time, 2.810, 2.962, 4.425, and 4.760 min. The results obtained showed two more major peaks than free enzymes (2.810 and 2.302 min) proving a higher degradation rate using immobilized laccase (Srinivasan et al., 2020). Besides, lignin peroxidase immobilized onto  $\text{Fe}_3\text{O}_4/\text{SiO}_2$ /polydopamine NPs have been identified to be promising for reducing organic compounds, such as dibutyl phthalate, phenol, tetracycline, and 5-chlorophenol, phenanthrene, fluoranthene, and benzo(a)pyrene, where the study showed 100%, 100%, 100%, 100%, 79%, 73% and 65% degradation and/or adsorption, respectively (Guo et al., 2019). Ranjan and team (2019) successfully synthesized cyanate hydratase onto an iron oxide/MWCNTs system to remove metal ions, such as copper (29.63%), iron (35.53%), chromium (39.31%), and lead (34.48%) from synthetic wastewater samples (Ranjan et al., 2018, 2019). Furthermore, Wang and coworkers (2019) developed metal-ion chelated magnetic NPs immobilized with bacterial prolidase to hydrolyze organophosphorus compounds such as methyl and ethyl paraoxon. The study showed His6-tagged *Escherichia coli* prolidase (His6-EcPepQ) immobilized onto silica-coated magnetic NPs had a maximum reaction rate of 17.48 and 6.31  $\mu\text{M min}^{-1}$  for methyl and ethyl paraoxon respectively (T.-F. Wang et al., 2019).

#### 5.4.2 | Dye removal

The textile industry contributes greatly to the production of dyes in wastewater effluents. Sarkar and team (2020) demonstrated that the



**FIGURE 7** Illustration of enzymatic treatment of wastewater effluent

most efficient and suitable enzymes in the degradation of dyes were laccase, azoreductase, and peroxidase, where these enzymes present as a multicopper oxidative, flavin mononucleotide (FMN) dependent, and  $\alpha - \beta$  with heme group, respectively. All these three enzymes showed excellent thermal stability and water interaction ability (Sarkar et al., 2020). For instance, Q. Zhu and coworkers (2022) developed immobilized laccase on  $\text{Fe}_3\text{O}_4@\text{SiO}_2$  NPs, which showed outstanding removal rate of malachite green, brilliant green, reactive blue 19, azophloxine, procion red MX-5B, and alizarin red dye at 98.7%, 99.3%, 88.8%, 79.0%, 78.7%, and 64.4% after 10 cycles whereby, the activity recovery of laccase was 109.7% (Q. Zhu et al., 2022). Z. Li and team (2020) have developed laccase immobilized on the surface of iron oxide/carbon/copper ( $\text{Fe}_3\text{O}_4/\text{C}/\text{Cu}^{2+}$ ) composite to degrade synthetic dyes, such as reactive blue 19, crystal violet, azophloxine, brilliant malachite green, and Procion MX-5B dye at 65%, 71%, 78%, 80%, 94%, and 60% after 10 cycles, respectively (Z. Li et al., 2020). Recently, a novel in-vault NP combined laccase system has shown good removal of reactive blue 19 and acid orange 7, whereby, the study showed 90% decolorization after 8 and 24 h, respectively (Y. Gao et al., 2022). Another study utilized laccase incorporated chitosan/ $\text{Fe}_3\text{O}_4$  nanosystem showed potential long-term efficiency in the removal of textile dyes; Reactive Blue 171 and Acid Blue 74 (Ulu et al., 2020).

Kalsoon and coworkers (2022) developed iron oxide NPs immobilized with manganese peroxidase to study the efficiency in the removal of acid black 234 and direct red 31 dye. The study showed after 24 h, the immobilized manganese peroxidase on iron oxide NPs showed 92% and 100% decolorization, respectively (Kalsoon et al., 2022). Lignin peroxidase immobilized on graphene oxide functionalized  $\text{MnFe}_2\text{O}_4$  NPs showed excellent removal of nigrosine, bromophenol blue, coomassie brilliant blue, and methylene blue at a decolorization percentage of 57.2%, 81.6%, 51.4%, and 88.2% within 1 h, respectively (Rathour et al., 2020). Another study involved the formation of peroxidase immobilized over  $\text{Fe}_3\text{O}_4$  NPs linked with glutaraldehyde to improve enzyme stability, catalytic activity, and reusability for the individual removal of green and red azo dyes (Darwesh et al., 2019). Enzyme immobilization through the formation of 3D NFs using graphite oxide, carbon nanotubes, and copper phosphate-laccase showed promising efficient dye removal (Li et al., 2017), whereas Ali et al. (2018) used ginger peroxidase on poly-pyrrole-cellulose-GO NF to remove blue 4 dye (Ali et al., 2018).

## 6 | THE FUTURE OF ENZYME-INCORPORATED NFS

### 6.1 | Current drawbacks and concerns

It is noteworthy from the previous sections that nanoscience and its applications were studied efficiently, and to yield a more environmentally clean, energy-efficient, and economically driven system. Some industries which have been improved, include the construction and infrastructure, pharmaceutical, energy production, and storage

industries (Iavicoli et al., 2014). Nevertheless, the incorporation of nanotechnology in an application may impose concerns on the environment, health, and safety, as well as ethical issues among consumers (OECD, 2013).

One of the major concerns of nanotechnology is the regulation and use of NPs in industries leading to ethical issues and/or potential abuse (Baran, 2016; Wood et al., 2003). For instance, the usage of nanotechnology to develop genetic weapons for redesigning human "spec," which may alter genetic traits altogether (Wood et al., 2003). Further, each country follows its regulation for using nanomaterials in biomedical applications, and hence there is no standard regulatory law. Thus, there should be a standardized regulation law and regulatory committee in each country for nanomaterials to be utilized in commercial biomedical applications (Barhoum et al., 2022). Moreover, NPs used in the cellular environment undergoes biodegradation which may lead to gene alterations and intracellular changes, that can be a potential threat to human life. Another issue raised is the usage of nanobots to enter the human body to target and destroy harmful microorganisms, however, concerns are raised regarding programming errors that can result in certain alterations/destruction. Some past incidents, such as drug resistance of viruses and/or bacteria, oil spills in oceans, and nuclear accidents are some of the examples, that may happen in the case of unregulated NP-based products in biomedical applications (Wood et al., 2003) as well as the presence of chemicals in the air due to the inability to breakdown (Buzea et al., 2007). The most crucial way of establishing legislation and addressing this challenge is by validating methods of synthesis, detection, and characterization as well as the development of risk assessment and hazard identifications of nanomaterials (Baran, 2016).

In addition, nanotechnology and its economic impact are closely related to community engagement as it determines the progress and success of nanotechnology due to its high cost. The public's point of view on the design of NPs would affect the final product and supply/demand ratio. Besides, investments are typically collaborations between the public, private, and government as a cooperative research and development agreement (Board et al., 2006). Therefore, the investment rate is fairly large and it would not be intellectually welcoming to go for huge investments (Wood et al., 2003), especially after a crucial economic slowdown due to the COVID-19 pandemic. Further, insurers also undergo the uncertainties of nanotechnology, where potentially affected line of work includes, employee compensation, general, environmental and product liability, product recall, and property (Allianz & OECD). Despite various pros of nanotechnology, it also affects job prospects and availability in several industries. For instance, the creation of nano-based lubricants may need lesser maintenance requirements and/or services (Board et al., 2006).

In terms of the environmental impacts of nanotechnology, the major challenge is the method of NPs synthesis, where the difference in shape, size, structure, and composition would affect their toxicity level. The different functional groups in an NP also would drastically alter its physical and chemical properties, which would result in their

inability to identify them in the environment, which can eventually cause potential harm to the atmosphere. Besides, there have been no quantitative and/or qualitative measurements to identify these NPs (Bitounis et al., 2016; Stone et al., 2009) in the environment, as detecting them in the air is rather tedious due to the size distribution between NPs and normal particulates in the air, whereas synthetic and natural NPs binds with each other in the water and sinks to the bottom. Certain methods also require a high amount of energy which is potentially harmful to the environment. Other potential risks include unclear life stages of NPs, as well as untrained professionals, which raises further concerns to utilize them in commercial applications and produce them in a large-scale (Dunphy Guzmán et al., 2006; Zhang et al., 2011).

## 6.2 | Future prospects

There are endless opportunities for NPs to grow industrially in various sectors due to their unique characteristics, such as good kinetic power, better insulation and reusability properties, low power consumption system as well as high sensitivity and precision quality (Khan et al., 2019). For instance, upcoming industries using NPs include photonics and optoelectronics. The nanophotonic term is currently being used in methods, such as optical systems, device doping, and ablation, optoelectronics devices, bio-nanophotonics, medical devices (e.g., lasers) as well as power physics (Y. Zhang et al., 2019). Advanced nanosystems such as molecular nanorobotics to grow artificial organs using nanomaterials, genetic therapy, as well as antiaging treatment in the future, is also being utilized in recent times (Khalid et al., 2020; Roco, 2006). Moreover, further studies on biomedical applications using NPs to develop wireless, high efficient devices, such as implantable biosensors, biomolecular sensors, and microelectromechanical devices, were also increasing rapidly (Y. Xu, Hu, et al., 2019). Other portable electronics, including home security devices and environmental sensors have been seen as promising applications of nanomaterials in the future (Vigneshvar & Senthilkumar, 2018).

Furthermore, the idea and advantages of collaboration among scientists, economists as well as governments can be very fruitful in developing revolutionary designs as nanotechnology is generally expensive (Scarazzati & Wang, 2019). For instance, in 2000, President Bill Clinton founded the National Nanotechnology Initiative (NNI) to act as support for nanotechnology research across governmental agencies, educational institutes, and research industries in the United States. The NNI research funding has increased more than 155-fold since 1997 (initial investment of \$116 million) and most recent data showed the research & development (R&D) investment at \$18.1 billion as of 2014 (Koshovets & Ganichev, 2017; Paull et al., 2003). However, there has been a steady decline in funding due to budget cuts since 2013 whereby, in 2016–2017, the proposed funding was cut to \$1.5 billion, which, could potentially be a barrier to the growth of the nanoindustry (National Nanotechnology Initiative, 2016). In Europe, the European Union (EU) research projects launched the Horizon 2020 program, one of the largest investments (€2 billion) in the development

of nanomaterials. Horizon 2020 aims to act as an important tool for gaining a better understanding of the possible safety concerns associated with nanotechnology, as well as exploring their capabilities (European Commission, 2020). On the other hand, the nanotechnology industry has been developing across Asia whereby, in China, the research development of nano-industry has been flourishing since the founding of Strategic Pioneering Programme in 2012 with a budget of \$152 million over 5 years. The long-term program which is led by the Chinese Academy of Sciences (CAS), is a key player in the nano-industry and is leading the ranks in terms of scientific papers and patents (J. Qiu, 2016). The government of India through the Department of Science and Technology funds the National Nanotechnology Program with nearly \$10 million as well as National Program on Smart Materials, which was funded with over \$15 million (funding from collaborations with five government agencies). In South Africa, the South African Nanotechnology Initiative (SANI) includes participation from 11 universities, 5 research organizers, and 10 private industries within the scope of research areas, such as chemicals, fuels, energy as well as telecommunications, whereas there have been 62 projects funded with nearly \$12.5 million by the National Council of Science and Technology, Mexico, in 19 institutions focusing on optics, microelectronics, coatings, and medical devices. The council also collaborated with EU and assigned \$1 million to call for research proposals on BisNano projects (Foladori et al., 2011; Singer et al., 2005). South Africa also founded 6 plans to reinforce R&D in the nano-industry over a 10-year research plan from 2008 to 2018 (e.g., National Nanotechnology Equipment Programme [NNEP]: Human and Infrastructure Capacity Development Strategic Framework), which indicates comprehensive investments and planning toward the growth of nanotechnology (Muhammad, 2022). The collaborative approach also improves risk management by identifying and prioritizing research ideas and potential risks of NPs. International collaborations also help industries to gain synergies and widen their databases.

Some precautionary steps which would aid high-priority stakeholders include independent research on the risks of nanotechnology on the human, environment as well as ensuring transparency to the public for obtaining public's trust and have adequate risk management ability (Allianz & OECD). In addition, nanotechnology may also reduce job opportunities in certain industries (Board et al., 2006) as mentioned in the previous subsection. According to Nance (2019), nanotechnology-based industries (e.g., nanomedicine and pharmaceutical industries) would have an upsurge in employment due to numerous career trajectories (e.g., start-ups, consulting, intellectual property and patenting, academia, venture capitalist, regulatory and policy writing) that the field could potentially provide over time (Nance, 2019).

## 7 | CONCLUSION

The evolution of nanotechnology in the 21st century has been interesting and proven to be useful and effective in various industries. Although the use of NPs started as a form of esthetic

pleasure in the 16th century, it has created a massive change in the current science, where its uses show effectiveness in pharmaceutical, medical, food, and wastewater industries.

The introduction of nanotechnology has created huge impacts on the community in terms of food safety, human health, and the environment. For instance, NPs have been vastly used in food industries to promote the longevity of foods, ensuring the taste, texture, odor, and coating are maintained as well as creating a safe and smart packaging system to ensure food safety. Further, nanotechnology has impacted human health in various ways. For instance, NPs synthesized using various methods and chemical precursors are applicable for the targeted and controlled delivery of drugs and/or genes, immobilize enzymes, biosensors to detect cancerous cells, and/or as biomarkers to detect specific diseases. Moreover, the application of NPs to preserve and conserve the environment has been the main focus of study among researchers in the past few years, where NPs are used in wastewater treatments to remove hazardous toxins (e.g., hydrocarbons, dye, and heavy metal ions) before entering the water system.

It is noteworthy that, there are few limitations and challenges in nanotechnology as well as enzyme-incorporated NPs. The major concern of nanotechnology is the abuse of NPs for ethical reasons, such as developing genetic weapons which may lead to gene alterations. Furthermore, the possibility of the existence of toxic chemical compounds in the environment due to the inability of NPs to breakdown, ill-defined life stages of NPs as well as the methods of synthesis, where certain methods consume more energy and lack environmental friendliness. Economically, the application of nanotechnology is rather costly, and therefore, large amounts of investments are required, however, it comes with uncertainties.

Thus, there are numerous applications of nanostructured materials across multiple industries (e.g., medicine, pharmaceutical, wastewater treatment, and electronics). Some advantages of nano-scale materials and enzyme-incorporated NPs are their smaller size, compactness, ability to increase stability, and physical and reproducibility properties, compared to conventional materials, whereas greener synthesis and their high efficacy rates were also utilized in various applications as mentioned in this article. Despite that, nanotechnology also possesses certain challenges, and therefore, scientists should focus on the synthesis and large-scale application of nanomaterials using a safer, sustainable, and cost-efficient approach. Once these challenges have been overcome, the future is irreducibly bright for the utilization of nanomaterials, especially enzyme-incorporated NPs in our daily lives.

## AUTHOR CONTRIBUTIONS

**Shamini Anboo:** Conceptualization, writing, original draft preparation. **Sie Yon Lau:** Supervision, validation, resources. **Jibrail Kansedo:** Writing, reviewing, and editing. **Pow-Seng Yap:** Writing, reviewing, and editing. **Tony Hadibarata:** Writing, reviewing, and editing. **Jaison Jeevanandam:** Writing, reviewing, and editing. **Azlina H. Kamaruddin:** Writing, reviewing, and editing.

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## CONFLICT OF INTEREST

The authors declare no conflict of interest.

## DATA AVAILABILITY STATEMENT

Data sharing is not applicable to this article as no new data were created or analyzed in this study.

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## REFERENCES

- Abd-Elsalam, K. A. (2020). *Multifunctional hybrid nanomaterials for sustainable agri-food and ecosystems: A note from the editor* (pp. 1–19). Elsevier.
- Abdollahi, K., Yazdani, F., Panahi, R., & Mokhtarani, B. (2018). Biotransformation of phenol in synthetic wastewater using the functionalized magnetic nano-biocatalyst particles carrying tyrosinase. *3 Biotech*, 8(10):419. <https://doi.org/10.1007/s13205-018-1445-2>
- Agarwal, P., Gupta, R., & Agarwal, N. (2016). A review on enzymatic treatment of phenols in wastewater. *Journal of Biotechnology & Biomaterials*, 6, 249. <https://doi.org/10.4172/2155-952X.1000249>
- Agostinelli, E., Belli, F., Tempera, G., Mura, A., Floris, G., Toniolo, L., Vavasori, A., Fabris, S., Momo, F., & Stevanato, R. (2007). Polyketone polymer: A new support for direct enzyme immobilization. *Journal of Biotechnology*, 127(4), 670–678. <https://doi.org/10.1016/j.jbiotec.2006.08.011>
- Ahmad, R., & Sardar, M. (2015a). Enzyme immobilization: An overview on nanoparticles as immobilization matrix. *Biochemistry & Analytical Biochemistry*, 4, 178. <https://doi.org/10.4172/2161-1009.1000178>
- Ahmad, R., & Sardar, M. (2015b). Enzyme immobilization: An overview on nanoparticles as immobilization matrix. *Biochemistry & Analytical Biochemistry*, 4, 178. <https://doi.org/10.4172/2161-1009.1000178>
- Al-Maqdi, K. A., Bilal, M., Alzamly, A., Iqbal, H., Shah, I., & Ashraf, S. S. (2021). Enzyme-loaded flower-shaped nanomaterials: A versatile platform with biosensing, biocatalytic, and environmental promise. *Nanomaterials*, 11(6), 1460.
- Alarcón-Correa, M., Günther, J.-P., Troll, J., Kadiri, V. M., Bill, J., Fischer, P., & Rothenstein, D. (2019). Self-assembled phage-based colloids for high localized enzymatic activity. *ACS Nano*, 13(5), 5810–5815. <https://doi.org/10.1021/acs.nano.9b01408>
- Ali, M., Husain, Q., Sultana, S., & Ahmad, M. (2018). Immobilization of peroxidase on polypyrrole-cellulose-graphene oxide nanocomposite via non-covalent interactions for the degradation of reactive blue 4 dye. *Chemosphere*, 202, 198–207. <https://doi.org/10.1016/j.chemosphere.2018.03.073>
- Allianz, A., & OECD. (2005). *Small sizes that matter: Opportunities and risks of nanotechnologies*. Organisation for Economic Co-operation and Development (OECD).

- An, J., Li, G., Zhang, Y., Zhang, T., Liu, X., Gao, F., Peng, M., He, Y., & Fan, H. (2020). Recent advances in enzyme-nanostructure biocatalysts with enhanced activity. *Catalysts*, 10, 338. <https://doi.org/10.3390/catal10030338>
- Arduini, F., Forchielli, M., Amine, A., Neagu, D., Cacciotti, I., Nanni, F., Moscone, D., & Palleschi, G. (2015). Screen-printed biosensor modified with carbon black nanoparticles for the determination of paraoxon based on the inhibition of butyrylcholinesterase. *Microchimica Acta*, 182(3), 643–651. <https://doi.org/10.1007/s00604-014-1370-y>
- Arnaldi, P., Pastorino, L., & Monticelli, O. (2020). On an effective approach to improve the properties and the drug release of chitosan-based microparticles. *International Journal of Biological Macromolecules*, 163, 393–401. <https://doi.org/10.1016/j.ijbiomac.2020.07.016>
- Astefanei, A., Núñez, O., & Galceran, M. T. (2015). Characterisation and determination of fullerenes: A critical review. *Analytica Chimica Acta*, 882, 1–21. <https://doi.org/10.1016/j.aca.2015.03.025>
- Asuri, P., Bale, S. S., Karajanagi, S. S., & Kane, R. S. (2006). The protein–nanomaterial interface. *Current Opinion in Biotechnology*, 17(6), 562–568. <https://doi.org/10.1016/j.copbio.2006.09.002>
- Asuri, P., Karajanagi, S. S., Yang, H., Yim, T.-J., Kane, R. S., & Dordick, J. S. (2006). Increasing protein stability through control of the nanoscale environment. *Langmuir*, 22(13), 5833–5836. <https://doi.org/10.1021/la0528450>
- Augustin, M. A., & Sanguansri, P. (2009). Nanostructured materials in the food industry. *Advances in Food and Nutrition Research*, 58, 183–213. [https://doi.org/10.1016/S1043-4526\(09\)58005-9](https://doi.org/10.1016/S1043-4526(09)58005-9)
- Azghandi, M. H. A., Farahani, B. V., & Dehghani, N. (2017). Encapsulation of methotrexate and cyclophosphamide in interpolymer complexes formed between poly acrylic acid and poly ethylene glycol on multi-walled carbon nanotubes as drug delivery systems. *Materials Science & Engineering*, 79, 841. <https://doi.org/10.1016/j.msec.2017.05.089>
- Bansal, V., Poddar, P., Ahmad, A., & Sastry, M. (2006). Room-temperature biosynthesis of ferroelectric barium titanate nanoparticles. *Journal of the American Chemical Society*, 128(36), 11958–11963. <https://doi.org/10.1021/ja063011m>
- Baran, A. (2016). Nanotechnology: Legal and ethical issues. *Ekonomia i Zarządzanie*, 8, 47–54. <https://doi.org/10.1515/emj-2016-0005>
- Barhoum, A., García-Betancourt, M. L., Jeevanandam, J., Hussien, E. A., Mekawy, S. A., Mostafa, M., Omran, M. M., S Abdalla, M., & Bechelany, M. (2022). Review on natural, incidental, bioinspired, and engineered nanomaterials: History, definitions, classifications, synthesis, properties, market, toxicities, risks, and regulations. *Nanomaterials*, 12(2), 177.
- Batule, B. S., Park, K. S., Kim, M. I., & Park, H. G. (2015). Ultrafast sonochemical synthesis of protein-inorganic nanoflowers. *International Journal of Nanomedicine*, 10(Special Issue on diverse applications in Nano-Theranostics), 137–142. <https://doi.org/10.2147/IJN.S90274>
- Bayda, S., Adeel, M., Tuccinardi, T., Cordani, M., & Rizzolio, F. (2019). The history of nanoscience and nanotechnology: From chemical-physical applications to nanomedicine. *Molecules*, 25(1), 112. <https://doi.org/10.3390/molecules25010112>
- Bekmezci, S. A., Soylemez, S., Yilmaz, G., Udum, Y. A., Yagci, Y., & Toppare, L. (2020). A new ethanol biosensor based on polyfluorene-g-poly(ethylene glycol) and multiwalled carbon nanotubes. *European Polymer Journal*, 122, 109300. <https://doi.org/10.1016/j.eurpolymj.2019.109300>
- Berekaa, M. M. (2015). Nanotechnology in food industry; advances in food processing, packaging and food safety. *International Journal of Current Microbiology and Applied Sciences*, 4(5), 345–357.
- Bhatia, S. (2016). Nanoparticles types, classification, characterization, fabrication methods and drug delivery applications. In S. Bhatia (Ed.), *Natural polymer drug delivery systems: Nanoparticles, plants, and algae* (33–93). Springer International Publishing [https://doi.org/10.1007/978-3-319-41129-3\\_2](https://doi.org/10.1007/978-3-319-41129-3_2)
- Bilgi, M., & Ayranci, E. (2016). Biosensor application of screen-printed carbon electrodes modified with nanomaterials and a conducting polymer: Ethanol biosensors based on alcohol dehydrogenase. *Sensors and Actuators B: Chemical*, 237, 849–855. <https://doi.org/10.1016/j.snb.2016.06.164>
- Bitounis, D., Pourchez, J., Forest, V., Boudard, D., Cottier, M., & Klein, J.-P. (2016). Detection and analysis of nanoparticles in patients: A critical review of the status quo of clinical nanotoxicology. *Biomaterials*, 76, 302–312. <https://doi.org/10.1016/j.biomaterials.2015.10.061>
- Blanco, A., & Blanco, G. (2017). Proteins. In A. Blanco & G. Blanco (Eds.), *Medical biochemistry* (pp. 21–71). Academic Press. <https://doi.org/10.1016/B978-0-12-803550-4.00003-3>
- Board, N., Sciences, D., & Council, N. (2006). *A matter of size: Triennial review of the national nanotechnology initiative*. National Academies Press (NAP). <https://doi.org/10.17226/11752>
- Bollella, P., & Katz, E. (2020). Biosensors—Recent advances and future challenges. *Sensors*, 20(22):6645. <https://doi.org/10.3390/s20226645>
- Brena, B. M., & Batista-Viera, F. (2006). Immobilization of enzymes. In J. M. Guisan (Ed.), *Immobilization of enzymes and cells* (pp. 15–30). Springer.
- Brodin, J. D., Sprangers, A. J., McMillan, J. R., & Mirkin, C. A. (2015). DNA-mediated cellular delivery of functional enzymes. *Journal of the American Chemical Society*, 137(47), 14838–14841. <https://doi.org/10.1021/jacs.5b09711>
- Brusnitsyn, D. V., Medyantseva, E. P., Varlamova, R. M., Sitdikova, R. R., Fattakhova, A. N., Kononova, O. A., & Budnikov, G. K. (2016). Carbon nanomaterials as electrode surface modifiers in development of amperometric monoamine oxidase biosensors. *Inorganic Materials*, 52(14), 1413–1419. <https://doi.org/10.1134/S002016851614003X>
- Buzea, C., Pacheco, I. I., & Robbie, K. (2007). Nanomaterials and nanoparticles: Sources and toxicity. *Biointerphases*, 2(4), 17–71. <https://doi.org/10.1116/1.2815690>
- Bäcker, M., Koch, C., Eiben, S., Geiger, F., Eber, F., Gliemann, H., Poghossian, A., Wege, C., & Schöning, M. J. (2017). Tobacco mosaic virus as enzyme nanocarrier for electrochemical biosensors. *Sensors and Actuators B: Chemical*, 238, 716–722. <https://doi.org/10.1016/j.snb.2016.07.096>
- Cao, J., Cai, Y., Yu, L., & Zhou, J. (2019). Dual physically crosslinked hydrogels based on the synergistic effects of electrostatic and dipole–dipole interactions. *Journal of Materials Chemistry B*, 7(4), 676–683. <https://doi.org/10.1039/C8TB03032D>
- Cao, M., Cai, R., Zhao, L., Guo, M., Wang, L., Wang, Y., Zhang, L., Wang, X., Yao, H., Xie, C., Cong, Y., Guan, Y., Tao, X., Wang, Y., Xu, S., Liu, Y., Zhao, Y., & Chen, C. (2021). Molybdenum derived from nanomaterials incorporates into molybdenum enzymes and affects their activities in vivo. *Nature Nanotechnology*, 16(6), 708–716.
- Cao, S., Wu, H., Pijpers, I. A. B., Shao, J., Abdelmohsen, L. K. E. A., Williams, D. S., & van Hest, J. C. M. (2021). Cucurbit-like polymersomes with aggregation-induced emission properties show enzyme-mediated motility. *ACS Nano*, 15(11), 18270–18278. <https://doi.org/10.1021/acsnano.1c07343>
- Castagnola, V., Cookman, J., De Araújo, J. M., Polo, E., Cai, Q., Silveira, C. P., Krpetić, Ž., Yan, Y., Boselli, L., & Dawson, K. A. (2017). Towards a classification strategy for complex nanostructures. *Nanoscale Horizons*, 2(4), 187–198. <https://doi.org/10.1039/c6nh00219f>
- Cavalcante, F. T. T., de A. Falcão, I. R., da S. Souza, J. E., Rocha, T. G., de Sousa, I. G., Cavalcante, A. L. G., de Oliveira, A. L. B., de Sousa, M. C. M., & dos Santos, J. C. S. (2021). Designing of nanomaterials-based enzymatic biosensors: Synthesis, properties, and applications. *Electrochem*, 2(1), 149–184. <https://doi.org/10.3390/electrochem2010012>
- Chagas, P. M. B., Torres, J. A., Silva, M. C., & Corrêa, A. D. (2015). Immobilized soybean hull peroxidase for the oxidation of phenolic compounds in coffee processing wastewater. *International Journal of*

- Biological Macromolecules*, 81, 568–575. <https://doi.org/10.1016/j.ijbiomac.2015.08.061>
- Chan, J. M., Zhang, L., Tong, R., Ghosh, D., Gao, W., Liao, G., Yuet, K. P., Gray, D., Rhee, J. W., Cheng, J., Golomb, G., Libby, P., Langer, R., & Farokhzad, O. C. (2010). Spatiotemporal controlled delivery of nanoparticles to injured vasculature. *Proceedings of the National Academy of Sciences*, 107(5), 2213–2218. <https://doi.org/10.1073/pnas.0914585107>
- Chatterjee, T., Chakraborti, S., Joshi, P., Singh, S. P., Gupta, V., & Chakrabarti, P. (2010). The effect of zinc oxide nanoparticles on the structure of the periplasmic domain of the *Vibrio cholerae* ToxR protein. *The FEBS journal*, 277(20), 4184–4194. <https://doi.org/10.1111/j.1742-4658.2010.07807.x>
- Chavali, M. S., & Nikolova, M. P. (2019). Metal oxide nanoparticles and their applications in nanotechnology. *SN Applied Sciences*, 1(6), 607. <https://doi.org/10.1007/s42452-019-0592-3>
- Chen, C.-C., Tsai, T.-H., Huang, Z.-R., & Fang, J.-Y. (2010). Effects of lipophilic emulsifiers on the oral administration of lovastatin from nanostructured lipid carriers: Physicochemical characterization and pharmacokinetics. *European Journal of Pharmaceutics and Biopharmaceutics*, 74(3), 474–482. <https://doi.org/10.1016/j.ejpb.2009.12.008>
- Chen, J., & Zou, X. (2019). Self-assemble peptide biomaterials and their biomedical applications. *Bioactive Materials*, 4, 120–131. <https://doi.org/10.1016/j.bioactmat.2019.01.002>
- Chen, M., Zeng, G., Xu, P., Lai, C., & Tang, L. (2017). How do enzymes 'meet' nanoparticles and nanomaterials? *Trends in Biochemical Sciences*, 42, 914–930. <https://doi.org/10.1016/j.tibs.2017.08.008>
- Chen, T., Xu, Y., Peng, Z., Li, A., & Liu, J. (2017). Simultaneous enhancement of bioactivity and stability of laccase by Cu<sup>2+</sup>/PAA/PPEGA matrix for efficient biosensing and recyclable decontamination of pyrocatechol. *Analytical Chemistry*, 89(3), 2065–2072. <https://doi.org/10.1021/acs.analchem.6b04691>
- Chen, W.-H., Vázquez-González, M., Zoabi, A., Abu-Reziq, R., & Willner, I. (2018). Biocatalytic cascades driven by enzymes encapsulated in metal-organic framework nanoparticles. *Nature Catalysis*, 1(9), 689–695. <https://doi.org/10.1038/s41929-018-0117-2>
- Chequer, F. M. D., de Oliveira, G. A. R., Ferraz, E. R. A. c., Cardoso, J. C., Zanoni, M. V. B., & de Oliveira, D. P. (2013). Textile dyes: Dyeing process and environmental impact. In M. Gunay (Ed.), *Eco-friendly textile dyeing and finishing*. IntechOpen. <https://doi.org/10.5772/53659>
- Cinti, S., Arduini, F., Moscone, D., Palleschi, G., Gonzalez-Macia, L., & Killard, A. J. (2015). Cholesterol biosensor based on inkjet-printed Prussian blue nanoparticle-modified screen-printed electrodes. *Sensors and Actuators B: Chemical*, 221, 187–190. <https://doi.org/10.1016/j.snb.2015.06.054>
- Clark, L. C., & Lyons, C. (1962). Electrode systems for continuous monitoring in cardiovascular surgery. *Annals of the New York Academy of Sciences*, 102(1), 29–45. <https://doi.org/10.1111/j.1749-6632.1962.tb13623.x>
- Colvin, V. L., & Kulinsky, K. M. (2007). Nanoparticles as catalysts for protein fibrillation. *Proceedings of the National Academy of Sciences*, 104(21), 8679–8680. <https://doi.org/10.1073/pnas.0703194104>
- Cui, J., & Jia, S. (2017). Organic-inorganic hybrid nanoflowers: A novel host platform for immobilizing biomolecules. *Coordination Chemistry Reviews*, 352, 249–263. <https://doi.org/10.1016/j.ccr.2017.09.008>
- Cui, J., Zhao, Y., Liu, R., Zhong, C., & Jia, S. (2016). Surfactant-activated lipase hybrid nanoflowers with enhanced enzymatic performance. *Scientific Reports*, 6(1), 27928. <https://doi.org/10.1038/srep27928>
- Darby, J. F., Atobe, M., Firth, J. D., Bond, P., Davies, G. J., O'Brien, P., & Hubbard, R. E. (2017). Increase of enzyme activity through specific covalent modification with fragments. *Chemical Science*, 8(11), 7772–7779. <https://doi.org/10.1039/C7SC01966A>
- Darwesh, O. M., Matter, I. A., & Eida, M. F. (2019). Development of peroxidase enzyme immobilized magnetic nanoparticles for bioremediation of textile wastewater dye. *Journal of Environmental Chemical Engineering*, 7(1), 102805. <https://doi.org/10.1016/j.jece.2018.11.049>
- Das, A., Jaswal, V., & Yogalakshmi, K. N. (2020). Degradation of chlorpyrifos in soil using laccase immobilized iron oxide nanoparticles and their competent role in deterring the mobility of chlorpyrifos. *Chemosphere*, 246, 125676. <https://doi.org/10.1016/j.chemosphere.2019.125676>
- Datta, S., Christena, L. R., & Rajaram, Y. R. S. (2013). Enzyme immobilization: An overview on techniques and support materials. *3 Biotech*, 3(1), 1–9. <https://doi.org/10.1007/s13205-012-0071-7>
- Deng, Y., Liu, K., Liu, Y., Dong, H., & Li, S. (2016). A novel acetylcholinesterase biosensor based on nano-porous pseudo carbon paste electrode modified with gold nanoparticles for detection of methyl parathion. *Journal of Nanoscience and Nanotechnology*, 16(9), 9460–9467. <https://doi.org/10.1166/jnn.2016.13059>
- Devasenathipathy, R., Mani, V., Chen, S.-M., Huang, S.-T., Huang, T.-T., Lin, C.-M., Hwa, K. Y., Chen, T. Y., & Chen, B. J. (2015). Glucose biosensor based on glucose oxidase immobilized at gold nanoparticles decorated graphene-carbon nanotubes. *Enzyme and Microbial Technology*, 78, 40–45. <https://doi.org/10.1016/j.enzmictec.2015.06.006>
- Din, F. U., Aman, W., Ullah, I., Qureshi, O. S., Mustapha, O., Shafique, S., & Zeb, A. (2017). Effective use of nanocarriers as drug delivery systems for the treatment of selected tumors. *International Journal of Nanomedicine*, 12, 7291–7309. <https://doi.org/10.2147/IJN.S146315>
- Ding, J., Liang, T., Zhou, Y., He, Z., Min, Q., Jiang, L., & Zhu, J. (2017). Hyaluronidase-triggered anticancer drug and siRNA delivery from cascaded targeting nanoparticles for drug-resistant breast cancer therapy. *Nano Research*, 10(2), 690–703. <https://doi.org/10.1007/s12274-016-1328-y>
- Ding, X., & Wang, Y. (2017). Weak bond-based injectable and stimuli responsive hydrogels for biomedical applications. *Journal of Materials Chemistry B*, 5(5), 887–906. <https://doi.org/10.1039/C6TB03052A>
- Duan, L., Li, H., & Zhang, Y. (2018). Synthesis of hybrid nanoflower-based carbonic anhydrase for enhanced biocatalytic activity and stability. *ACS Omega*, 3(12), 18234–18241. <https://doi.org/10.1021/acsomega.8b02247>
- Dunphy Guzmán, K. A., Taylor, M. R., & Banfield, J. F. (2006). Environmental risks of nanotechnology: National nanotechnology initiative funding, 2000–2004. *Environmental Science & Technology*, 40(5), 1401–1407. <https://doi.org/10.1021/es0515708>
- El Harrad, L., Bourais, I., Mohammadi, H., & Amine, A. (2018). Recent advances in electrochemical biosensors based on enzyme inhibition for clinical and pharmaceutical applications. *Sensors*, 18(1):164. <https://doi.org/10.3390/s18010164>
- Elliott, J. A., Shibuta, Y., Amara, H., Bichara, C., & Neyts, E. C. (2013). Atomistic modelling of CVD synthesis of carbon nanotubes and graphene. *Nanoscale*, 5(15), 6662–6676. <https://doi.org/10.1039/C3NR01925J>
- End, N., & Schöning, K.-U. (2004). Immobilized biocatalysts in industrial research and production. *Immobilized Catalysts*, 242, 273–317.
- European Commission. (2020). *Horizon 2020*.
- Eş, I., Vieira, J. D. G., & Amaral, A. C. (2015). Principles, techniques, and applications of biocatalyst immobilization for industrial application. *Applied Microbiology and Biotechnology*, 99(5), 2065–2082. <https://doi.org/10.1007/s00253-015-6390-y>
- Fang, X., Zhang, C., Qian, X., & Yu, D. (2018). Self-assembled 2,4-dichlorophenol hydroxylase-inorganic hybrid nanoflowers with enhanced activity and stability. *RSC Advances*, 8(37), 20976–20981. <https://doi.org/10.1039/C8RA02360C>
- Foladori, G., Zayago, É., Appelbaum, R., & Parker, R. (2011). Mexico–U.S. Scientific collaboration in nanotechnology. *Frontera Norte*, 24(48),

- 145–164. [http://www.scielo.org.mx/scielo.php?script=sci\\_arttext&pid=S0187-73722012000200006#notes](http://www.scielo.org.mx/scielo.php?script=sci_arttext&pid=S0187-73722012000200006#notes)
- Fu, I. W., Markegard, C. B., Chu, B. K., & Nguyen, H. D. (2014). Role of hydrophobicity on self-assembly by peptide amphiphiles via molecular dynamics simulations. *Langmuir*, 30(26), 7745–7754. <https://doi.org/10.1021/la5012988>
- Gao, H.-Y., Wagner, H., Held, P. A., Du, S., Gao, H.-J., Studer, A., & Fuchs, H. (2015). In-plane Van der Waals interactions of molecular self-assembly monolayer. *Applied Physics Letters*, 106(8), 081606. <https://doi.org/10.1063/1.4907777>
- Gao, Y., Wang, M., Shah, K., Singh Kalra, S., Rome, L. H., & Mahendra, S. (2022). Decolorization and detoxification of synthetic dye compounds by laccase immobilized in vault nanoparticles. *Bioresource Technology*, 351, 127040. <https://doi.org/10.1016/j.biortech.2022.127040>
- García-Galan, C., Berenguer-Murcia, Á., Fernández-Lafuente, R., & Rodrigues, R. C. (2011). Potential of different enzyme immobilization strategies to improve enzyme performance. *Advanced Synthesis & Catalysis*, 353(16), 2885–2904. <https://doi.org/10.1002/adsc.201100534>
- Ge, J., Lei, J., & Zare, R. N. (2012). Protein–inorganic hybrid nanoflowers. *Nature Nanotechnology*, 7(7), 428–432. <https://doi.org/10.1038/nnano.2012.80>
- Gokoglan, T. C., Kesik, M., Soylemez, S., Yuksel, R., Unalan, H. E., & Toppare, L. (2017). Paper based glucose biosensor using graphene modified with a conducting polymer and gold nanoparticles. *Journal of the Electrochemical Society*, 164(6), G59–G64. <https://doi.org/10.1149/2.0791706jes>
- Gomez, A., Narayan, M., Zhao, L., Jia, X., Bernal, R. A., Lopez-Moreno, M. L., & Peralta-Videa, J. R. (2021). Effects of nano-enabled agricultural strategies on food quality: Current knowledge and future research needs. *Journal of Hazardous Materials*, 401, 123385. <https://doi.org/10.1016/j.jhazmat.2020.123385>
- Greschner, A. A., Bujold, K. E., & Sleiman, H. F. (2013). Intercalators as molecular chaperones in DNA self-assembly. *Journal of the American Chemical Society*, 135(30), 11283–11288. <https://doi.org/10.1021/ja404402b>
- Guo, J., Liu, X., Zhang, X., Wu, J., Chai, C., Ma, D., Chen, Q., Xiang, D., & Ge, W. (2019). Immobilized lignin peroxidase on Fe<sub>3</sub>O<sub>4</sub>@SiO<sub>2</sub>@polydopamine nanoparticles for degradation of organic pollutants. *International Journal of Biological Macromolecules*, 138, 433–440. <https://doi.org/10.1016/j.ijbiomac.2019.07.105>
- Guo, S., Li, H., Liu, J., Yang, Y., Kong, W., Qiao, S., Huang, H., Liu, Y., & Kang, Z. (2015). Visible-light-induced effects of Au nanoparticle on laccase catalytic activity. *ACS Applied Materials & Interfaces*, 7(37), 20937–20944. <https://doi.org/10.1021/acsami.5b06472>
- Górecka, E., & Jastrzębska, M. (2011). Immobilization techniques and biopolymer carriers. *Biotechnology and Food Science*, 75(1), 65–86. <https://doi.org/10.34658/bfs.2011.75.1.65-86>
- Haghju, S., Bari, M. R., & Khaled-Abad, M. A. (2018). Affecting parameters on fabrication of β-D-galactosidase immobilized chitosan/poly (vinyl alcohol) electrospun nanofibers. *Carbohydrate Polymers*, 200, 137–143. <https://doi.org/10.1016/j.carbpol.2018.07.096>
- Haider, T., & Husain, Q. (2008). Concanavalin A layered calcium alginate–starch beads immobilized β galactosidase as a therapeutic agent for lactose intolerant patients. *International Journal of Pharmaceutics*, 359(1–2), 1–6. <https://doi.org/10.1016/j.ijpharm.2008.03.013>
- Han, H., Valdepérez, D., Jin, Q., Yang, B., Li, Z., Wu, Y., Pelaz, B., Parak, W. J., & Ji, J. (2017). Dual enzymatic reaction-assisted gemcitabine delivery systems for programmed pancreatic cancer therapy. *ACS Nano*, 11(2), 1281–1291. <https://doi.org/10.1021/acs.nano.6b05541>
- Han, L., & Liu, A. (2017). Novel cell–inorganic hybrid catalytic interfaces with enhanced enzymatic activity and stability for sensitive biosensing of paraoxon. *ACS Applied Materials & Interfaces*, 9(8), 6894–6901. <https://doi.org/10.1021/acsami.6b15992>
- Handford, C. E., Dean, M., Henchion, M., Spence, M., Elliott, C. T., & Campbell, K. (2014). Implications of nanotechnology for the agri-food industry: Opportunities, benefits and risks. *Trends in Food Science & Technology*, 40(2), 226–241. <https://doi.org/10.1016/j.tifs.2014.09.007>
- Hanefeld, U., Gardossi, L., & Magner, E. (2009). Understanding enzyme immobilisation. *Chemical Society Reviews*, 38(2), 453–468. <https://doi.org/10.1039/B711564B>
- Hao, Y., Li, H., Cao, Y., Chen, Y., Lei, M., Zhang, T., Xiao, Y., Chu, B., & Qian, Z. (2019). Uricase and horseradish peroxidase hybrid CaHPO<sub>4</sub> nanoflower integrated with transcutaneous patches for treatment of hyperuricemia. *Journal of Biomedical Nanotechnology*, 15(5), 951–965. <https://doi.org/10.1166/jbn.2019.2752>
- He, X., Deng, H., & Hwang, H.-m (2019). The current application of nanotechnology in food and agriculture. *Journal of Food and Drug Analysis*, 27(1), 1–21. <https://doi.org/10.1016/j.jfda.2018.12.002>
- Henchion, M., McCarthy, M., Dillon, E. J., Greehy, G., & McCarthy, S. N. (2019). Big issues for a small technology: Consumer trade-offs in acceptance of nanotechnology in food. *Innovative Food Science & Emerging Technologies*, 58, 102210. <https://doi.org/10.1016/j.ifset.2019.102210>
- Hiratsuka, A., Fujisawa, K., & Muguruma, H. (2008). Amperometric biosensor based on glucose dehydrogenase and plasma-polymerized thin films. *Analytical Sciences: The International Journal of the Japan Society for Analytical Chemistry*, 24(4), 483–486. <https://doi.org/10.2116/analsci.24.483>
- Homaei, A. A., Sariri, R., Vianello, F., & Stevanato, R. (2013). Enzyme immobilization: An update. *Journal of Chemical Biology*, 6(4), 185–205. <https://doi.org/10.1007/s12154-013-0102-9>
- Hu, Q., Sun, W., Lu, Y., Bomba, H. N., Ye, Y., Jiang, T., Isaacson, A. J., & Gu, Z. (2016). Tumor microenvironment-mediated construction and deconstruction of extracellular drug-delivery depots. *Nano Letters*, 16(2), 1118–1126. <https://doi.org/10.1021/acs.nanolett.5b04343>
- Huang, W.-C., Wang, W., Xue, C., & Mao, X. (2018). Effective enzyme immobilization onto a magnetic chitin nanofiber composite. *ACS Sustainable Chemistry & Engineering*, 6(7), 8118–8124. <https://doi.org/10.1021/acssuschemeng.8b01150>
- Huang, Y., Cui, L., Xue, Y., Zhang, S., Zhu, N., Liang, J., & Li, G. (2017). Ultrasensitive cholesterol biosensor based on enzymatic silver deposition on gold nanoparticles modified screen-printed carbon electrode. *Materials Science and Engineering: C*, 77, 1–8. <https://doi.org/10.1016/j.msec.2017.03.253>
- Hwang, E. T., & Gu, M. B. (2013). Enzyme stabilization by nano/microsized hybrid materials. *Engineering in Life Sciences*, 13(1), 49–61. <https://doi.org/10.1002/elsc.201100225>
- Hwang, E. T., & Lee, S. (2019). Multienzymatic cascade reactions via enzyme complex by immobilization. *ACS Catalysis*, 9(5), 4402–4425. <https://doi.org/10.1021/acscatal.8b04921>
- Iavicoli, I., Leso, V., Ricciardi, W., Hodson, L. L., & Hoover, M. D. (2014). Opportunities and challenges of nanotechnology in the green economy. *Environmental Health*, 13(1), 78. <https://doi.org/10.1186/1476-069X-13-78>
- Ibrahim, K. S. (2013). Carbon nanotubes-properties and applications: A review. *Carbon letters*, 14(3), 131–144. <https://doi.org/10.1016/j.arabjc.2017.05.011>
- Ispas, C., Sokolov, I., & Andreescu, S. (2009). Enzyme-functionalized mesoporous silica for bioanalytical applications. *Analytical and Bioanalytical Chemistry*, 393(2), 543–554. <https://doi.org/10.1007/s00216-008-2250-2>
- Jeevanandam, J., Patel, P., Ponnuchamy, K., Manchala, S., Acquah, C., & Danquah, M. K. (2022). Biocatalytic nanomaterials as an alternative to peroxidase enzymes, *Nanomaterials for biocatalysis* (pp. 513–542). Elsevier.



- Jiang, Q., Song, C., Nangreave, J., Liu, X., Lin, L., Qiu, D., Wang, Z. G., Zou, G., Liang, X., Yan, H., & Ding, B. (2012). DNA origami as a carrier for circumvention of drug resistance. *Journal of the American Chemical Society*, 134(32), 13396–13403. <https://doi.org/10.1021/ja304263n>
- Johnson, B. J., Russ Algar, W., Malanoski, A. P., Ancona, M. G., & Medintz, I. L. (2014). Understanding enzymatic acceleration at nanoparticle interfaces: Approaches and challenges. *Nano Today*, 9(1), 102–131. <https://doi.org/10.1016/j.nantod.2014.02.005>
- Justino, C. I. L., Duarte, A. C., & Rocha-Santos, T. A. P. (2017). Recent progress in biosensors for environmental monitoring: A review. *Sensors*, 17(12), 2918. <https://doi.org/10.3390/s17122918>
- Juul, S., Iacovelli, F., Falconi, M., Kragh, S. L., Christensen, B., Frøhlich, R., Franch, O., Kristoffersen, E. L., Stougaard, M., Leong, K. W., Ho, Y. P., Sørensen, E. S., Birkedal, V., Desideri, A., & Knudsen, B. R. (2013). Temperature-controlled encapsulation and release of an active enzyme in the cavity of a self-assembled DNA nanocage. *ACS Nano*, 7(11), 9724–9734. <https://doi.org/10.1021/nn4030543>
- Kahraman, E., Güngör, S., & Ozsoy, Y. (2017). Potential enhancement and targeting strategies of polymeric and lipid-based nanocarriers in dermal drug delivery. *Therapeutic Delivery*, 8, 967–985. <https://doi.org/10.4155/tde-2017-0075>
- Kalsoom, U., Ahsan, Z., Bhatti, H. N., Amin, F., Nadeem, R., Aftab, K., & Bilal, M. (2022). Iron oxide nanoparticles immobilized *Aspergillus flavus* manganese peroxidase with improved biocatalytic, kinetic, thermodynamic, and dye degradation potentialities. *Process Biochemistry*, 117, 117–133. <https://doi.org/10.1016/j.procbio.2022.04.002>
- Kamaly, N., Yameen, B., Wu, J., & Farokhzad, O. C. (2016). Degradable controlled-release polymers and polymeric nanoparticles: Mechanisms of controlling drug release. *Chemical Reviews*, 116(4), 2602–2663. <https://doi.org/10.1021/acs.chemrev.5b00346>
- Karajanagi, S. S., Vertegel, A. A., Kane, R. S., & Dordick, J. S. (2004). Structure and function of enzymes adsorbed onto single-walled carbon nanotubes. *Langmuir*, 20(26), 11594–11599. <https://doi.org/10.1021/la047994h>
- Karigar, C. S., & Rao, S. S. (2011). Role of microbial enzymes in the bioremediation of pollutants: A review. *Enzyme Research*, 2011, 805187. <https://doi.org/10.4061/2011/805187>
- Kaçar, C., Erden, P. E., Dalkiran, B., İnal, E. K., & Kiliç, E. (2020). Amperometric biogenic amine biosensors based on Prussian blue, indium tin oxide nanoparticles and diamine oxidase–or monoamine oxidase–modified electrodes. *Analytical and Bioanalytical Chemistry*, 412(8), 1933–1946. <https://doi.org/10.1007/s00216-020-02448-4>
- Kerry, J. P., O'grady, M. N., & Hogan, S. A. (2006). Past, current and potential utilisation of active and intelligent packaging systems for meat and muscle-based products: A review. *Meat Science*, 74(1), 113–130. <https://doi.org/10.1016/j.meatsci.2006.04.024>
- Khalid, K., Tan, X., Mohd Zaid, H. F., Tao, Y., Lye Chew, C., Chu, D. -T., Lam, M. K., Ho, Y. C., Lim, J. W., & Chin Wei, L. (2020). Advanced in developmental organic and inorganic nanomaterial: A review. *Bioengineered*, 11(1), 328–355. <https://doi.org/10.1080/21655979.2020.1736240>
- Khan, I., Saeed, K., & Khan, I. (2019). Nanoparticles: Properties, applications and toxicities. *Arabian Journal of Chemistry*, 12(7), 908–931. <https://doi.org/10.1016/j.arabj.2017.05.011>
- Kim, J., Grate, J. W., & Wang, P. (2008). Nanobiocatalysis and its potential applications. *Trends in Biotechnology*, 26(11), 639–646. <https://doi.org/10.1016/j.tibtech.2008.07.009>
- Kim, J. H., Jun, S.-A., Kwon, Y., Ha, S., Sang, B.-I., & Kim, J. (2015). Enhanced electrochemical sensitivity of enzyme precipitate coating (EPC)-based glucose oxidase biosensors with increased free CNT loadings. *Bioelectrochemistry*, 101, 114–119. <https://doi.org/10.1016/j.bioelechem.2014.08.017>
- Kim, K. H., Lee, O. K., & Lee, E. Y. (2018). Nano-immobilized biocatalysts for biodiesel production from renewable and sustainable resources. *Catalysts*, 8(2). <https://doi.org/10.3390/catal8020068>
- Kohman, R. E., Cha, S. S., Man, H.-Y., & Han, X. (2016). Light-triggered release of bioactive molecules from DNA nanostructures. *Nano Letters*, 16(4), 2781–2785. <https://doi.org/10.1021/acs.nanolett.6b00530>
- Koshovets, O. B., & Ganichev, N. A. (2017). Nanotechnology and the new technological revolution: Expectations and reality. *Studies on Russian Economic Development*, 28(4), 391–397. <https://doi.org/10.1134/S1075700717040104>
- Kuhad, R. C., & Singh, A. (2013). *Biotechnology for environmental management and resource recovery*. Springer.
- Kumar, S., Dilbaghi, N., Rani, R., & Bhanjana, G. (2012). Nanotechnology as emerging tool for enhancing solubility of poorly water-soluble drugs. *BioNanoScience*, 2, 227–250. <https://doi.org/10.1007/S12668-012-0060-7>
- Kurbanoglu, S., Mayorga-Martinez, C. C., Medina-Sánchez, M., Rivas, L., Ozkan, S. A., & Merkoçi, A. (2015). Antithyroid drug detection using an enzyme cascade blocking in a nanoparticle-based lab-on-a-chip system. *Biosensors and Bioelectronics*, 67, 670–676. <https://doi.org/10.1016/j.bios.2014.10.014>
- Kurbanoglu, S., Rivas, L., Ozkan, S. A., & Merkoçi, A. (2017). Electrochemically reduced graphene and iridium oxide nanoparticles for inhibition-based angiotensin-converting enzyme inhibitor detection. *Biosensors and Bioelectronics*, 88, 122–129. <https://doi.org/10.1016/j.bios.2016.07.109>
- Labhassetwar, V., Song, C., & Levy, R. J. (1997). Nanoparticle drug delivery system for restenosis. *Advanced Drug Delivery Reviews*, 24(1), 63–85. [https://doi.org/10.1016/S0169-409X\(96\)00483-8](https://doi.org/10.1016/S0169-409X(96)00483-8)
- Lang, Q., Han, L., Hou, C., Wang, F., & Liu, A. (2016). A sensitive acetylcholinesterase biosensor based on gold nanorods modified electrode for detection of organophosphate pesticide. *Talanta*, 156, 34–41. <https://doi.org/10.1016/j.talanta.2016.05.002>
- Lee, H., Song, C., Hong, Y. S., Kim, M. S., Cho, H. R., Kang, T., Shin, K., Choi, S. H., Hyeon, T., & Kim, D. H. (2017). Wearable/disposable sweat-based glucose monitoring device with multistage transdermal drug delivery module. *Science Advances*, 3(3), Article e1601314. <https://doi.org/10.1126/sciadv.1601314>
- Lee, S., Cheon, S., Kim, M. I., & Park, T. J. (2015). Organic-inorganic hybrid nanoflowers: Types, characteristics, and future prospects. *Journal of Nanobiotechnology*, 13, 54. <https://doi.org/10.1186/s12951-015-0118-0>
- Lellis, B., Fávoro-Polonio, C. Z., Pamphile, J. A., & Polonio, J. C. (2019). Effects of textile dyes on health and the environment and bioremediation potential of living organisms. *Biotechnology Research and Innovation*, 3(2), 275–290. <https://doi.org/10.1016/j.biori.2019.09.001>
- Lewis, T., & Stone, W. L. (2022). *Biochemistry, proteins enzymes*. Statpearls Publishing. <https://www.ncbi.nlm.nih.gov/books/NBK554481/>
- Li, G., Ma, P., He, Y., Zhang, Y., Luo, Y., Zhang, C., & Fan, H. (2018). Enzyme–Nanowire mesocrystal hybrid materials with an extremely high biocatalytic activity. *Nano Letters*, 18(9), 5919–5926. <https://doi.org/10.1021/acs.nanolett.8b02620>
- Li, H., Gao, M., Gao, Q., Wang, H., Han, B., Xia, K., & Zhou, C. (2019). Palladium nanoparticles uniformly and firmly supported on hierarchical flower-like TiO<sub>2</sub> nanospheres as a highly active and reusable catalyst for detoxification of Cr(VI)-contaminated water. *Applied Nanoscience*, 10, 1–11. <https://doi.org/10.1007/s13204-019-01164-5>
- Li, H., Guo, S., Li, C., Huang, H., Liu, Y., & Kang, Z. (2015). Tuning laccase catalytic activity with phosphate functionalized carbon dots by visible light. *ACS Applied Materials & Interfaces*, 7(18), 10004–10012. <https://doi.org/10.1021/acsami.5b02386>

- Li, H., Hou, J., Duan, L., Ji, C., Zhang, Y., & Chen, V. (2017). Graphene oxide-enzyme hybrid nanoflowers for efficient water soluble dye removal. *Journal of Hazardous Materials*, 338, 93–101. <https://doi.org/10.1016/j.jhazmat.2017.05.014>
- Li, J., Wu, Q., & Wu, J. (2016). Synthesis of nanoparticles via solvothermal and hydrothermal methods. In M. Aliofkharzraei (Ed.), *Handbook of nanoparticles* (pp. 295–328). Springer International Publishing. [https://doi.org/10.1007/978-3-319-15338-4\\_17](https://doi.org/10.1007/978-3-319-15338-4_17)
- Li, K., Wang, J., He, Y., Abdulrazaq, M. A., & Yan, Y. (2018). Carbon nanotube-lipase hybrid nanoflowers with enhanced enzyme activity and enantioselectivity. *Journal of Biotechnology*, 281, 87–98. <https://doi.org/10.1016/j.jbiotec.2018.06.344>
- Li, Y., Li, Y., Yu, X., & Sun, Y. (2019). Electrochemical determination of carbofuran in tomatoes by a concanavalin A (con A) polydopamine (PDA)-Reduced graphene oxide (RGO)-Gold nanoparticle (GNP) glassy carbon electrode (GCE) with immobilized acetylcholinesterase (AChE). *Analytical Letters*, 52(14), 2283–2299. <https://doi.org/10.1080/00032719.2019.1609490>
- Li, Z., Chen, Z., Zhu, Q., Song, J., Li, S., & Liu, X. (2020). Improved performance of immobilized laccase on Fe<sub>3</sub>O<sub>4</sub>@C-Cu<sup>2+</sup> nanoparticles and its application for biodegradation of dyes. *Journal of Hazardous Materials*, 399, 123088. <https://doi.org/10.1016/j.jhazmat.2020.123088>
- Li, Z., Ding, Y., Li, S., Jiang, Y., Liu, Z., & Ge, J. (2016). Highly active, stable and self-antimicrobial enzyme catalysts prepared by biomimetic mineralization of copper hydroxysulfate. *Nanoscale*, 8(40), 17440–17445. <https://doi.org/10.1039/C6NR06115J>
- Liu, W.-T. (2006). Nanoparticles and their biological and environmental applications. *Journal of Bioscience and Bioengineering*, 102(1), 1–7. <https://doi.org/10.1263/jbb.102.1>
- Lin, Z., Xiao, Y., Yin, Y., Hu, W., Liu, W., & Yang, H. (2014). Facile synthesis of enzyme-inorganic hybrid nanoflowers and its application as a colorimetric platform for visual detection of hydrogen peroxide and phenol. *ACS Applied Materials & Interfaces*, 6(13), 10775–10782. <https://doi.org/10.1021/am502757e>
- Liu, M., Fu, J., Qi, X., Wootten, S., Woodbury, N. W., Liu, Y., & Yan, H. (2016). A three-enzyme pathway with an optimised geometric arrangement to facilitate substrate transfer. *ChemBioChem*, 17(12), 1097–1101. <https://doi.org/10.1002/cbic.201600103>
- Liu, Q., Fei, A., Huan, J., Mao, H., & Wang, K. (2015). Effective amperometric biosensor for carbaryl detection based on covalent immobilization of acetylcholinesterase on multiwall carbon nanotubes/graphene oxide nanoribbons nanostructure. *Journal of Electroanalytical Chemistry*, 740, 8–13. <https://doi.org/10.1016/j.jelechem.2014.12.037>
- Liu, X., Fang, Y., Yang, X., Li, Y., & Wang, C. (2018a). Electrospun epoxy-based nanofibrous membrane containing biocompatible feather polypeptide for highly stable and active covalent immobilization of lipase. *Colloids and Surfaces B: Biointerfaces*, 166, 277–285. <https://doi.org/10.1016/j.colsurfb.2018.03.037>
- Liu, X., Fang, Y., Yang, X., Li, Y., & Wang, C. (2018b). Electrospun nanofibrous membranes containing epoxy groups and hydrophilic polyethylene oxide chain for highly active and stable covalent immobilization of lipase. *Chemical Engineering Journal*, 336, 456–464. <https://doi.org/10.1016/j.cej.2017.12.048>
- Liu, X., Yan, H., Liu, Y., & Chang, Y. (2011). Targeted cell–cell interactions by DNA nanoscaffold-templated multivalent bispecific aptamers. *Small*, 7(12), 1673–1682. <https://doi.org/10.1002/sml.201002292>
- Lv, Y., Hu, R., Zhu, G., Zhang, X., Mei, L., Liu, Q., Qiu, L., Wu, C., & Tan, W. (2015). Preparation and biomedical applications of programmable and multifunctional DNA nanoflowers. *Nature Protocols*, 10(10), 1508–1524. <https://doi.org/10.1038/nprot.2015.078>
- Ma, J., Ren, W., Zhao, J., & Yang, H. (2017). Growth of TiO<sub>2</sub> nanoflowers photoanode for dye-sensitized solar cells. *Journal of Alloys and Compounds*, 692, 1004–1009. <https://doi.org/10.1016/j.jallcom.2016.09.134>
- Ma, L., He, Y., Wang, Y., Wang, Y., Li, R., Huang, Z., Jiang, Y., & Gao, J. (2019). Nanocomposites of Pt nanoparticles anchored on UiO66-NH<sub>2</sub> as carriers to construct acetylcholinesterase biosensors for organophosphorus pesticide detection. *Electrochimica Acta*, 318, 525–533. <https://doi.org/10.1016/j.electacta.2019.06.110>
- Mahdieh, N., & Rabbani, B. (2013). An overview of mutation detection methods in genetic disorders. *Iranian Journal of Pediatrics*, 23(4), 375–388.
- Maleki, N., Khashanian, S., Maleki, E., & Nazari, M. (2017). A novel enzyme based biosensor for catechol detection in water samples using artificial neural network. *Biochemical Engineering Journal*, 128, 1–11. <https://doi.org/10.1016/j.bej.2017.09.005>
- Mansour, H. B., Houas, I., Montassar, F., Ghedira, K., Barillier, D., Mosrati, R., & Chekir-Ghedira, L. (2012). Alteration of in vitro and acute in vivo toxicity of textile dyeing wastewater after chemical and biological remediation. *Environmental Science and Pollution Research*, 19(7), 2634–2643. <https://doi.org/10.1007/s11356-012-0802-7>
- Martín-Timón, I., Sevillano-Collantes, C., Segura-Galindo, A., & Del Cañizo-Gómez, F. J. (2014). Type 2 diabetes and cardiovascular disease: Have all risk factors the same strength. *World Journal of Diabetes*, 5(4), 444–470. <https://doi.org/10.4239/wjd.v5.i4.444>
- Mauri, E., Chincarini, G. M. F., Rigamonti, R., Magagnin, L., Sacchetti, A., & Rossi, F. (2017). Modulation of electrostatic interactions to improve controlled drug delivery from nanogels. *Materials Science and Engineering: C*, 72, 308–315. <https://doi.org/10.1016/j.msec.2016.11.081>
- Medyantseva, E. P., Brusnitsyn, D. V., Varlamova, R. M., Beshevets, M. A., Budnikov, H. C., & Fattakhova, A. N. (2015). Capabilities of amperometric monoamine oxidase biosensors based on screen-printed graphite electrodes modified with multiwall carbon nanotubes in the determination of some antidepressants. *Journal of Analytical Chemistry*, 70(5), 535–539. <https://doi.org/10.1134/S106193481505010X>
- Medyantseva, E. P., Brusnitsyn, D. V., Varlamova, R. M., Maksimov, A. A., Fattakhova, A. N., Konovalova, O. A., & Budnikov, G. K. (2015). Effect of nanostructured materials as electrode surface modifiers on the analytical capacity of amperometric biosensors. *Russian Journal of Applied Chemistry*, 88(1), 40–49. <https://doi.org/10.1134/S1070427215010073>
- Medyantseva, E. P., Brusnitsyn, D. V., Varlamova, R. M., Maksimov, A. A., Konovalova, O. A., & Budnikov, H. C. (2017). Surface modification of electrodes by carbon nanotubes and gold and silver nanoparticles in monoaminoxidase biosensors for the determination of some antidepressants. *Journal of Analytical Chemistry*, 72(4), 362–370. <https://doi.org/10.1134/S1061934817040086>
- Mi, L., Yu, J., He, F., Jiang, L., Wu, Y., Yang, L., Han, X., Li, Y., Liu, A., Wei, W., Zhang, Y., Tian, Y., Liu, S., & Jiang, L. (2017). Boosting gas involved reactions at nanochannel reactor with joint gas–solid–liquid interfaces and controlled wettability. *Journal of the American Chemical Society*, 139(30), 10441–10446. <https://doi.org/10.1021/jacs.7b05249>
- Mo, Y., & Lim, L.-Y. (2004). Mechanistic study of the uptake of wheat germ agglutinin-conjugated PLGA nanoparticles by A549 cells. *Journal of Pharmaceutical Sciences*, 93(1), 20–28. <https://doi.org/10.1002/jps.10507>
- Mohamad, N. R., Marzuki, N. H. C., Buang, N. A., Huyop, F., & Wahab, R. A. (2015). An overview of technologies for immobilization of enzymes and surface analysis techniques for immobilized enzymes. *Biotechnology, Biotechnological Equipment*, 29(2), 205–220. <https://doi.org/10.1080/13102818.2015.1008192>
- Mousavi, S. R., & Rezaei, M. (2011). Nanotechnology in agriculture and food production. *Journal of Applied Environmental and Biological Sciences*, 1, 414–419.
- Muhammad, I. D. (2022). A comparative study of research and development related to nanotechnology in Egypt, Nigeria and South

- Africa. *Technology in Society*, 68, 101888. <https://doi.org/10.1016/j.techsoc.2022.101888>
- Müller, R. H., Petersen, R. D., Hommos, A., & Pardeike, J. (2007). Nanostructured lipid carriers (NLC) in cosmetic dermal products. *Advanced Drug Delivery Reviews*, 59(6), 522–530. <https://doi.org/10.1016/j.addr.2007.04.012>
- Nance, E. (2019). Careers in nanomedicine and drug delivery. *Advanced Drug Delivery Reviews*, 144, 180–189. <https://doi.org/10.1016/j.addr.2019.06.009>
- National Nanotechnology Initiative. (2016). *NNI supplement to the President's budget for the fiscal year 2017*.
- Navarro, J., de Marcos, S., & Galbán, J. (2020). Colorimetric-enzymatic determination of tyramine by generation of gold nanoparticles. *Microchimica Acta*, 187(3), 174. <https://doi.org/10.1007/s00604-020-4141-y>
- Ngo, T. A., Nakata, E., Saimura, M., & Morii, T. (2016). Spatially organized enzymes drive cofactor-coupled cascade reactions. *Journal of the American Chemical Society*, 138(9), 3012–3021. <https://doi.org/10.1021/jacs.5b10198>
- Nguyen, H. H., Lee, S. H., Lee, U. J., Fermin, C. D., & Kim, M. (2019). Immobilized enzymes in biosensor applications. *Materials*, 12(1):121. <https://doi.org/10.3390/ma12010121>
- Obzturk, B. (2001). *Immobilization of lipase from Candida rugosa on hydrophobic and hydrophilic supports* [MSc dissertation]. Izmir Institute of Technology.
- OECD. (2013). *Nanotechnology for green innovation*. <https://doi.org/10.1787/5k450q9j8p8q-en>
- Pang, Y. Y., Zhang, P., Gao, D. Y., Mo, F., & Yuan, J. S. (2013). The influence of the rib depth on the tensile properties of GFRP bar. *Applied Mechanics and Materials*, 438–439, 361–364.
- Parisi, C., Vigani, M., & Rodríguez-Cerezo, E. (2015). Agricultural nanotechnologies: What are the current possibilities. *Nano Today*, 10(2), 124–127. <https://doi.org/10.1016/j.nantod.2014.09.009>
- Park, J.-M., Kim, M., Park, H.-S., Jang, A., Min, J., & Kim, Y.-H. (2013). Immobilization of lysozyme-CLEA onto electrospun chitosan nanofiber for effective antibacterial applications. *International Journal of Biological Macromolecules*, 54, 37–43. <https://doi.org/10.1016/j.ijbiomac.2012.11.025>
- Patel, R. N. (2008). Chemo-enzymatic synthesis of pharmaceutical intermediates. *Expert Opinion on Drug Discovery*, 3(2), 187–245. <https://doi.org/10.1517/17460441.3.2.187>
- Patel, S. K. S., Otari, S. V., Chan Kang, Y., & Lee, J.-K. (2017). Protein-inorganic hybrid system for efficient his-tagged enzymes immobilization and its application in l-xylulose production. *RSC Advances*, 7(6), 3488–3494. <https://doi.org/10.1039/C6RA24404A>
- Paull, R., Wolfe, J., Hébert, P., & Sinkula, M. (2003). Investing in nanotechnology. *Nature Biotechnology*, 21(10), 1144–1147. <https://doi.org/10.1038/nbt1003-1144>
- Pham, D. T., Lee, T. H., Luong, D. H., Yao, F., Ghosh, A., Le, V. T., Kim, T. H., Li, B., Chang, J., & Lee, Y. H. (2015). Carbon nanotube-bridged graphene 3D building blocks for ultrafast compact supercapacitors. *ACS Nano*, 9(2), 2018–2027. <https://doi.org/10.1021/nn507079x>
- Prabaharan, M., & Mano, J. F. (2004). Chitosan-based particles as controlled drug delivery systems. *Drug Delivery*, 12(1), 41–57. <https://doi.org/10.1080/10717540590889781>
- 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. *Critical Reviews in Therapeutic Drug Carrier Systems*, 26(6), 523–580. <https://doi.org/10.1615/critrevtherdrugcarriersyst.v26.i6.10>
- Qiu, J. (2016). Nanotechnology development in China: Challenges and opportunities. *National Science Review*, 3(1), 148–152. <https://doi.org/10.1093/nsr/nww007>
- Qiu, X., Wang, S., Miao, S., Suo, H., Xu, H., & Hu, Y. (2021). Co-immobilization of laccase and ABTS onto amino-functionalized ionic liquid-modified magnetic chitosan nanoparticles for pollutants removal. *Journal of Hazardous Materials*, 401, 123353. <https://doi.org/10.1016/j.jhazmat.2020.123353>
- Qiu, X., Wang, Y., Xue, Y., Li, W., & Hu, Y. (2020). Laccase immobilized on magnetic nanoparticles modified by amino-functionalized ionic liquid via dialdehyde starch for phenolic compounds biodegradation. *Chemical Engineering Journal*, 391, 123564. <https://doi.org/10.1016/j.cej.2019.123564>
- Rathour, K., R. K., Bhatia, R. K., Rana, D. S., Bhatt, A. K., & Thakur, N. (2020). Fabrication of the most stable and reusable nanobiocatalyst for dye decolorization by immobilization of lignin peroxidase on graphene oxide functionalized MnFe<sub>2</sub>O<sub>4</sub> superparamagnetic nanoparticles. *Bioresource Technology*, 317, 124020. <https://doi.org/10.1016/j.biortech.2020.124020>
- Ranjan, B., Pillai, S., Permaul, K., & Singh, S. (2018). A novel strategy for the efficient removal of toxic cyanate by the combinatorial use of recombinant enzymes immobilized on aminosilane modified magnetic nanoparticles. *Bioresource Technology*, 253, 105–111. <https://doi.org/10.1016/j.biortech.2017.12.087>
- Ranjan, B., Pillai, S., Permaul, K., & Singh, S. (2019). Simultaneous removal of heavy metals and cyanate in a wastewater sample using immobilized cyanate hydratase on magnetic-multiwall carbon nanotubes. *Journal of Hazardous Materials*, 363, 73–80. <https://doi.org/10.1016/j.jhazmat.2018.07.116>
- Reis, R. L., & San Román, J. (2004). *Biodegradable systems in tissue engineering and regenerative medicine*. CRC press.
- Revenga-Parra, M., Villa-Manso, A. M., Briones, M., Mateo-Martí, E., Martínez-Periñán, E., Lorenzo, E., & Pariente, F. (2020). Bioelectrocatalytic platforms based on chemically modified nanodiamonds by diazonium salt chemistry. *Electrochimica Acta*, 357, 136876. <https://doi.org/10.1016/j.electacta.2020.136876>
- Rho, J. Y., Cox, H., Mansfield, E., Ellacott, S. H., Peltier, R., Brendel, J. C., Hartlieb, M., Waigh, T. A., & Perrier, S. (2019). Dual self-assembly of supramolecular peptide nanotubes to provide stabilisation in water. *Nature Communications*, 10(1), 4708. <https://doi.org/10.1038/s41467-019-12586-8>
- Roco, M. C. (2006). *Nanotechnology's future*. Scientific American, A Division of Springer Nature America, Inc.
- Rodrigues, R. C., Ortiz, C., Berenguer-Murcia, Á., Torres, R., & Fernández-Lafuente, R. (2013). Modifying enzyme activity and selectivity by immobilization. *Chemical Society Reviews*, 42(15), 6290–6307. <https://doi.org/10.1039/C2CS35231A>
- Rong, J., Zhang, T., Qiu, F., & Zhu, Y. (2017). Preparation of efficient, stable, and reusable laccase-Cu<sub>3</sub>(PO<sub>4</sub>)<sub>2</sub> hybrid microspheres based on copper foil for decoloration of Congo Red. *ACS Sustainable Chemistry & Engineering*, 5(5), 4468–4477. <https://doi.org/10.1021/acssuschemeng.7b00820>
- Sadegh, H., Ghoshekandi, R. S., Masjedi, A., Mahmoodi, Z., & Kazemi, M. (2016). A review on carbon nanotubes adsorbents for the removal of pollutants from aqueous solutions. *Int J Nano Dimension*, 7, 109.
- Salvador-Morales, C., Zhang, L., Langer, R., & Farokhzad, O. C. (2009). Immunocompatibility properties of lipid-polymer hybrid nanoparticles with heterogeneous surface functional groups. *Biomaterials*, 30(12), 2231–2240. <https://doi.org/10.1016/j.biomaterials.2009.01.005>
- Samphao, A., Kunpatee, K., Prayoonpokarach, S., Wittayakun, J., Švorc, L., Stankovic, D. M., Zagar, K., Ceh, M., & Kalcher, K. (2015). An ethanol biosensor based on simple immobilization of alcohol dehydrogenase on Fe<sub>3</sub>O<sub>4</sub>@Au nanoparticles. *Electroanalysis*, 27(12), 2829–2837. <https://doi.org/10.1002/elan.201500315>
- Saptarshi, S. R., Duschl, A., & Lopata, A. L. (2013). Interaction of nanoparticles with proteins: Relation to bio-reactivity of the nanoparticle. *Journal of Nanobiotechnology*, 11(1), 26. <https://doi.org/10.1186/1477-3155-11-26>

- Sarkar, S., Banerjee, A., Chakraborty, N., Soren, K., Chakraborty, P., & Bandopadhyay, R. (2020). Structural-functional analyses of textile dye degrading azoreductase, laccase and peroxidase: A comparative in silico study. *Electronic Journal of Biotechnology*, 43, 48–54. <https://doi.org/10.1016/j.ejbt.2019.12.004>
- Scarazzati, S., & Wang, L. (2019). The effect of collaborations on scientific research output: The case of nanoscience in Chinese regions. *Scientometrics*, 121(2), 839–868. <https://doi.org/10.1007/s11192-019-03220-x>
- Schwizer, F., Okamoto, Y., Heinisch, T., Gu, Y., Pellizzoni, M. M., Lebrun, V., Reuter, R., Köhler, V., Lewis, J. C., & Ward, T. R. (2018). Artificial metalloenzymes: Reaction scope and optimization strategies. *Chemical Reviews*, 118(1), 142–231. <https://doi.org/10.1021/acs.chemrev.7b00014>
- Shandilya, R., Bhargava, A., Bunkar, N., Tiwari, R., Goryacheva, I. Y., & Mishra, P. K. (2019). Nanobiosensors: Point-of-care approaches for cancer diagnostics. *Biosensors and Bioelectronics*, 130, 147–165. <https://doi.org/10.1016/j.bios.2019.01.034>
- Sheldon, R. A. (2007). Cross-linked enzyme aggregates (CLEARs): Stable and recyclable biocatalysts. *Biochemical Society Transactions*, 35(6), 1583–1587. <https://doi.org/10.1042/BST0351583>
- Shen, Q., Yang, R., Hua, X., Ye, F., Zhang, W., & Zhao, W. (2011). Gelatin-templated biomimetic calcification for  $\beta$ -galactosidase immobilization. *Process Biochemistry*, 46(8), 1565–1571. <https://doi.org/10.1016/j.procbio.2011.04.010>
- Shende, P., Kasture, P., & Gaud, R. S. (2018). Nanoflowers: The future trend of nanotechnology for multi-applications. *Artificial Cells, Nanomedicine, and Biotechnology*, 46(Suppl. 1), 413–422. <https://doi.org/10.1080/21691401.2018.1428812>
- Shkodra, B., Demelash Abera, B., Cantarella, G., Douaki, A., Avancini, E., Petti, L., & Lugli, P. (2020). Flexible and printed electrochemical immunosensor coated with oxygen plasma treated SWCNTs for histamine detection. *Biosensors*, 10(4):35. <https://doi.org/10.3390/bios10040035>
- Siegrist, M., Stampfli, N., Kastenholz, H., & Keller, C. (2008). Perceived risks and perceived benefits of different nanotechnology foods and nanotechnology food packaging. *Appetite*, 51(2), 283–290. <https://doi.org/10.1016/j.appet.2008.02.020>
- Silva-Torres, O., Bojorquez-Vazquez, L., Simakov, A., & Vazquez-Duhalt, R. (2019). Enhanced laccase activity of biocatalytic hybrid copper hydroxide nanocages. *Enzyme and Microbial Technology*, 128, 59–66. <https://doi.org/10.1016/j.enzmictec.2019.05.008>
- Singer, P. A., Daar, A. S., & Salamanca-Buentello, F. (2005). *Harnessing nanotechnology to improve global equity* (Vol. 21). Issues in Science and Technology.
- Somturk, B., Yilmaz, I., Altinkaynak, C., Karatepe, A., Özdemir, N., & Ocsoy, I. (2016). Synthesis of urease hybrid nanoflowers and their enhanced catalytic properties. *Enzyme and Microbial Technology*, 86, 134–142. <https://doi.org/10.1016/j.enzmictec.2015.09.005>
- Srinivasan, P., Selvankumar, T., Paray, B. A., Rehman, M. U., Kamala-Kannan, S., Govarthanan, M., Kim, W., & Selvam, K. (2020). Chlorpyrifos degradation efficiency of *Bacillus* sp. laccase immobilized on iron magnetic nanoparticles. *3 Biotech*, 10(8), 366. <https://doi.org/10.1007/s13205-020-02363-6>
- Stevens, P., Sekido, M., & Lee, R. (2004). Synthesis and evaluation of a hematoporphyrin derivative in a folate receptor-targeted solid-lipid nanoparticle formulation. *Anticancer Research*, 24(1), 161–165.
- Stevens, P. J., Sekido, M., & Lee, R. J. (2004). A folate receptor-targeted lipid nanoparticle formulation for a lipophilic paclitaxel prodrug. *Pharmaceutical Research*, 21(12), 2153–2157. <https://doi.org/10.1007/s11095-004-7667-5>
- Stone, V., Hankin, S., Aitken, R., Aschberger, K., Baun, A., Christensen, F., & Tran, L. (2009). *ENRHES—Engineered nanoparticles: Review of health and environmental safety*. Technical University of Denmark (DUT).
- Talbert, J. N., & Goddard, J. M. (2012). Enzymes on material surfaces. *Colloids and Surfaces*, 93, 8–19. <https://doi.org/10.1016/j.colsurfb.2012.01.003>
- Tang, H., Chen, J., Yao, S., Nie, L., Deng, G., & Kuang, Y. (2004). Amperometric glucose biosensor based on adsorption of glucose oxidase at platinum nanoparticle-modified carbon nanotube electrode. *Analytical Biochemistry*, 331(1), 89–97. <https://doi.org/10.1016/j.ab.2004.05.005>
- Tao, Y., Ju, E., Ren, J., & Qu, X. (2014). Polypyrrole nanoparticles as promising enzyme mimics for sensitive hydrogen peroxide detection. *Chemical Communications*, 50(23), 3030–3032. <https://doi.org/10.1039/c4cc00328d>
- Thevenot, J., Troutier, A.-L., David, L., Delair, T., & Ladavière, C. (2007). Steric stabilization of lipid/polymer particle assemblies by poly(ethylene glycol)-lipids. *Biomacromolecules*, 8(11), 3651–3660. <https://doi.org/10.1021/bm700753q>
- Turner, A. P. F. (2000). Biochemistry: Biosensors-sense and sensitivity. *Science*, 290(5495), 1315–1317. <https://doi.org/10.1126/science.290.5495.1315>
- Turner, N. J. (2009). Directed evolution drives the next generation of biocatalysts. *Nature Chemical Biology*, 5(8), 567–573. <https://doi.org/10.1038/nchembio.203>
- Ulu, A., Birhanli, E., Boran, F., Köytepe, S., Yesilada, O., & Ateş, B. (2020). Laccase-conjugated thiolated chitosan-Fe<sub>3</sub>O<sub>4</sub> hybrid composite for biocatalytic degradation of organic dyes. *International Journal of Biological Macromolecules*, 150, 871–884. <https://doi.org/10.1016/j.ijbiomac.2020.02.006>
- Updike, S. J., & Hicks, G. P. (1967). The enzyme electrode. *Nature*, 214(5092), 986–988. <https://doi.org/10.1038/214986a0>
- Verma, M. L., Rao, N. M., Tsuzuki, T., Barrow, C. J., & Puri, M. (2019). Suitability of recombinant lipase immobilised on functionalised magnetic nanoparticles for fish oil hydrolysis. *Catalysts*, 9(5), 420. <https://doi.org/10.3390/catal9050420>
- Vidali, M. (2001). Bioremediation: An overview. *Pure and Applied Chemistry*, 73(7), 1163–1172. <https://doi.org/10.1351/pac200173071163>
- Vigneshvar, S., & Senthilkumaran, B. (2018). Current technological trends in biosensors, nanoparticle devices and biolabels: Hi-tech network sensing applications. *Medical Devices and Sensors*, 1(2):e10011. <https://doi.org/10.1002/mds3.10011>
- Vranish, J. N., Ancona, M. G., Oh, E., Susumu, K., & Medintz, I. L. (2017). Enhancing coupled enzymatic activity by conjugating one enzyme to a nanoparticle. *Nanoscale*, 9(16), 5172–5187. <https://doi.org/10.1039/C7NR00200A>
- Vukojević, V., Djurdjić, S., Ognjanović, M., Fabián, M., Samphao, A., Kalcher, K., & Stanković, D. M. (2018). Enzymatic glucose biosensor based on manganese dioxide nanoparticles decorated on graphene nanoribbons. *Journal of Electroanalytical Chemistry*, 823, 610–616. <https://doi.org/10.1016/j.jelechem.2018.07.013>
- Wang, C., Zhang, Q., Wang, X., Chang, H., Zhang, S., Tang, Y., Xu, J., Qi, R., & Cheng, Y. (2017). Dynamic modulation of enzyme activity by near-infrared light. *Angewandte Chemie International Edition*, 56(24), 6767–6772. <https://doi.org/10.1002/anie.201700968>
- Wang, H., Zhao, Z., Liu, Y., Shao, C., Bian, F., & Zhao, Y. (2018). Biomimetic enzyme cascade reaction system in microfluidic electrospray microcapsules. *Science Advances*, 4(6), 2816. <https://doi.org/10.1126/sciadv.aat2816>
- Wang, J., Zhu, H., Xu, Y., Yang, W., Liu, A., Shan, F., Cao, M., & Liu, J. (2015). Graphene nanodots encaged 3-D gold substrate as enzyme loading platform for the fabrication of high performance biosensors. *Sensors and Actuators B: Chemical*, 220, 1186–1195. <https://doi.org/10.1016/j.snb.2015.06.044>
- Wang, L., Gong, C., Yuan, X., & Wei, G. (2019). Controlling the self-assembly of biomolecules into functional nanomaterials through internal interactions and external stimulations: A review. *Nanomaterials*, 9(2), 285. <https://doi.org/10.3390/nano9020285>

- Wang, L., Wei, L., Chen, Y., & Jiang, R. (2010). Specific and reversible immobilization of NADH oxidase on functionalized carbon nanotubes. *Journal of Biotechnology*, 150(1), 57–63. <https://doi.org/10.1016/j.jbiotec.2010.07.005>
- Wang, L.-B., Wang, Y.-C., He, R., Zhuang, A., Wang, X., Zeng, J., & Hou, J. G. (2013). A new nanobiocatalytic system based on allosteric effect with dramatically enhanced enzymatic performance. *Journal of the American Chemical Society*, 135(4), 1272–1275. <https://doi.org/10.1021/ja3120136>
- Wang, M., Abad, D., Kickhoefer, V. A., Rome, L. H., & Mahendra, S. (2015). Vault nanoparticles packaged with enzymes as an efficient pollutant biodegradation technology. *ACS Nano*, 9(11), 10931–10940. <https://doi.org/10.1021/acs.nano.5b04073>
- Wang, M., Huang, S.-W., Xu, D., Bao, W.-J., & Xia, X.-H. (2015). Ultrahigh enzyme activity assembled in layered double hydroxides via Mg<sup>2+</sup>-allosteric effector. *Analytical Chemistry*, 87(11), 5831–5836. <https://doi.org/10.1021/acs.analchem.5b01427>
- Wang, T.-F., Lo, H.-F., Chi, M.-C., Lai, K.-L., Lin, M.-G., & Lin, L.-L. (2019). Affinity immobilization of a bacterial prolidase onto metal-ion-chelated magnetic nanoparticles for the hydrolysis of organophosphorus compounds. *International Journal of Molecular Sciences*, 20(15), 3625. <https://doi.org/10.3390/ijms20153625>
- Wang, X., Shi, J., Li, Z., Zhang, S., Wu, H., Jiang, Z., Yang, C., & Tian, C. (2014). Facile one-pot preparation of chitosan/calcium pyrophosphate hybrid microflowers. *ACS Applied Materials & Interfaces*, 6, 6–32. <https://doi.org/10.1021/am503787h>
- Wang, Y., Chen, D., Wang, G., Zhao, C., Ma, Y., & Yang, W. (2018). Immobilization of cellulase on styrene/maleic anhydride copolymer nanoparticles with improved stability against pH changes. *Chemical Engineering Journal*, 336, 152–159. <https://doi.org/10.1016/j.cej.2017.11.030>
- Wang, Y., Wang, X., Lu, W., Yuan, Q., Zheng, Y., & Yao, B. (2019). A thin film polyethylene terephthalate (PET) electrochemical sensor for detection of glucose in sweat. *Talanta*, 198, 86–92. <https://doi.org/10.1016/j.talanta.2019.01.104>
- Wei, M., & Wang, J. (2015). A novel acetylcholinesterase biosensor based on ionic liquids-AuNPs-porous carbon composite matrix for detection of organophosphate pesticides. *Sensors and Actuators B: Chemical*, 211, 290–296. <https://doi.org/10.1016/j.snb.2015.01.112>
- Wong, J. K. H., Tan, H. K., Lau, S. Y., Yap, P.-S., & Danquah, M. K. (2019). Potential and challenges of enzyme incorporated nanotechnology in dye wastewater treatment: A review. *Journal of Environmental Chemical Engineering*, 7(4), 103261. <https://doi.org/10.1016/j.jece.2019.103261>
- Wood, S., Jones, R., & Geldart, A. (2003). *The social and economic challenges of nanotechnology*. Economic and Social Research Council.
- Wu, D., Feng, Q., Xu, T., Wei, A., & Fong, H. (2018). Electrospun blend nanofiber membrane consisting of polyurethane, amidoxime polyacrylonitrile, and  $\beta$ -cyclodextrin as high-performance carrier/support for efficient and reusable immobilization of laccase. *Chemical Engineering Journal*, 331, 517–526. <https://doi.org/10.1016/j.cej.2017.08.129>
- Wu, F., Su, L., Yu, P., & Mao, L. (2017). Role of organic solvents in immobilizing fungus laccase on single-walled carbon nanotubes for improved current response in direct bioelectrocatalysis. *Journal of the American Chemical Society*, 139(4), 1565–1574. <https://doi.org/10.1021/jacs.6b11469>
- Xia, T.-T., Liu, C.-Z., Hu, J.-H., & Guo, C. (2016). Improved performance of immobilized laccase on amine-functionalized magnetic Fe<sub>3</sub>O<sub>4</sub> nanoparticles modified with polyethylenimine. *Chemical Engineering Journal*, 295, 201–206. <https://doi.org/10.1016/j.cej.2016.03.044>
- Xiang, X., Ding, S., Suo, H., Xu, C., Gao, Z., & Hu, Y. (2018). Fabrication of chitosan-mesoporous silica SBA-15 nanocomposites via functional ionic liquid as the bridging agent for PPL immobilization. *Carbohydrate Polymers*, 182, 245–253. <https://doi.org/10.1016/j.carbpol.2017.11.031>
- Xiang, X., Suo, H., Xu, C., & Hu, Y. (2018). Covalent immobilization of lipase onto chitosan-mesoporous silica hybrid nanomaterials by carboxyl functionalized ionic liquids as the coupling agent. *Colloids and Surfaces B: Biointerfaces*, 165, 262–269. <https://doi.org/10.1016/j.colsurfb.2018.02.033>
- Xiao, Y., Shi, K., Qu, Y., Chu, B., & Qian, Z. (2018). Engineering nanoparticles for targeted delivery of nucleic acid therapeutics in tumor. *Molecular Therapy: Methods & Clinical Development*, 12, 1–18. <https://doi.org/10.1016/j.omtm.2018.09.002>
- Xiong, R., Zhang, W., Zhang, Y., Zhang, Y., Chen, Y., He, Y., & Fan, H. (2019). Remote and real time control of an FVIO-enzyme hybrid nanocatalyst using magnetic stimulation. *Nanoscale*, 11(39), 18081–18089. <https://doi.org/10.1039/C9NR04289J>
- Xu, C., Yu, Y., Sun, Y., Kong, L., Yang, C., Hu, M., Yang, T., Zhang, J., Hu, Q., & Zhang, Z. (2019). Transformable nanoparticle-enabled synergistic elicitation and promotion of immunogenic cell death for triple-negative breast cancer immunotherapy. *Advanced Functional Materials*, 29(45), 1905213. <https://doi.org/10.1002/adfm.201905213>
- Xu, Y., Hu, X., Kundu, S., Nag, A., Afsarimanesh, N., Sapra, S., Mukhopadhyay, S. C., & Han, T. (2019). Silicon-based sensors for biomedical applications: A review. *Sensors*, 19, 2908. <https://doi.org/10.3390/s19132908>
- Yang, Y., & Hobbs, J. E. (2020). Food values and heterogeneous consumer responses to nanotechnology. *Canadian Journal of Agricultural Economics*, 68(3), 289–313. <https://doi.org/10.1111/cjag.12225>
- Yang, Y., Wang, S., Zhou, Z., Zhang, R., Shen, H., Song, J., Su, P., & Yang, Y. (2018). Enhanced reusability and activity: DNA directed immobilization of enzyme on polydopamine modified magnetic nanoparticles. *Biochemical Engineering Journal*, 137, 108–115. <https://doi.org/10.1016/j.bej.2018.05.019>
- Yaqoob, A. A., Ahmad, H., Parveen, T., Ahmad, A., Oves, M., Ismail, I., Qari, H. A., Umar, K., & Mohamad Ibrahim, M. N. (2020). Recent advances in metal decorated nanomaterials and their various biological applications: A review. *Frontiers in Chemistry*, 8(341), 341. <https://doi.org/10.3389/fchem.2020.00341>
- Ye, R., Zhu, C., Song, Y., Lu, Q., Ge, X., Yang, X., Zhu, M. J., Du, D., Li, H., & Lin, Y. (2016). Bioinspired synthesis of all-in-one organic-inorganic hybrid nanoflowers combined with a handheld pH meter for on-site detection of food pathogen. *Small: Nano Micro*, 12(23), 3094–3100. <https://doi.org/10.1002/sml.201600273>
- Yin, Y., Xiao, Y., Lin, G., Xiao, Q., Lin, Z., & Cai, Z. (2015). An enzyme-inorganic hybrid nanoflower based immobilized enzyme reactor with enhanced enzymatic activity. *Journal of Materials Chemistry B*, 3(11), 2295–2300. <https://doi.org/10.1039/C4TB01697A>
- Yoo, E.-H., & Lee, S.-Y. (2010). Glucose biosensors: An overview of use in clinical practice. *Sensors*, 10(5), 4558–4576. <https://doi.org/10.3390/s100504558>
- Yu, L., Banerjee, I. A., Gao, X., Nuraje, N., & Matsui, H. (2005). Fabrication and application of enzyme-incorporated peptide nanotubes. *Bioconjugate Chemistry*, 16(6), 1484–1487. <https://doi.org/10.1021/bc050199a>
- Yu, X., Gou, X., Wu, P., Han, L., Tian, D., Du, F., Chen, Z., Liu, F., Deng, G., Chen, A. T., Ma, C., Liu, J., Hashmi, S. M., Guo, X., Wang, X., Zhao, H., Liu, X., Zhu, X., Sheth, K., ... Zhou, J. (2018). Activatable protein nanoparticles for targeted delivery of therapeutic peptides. *Advanced Materials*, 30(7), 1705383. <https://doi.org/10.1002/adma.201705383>
- Zhai, R., Zhang, B., Wan, Y., Li, C., Wang, J., & Liu, J. (2013). Chitosan-halloysite hybrid-nanotubes: Horseradish peroxidase immobilization and applications in phenol removal. *Chemical Engineering Journal*, 214, 304–309. <https://doi.org/10.1016/j.cej.2012.10.073>
- Zhang, B., Li, P., Zhang, H., Wang, H., Li, X., Tian, L., Ali, N., Ali, Z., & Zhang, Q. (2016). Preparation of lipase/Zn<sub>3</sub>(PO<sub>4</sub>)<sub>2</sub> hybrid nanoflower

- and its catalytic performance as an immobilized enzyme. *Chemical Engineering Journal*, 291, 287–297. <https://doi.org/10.1016/j.cej.2016.01.104>
- Zhang, B., Misak, H., Dhanasekaran, P. S., Kalla, D., & Asmatulu, R. (2011). *Environmental impacts of nanotechnology and its products*. American Society for Engineering Education (ASEE).
- Zhang, D.-H., Yuwen, L.-X., & Peng, L.-J. (2013). Parameters affecting the performance of immobilized enzyme. *Journal of Chemistry*, 2013, 1–7. <https://doi.org/10.1155/2013/946248>
- Zhang, J., Hu, H., & Yang, L. (2021). Ultra-highly sensitive and stable acetylcholinesterase biosensor based on TiO<sub>2</sub>-NRs and rGO. *Microchemical Journal*, 168, 106435. <https://doi.org/10.1016/j.microc.2021.106435>
- Zhang, K., Yang, W., Liu, Y., Zhang, K., Chen, Y., & Yin, X. (2020). Laccase immobilized on chitosan-coated Fe<sub>3</sub>O<sub>4</sub> nanoparticles as reusable biocatalyst for degradation of chlorophenol. *Journal of Molecular Structure*, 1220, 128769. <https://doi.org/10.1016/j.molstruc.2020.128769>
- Zhang, L., Chan, J. M., Gu, F. X., Rhee, J. -W., Wang, A. Z., Radovic-Moreno, A. F., Alexis, F., Langer, R., & Farokhzad, O. C. (2008). Self-assembled lipid-polymer hybrid nanoparticles: A robust drug delivery platform. *ACS Nano*, 2(8), 1696–1702. <https://doi.org/10.1021/nn800275r>
- Zhang, S., Wu, Z., Chen, G., & Wang, Z. (2018). An improved method to encapsulate laccase from *Trametes versicolor* with enhanced stability and catalytic activity. *Catalysts*, 8(7), 286. <https://doi.org/10.3390/catal8070286>
- Zhang, Y., Lim, C.-K., Dai, Z., Yu, G., Haus, J. W., Zhang, H., & Prasad, P. N. (2019). Photonics and optoelectronics using nano-structured hybrid perovskite media and their optical cavities. *Physics Reports*, 795, 1–51. <https://doi.org/10.1016/j.physrep.2019.01.005>
- Zhao, X., Hao, F., Lu, D., Liu, W., Zhou, Q., & Jiang, G. (2015). Influence of the surface functional group density on the carbon-nanotube-induced  $\alpha$ -chymotrypsin structure and activity alterations. *ACS Applied Materials & Interfaces*, 7(33), 18880–18890. <https://doi.org/10.1021/acsami.5b05895>
- Zhao, Z., Fu, J., Dhakal, S., Johnson-Buck, A., Liu, M., Zhang, T., Woodbury, N. W., Liu, Y., Walter, N. G., & Yan, H. (2016). Nanocaged enzymes with enhanced catalytic activity and increased stability against protease digestion. *Nature Communications*, 7(1), 10619. <https://doi.org/10.1038/ncomms10619>
- Zheng, J.-N., He, L.-L., Chen, C., Wang, A.-J., Ma, K.-F., & Feng, J.-J. (2014). One-pot synthesis of Pt<sub>3</sub>Co nanoflowers with enhanced oxygen reduction and methanol oxidation. *Journal of Power Sources*, 268, 744–751. <https://doi.org/10.1016/j.jpowsour.2014.06.109>
- Zheng, L., Sun, Y., Wang, J., Huang, H., Geng, X., Tong, Y., & Wang, Z. (2018). Preparation of a flower-like immobilized D-Psicose 3-Epimerase with enhanced catalytic performance. *Catalysts*, 8(10), 468. <https://doi.org/10.3390/catal8100468>
- Zheng, Y., Liu, Z., Jing, Y., Li, J., & Zhan, H. (2015). An acetylcholinesterase biosensor based on ionic liquid functionalized graphene-gelatin-modified electrode for sensitive detection of pesticides. *Sensors and Actuators B: Chemical*, 210, 389–397. <https://doi.org/10.1016/j.snb.2015.01.003>
- Zhu, J., Zhang, Y., Lu, D., Zare, R. N., Ge, J., & Liu, Z. (2013). Temperature-responsive enzyme-polymer nanoconjugates with enhanced catalytic activities in organic media. *Chemical Communications*, 49(54), 6090–6092. <https://doi.org/10.1039/C3CC42493F>
- Zhu, Q., Song, J., Liu, Z., Wu, K., Li, X., Chen, Z., & Pang, H. (2022). Photothermal catalytic degradation of textile dyes by laccase immobilized on Fe<sub>3</sub>O<sub>4</sub>@SiO<sub>2</sub> nanoparticles. *Journal of Colloid and Interface Science*, 623, 992–1001. <https://doi.org/10.1016/j.jcis.2022.05.083>
- Zhuang, L., Song, Q., Lei, C., & Shuping, Z. (2017). Detection of carbofuran pesticide in seawater by using an enzyme biosensor. *Journal of Coastal Research*, 80(Suppl. 1), 1–5. <https://doi.org/10.2112/SI80-001.1>

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